Journal of Food Protection

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ABSTRACTS

This is a collection of the abstracts from IAFP 2019, held in Louisville, Kentucky.
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Ivan Parkin Lecture Abstract
John H. Silliker Lecture Abstract
Abstracts
Symposium
Roundtable
Poster
Author and Presenter Index
Developing Scientist Competitors
Undergraduate Student Competitors

13
23
33
39
329
363
Don't Wash Your Chicken!

donwashyourchicken.org

Videos, animation, recipes, and printable fotonovelas reinforce the importance of not washing raw poultry.

Don't Be Gross
don'tbegross.org

These short, shareable animations convey the importance of hand washing and other health issues.

Produce Safety Matters

producesafetymatters.org

Designed for extension training and outreach, growers, packers, and retailers learn tips to prevent contamination from farmer’s fork in these crisp animations.

The Transformational Design Model is an educational design model based on five key ways to change people: their knowledge (what they know), skill (what they can do), behavior (how they act), emotion, (how they feel) and physiology (how they are). When educators design the specific ways in which they want a learner to change, the next step is to design the activities that will lead to that change.

Activity design is more complex, as there are hundreds of ways to learn, experience, develop and grow; such as, receiving information, failing, observing, planning, communicating, thinking, and solving problems. This range of activities includes moving a learner from activities that provide simple exposition, through different types of activities to more inquiry-based learning. This range of activities is helpful in guiding designers through a learning experience. The range provided doesn’t offer a continuum of good through bad; rather, it is designed to help developers think through the needs of the player. Sometimes simple exposure to knowledge is useful, when other kinds of learning and change demands reflection, creative activity and building, or learner-centered project development.

Additional resources developed by the Learning Games Lab are available at learninggameslab.org.

Public health plays an important role in food safety, in the changing landscape of foods, tastes and processes, pathogens can find a niche, persist and emerge. Public health surveillance and investigations can identify problems and help target solutions to prevent foodborne illnesses. The tools public health uses for surveillance have also been evolving. Better microbiological methods improve definition of individual strains, separating “signal” from “noise.” These improvements mean finding more outbreaks, helping to drive immediate control efforts and longer term prevention policies.

The transition to whole genome sequencing is now underway in our public health surveillance network PulseNet. This new tool already provides better strain resolution and new ways of looking at food safety problems. Whole genome sequencing differs in several important ways from the standard PFGE subtyping PulseNet used for the past 23 years. Resolving differences down to single nucleotides provides a scale of similarity that can be as precise as needed. From sequence, other strain-characteristics can be predicted including serotype, antibiotic-resistance profile, and virulence. This is changing the workflow in our public health labs, so more characteristics are known when a cluster of related infections is detected. Unlike the previous closed PFGE database of PulseNet, the sequence database is open access. As public health scientists and partners at FDA and USDA will add ~60,000 bacterial sequences a year, the database will be a rich resource for future research.

We anticipate that as sequencing is applied to surveillance, investigation of the many smaller outbreaks detected should find more specific control points and guide prevention, including harborage in processing, reservoirs in production and new sources from other countries. As we find even smaller outbreaks, the line blurs between traditional outbreaks and the background of individual “sporadic” cases.

Beyond the traditional role of helping public health find and stop outbreaks, this new surveillance system can do much more to prevent illnesses.

• We can more easily track “clades of concern,” investigating them even in the absence of an outbreak. For example, we can track strains with greater confidence that caused major repeated outbreaks in the past, are still present at lower incidence now, and could yet cause future outbreaks. We can see other strains that emerge, increase over time, and may be investigated and controlled before they cause a large traditional outbreak, preventing more foodborne infections.

• Other countries are rapidly adopting similar surveillance strategies. Canada, the European Union, and Australia are in the vanguard with the U.S., and many more are starting soon. By comparing sequences across borders, all can better understand the spread of pathogens through travel and trade.

• As tools for interpreting sequences become more accessible, many in food science will find tracking specific strains useful to examine the ecology of bacterial pathogens in food production and processing. By comparing them with strains in the public database, internal control efforts can be focused.

• It will be possible to use more genetic markers for surveillance, persistence, or adaptation to specific reservoirs and hosts. The potential to understand better the biology of these bacteria is growing rapidly.

The next transition, building on sequencing experience, will someday bypass traditional culture and go directly to metagenic analyses to construct genomes directly from specimens. We stand at a threshold in microbial food safety, with the opportunity to accelerate research, investigation and prevention. I hope to learn and reframe much more, together with all of you.
Symposium Abstracts

S1  Tracking FSMA Quantitative and Qualitative Impacts on the Food Industry Under Full FDA Enforcement – Stats, Trends, Challenges and Lessons Learned

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KARLEIGH BACON: The Kroft Heinz Company, Chicago, IL, USA
GREGORY PRITCHARD: Nestlé USA, Glendale, CA, USA
ALLEN SAYLER: EAS Consulting Group, Alexandria, VA, USA
PURNENDU VASAVADA: University of Wisconsin-River Falls, River Falls, WI, USA
ELIZABETH FAWELL: Hogan Lovells, Washington, DC, USA

The FDA Food Safety Modernization Act (FSMA) was designed to significantly expand the enforcement tools available to the U.S. Food & Drug Administration. The primary intent was to shift both FDA and the food manufacturing and processing focus from reactionary after a contamination event occurred to preventive by emphasizing known approaches, systems and technology that together will likely reduce the incidents of food contamination during manufacturing, packaging and distribution. The two most impactful of the FSMA regulations for most food manufacturers were the “Preventive Controls for Human Foods (PCHF)” and the “Foreign Supplier Verification Program (FSVP)” which were published in final form in September of 2015 and late November of 2016. The PCHF has reached full implementation and enforcement by the FDA, with FSVP not far behind.

This full symposium will review published and unpublished data on the number of FDA FSMA enforcement inspections, the number of 483s, U.S. Customs food rejection and hold data, identify the data for each major food sector and then compare it to FDA recall data as well as CDC illness outbreak data, both historically as well as current to see if there are any trends to identify if FSMA is having a positive impact on food safety in the U.S.

In addition to the quantitative data, speakers will be providing qualitative date case studies on the actual content of the FDA FSMA-based 483s as well as private food manufacturer’s own internal auditing summaries to share specific issues that are being identified by FDA and internally by food companies as needed more attention in this the “FSMA Era” of food safety.

Finally, there will be an assessment of whether the efforts of the FDA-supported Food Safety Preventive Controls Alliance (FSPCA) and the Produce Safety Alliance (PSA) to improve the knowledge and skills at all levels in the food processing industry has been effective.

S2  Seek and You Shall Find: The Intricacies of a Robust Listeria Environmental Monitoring Plan

JENNY SCOTT: U.S. Food and Drug Administration – CFSAN, College Park, MD, USA
LINDSAY WARD-GOKHALE: U.S. Department of Agriculture – FSIS, Washington, DC, USA
JOHN DONAGHY: Nestec Ltd., Vevey, Switzerland

Listeria is a ubiquitous microorganism and for many food establishments, it is an inescapable food safety matter. This symposium will highlight the components of a strong and effective EMP with special considerations for Listeria. Focus topics will include FDA and USDA guidance documents, pathogen harborage points, sampling devices and methodology, detection methods, implementation of preventive controls, and corrective actions.

The symposium will begin with an introduction and overview of the current FDA guidance for ready-to-eat foods and prerequisite programs necessary for Listeria control. The FDA will then provide insights into the FSIS and their experiences with risk assessment of Listeria. An industry expert will subsequently share their perspectives and engage the audience with case studies, lessons learned, and personal insights. Novel technologies for monitoring Listeria, including whole genome sequencing and agent-based methods, will be provided. The symposium will conclude with a presentation on current data gaps and research needs for Listeria. The overall goal of this symposium is to outline the current FDA and USDA guidance and examine the ideology of a robust EMP for Listeria with viewpoints from these government agencies and industry.

S3  Tracing Produce: Where We are and What’s Next?

ED TREACY: PMA, Newark, DE, USA
KATHERINE VIERK: U.S. Food and Drug Administration, College Park, MD, USA
TEJAS BHATT: Walmart, Bentonville, AR, USA

After nationwide outbreaks of E. coli O157: H7 and Salmonella spp. linked to produce items, many questions exist on how to improve traceability of produce from farm to fork and fork to farm. This session will provide information on current industry initiatives underway to address produce traceability, discuss the status of regulations impacting traceability, and introduce technological advances that are making an impact.

S4  Water Management in Food Manufacturing: Be Prepared for Problems

MIEKE UYTENDAELE: Laboratory of Food Microbiology and Food Preservation, Ghent University, Ghent, Belgium
PHYLLIS POSY: Strategic Services & Regulatory Affairs Atlantium Technologies, Har Tuv Industrial Park, Israel
ANETT WINKLER: Cargill, Inc., Munich, Germany

Food and Beverage industries around the Globe differ enormously in their size, products and capabilities. However, there is a common dependence on water, especially on the water that is safe and suitable for use in a particular operation. Also in common are many challenges in water management, partially driven by climate/weather changes (e.g., hotter summers in moderate climates). The symposium will look at choosing/managing incoming water supplies, how to match water sourcing with treatments that are suitable for which hazards, as well as following the water stream in the manufacturing to ensure adequate quality at the point of use. The symposium would start setting the scene by discussing recent food safety issues where water was involved and how to identify water hazards in overall hazard analysis. Different Water treatment technologies will be outlined and practical approaches to delivering water suitable for use presented. The last segment focuses on maintaining microbial integrity within a plant providing practical insights into storage and distribution challenges and water testing regimes.
S9 – S13

Symposium

S5 Does Zero Risk Really Exist: How to Communicate Variability and Uncertainty to Government and Industry Managers
LAURENT GULLIER: AVIS Food Laboratory for Food Safety, University of Paris-Est, Marne-la-Vallée, France
LEON GORRIS: Food Safety Expert, Nijmegen, Netherlands
KRIS DE SMET: European Commission, Gên, Belgium
Standard methods, such as microbiological analysis, take years to standardize and are not widely regarded as the go-to standard to ensure food safety. The sources of uncertainty and variability in the available information and parameters used in the risk assessment should be fully characterized and documented in order to provide a more complete account for risk. Hence, communicating the output of such assessments to decision makers from the government and the industry can be challenging especially in the absence of a clear risk acceptable level. It is not clear what level of uncertainty and variability approaches and guidance is crucial to facilitate risk managers to incorporate properly the uncertainty in the decision making process thus enhancing the choice of more effective mitigation strategies.

The purpose of this symposium is to give the audience:
- an introduction to the concepts of variability and uncertainty, how to differentiate and use them to ensure food safety
- a presentation on the industry challenges to take a decision in a variable and an uncertain world
- a presentation on how variability and uncertainty are managed by risk managers and how they are taken into account in regulation
- ended audience: Risk managers and assessors in food safety authorities and food industry

S6 Impact of Robotics and Artificial Intelligence on Food Safety
ALAN CHAN: Alibaba Inc, Hangzhou, China
MIKE HARPER: Soft Robotics, Bedford, MA, USA
IAN JENSON: Meat & Livestock Australia, North Sydney, Australia
Roboticization, artificial intelligence (AI) and Machine Learning (ML) are rapidly developing and revolutionizing the agriculture and food industry, from optimizing production and product innovation to tailoring product design and addressing consumer preference. In September 2017, the world’s first entirely machine-operated crop was harvested without a human ever entering the field where it was produced. However, while robots typically do what their designers intend, any decision making coming from a robot needs to be validated on the program computers. While speed, efficiency and non-stop working may be benefits, the question is whether there are any new governance or operational risks to consider. So, what is happening in the food industry at this point, do we gain experience and do we learn about managing possible risks?

Food and beverage companies are typically among the least prepared to exploit the power of AI and ML, essentially because they lack relevant data. Nevertheless, amongst the few industry players starting to move, in the coming years, new machines making independent decisions will increasingly produce outcomes contributing to health, food safety and other areas.

The short symposium will bring together a few experts in this field to 1) give an update of how far AI and robots have developed in primary food production and in food production; 2) the (existing or absence of) consideration of food safety concepts in the design of these machines; and 3) current or planned initiatives on the regulatory and standard-setting side to accompany this revolution. Speakers will be asked to contribute on topics such as: optimizing safe food designs using artificial intelligence; artificial intelligence impact on manufacturing operations and food safety assurance; and challenges in embracing artificial intelligence for food safety.

S7 New Methods in Analytical and Bioanalytical Sensing for Food Safety and Quality
ULLI HE: University of Missouri at Citzken, Anheuser, MA, USA
EMMA FARQUHARSON: Cornell University, Ithaca, NY, USA
JOEY TALBERT: Iowa State University, Amst, IA, USA
The growing demand for rapid and cost-effective methods to rapidly detect food contaminants has led to several promising techniques. These methods could soon allow portable sensing for point-of-use analysis. The International Association of Environmental Analytical Chemistry (IAEAC) is co-sponsoring a symposium to discuss some of the innovative research designed toward improving food safety. We will hear from several early stage investigators who will discuss their microbial and chemical sensing efforts. These methods include Surface Enhanced Raman Spectroscopy (SERS), synthetic biology, enzyme engineering, and phage engineering. Audience members will gain a background as well as specific knowledge of what lies on the horizon for rapid methods.

S8 Ensuring Safety by Design: Connecting the Dots of Food Protection throughout the Farm-to-Fork Continuum – A Poultry Case Study
WILLIAM CHANEY: Diamond V, Cedar Rapids, IA, USA
JERRY LINDY PICKETT: IDBI-Analytical Laboratories, Springfield, AR, USA
STEPHANIE COLLARD: Clear Labs Inc, Menlo Park, CA, USA
Food systems are comprised of many interconnected and moving parts, which can quickly become very complex and overwhelming when considering that food producers are ultimately responsible for ensuring the safety of their products. With the shift from a reactive approach to food safety issues to more of a preventative approach sparked by the new Food Safety Modernization Act, companies are being challenged to ensure food safety by design from farm to fork. This symposium seeks to connect the food safety dots and considerations between suppliers, food producers, pathogen diagnostic developers, and regulators with an emphasis on the poultry industry. Experts in these respective areas will share their knowledge on topics such as pathogen identification in the live pre-harvest environment, product formulation, integrating food safety hurdles in manufacturing, product sampling and testing strategies, challenges and considerations for pathogen detection in challenging matrices, and a regulatory perspective on pathogen reduction in the poultry industry.

S9 Making Sense of Food Allergen Analysis
RAKHI PANDA: U.S. Food and Drug Administration, College Park, MD, USA
JOSEPH BAUMERT: University of Nebraska-Lincoln, Lincoln, NE, USA
Food allergens are critical food safety hazards and must be considered in the development of a food safety plan. Robust allergen control programs focus on the identification of food allergens to prevent the presence of undeclared allergens in products. Many manufacturers and auditors look to data from allergen testing to provide evidence of allergen control effectiveness. Analytical methods are also important for the evaluation of whether allergen contaminants in products can enter the food chain. Numerous methods, including commercial kits and procedures are widely used for the detection of allergens, but a substantial amount of confusion remains about choosing the right method for a particular application and interpreting the results of different types of methods. The objective of this symposium is to give attendees a fundamental understanding of how allergen methods work, why different methods can be used to deliver different results, how to incorporate analytical methods into an allergen control program, and what to do with positive qualitative results.

S10 Listeria monocytogenes and the Produce Industry: Best Practices for Sanitary Design, Control and Monitoring
JENNIFER MCENTIRE: United Fresh, Washington, DC, USA
Teresa Hajek: California-Dongola, CA, USA
ROBERT DONOFRO: Neogen Corporation, Lansing, MI, USA
This session will focus on the impact of listeria monocytogenes on the produce industry and the associated control measures and monitoring ap- proaches that are currently being applied or are in development. A review of the current regulatory requirements will be covered. Industry and associated agencies will discuss how they are collaborating to leverage industry/academic expertise to translate this science into tools people can use, and have a positive impact on pathogen control. The efficacy of various sanitation approaches and sanitary design on l. monocytogenes will be discussed. Approaches to validating and verifying the performance and claims of methods for monitoring and detection of l. monocytogenes and l. sippp will be presented.

S11 Why are We Still Having Food Safety Failures If We All Have Food Safety Systems?
GALE PRINCE: Sage Food Consulting, Cincinnati, OH, USA
SALLY CROWLEY: Coralg, Inc., Hopkins, MN, USA
NATHAN ANDERSON: U.S. Food and Drug Administration, Bedford Park, IL, USA
While there is a tremendous diversity of human food in the global food supply, history teaches that there are some foods in which there is just starting to be adopted by some organizations in the food industry (in areas such as automation of processing and supply chain optimization) but there are still some in which there are no definitive preventive controls that will eliminate consumer risk illustrating the importance of investments in research that will help food safety professionals better understand the hazard of our food supply and technologies to reduce or eliminate these hazards.

S12 Water Re-use in the Food Processing Industry: Risk-based Approaches in Practice
KANG ZHOU: FAO, Rome, Italy
LEON GORRIS: Food Safety Expert, Nijmegen, Netherlands
PHILYS POST: Strategic Services & Regulatory Affairs Atlantic Technologies, Har Tув Industrial Park, Israel
ANNA LEDDING: Mactec, Spain
While access to reliable sources of potable water varies dramatically around the globe and such sources are often scarce already, global trends including food security and global warming increasingly exacerbate water supply shortages to the extent that water security is under threat. Among the possible other sources of water available to the food industry could be water that is recovered from food or from operations in a food processing/ handling facility. Such water could be re-used in different ways and for different purposes, but the possible occurrence of microbiological and other hazards needs to be considered. There is yet little science and operational best practices for responsible water re-use in key segments of the food industry. When deciding whether a re-use water source can be used for a specific food application and whether a treatment or other type of reconditioning is required to make this water fit-for-purpose, the key criterion is that the re-use water does not pose a health risk to consumers. A risk-based treatment decision-model is currently being developed and is designed to help water re-use operations be carefully assessed and managed in the context of the specific food facility, and risk mitigation measures must be managed within that facility’s food safety management system (e.g., GHPL/ACPS). As food industry sectors in more countries develop or improve water re-use solutions, and more data become available, there is a need to establish a regulatory and industry expertise from around the world to share practical lessons learned in developing and implementing risk-based operational guidelines.

S13 Artificial Intelligence and Machine Learning: What they are and Their Potential Applications for Food Safety
KATHLEEN BAKER: Center for Food Safety, University of Georgia, Griffin, GA, USA
WENDY WHITE: Georgia Tech, Greensboro, GA, USA
ABIGAIL HORN: Center for Applied Network Analysis, Rock School of Medicine, University of Southern California, Los Angeles, CA, USA
While the term was first coined by a computer scientist in 1956, the field of artificial intelligence (AI) has been expanding in the last few years due to improvements in processing power and data storage capacity, increasing data availability, and rapid progress of analytical techniques. AI is a broad and encompasses many different subfields (e.g., machine learning, natural language processing, and robotics). There is a great deal of excitement surrounding AI across public and private sectors and it is has already been transforming many industries such as healthcare and finance, and retail. AI is just starting to be adopted by some organizations in the food industry (in areas such as automation of processing and supply chain optimization) but is not widely utilized. Recently, there have been efforts to apply AI technologies in the area of food safety, yet there is a great deal of unexploited potential. For example, in the last year many organizations have been exploring blockchain technologies from farm to consumer – this presents an opportunity for companies to then apply AI technologies to improve food safety. It is time to discuss how AI can be used
to improve food safety. The overall goal of this symposium is to make AI accessible across audiences, in order to foster discussion and encourage the application of AI to food safety. Thus, this symposium is intended as a forum and forum towards this initiative. The symposium includes (i) review of the key AI and AI-related understanding and framework for discussion, (ii) discuss case-studies representing diverse applications, and (iii) present opportunities and challenges of AI in the food industry.

S14 Food Microbiome Transfer Dynamics from Farm to Processing - What Can Metagenomics Add to the Picture?

CHRISTOPHER GRIMM: U.S. Food and Drug Administration – CFSA, Laurel, MD, USA

STEVEN RICKE: University of Arkansas, Fayetteville, AR, USA

KEITH BELK: Colorado State University, Department of Animal Sciences, Fort Collins, CO, USA

The Human and Animal Gut Microbiome projects have provided critical information about the impact of human gut microflora on health and disease. These projects have the framework for studying the influence of microbiomes associated with human and animal foods and their production environments on food safety and quality. Metagenomic sequencing of foods and food environments can provide microbial community profiles and identify genetic markers of microbial resistance, pathogen serotypes, and virulence genes. For example, recent studies describing bovine fecal microbiota composition and beef production facilities have revealed potential associations between pathogen transfer and microbiota composition in processing facilities. This information could facilitate an understanding of the ecology of microbial communities in each habitat as well as transport of pathogens and antimicrobial resistance genes between foods and food environments. The goal of this symposium is to describe current microbiome research projects focused on pathogen presence and persistence in a variety of farm environments (i.e., the farm-to-fork transfer dynamics) along the processing sectors. We will provide an overview of microbial community profiles in foods and farm environments and describe microbiome shifts induced by preventive controls employed at meat and poultry processing facilities to control foodborne pathogens and spread of antimicrobial resistance genes.

Presenters will discuss the use of microbiome profiling to identify potential indicators of pathogen presence, persistence and transfer on produce and across the meat and poultry supply chains. Together, these talks will provide information about the risks associated with the dissemination of antimicrobial resistance and foodborne pathogens imposed by farming and production practices.

S15 Science and Regulatory Update: Lethality and Stabilization of Meat and Poultry Products

SUSAN HAMMONS: U.S. Department of Agriculture – FSIS, Washington, DC, USA

BRADLEY MARKS: Michigan State University, East Lansing, MI, USA

KEITH BELK: Colorado State University, Department of Animal Sciences, Fort Collins, CO, USA

STEVEN RICKE: University of Nebraska-Lincoln, Lincoln, NE, USA

In 2017, the U.S. Food and Drug Administration (FDA) and the U.S. Department of Agriculture (USDA) Food Safety and Inspection Service (USDA FSIS) released updated versions of the commonly used lethality and stabilization guidelines and safe harbors known as Appendices B and A. Appendices A and B are used by numerous meat and poultry processors both large and very small. The changes from the previous 1999 versions to the 2017 versions identified opportunities for additional research and increased specific details for the control of Salmonella, C. perfringens, C. botulinum, and E. coli. As part of the update of Appendices A and B, both industry and government also identified components that proved to be problematic for the industry to implement and an opportunity for improvement. Shortly after their release both USDA FSIS and Agricultural Research Service (ARS) and industry have commissioned numerous efforts targeting the gaps in the current scientific body of knowledge. The theme of this symposium is to update the scientific community on the ongoing and soon to be completed research and guidance targeting Salmonella, C. perfringens, C. botulinum in heat-treated meat and poultry products.

S16 May Contain Allergens – A Risk-based Approach for Determining the Use of Precautionary Allergen Labelling (PAL)

JOSEPH BAUMERT: University of Nebraska-Lincoln, Lincoln, NE, USA

BRENT KOBELUSH: Colorado State University, Fort Collins, CO, USA

DAVID CLIFFORD: Nestle USA, Inc., Dublin, OH, USA

The use of Precautionary Allergen Labelling (PAL) statements such as May Contain on Shared Equipment or Contains Traces on food labels is a source of confusion for consumers with allergies, and the clinicians that treat them. This symposium begins with a discussion on the potential sources of allergen cross-contact in the supply chain (agricultural comingling, distribution, storage) and the potential risk this scenario presents. The second presenter will focus on the cleaning of manufacturing equipment with specific a focus on allergen cleaning validations in wet cleaning protocols as a means to minimize the foodborne pathogens and spread of allergens within a cross contact. Lastly, a speaker will present on allergen thresholds and their application when using VITAs (Voluntary Incidental Trace Allergen Labeling) to determine if PAL is warranted based on risk.

S17 Managing Large Multidisciplinary/Multi-Institutional Food Safety Projects – Effectively, Impactfully, and with Integrity

DENIS GRAY: North Carolina State University, Raleigh, NC, USA

KIMBERLY COOK: USDA-ARS, Beltsville, MD, USA

LEE-ANN JAYKUS: Department of Food, Bioprocessing, and Nutritional Sciences, North Carolina State University, Raleigh, NC, USA

EDITH WILKIN: Lempicki Foods, Denver, CO, USA

LINDA J. HARRIS: University of California-Davis, Department of Food Science and Technology, Davis, CA, USA

Food safety is a complex and multidisciplinary challenge. Therefore, federally funded food safety projects, and even industry-centered projects, increasingly involve large, multidisciplinary, multi-institutional collaborative teams. The leaders of these collaborative teams often have little or no education or training in managing such projects. This symposium brings together a unique and diverse cohort of presenters, ranging from an expert on assessing the readiness of centers with experience in managing multisite projects to an expert on bringing in industry leaders to understand the unique challenges of managing such projects (in government, academic, and industry) to a representative of the Scientific Integrity Consortium. The speakers will describe measures for evaluating the effectiveness of such large-scale collaborations, identify common features of successful collaborations, share best practices for managing and forming such teams, and outline essential foundational principles for ensuring the safety and integrity of the resulting research. A panel discussion is included to maximize opportunities for attendance interaction with the multiple perspectives provided by the speakers. After this session, attendees will have a better appreciation of how to play together well in the research sandbox.

S18 Is Cell Cultured Meat Really Meaty?

PAUL MOZZIKA: North Carolina State University, Raleigh, NC, USA

ROBERTA WAGNER: Food and Drug Administration Center for Food Safety and Applied Nutrition, College Park, MD, USA

JEFFREY PASANO: Food and Drug Administration Center for Food Safety and Applied Nutrition, Washington, DC, USA

November 16, 2018, a joint statement was issued by FDA Commissioner Gottlieb and USDA Secretary Perdue on the regulation of cell-cultured food products. The memorandum outlines FDA and USDA met regulatory alignment and are building innovative food products and maintain the highest standards of public health.” The two agencies determined that “the USDA and the FDA should jointly oversee the production of cell-cultured foods products derived from livestock and poultry. Drawing on the expertise of both USDA and FDA, the agencies are being called to “review agreement framework within the FDA oversees cell culture technologies, and USDA is working on the regulatory framework.”

A transition from FDA to USDA oversight will occur during the cell harvest stage. USDA will then oversee the production and labeling of food products derived from the cells of livestock and poultry. “FDA and USDA are confident that this regulatory framework can be successfully implemented and assure the safety of these products.”

Biotechnology is making possible meat production through non-traditional means. This symposium was designed to provide baseline knowledge to attendees regarding what cell-cultured meat is, an overview of animal-derived cell culture technology, related food safety concerns particular to these new methods of production, and information regarding regulation and oversight of “clean meat” facilities which produce varieties of beef, chicken, pork, and fish using this new technology. These products present certain regulatory challenges, which will be discussed, as they are not derived from animal carcasses, and will require some adjustments to statutory and regulatory definitions.

Is there a strategy for regulating these novel food products? What role will USDA-FSIS play?

As a first step toward addressing how Federal regulatory agencies will assure the safety and accurate labeling of human food produced using animal cell culture technology and the inspection of establishments involved in its production, FDA and FSIS have worked collaboratively to develop an agreement framework. This symposium will provide attendees regarding what cell-cultured meat is, an overview of animal-derived cell culture technology, related food safety concerns particular to these new methods of production, and information regarding regulation and oversight of “clean meat” facilities which produce varieties of beef, chicken, pork, and fish using this new technology. These products present certain regulatory challenges, which will be discussed, as they are not derived from animal carcasses, and will require some adjustments to statutory and regulatory definitions.

What is it? Where is it from?

How are cells harvested from animals and is how meat grown from those cells? A clean meat facility is like a clean, fermentation-based food processing plant and allows meat to be harvested from cells rather than from raising animals to provide animal products to our growing population.

Is there a strategy for regulating these novel food products? What role will FDA play?

FDA is responsible for implementing and enforcing the Federal Food, Drug, and Cosmetic Act, the Public Health Service Act, and the Fair Packaging and Labeling Act. FSIS is responsible for implementing and enforcing the Federal Meat Inspection Act, the Poultry Products Inspection Act, and the Egg Products Inspection Act. Each agency has an important role in the oversight of human food derived from cell lines of USDA-amenable species and required to bear a USDA mark of inspection. A previous talk described the joint FDA/USDA agreement outlining the roles and responsibilities for oversight of such food as well as FSIS’s plans for application of their statutory authorities under the agreement. This talk will discuss FDA’s statutory authorities, prior relevant experience in multiple regulatory domains, and considerations informing the development of the premarket assessment process and postmarket oversight of cell culture described in the joint agreement.

S19 Beyond Slide Decks and Classrooms: Novel Approaches to Food Safety Learning

ANDY YEOMAN: Focus Games, Glasgow, United Kingdom

CAROL LEAMON: Asansify, Inc., Waterloo, ON, Canada

AUSTIN WELCH: Sage Media, Thornton, CO, USA

MEGAN RENDORA: The Hershey Company, Hershey, PA, USA

The old-style lecture using slide decks is a boring approach to food safety training. Adding videos, in-class discussions and clicker response systems can reduce monotony and increase learner interest and engagement, yet there’s so much more that can improve learning and, ultimately, food safety culture. Novel uses of food in the food industry to enhance learning, interest and engagement at different levels. This symposium will raise awareness about these new approaches.

Board games which have been used for millennia to pass the time have been developed and used to transfer and reinforce food safety messages while encouraging interaction with team members during training sessions and breaks. Companies are also changing the dynamics of learning by including micro-learning approaches, often in combination with internal computer or online games, to improve engagement of associates and positive feedback. As food safety learning builds on evidence from training that shows knowledge retention increases when information is presented in short targeted training, online/off-line, which addresses different learning styles, Employee behavior changes when learning is effectively reinforced within the learning platform and on-the-job.

Of course, senior leaders have different mindsets and needs for food safety information so learning approaches must engage their brains in a way that works for them. Furthermore, senior leaders coming from outside the food industry often have limited understanding of food safety risks. The ground-breaking use of escape rooms, investigative approaches and other experiential learning techniques are building essential learning and stepping into critical thinking skills to change behavior, thereby protecting brands and consumers.

Different options are available to the food industry to improve learning outcomes and change behavior to ultimately strengthen food safety culture. Presenters in this symposium will spark ideas about the potential application of novel approaches in workplaces to advance food safety practices and cultures.
S20 International Food Defense Preparation for FSMA and Beyond
AMY KIRCHER: Food Protection and Defense Institute, University of Minnesota, St. Paul, MN, USA
RYAN NEWBORN: U.S. Food and Drug Administration, College Park, MD, USA
KARLEIGH BACON: Kraft Heinz Company, Glendale, IL, USA

The first FSMA Intentional Adulteration Rule compliance date is July 26, 2019. With this deadline fast approaching, the food industry is working to prepare for food defense programs that comply with the regulation and that protect the public from the threat of intentional adulteration. The food system is a globally connected supply chain where a disturbance at one point could resonate throughout the world. Therefore, collective global vigilance is necessary to protect the system from intentional adulteration. The objective of this symposium is to share best practices and lessons learned from international, federal, and state food defense initiatives. The intended audience includes food safety, quality, and regulatory personnel in the food industry, government, and academia responsible for contributing to global food defense efforts. The session will start by describing the need and current status and discuss how challenges and opportunities are being addressed in the industry. The platform will involve speakers representing the food industry, government, and academia from multiple countries. The session will include case studies and address how to remove the subjectivity from the process and assign accountability beyond that of the technical lead to engage the entire site.

S21 Applying Lessons Learned: Keeping STEC Off Our Lettuce
MICHÈLE SMITH: U.S. Food and Drug Administration, College Park, MD, USA
MIRA MATTIOLI: Centers for Disease Control and Prevention, Division of Foodborne, Waterborne and Environmental Diseases, Atlanta, GA, USA
TERESA LOPEZ: Arizona LGMA, Phoenix, AZ, USA

Recent outbreaks in foodborne illness associated with romaine lettuce are of great concern to both the food industry and the public. Many of these outbreaks are occurring in the spring and have been associated with Escherichia coli O157:H7. In response to the outbreak, key industry partners formed a Leaflly Greens Task Force designed to assess and address issues associated with recent foodborne illnesses attributed to consumption of leafy greens and to prevent such a tragedy from occurring in the future. This symposium will provide a brief overview of the 2018 E. coli O157 outbreak linked to romaine lettuce grown in the Yuma region and focus more on the environmental assessment findings including water testing, lessons learned, and both federal and industry prevention strategies moving forward.

S22 Breaking the Mold: Using Foods to Protect Against Food Allergy
WESLEY SIBLEY SUBBUT: University of Louisville School of Medicine, Louisville, KY, USA
MARTIN CHAUDY: Indoor Biotechnologies, Inc., Charlottesville, VA, USA
SCOTT COFFMAN: Indoor Biotechnologies, Inc., Chapel Hill, NC, USA

Sensory breakthroughs in food allergy from the Learning Early About Peanut Allergy (LEAP) trial show that introducing food (peanut) into the diet during weaning can prevent peanut allergy. This session will describe how these studies have transformed treatment of food allergy. A variety of peanut foods are now being developed for allergy prevention in infancy. The approach is also being applied to other food allergens. The symposium will provide a primer on what makes a food an allergen and discuss the allergenic composition of foods and food ‘prevention’ products. New technology for multiple food allergens simultaneously. This innovative technology can be used to monitor therapeutic doses of allergens in foods and to improve safety. This symposium will provide an overview of the 2018 E. coli O157 outbreak linked to romaine lettuce grown in the Yuma region and focus more on the environmental assessment findings including water testing, lessons learned, and both federal and industry prevention strategies moving forward.
The Use of Radical Microbial Methods by Government Agencies for “Official” Testing

PAUL INT VELD: Netherlands Food and Food Product Safety Authority, Utrecht, Netherlands

PATIMA FIRIEE: Food Safety Dubai, Dubai, United Arab Emirates

BOBBY KRISHNA: Food Safety Dubai, Dubai, United Arab Emirates

THOMAS HAMMACK: U.S. Food and Drug Administration - Center for Food Safety and Applied Nutrition, College Park, MD, USA

MARGO HAMLING: Animal Health Protection for Primary Industries, Wellington, New Zealand

CONSTANZA VERGARA ESCOBAR: Chilean Food Safety and Quality Agency, AICHAPIA, Ministry of Agriculture, Santiago, CH, Chile

JOSE EMMO EMOSIO: FDA, Center for Food Safety and Applied Nutrition, College Park, MD, USA

Microbiological methods used by government agencies for “Official” testing have traditionally been cultural-based methods – many, which are decades old, time-consuming, and have many procedural steps that can be challenging for members of a laboratory to conduct with consistency.

Over the last 30+ years, method developers have introduced newer, faster methods (technologies) that have improved: time-to-result, method repeatability, sensitivity, etc. In addition, several globally validated registration/verification bodies/certification programs have adopted and harmonized well-defined scientific standards that have been used by many method developers to validate these rapid methods and demonstrate performance equivalence to recognized reference official methods.

Many food manufacturers and testing labs around the globe, would like to use these validated/certified rapid methods more routinely, but an important question remains:

Will government agencies accept the use of these methods for “Official” testing?

This IAPP symposium will focus on speakers from government agencies who will be asked to follow a defined presentation format, so country criteria can more easily be compared across each global region being presented. Each speaker will answer the following key questions.

Does their agency:

1. Allow for the use of rapid microbial methods for Official testing?

2. Require these rapid microbial methods to be first validated through recognized global certification bodies such as: AOAC INTERNATIONAL, Microbial Certification, NV, Validation by AO/NFQ, NorCal Certification?

3. Have additional requirements that these rapid microbial methods must meet?

4. Have any defined restrictions/limitations for the use of these rapid microbial methods?

A final slide summarizing the answers to these 4 questions from each of six participating agencies will be projected, with time planned for Q&A.

New Research Findings – Control of Listeria in Dairy

KATHLEEN GLASS: University of Wisconsin-Madison, Madison, WI, USA

DENNIS D’AMICO: University of Connecticut, Department of Animal Science, Storrs, CT, USA

KEVIN KEENER: Iowa State University, Ames, IA, USA

VENEDA RIGHTNGALE: Texas Tech University, Lubbock, TX, USA

STEPHANIE RICHARD: University of Alabama, Tuscaloosa, AL, USA

MARTIN WIEDMANN: Cornell University, Ithaca, NY, USA

The Innovation Center for U.S. Dairy (IC) provides a forum for the dairy industry to work together pre-competitively and is focused on broadening the adoption of current tools and best practices. In 2015 the IC initiated a research consortium to expand dairy’s tool kit by leveraging both industry and academic expertise to identify gaps and develop pre-competitively. These research efforts are creating new tools and practices to control Listeria in finished product and plant environments with the goal of protecting consumers and the dairy industry. This symposium will highlight the research findings of the consortium which range from fundamental knowledge of understanding the cell envelope of Listeria to methods to control Listeria in high-risk cheeses.

A Precarious Balancing Act: Co-managing Preharvest Environments to Maximize Food Safety, Sustainability, and Economic Viability of Farm Operations

MICHELE JAY-RUSSELL: Western Center for Food Safety, University of California-Davis, Davis, CA, USA

PATRICK MAUR: University of California, Berkeley, Berkeley, CA, USA

MATTHEW JONES: Washington State University, Pullman, WA, USA

Recent decades have had important implications for food safety measures on ecosystem health in agricultural environments. For example, following the 2006 E.coli outbreak in spinach, growers reported increased pressure from buyers, auditors, and other groups to implement new measures for food safety, including those related to preharvest pest management, dust management, and the like. The goal of this research is to provide answers to the following questions:

• What are the preharvest pest management practices that are most effective for reducing the risk of pathogenic foodborne waterborne pathogens and other adhesions (e.g., non-crop vegetation removal)?

• How can these practices be implemented in ways that are economically viable for farms and farm communities?

• How do these practices interact with preharvest pest management practices?

Developing tools and approaches for co-managing (i.e., managing a farm for multiple aims, such as food safety and conservation) farms is a complex problem that requires input from all stakeholders, including including government officials and extension agents. It is therefore critical that members of the IAPP community who interface with farmers are at the forefront of co-management research. Much of the current research on co-management is currently being pursued by researchers from outside the IAPP community. Therefore, this symposium is a unique opportunity to bridge the gap between the research and the forefront of co-management and food safety specialists, as well as introduce a conceptual framework to facilitate the development of science-based co-management strategies. The objectives of this proposal are to (i) provide an overview of the impact of food safety practices on ecosystem health, (ii) present perspective on grower needs and current co-management efforts, (iii) present a case study on co-managing preharvest environments, and (iv) provide a checklist of suggestions for addressing the impact of food safety practices on ecosystem health.

Emerging Hazards Associated with Seafood

MELANIE GAY: ANSES, Boulogne sur Mer, France

JACQUELINE WOODS: U.S. Food and Drug Administration, Dauphin Island, AL, USA

STACEY MCLEROY: U.S. Food and Drug Administration, Center for Food Safety and Applied Nutrition, College Park, MD, USA

Seafood presents a unique challenge from a food safety perspective. There are a wide variety of seafood products consumed, including fish, crustaceans, and molluscan bivalves. Each has food safety hazards associated with them. And, some are known to be riskier than others.

Sometimes that is because of where the seafood is harvested; sometimes because of how it is consumed. While some hazards, such as bacteria, can be reduced by cooking; others, such as toxins, are heat resistant. Parasites can be killed by freezing the seafood before reaching the consumer, while the freezing preserves many of the parasites antigenic components any enteric viruses that may be present. So, it is no wonder that seafood is a common route of foodborne illness. Even with all we know, ever-changing consumption patterns, the spread of marine organisms, and diversity of harvest give rise to new sources of seafood safety concerns. This session will highlight some examples of recently emerged seafood safety hazards. In addition, the effectiveness of current food safety controls for these emerging hazards and the need for new, or modified, control measures will be discussed.

Fact or Fiction: Combating Consumer Perceptions of Food Safety Myths with Data

DONALD W. SCHAFER: Rutgers University, New Brunswick, NJ, USA

LINDA BETHENY ANDREW: University of Georgia, Athens, GA, USA

AARON LAVallee: USDA Food and Safety Inspection Service, Washington, DC, USA

In today’s internet is rife with messages around safe food handling, some correct, but many created without scientific data or support to back them up. These messages have been fed into social media channels and other digital outlets perpetuating misinformation on proper food safety practices and allowing them to persist or even spread among consumers. As the debate around food safety evolves to more targeted discussions, food safety professionals need to be generating data and developing sound evidence-based messages to engage with the public on these issues. Changing consumer’s minds is difficult, though not impossible, task to undertake and requires strong data on the myth in question along with effective communication strategies and repetition of the message itself. Performing research to generate data regarding the validity of such myths could allow food safety professionals more effectively engage with consumers that believe or practice behaviors inspired by such narratives. However, there is an abundance of food safety myths and relatively few of them have been tackled and debunked effectively. Those that have are often initially met with resistance by consumers and may lack the scientific and consumer trust to be effective. This session will focus on employing these food safety narratives popular in the web and discuss several examples of where specific food safety myths were researched, such as the S-5 rule, double dipping, and home food preparation-based myths, followed by how such data can be generated, how messaging is developed from that data, and how these messages are ultimately used in a packaging context to raise consumer awareness and change behavior.

Future Pains: Assessing the Long-term Consequences of Foodborne Exposure to Microbial and Chemical Hazards

KRISTEN POGEREA BROWN: University of Arizona, Tucson, AZ, USA

BARBARA KOWALCYK: The Ohio State University, Columbus, OH, USA

SUZANNE FITZPATRICK: U.S. Food and Drug Administration, College Park, MD, USA

Foodborne exposures to microbial and chemical hazards are major causes of morbidity and mortality globally. In the U.S., foodborne pathogens cause an estimated 76.6 million illnesses, 325,000 hospitalizations, and 6,748 deaths annually. In addition, these acute illnesses, motion, infant mortality, and chronic diseases are associated with multiple types of cancer, as well as developmental, neurological, respiratory, and other effects. Because many of these manifestations may not arise for weeks, months, or even years after infection, they can be difficult to characterize and are too often neglected by clinicians and public health officials.

Long-term health outcomes caused by microbial and chemical hazards cause considerable burden and should be considered in food safety prioritization and policy development. To do so, we need sustained research and tools to identify characteristic, and quantifying the likelihood, durations, and severities of long-term health consequences. This symposium will present a number of different approaches to assessing these impacts, including the use of literature review, cohort surveys, analysis of medical claims data, and novel predictive toxicology tools such as computer-based.
S37 Campylobacter, Health Impact, Performance Objectives and Effectiveness of Sampling Plans
JEFFREY FABER: University of Guelpgh, CRIFS, Department of Food Science, Guelpgh, ON, Canada
LEON GORRIS: Food Safety Expert, NJmegen, Netherlands
MARCEL ZWIEUIT: Wageningen University, Wageningen, Netherlands

Campylobacter is the zoonotic bacterium with the largest public health impact in many countries. Since the organism does not grow in the food chain, many investigations focus on the production and slaughter of animals. In susceptible biological hazard is identified. The food industry has also enhanced efforts to ensure that there is scientific justification for the process controls used to mitigate potential biological hazards, including in-plant validation studies using surrogate organisms. However, conducting in-plant validation studies may not be feasible for some pathogen inactivation strategies due to the size and layout of industrial operations. The lack of suitable surrogate for certain processes, challenges in designing appropriate inoculation techniques and procedures for recovery of the surrogate, and the financial burden of completing multiple validation studies can be daunting and may hinder the plant’s ability to maintain a robust food safety program. Many supplier verification audits have already included audit criteria to assess the adequacy of preventive controls to deliver the target lethality. Food recalls, however, continue to occur for products with well-recognized thermal or chemical preventive measures. Pathogen recontamination along the chain and the levels of pathogens in the food product, as well as the heterogeneity of pathogen distribution, it is critical to design appropriate sampling plans.

S38 When the Enterobacteriaceae Hits the Fan: Wind and Particulate-associated Distribution of Foodborne Pathogens
UMITRUI MACARISIN: U.S. Food and Drug Administration, College Park, MD, USA
DE ANN DAVIS: Church Brothers Farms, Salinas, CA, USA
DAVID INGRAM: U.S. Food and Drug Administration – CFSAW, College Park, MD, USA

Wind, dust and particulate matter suspended in the air can be an understudied vehicle for foodborne pathogen dispersal in agricultural animal operations and food processing facilities. Dust storms, farm operations, soil erosion and other factors can result in the redistribution of foodborne pathogens across vast distances. The consequences of these events can amount to outbreaks and recalls, with the vehicle and source of the pathogen remaining elusive. Additionally, microbial physical characteristics render these contaminants amenable to transmission. The ability to transport dispersed foodborne pathogens can affect a wide variety of produce such as fruits, leafy greens and tree fruit. The role of dust, wind and climatic conditions that can cause their deposition on foodborne pathogens on the surfaces of produce is of high importance as it can help in better risk evaluation, mitigation and preventative control measures.

The talk presented in this symposium will focus on:
- Role of particulate distribution of foodborne pathogens in tree fruit production environments.
- Grower’s concerns associated with dust storms and dust dispersal.
- Regulatory adaptations that can help mitigate risks associated with dust dispersal.

S39 What Do We Know about Microplastics in Foods and Their Impact on Human Health?
GARTH COVERNTON: University of Victoria, Dept. of Biology, Victoria, BC, Canada
J. EVAN WARD: University of Connecticut Dept. of Marine Sciences, Groton, CT, USA
BART KOELMANS: U.S. Food and Drug Administration Research and Wageningen, Netherlands

Plastic waste and pollution are becoming a huge global problem. It is estimated that by 2030, 111 million metric tons of plastic will be displaced by China’s January 1, 2018 import ban. (China previously used or reused recycled plastic and was the largest importer of plastic waste.) The food industry uses large amounts of single-use plastics to package food and beverages. Plastic waste slowly degrades into small particles (micro- and nanoparticles) which can find their way into food and consumers. While the risk of microplastics in food products is probably highest in seafood because of the plastic pollution in oceans and freshwater, microplastics have been found in beer and honey, and pig and poultry feed contains fishmeal.

Are you frightened, curious or skeptical about reports of plastic contaminants in foods and beverages? Do you communicate with consumers about these issues? Do you have plans to communicate with consumers about the risks and precautions of these contaminants? The session will focus on characterizing the potential risks and benefits of food-related microplastics.

S40 The Mitigation and Regulation of Heat-formed Substances Produced in Foods during Cooking: What Have We Learned and How Can We Use This Knowledge to Inform Risk Assessment?
J. EVAN WARD: University of Connecticut Dept. of Marine Sciences, Groton, CT, USA
DAVID INGRAM: U.S. Food and Drug Administration – CFSAW, College Park, MD, USA

A growing field in food safety is the focus on the potential risk of heat-formed substances produced during cooking. Compounds that are known as heat-stable, but unstable at the lower temperatures practiced in many countries. It will address how to assess and communicate these risks to food processors and consumers. The potential impact and implications on the food industry and, ultimately, the end consumer, of using current approaches to assess the potential public health impact of compounds formed during routine cooking of food will be debated.

S41 Strategies to Prevent Pathogen Contamination in Post-harvest Dry and Wet Environments
LORALYN LEDENBACH: Kraft Heinz Company, Glenview, IL, USA
LILA WILSON: Kellogg’s, Battle Creek, MI, USA
JOHN HOLAH: UKIE NEHGD & Holchem Laboratories Ltd., Bury, United Kingdom

The enactment of the Food Safety Modernization Act has paved the way to modern food safety standards and prevention practices. Many food chains have regulatory bodies in place to enforce these standards. The food industry has also enhanced efforts to ensure that there is scientific justification for the process controls used to mitigate potential biological hazards, including in-plant validation studies using surrogate organisms. However, conducting in-plant validation studies may not be feasible for some pathogen inactivation strategies due to the size and layout of industrial operations. The lack of suitable surrogate for certain processes, challenges in designing appropriate inoculation techniques and procedures for recovery of the surrogate, and the financial burden of completing multiple validation studies can be daunting and may hinder the plant’s ability to maintain a robust food safety program. Many supplier verification audits have already included audit criteria to assess the adequacy of preventive controls to deliver the target lethality. Food recalls, however, continue to occur for products with well-recognized thermal or chemical preventive measures. Pathogen recontamination along the chain is troublesome and there is a clear relationship between the prevalence of Campylobacter in broiler flocks and public health risk. Chickens serve as reservoirs, and Campylobacter’s ability to survive on chicken flocks depends on its ability to survive the cooking process (i.e., heating). Heating and subsequent storage conditions, survival ability and cell history affect the robustness of Campylobacter along the chain and quantitative knowledge is needed to refine microbiological exposure assessment models and to evaluate the impact of industrial control interventions. The effectiveness of control measures is verified by microbiological testing. Testing efficacy is however hampered by the fact that Campylobacter is often damaged in food and may only represent a small fraction of the total microflora in food. Analytical testing methods, therefore, incorporate an enrichment procedure to recover and selectively amplify Campylobacter to higher numbers and quantitation following enrichment. These enrichment-based detection systems are used for surveillance and can be used for rapid detection systems. This session focuses on the challenges on the control and detect Campylobacter along the food chain. It will highlight the advances to predict the robustness of Campylobacter along the food chain using molecular markers taking into account cell history and strain variability. It will also discuss the effects of strain variability and competitive flora on the outcome of enrichment-based detection procedures, and it will give an update about the lessons learned and the difference between qualitative and quantitative pathogen detection methods in Campylobacter products.

S43 Are There Instructions Included? The Role of Regionality and Experimental Choices on the Survival of Foodborne Pathogens in Manure-amended Soils
MANAN SHARMA: U.S. Department of Agriculture – ARS, Environmental Microbial and Food Safety Laboratory, Beltsville, MD, USA
MICHAEL JAV-RUSSELL: Western Center for Food Safety, University of California-Davis, CA, USA
KEITH SCHNEIDER: University of Florida, Gainesville, FL, USA

Animal manure is a commonly used organic fertilizer for cultivation of fruits and vegetables. Land application of manure is a useful management strategy for increasing organic carbon and soil microbial biomass. However, the placement of manure in soils can introduce bacterial pathogens into food chains. A number of studies have been conducted to evaluate the survival of foodborne pathogens in manure-amended soils. These factors, along with manure types commonly used, application method and rate, vary among geographic region in the U.S. and other countries. What food safety programs and/or strategies can be used to control these pathogens in manure-amended soils? How should risk assessment and control strategies be developed? The session will begin by presenting the current state of knowledge on the survival of foodborne pathogens in manure-amended soils. The session will then go on to explore recent advances in fundamental and emerging technologies in the monitoring of pathogens in manure-amended soils. The session will highlight the progress that presentations and discussions during this symposium will spark meaningful dialogue on how best to evaluate the threat of microplastics to our food supply and trigger plans and actions to minimize it.

S42 Challenges in Campylobacter Detection and Control
HEIDY DEN BESTEN: Wageningen University, Wageningen, Netherlands
BENJAMIN DUQUE: UMBR1014 Seattle, WA, On, Nantes, France
MICHAEL WILLIAMS: U.S. Department of Agriculture-FSIS, Washington, DC, USA

Campylobacteriosis is the most frequently reported zoonotic worldwide. Control of Campylobacter along the food chain is troublesome and there is a clear relationship between the prevalence of Campylobacter in broiler flocks and public health risk. Chickens serve as reservoirs, and Campylobacter’s ability to survive on chicken flocks depends on its ability to survive the cooking process (i.e., heating), heating and subsequent storage conditions. Survival ability and cell history affect the robustness of Campylobacter along the chain and quantitative knowledge is needed to refine microbiological exposure assessment models and to evaluate the impact of industrial control interventions. The effectiveness of control measures is verified by microbiological testing. Testing efficacy is however hampered by the fact that Campylobacter is often damaged in food and may only represent a small fraction of the total microflora in food. Analytical testing methods, therefore, incorporate an enrichment procedure to recover and selectively amplify Campylobacter to higher numbers and quantitation following enrichment. These enrichment-based detection systems are used for surveillance and can be used for rapid detection systems. This session focuses on the challenges on the control and detect Campylobacter along the food chain. It will highlight the advances to predict the robustness of Campylobacter along the food chain using molecular markers taking into account cell history and strain variability. It will also discuss the effects of strain variability and competitive flora on the outcome of enrichment-based detection procedures, and it will give an update about the lessons learned and the difference between qualitative and quantitative pathogen detection methods in Campylobacter products.

S44 Updates on the Impact of Sampling Plans on Food Safety
JOHN HOLAH: UKIE NEHGD & Holchem Laboratories Ltd., Bury, United Kingdom

The session will provide an overview of the current understanding regarding the impact of sampling plans on food safety. It will cover a broad range of topics, including the importance of sampling plans in food safety, factors that can affect the effectiveness of sampling plans, and the latest research in this area. The session will also provide practical advice on how to develop and implement effective sampling plans.
45 Updates to the Conference for Food Protection and the Food Code

DAVID MCSWANE: Conference for Food Protection, Martinsville, IN, USA

GIRVIN LIGGANS: U.S. Food and Drug Administration, College Park, MD, USA

BRENDA BACON: Harris Teeter, Matthew, NC, USA

The Conference for Food Protection (CFP) originated in 1971 to provide members of industry, regulatory, academic, consumer and professional organizations with a forum to discuss food safety issues and the need for rational modifications of Food Safety Guidance. Such guidance is incorporated into food safety laws and regulations at all levels of government throughout the United States. The Food Code defines criteria that are critical to reducing the risk of foodborne illness in food establishments, provide uniform standards to recognize levels of food safety that are adequate, and ensure compliance and enforcement structures that can achieve these goals. The CFP symposium will outline important changes to the Food Code over recent years included in the 2017 edition, and what the CFP is doing for its 2020 meeting in Denver.

46 Is Bacillus cereus the Next Big Thing to Worry about in the Food Industry?

FLORENCE POSTOLESIO: ADRA - UMT ACTA 01 ALTERQUIM, Quimper, France

JASINA KOVAC: The Pennsylvania State University, University Park, PA, USA

SANDRA TALLENT: U.S. Food and Drug Administration, College Park, MD, USA

Bacillus cereus was long known as the B. cereus group, which includes closely related Gram-positive, spore-forming and aerobic bacilli, widely distributed in the environment and food matrices. Beside characteristic colonies on Mold agar, these species exhibit highly divergent properties and their distinction remains challenging. Presently their classification relies mainly on distinctive phenotypic traits, such as pathogenic potential to mammals (B. anthracis, B. anthracis) or pathogenic potential to insects (B. thuringiensis), the production of B. cereus enterotoxin (B. cereus, B. thuringiensis, B. weihenstephanensis, B. weihenstephanensis), as well as colony morphology (B. pseudotuberculosis). Recently, Bacillus aestuarii, Bacillus maricopa, Bacillus glekemeri, and Bacillus variabilis have been recognized as plausible members of this group.

While food and raw material generally show low spore contamination, food poisoning outbreaks are mainly due to improper conditions of use and storage of food after cooking. Careless food handling, especially time and temperature abuse, of products such as cooked rice, sauces, and soups, and ready-to-eat products allow production of bacterial toxins associated with B. cereus foodborne illnesses.

The issue that continuously circulates the food industry and food safety officials is how to separate the bacteria that can cause food spoilage from the strains that can cause human illness.

47 Advancing the Science of Risk-based Criteria for Agricultural Water Quality

EMILY GREP: United Fresh Produce Association, Washington, DC, USA

DON STOECKEL: Rutgers University, New Brunswick, NJ, USA

DOMINIQUE WOLD: University of Arizona, Maricopa, AZ, USA

ADRIA - UMT ACTIA19 .03 ALTER'iX, Quimper, France

SANDRA TALLENT: U.S. Food and Drug Administration, College Park, MD, USA

Through FHA, Congress directed USDA to develop a Produce Safety Rule (PSR) including science-based minimum standards for the safe growing, harvesting, packing, and holding of produce. The water quality criteria in the PSR, that originate with USEPA recreational water quality criteria, have been challenged. In response, the FDA is re-evaluating the water quality requirements of the PSR and compliance dates for these regulations have been pushed back to January 2020 or 2022, respectively. The purpose of this symposium is to discuss a potential path toward a sound scientific basis for criteria governing the quality of water used in produce production that is protective of public health.

48 Determining Preventive Controls for Viruses and Parasites

SOPHIE ZUBER: Nestle Research Center, Lausanne, Switzerland

KALI KNEIL: University of Delaware, Newark, DE, USA

TIMOTHY JACKSON: Driscolls, Watsonville, CA, USA

There has been an increasing number of foodborne outbreaks and recalls associated with fresh produce produced with contaminated viruses and parasites both in the United States and worldwide. When pathogens are recognized as contaminants of a particular type of fruit or vegetable they need to be identified and characterized. The hazard analysis in a HACCP plan and addressable hazards will identify some of the possible preventive controls that could be developed for the most epidemiologically important foodborne viruses and parasites related to the fruits and vegetable sector. It will also address advances and challenges in control methods currently in use for safe produce as they relate to viruses/parasites.

Experts from the fields of food parasitology and virology will present in detail possible preventive controls for such agents and clarify any misconceptions. In addition, speakers from industry will discuss potential or already in place systems to control such emerging pathogens.
553 The Impact of Packaging Materials on Food Safety: Testing, Modeling and Regulation
LUKE ACKERMAN: U.S. Food and Drug Administration, College Park, MD, USA
MELVYN PASCAL: The Ohio State University, Columbus, OH, USA
TBD TBD: TBD, TBD, AL, USA
NAEM MADY: InterTech, Boca Raton, FL, USA
CHARLES NESLUND: Eurofins, Lancaster, PA, USA
MAEVE CUSHEN: CreameGlobal, Dublin, Ireland

Packaging materials continue to play a critical role in maintaining the quality and ensuring the safety of food products. New research and applications have challenged the traditional view of packaging that offers improved protection from oxygen, light, heat, and microbial contamination. For example, the use of nanomaterials and antimicrobial coatings are gaining wider use and acceptance because of the benefits offered.

With the advancement in materials science and engineering, the use of new chemical formulations in packaging comes a greater concern about the unintended consequences of chemicals migrating from packaging to food. This has resulted in increased regulation and testing of existing and new packaging materials.

This symposium will focus on the following topics:
- Assessment of chemical contaminants in food packages, current regulations and hot-topic compounds like BPA and PFAS
- Risk assessment for new and old food packaging materials, packaging chemicals, chemical exposure assessment, risk communication
- Testing materials in contact with food and the effectiveness of test methods for new materials
- Chemical migration modeling as guidance or an alternative to testing
- Regulatory aspects of food packaging materials and safety

Attendees will learn, from a diverse collection of experts in this field, how advances in science and technology can be used to improve the impact of packaging materials on food safety.

554 Agricultural Water and Emerging Pathogens in the Age of FSMA: Do We Need to Worry?
KALI KNIEL: University of Delaware, Newark, DE, USA
SOTRACES TRUJILLO: U.S. Food and Drug Administration, College Park, MD, USA
JENNY KROON: University of Delaware, Newark, DE, USA

The contamination of agricultural water with foodborne pathogens represents a major concern in produce safety. Several disease outbreaks have been reported worldwide which could have been linked to contaminated agricultural water used in unpacked or downstream farm operations. This symposium will provide updated information about the status of agricultural water regarding the prevalence of viruses and parasites. In addition, we will discuss how effective the current safety standards are in assuring that the agricultural water is free of such pathogens. Speakers from Federal agencies, academia and industry will participate in this symposium.

555 Environmental Monitoring – A Cost-effective Tool or Expensive Waste of Resource?
ANET WINKLER: Cogitii, Munich, Germany
ROY BETTS: Cargill, Inc., Munich, Germany
BRUCE BOURJOURLE: Commercial Food Sanitation, South Burlington, VT, USA

It is clear that Environmental Monitoring (EM) costs the industry much time and money and is widely practiced. However whilst well thought out and well designed EM systems are of great value and can be an integral part of our food safety programmes, however, EM will only be of real value if the environment we are trying to monitor is well understood. How do we get the most out of what we invest in that activity? How to set meaningful sampling strategies and criteria? How useful are the indicators? How to interpret the results? Are there adequate corrective actions defined? How can these be verified?

Over the last year’s environmental monitoring (EM) gained a lot of attention from both the industry and regulators. Many documents (e.g., published by Codex Alimentarius, FDA, GMA) underline its importance, provide general guidance and even require EM as part of Food Safety management. Over the past year, EM was used to demonstrate lack of presence or ensure EM as part of Food Safety management. However, EM is not the only part of EM practice and a broad based understanding of EM practice is required. It will also pose questions such as: How do we get the most information out of the time and money we invest in that activity? How to set meaningful sampling strategies and criteria? How useful are the indicators? How to interpret the results? Are there adequate corrective actions defined? How can these be verified?

556 Poultry Vaccines: What is Working, What are the Gaps and What is on the Horizon?
CONNIE SCHMELIK-SANDAGE: U.S. Department of Agriculture – FSIS, Washington, DC, USA
CHARLES NESLUND: Eurofins, Lancaster, PA, USA
FRANCISCO GARCÉS-VEGA: Iowa State University, Ames, IA, USA

The earliest probiotics were fermented dairy products such as yogurt and kefir. Elie Metchnikoff published observations on the longevity of Bulgarian peasants who regularly consumed these foods as long ago as 1908. Today, cultured dairy products remain a significant portion of the probiotics market, but other forms including dietary supplements and foods with added probiotic cultures have become increasingly popular: so much so that the global probiotics market in all forms is estimated at $41 bn (USD) in 2017. The market is growing rapidly with a compound annual growth rate (CAGR) of over 15% expected to remain the same over the upcoming decade. The demand for probiotics is driven by the need to maintain their excellent safety profile, as well as the need to develop unique challenges for safety and quality managers because detecting pathogens against the background probiotic population is difficult, and culture methods may not be adequate, particularly for multiple antibiotics, including fluoroquinolones and extended spectrum β-lactamases. They are present in many foods, such as poultry meat, red meat, and fresh produce. Research has connected poultry and poultry meat to disease (e.g., UTI) in humans. The objective of this symposium is to explore some of the issues and opportunities involved in protecting poultry consumers and brands.

557 Extraintestinal Pathogenic Escherichia coli (ExPEC): Urinary Tract Infections, Sepsis, and Avian Colibacillosis
JAMES JOHNSON: University of Minnesota, Minneapolis, MN, USA
LEE RILEY: University of California, Berkeley, Berkeley, CA, USA
MEHUL MELNATA: Iowa State University, Ames, IA, USA

Extraintestinal Pathogenic Escherichia coli (ExPEC) are responsible for various diseases in humans and animals. The proposed subtypes of ExPEC include avian pathogenic E. coli (APEC), uropathogenic E. coli (UPEC), sepsis-associated E. coli (SEP), and meningitis-associated E. coli. APEC is responsible for avian colibacillosis, a significant disease in poultry flocks. Community-acquired urinary tract infection caused by UPEC is one of the most common infectious diseases in the U.S. affecting approximately seven million women and costing approximately $1.1 billion dollars annually. In 2010, it is estimated that in the U.S., 33,000 deaths due to sepsis are caused by ExPEC. Meanwhile, meningitis-associated E. coli is one of the leading causes of bacterial meningitis. Sepsis is the 6th leading cause of death in the U.S. The ExPEC are often resistant to multiple antimicrobials. These antimicrobials are used to treat infections such as foodborne infections, such as campylobacteriosis, and newborns and fresh produce. Research has connected poultry and poultry meat to disease (e.g., UTI) in humans. The objective of this symposium is to review the evidence connecting food to human disease, describe the genetic characteristics of ExPEC, discuss the latest research on vaccine development, and the methods for post-harvest control of ExPEC in food and water. This symposium will be of benefit to poultry breeders, meat and poultry processors, as well as the public health community and consumers.

560 A New Paradigm: Cutting Pathogens Off at the Pass by Understanding Their Evolution Dynamics
MANNAN SHARMA: U.S. Department of Agriculture – ARS, Environmental Microbial and Food Safety Laboratory, Beltsville, MD, USA
TERESA BERGHOFF: North Dakota State University, Fargo, ND, USA
FRANCISCO GARCÉS-VEGA: Independent Consultant, Cali, Colombia

Understanding the evolution and dissemination dynamics of foodborne pathogens in food environment is critical to gain insights into the likeness of the emerging hazards of new pathogens and preventing or reducing their occurrence. This symposium will explore the history and current status of live vaccines, the vulnerability of attenuated vaccines with regard to regulatory testing, and the future of vaccines to improve the range of protection and strategies to better control pertussis of vaccine strains in birds headed for harvest.

Biofilm Removal as a Critical Part of Spillag and Pathogen Contamination Prevention
ABIGAIL SNYDER: The Ohio State University, Columbus, OH, USA
LAURENT DEHALLE: REALCO, Ottignies-Louvain-la-Neuve, Belgium
CHRISTOPHER MCNAMARA: Ocean Spray Cranberries, Inc., Lakeville-Middleboro, MA, USA

Biofilm is a community of microorganisms adhered to surfaces in a porous extracellular polymeric matrix. Each biofilm represents an ecosystem that are pathogenic and spoilage bacteria, yeasts and molds coexist and interact, increasing their resistance to sanitizers and cleaning operations. Biofilm contamination of foods and beverages can lead to spoilage and represents a food safety hazard. There is a lack of targeted strategies for efficient removal of biofilms in industrial plants. Biofilm formation in creams, corners, daod zones, valves or areas where the mixing rate is low is almost inevitable. In industrial settings, surface disinfection is usually focused on the use of biocides, aiming to inactivate the microorganisms. Since biofilms are complex biological structures adhered to a surface, these strategies often fail, as the removal of the base layer is neglected. Eliminating and preventing formation of the base layer is fundamental for controlling biofilm development. Currently, strong oxidizing agents such as chlorine, citric acid and bromide or organic biocides such as isothiazole butone, which are used, generate side effects. An alternative for biofilm detachment is the use of various physical treatments such as ultrasound, thermal shock, or mechanical treatments using jets or shear stress induced by fluid hydrodynamics. Enzymatic treatment is an emerging intervention that functions by decreasing the biofilm cohesion and destroying the physical integrity of the cell wall. It is essential to the eradication of the negative impact of biofilm on the environment. In addition, it is necessary to find solutions that promote significant detachment including the basal layer. The combination of two or more of these treatments may be necessary for the complete removal, and verification of efficient biofilm removal and prevention as well as natural compounds for the control of biofilms on food contact surfaces will be discussed in depth.

Symposium Schedule

S55 – S57

S53 – S57

S58 – S60
S61 Resurgence of Less Recognized and Presumptive Pathogens: Food Safety Implications

ALVIN LEE: Institute for Food Safety and Health, Illinois Institute of Technology, Bedford Park, IL, USA
PURNENDU VASAVADA: University of Wisconsin-River Falls, River Falls, WI, USA
BOY BETTS: Campden BRI, Chipping Campden, United Kingdom
KEITH LAMPEL: U.S. Food and Drug Administration (retired), Louisville, MD, USA

It is increasingly recognized that many otherwise commensal organisms can become pathogens under right conditions, in a right host and, if consumed in sufficient quantities. Recent outbreaks linked to Cyclospora cyttoems, Listeria innocua, Hepatitis E, and other less recognized presumptive foodborne pathogens and their food safety implications. The global public health burden of foodborne diseases is increasing, and the cost of identifying and controlling new or emerging pathogenic foodborne organisms is increasing. Risk assessment and risk management practices are needed to manage the risk. More troubling is the incidence of these pathogens in novel food sources. Speakers from academia, industry and regulatory agencies will review less recognized emerging pathogens and discuss their food safety implications.

S62 Novel and Emerging Technologies for Improving Sanitation

JULIE GOODARD: Cornell University, Ithaca, NY, USA
DALE GRINSTEAD: Diversify, Racine, WI, USA
IMA HUSSEIN: Ecolab Inc., Minneapolis, MN, USA

Novel and Emerging Technologies for Improving Sanitation: The way that cleaning and sanitation (C&S) is conducted has not changed extensively over the past 50 years. Commonly used technology, products, and practices that are in use today would be recognizable by someone who was conducting C&S programs in the 1960s or 1970s. Most cleaning is conducted with traditional methods using reactive or inactivant cleaners. While there are some differences in the specific chemistry of those products, those differences are largely variations on a theme. Similarly, the suite of antimicrobial agents used has not changed extensively for many decades. Quaternary ammonium chloride, peracetic acid, chlorine bleach, and a few others are still the “go to” agents used to control microorganisms. Although the common practice has largely stood still for more than 2 generations, science has not. There have been many technological improvements that can improve sanitation results. This session will focus on several of those improvements and how they may be used to improve sanitation practices and food safety outcomes. Technologies that will be processes are detected and assessed in practice and new technology to control biofilms including use of beneficial or harmless bacteria to out-compete harmful bacteria and synergists that can make biofilms more effective against biofilms.

LILIA SANTIAGO: University of Florida, Gainesville, FL, USA
AARON UESUGI: Merck Animal Health, Madison, NJ, USA
KELSEY SHERWIN: Ecolab Inc., Greensboro, NC, USA
DEEPAK LILLEY: Kraft Heinz Company, Glenview, IL, USA

S63 Application of Principles of Failure Mode Effects Analysis (FMEA) for Effective Verification and Implementation of Food Safety Plans

BALASUBRAMANYAM KOTTAPPA: Congra Brands, Omaha, NE, USA
渗润 LILIA SANTIAGO: University of Florida, Gainesville, FL, USA
AARON UESUGI: Merck Animal Health, Madison, NJ, USA

The Food Safety Modernization Act (FSMA) requires hazard analysis, a key component that manufacturers need to consider for effective implementation of risk-based preventive controls. Hazard analysis consists of hazard identification and hazard evaluation, both of which are essential to determine if an identified reasonable foreseeable hazard requires a preventive control. Current HACCP based systems often rely on reactive approaches to understand if food safety risks are controlled across the supply chain and hence there is a need for a powerful systematic preventive method for food safety management to verify if the preventive controls are significantly minimizing or preventing food safety hazards. Failure Modes and Effects Analysis (FMEA) is a semi-quantitative risk assessment methodology designed to identify and address all potential failure modes during manufacturing of a product or process. A FMEA risk assessment tool can be used to effectively the implementation of the preventive controls in a food safety system by prioritizing corrective actions based on the potential seriousness of the failure mode, failure severity, and the probability of detection. A FMEA methodology can be used to create a proactive sanitation plan based on knowledge of occurrence of the hazard (O), severity of the hazard (S) and possibility to detect the failure (D) before affecting the customer or consumer, ideally before consumption. A Risk Priority Number (RPN = O * S * D) is calculated and corrective actions are suggested for potential failures that have a substantial risk to reduce and eliminate the potential failures from the system. Thus, incorporation of FMEA analysis within the verification procedure of HACCP based system may be a convenient tool for better food safety assurance. The symposium will present overviews of FMEA process and practical example demonstrating the application of FMEA principles to control biological, chemical and physical hazards. We anticipate that the examples that shared will help to provide directions to comprehensively current practices among participants.

S64 Attributing Illnesses to Food Sources in the Face of Uncertainty

MARCEL ZWIETERING: Wageningen University, Wageningen, Netherlands
MICHAEL BATZ: U.S. Food and Drug Administration, Silver Spring, MD, USA
CARY CHEN PARKER: University of Florida, Gainesville, FL, USA

A current research collaboration between Health Canada, the University of Guelph and the University of North Carolina are investigating the survival and inactivation of Listeria monocytogenes, Salmonella, and foodborne viruses during the storage of low-moisture foods. This is a wide-ranging research consortium funded by the ILSI North America Food Microbiology Committee and includes a number of developing research scientists, who will present their findings. The ILSI North America Food Microbiology Committee is committed to proactively improving the understanding and control of foodborne pathogens and is presenting this symposium to help achieve that goal.

S65 Safety of Animal Source Foods in Low and Middle Income Countries

ARIE HAVERLAA: University of Florida, Gainesville, FL, USA
KEITH LAMPEL: University of North Carolina at Chapel Hill, Chapel Hill, NC, USA
WILLIAM CHANEY: University of Georgia, College of Agricultural and Environmental Sciences, College Park, GA, USA
LINNEA NEWMAN: Health Canada, Ottawa, ON, Canada
KRAFHE WAMANGITUKA: Feed the Future Innovation Labs and a program jointly sponsored by the Bill and Melinda Gates Foundation and the UK Department for International Development. This will also offer new opportunities for researchers from LMIC to present their results and expand their network at the major global food science conferences in 2019.

S66 Let’s Hear from Next Generation Food Safety Scientists on Pathogen Behavior in Ready-to-Eat Foods

NEVA NASHIER: Health Canada, Ottawa, ON, Canada
VIVIAN LY: Health Canada, Ottawa, ON, Canada

S67 Antibiotic Reduction, Alternatives, and the Relationship to Food Pathogen Outbreaks

RICHARD GRIFFITHS: UK Public Health Association, London, United Kingdom
JASON PURCELL: U.S. Food and Drug Administration, Silver Spring, MD, USA

This symposium will include a diverse and international set of speakers from academia, public health agencies, and regulatory authorities. It will include presentations on new and emerging methods of source attribution and the challenges and implications of uncertainty in sampling and attribution methods. The presentation topics will include a scoping review of the literature, structured expert elicitation, and various analyses of outbreak data. This session is being organized by the Interagency Food Safety Analytics Collaboration (FSAC), a partnership between the U.S. Centers for Disease Control and Prevention (CDC), Food and Drug Administration (FDA), and USDA Food Safety and Inspection Service (FSIS).

S68 Using Food Microbiomes

NUR HANIF: Cosmostd, Rockville, MD, USA
MENI LEDDY: Orange County Water District, Orange, CA, USA
CLAUDIO PAULIS: University of Quebec, Guelph, ON, Canada
GREGORY SIRAGUSA: Eurofins Microbiology, New Berlin, WI, USA

Microbiomes are now a mainstay across almost all fields of microbiology and becoming more so in food microbiology. From a single assay, a microbiome tells us identity and proportions of microorganisms and can identify specific genes in samples.
This symposium will present microbiome case examples and uses from the perspectives of microbiome users. It is our goal that attendees will begin to feel comfortable adapting this cutting-edge technology to answer contemporary issues in their respective industries.

S69  |  Biofilm and Low-water Activity Foods
DIANE WALKER: LSU Center for Biofilm Engineering, Baton Rouge, MT, USA
KATARZYNA LEDWOCH: Cardiff University, Cardiff, United Kingdom
KARI THORSON: General Mills, Minneapolis, MN, USA
ALYX ROBERTSON: Public Health Microbiology Laboratory, Tennessee State University, Nashville, TN, USA

Dry-surface biofilms are an increasing concern in the food industry especially related to low-moisture foods. Although biofilms have traditionally been associated with wet surfaces, research has shown that microorganisms can survive for extended periods in a desiccated state on dry surfaces. The healthcare industry has also been tackling this issue on medical equipment and environmental surfaces, indicating that bacteria can survive and then be transferred from dry surface biofilms to the hands of healthcare workers and patients as an important role in transmission. In the food industry with the current Good Manufacturing Practice, Hazard Analysis, and Risk-based Preventive Control for Food, the focus is on sanitation as a preventive control as an increasing concern. This symposium will focus on the characteristics and approaches to control of dry-surface biofilms on surfaces processing low-water activity.

S70  |  Polypropylene Permaculture? Microplastics in Terrestrial Agricultural Systems
ESPERANZA MUERTA LUNAWA: El Colegio de la Frontera Sur/Wageningen University and Research, Campeche, Mexico
SHANNON BARTLETT-HUNT: University of Nebraska-Lincoln, Omaha, NE, USA
MARION BRODHAGEN: Western Washington University, Bellingham, WA, USA

Microplastics are a well-documented source of contamination in surface water and soil, but little research has investigated the potential of microplastics to influence terrestrial food systems. The hydrophobic nature of these compounds in the environment can cause them to absorb toxic pollutants such as polybrominated diphenyl ethers, endocrine-disruptors, and pharmaceuticals and personal care products. Furthermore, biomagnification of these contaminants through terrestrial food chains may exacerbate the exposure potential to humans. However, research is only beginning to explore the consequences of their deposition in agricultural systems. Concern about environmental impacts of traditional nutrient and pest management approaches, as well as reduced availability of traditional irrigation water sources, has led to increased interest in alternative agricultural inputs. As a result, adoption of reclaimed wastewater for irrigation, biostimulants and composts for nutrient management, and biodegradable plastic row cover for weed suppression has increased. However, these inputs are potential reservoirs for microplastics. Additionally, the risk that microplastics and their associated chemical contaminants pose to consumers if present in agricultural systems is not well understood.

This symposium will bring together cutting-edge research working to address the potential of microplastics occurring in terrestrial agricultural systems and the long-term consequences of their deposition including relevance to human health. By doing so, we hope to create a platform to address and refine the questions concerning mitigation of chemical contaminant risk in the agricultural environment. The specific objectives of this symposium are to: 1) present an overview of sources of microplastics, transmission routes to terrestrial systems, and human health risks associated with exposure to this xenobiotic; 2) evaluate current research on microplastic transfer to agricultural systems via alternative inputs such as biostimulants and biodegradable plastic row cover; 3) explore current efforts to mitigate terrestrial food production systems exposure to microplastics.

S71  |  Revolutionary Diagnostic Changes are Shifting the Epidemiological Landscape and Posing Challenges for Outbreak Identification
MELISSA MILLER: Cornell University, Ithaca, NY, USA
ROBYN ATKINSON-DUNN: CDC Public Health Laboratory, Salt Lake City, UT, USA
SHANNON BARTLETT-HUNT: University of Nebraska-Lincoln, Omaha, NE, USA
DIANE WALKER: LSU Center for Biofilm Engineering, Baton Rouge, MT, USA
MICHAEL ROBERSON: LSU Center for Biofilm Engineering, Baton Rouge, MT, USA
MARC ALLARD: U.S. Food and Drug Administration, Center for Food Safety & Applied Nutrition, College Park, MD, USA

Foodborne disease outbreak surveillance plays a pivotal role in our nation’s food safety system. It has primary responsibility for identifying problems in the food supply chain that would not otherwise be recognized, limiting the extent and impact of outbreaks, and providing key information to the food industry for risk management. The primary source of data for surveillance comes from clinical diagnostic laboratories. The detection methods for foodborne pathogens in stool samples have been evolving from traditional microbiological cultivation to novel culture-independent diagnostic tests (CIDT). This trend accelerated recently with the introduction of CIDT syndromic panels that test for multiple pathogens. The shift has been driven by the demand for faster protocols that can provide physicians information to treat patients. The adoption of CIDT by clinical laboratories has increased dramatically and it is now estimated that more than 20% of all bacterial infections are tested using only a CIDT. This major technological move has multiple epidemiological implications. Without isolates, the detection of clusters and outbreaks as well as the identification of resistance and virulence trends will be notably diminished. The third important consequence of increased CIDT adoption is the difficulty of measuring disease incidence rates due to different performance compared to culture methods. The proposed symposium is intended to: 1) provide an overview of the commercially available CIDT instruments and kits, 2) describe benefits and extent of CIDT adoption by the diagnostic and public health sector, 3) report on the already observed epidemiological trends resulting from CIDT, and 4) explore a selection of possible solutions that the CDC, other public sector agencies and industries are pursuing to minimize their impact on outbreak detection and etiological agent characterization. The topic of CIDT has been largely missing from previous IAFP meetings and because of its importance, it is critical to include in 2019.

S72  |  Distribution of Foodborne Pathogens – Geographical Insight from the Use of WGS
MARTIN WIEDMANN: Cornell University, Ithaca, NY, USA
HEATHER CARLETON: Centers for Disease Control and Prevention, Atlanta, GA, USA
MARC ALLARD: U.S. Food and Drug Administration, Center for Food Safety & Applied Nutrition, College Park, MD, USA

The use of Whole Genome Sequencing (WGS) techniques for food safety is increasing around the world and a greater number of institutions are pursuing to minimize their impact on outbreak detection and etiological agent characterization. The topic of CIDT has been largely missing from previous IAFP meetings and because of its importance, it is critical to include in 2019.

S73  |  Predictive Microbiology and Risk Assessment Software Fair: Spotlight on Case Studies
YUHUAN CHEN: U.S. Food and Drug Administration – CFSAF, College Park, MD, USA
MARC TAMPLIN: Centre for Food Safety & Innovation, Tasmanian Institute of Agriculture, University of Tasmania, Hobart, TAS, Australia
JOSE FERRETER RODRIGUEZ: University of Cordoba, Cordoba, Spain

Predictive microbiology and risk assessment tools are useful to accelerate and enhance microbial food safety assessments. The application of predictive models has been growing in recent years and can be valid as rapid response tools and the approach is recognized by the Codex alimentarius, national and international regulations to assess the exposure to microbiological contaminants and support management decisions. The International Committee on Predictive Modelling in Foods (ICPM) with the support of the MMRA POG is dedicated to organize a Software Fair in the IAFP Annual Meeting in 2019 to encourage the dissemination and the use of predictive microbiology and risk assessment tools by the food industry. The symposium will be divided into two parts. In the first part (1.5h), the six software fair will be briefly presented focusing on each specific tool based on real industrial case studies including shelf-life determination, thermal processing, HACCP and formulation, sampling and full exposure or risk assessments. Then, in the second part of the symposium (1.5h), the participants will get the chance to practice using the software of their choice and their preferred case study. This live demo will take place in a dedicated and equipped space with 6 different areas: one for each software (tables for small groups and screens). Each developer will have a small group and will perform the demo on the previously presented software tool and interested participants could also try on their own laptops.
Roundtable Abstracts

RT1 Is It Time for Food Safety Performance Standards Since Zero Risk Is Not an Option?
CANDACE DOEPPER: ToxTactics, Newport, KY, USA
DONNA GARREN: American Frozen Food Institute, McLean, VA, USA
SCOTT HODD: General Mills, Golden Valley, MN, USA
ANGELA SIEMENS: Cargill, Inc., Towanda, KS, USA
CRAIG HEDBERG: University of Minnesota, School of Public Health, Minneapolis, MN, USA

Food safety systems rely on verification activities to determine if the system is working as designed and validated. Microbiological performance standards can be used to verify if a processing system is adequately controlling a specific hazard. Performance standards should be set to protect public health. Sampling protocols and microbiological testing methods must be appropriate for the food being tested. In the U.S. poultry industry, performance standards have been in place to measure the prevalence of Salmonella. Over time, the performance standards have changed to reflect the improved conditions in the industry. Prevalence-based performance standards may work for other product categories, especially in dry products of raw agricultural products such as wheat flour and the produce area, especially for frozen fruits and vegetables. This roundtable discussion will explore the current and potential future uses of performance standards in foods where it is not reasonable to expect zero presence of pathogens.

RT2 Today’s RTE Redefined – Managing Environmental Controls and the Risk of the “Reasonably Foreseeable”
SEAN LEIGHTON: Cargill, Inc., Wayzata, MN, USA
PSEYMUN FATAMI: The Acheson Group, Pleasanton, CA, USA
SCOTT HODD: General Mills, St. Paul, MN, USA
MARTIN WIEDMANN: Cornell University, Ithaca, NY, USA
ERIC BROWN: U.S. Food and Drug Administration, Center for Food Safety & Applied Nutrition, College Park, MD, USA
DAVID ACHESON: The Acheson Group, Big Fork, MT, USA

Today’s RTE includes baked, raw potato salad, emojis. If a person ate it without further processing, what is RTE? What is required of these foods? And where and how should you establish an environmental monitoring and control program to manage that risk? Today’s food manufacturers are being held to higher food safety regulatory and customer controls while, at the same time, consumer uses of foods continually expand beyond standard definitions. Even while the food industry is seeing emerging risks in pathogenic contamination, the trending consumer belief toward less processed-more healthy is taking a vast array of raw and minimally processed foods closer and closer to RTE. This ambiguity and ever-emerging food trends put the food industry at increasing risk which many don’t realize they are facing. Bringing in industry-leading regulatory, manufacturing, legal, and consultation experts, this roundtable will focus on the challenges of defining RTE and non-RTE, the regulatory expectations; how to manage an FDA “swab-a-thon” and how to conduct your own; use of whole genome sequencing (WGS); and designing a good environmental monitoring and control program, including equipment and plant design, to reduce risk.

RT3 Emerging Foods: Seaweed, Superfood, Health & Safety, Challenges & Opportunities
PATRICIA BIANCHI: Aquarium Stewardship Council, Utrecht, Netherlands
BALUNKESWAR (BALU) NAYAK: School of Food & Agriculture, University of Maine, Orono, ME, USA
WILLIAM BURKHARDT: U.S. Food and Drug Administration – Center for Food Safety and Applied Nutrition, Mobile, AL, USA
ANUSHKA CONECON: Connecticut Sea Grant and Department of Extension, University of Connecticut, Groton, CT, USA
ANDREA (TREY) ANGERA: President, Springfield Seaweed, LLC, Gouldsboro, ME, USA

Seaweed aquaculture makes up a significant portion of organic cultivated worldwide, ~28 million metric tons, with a value ~US $10 billion. In addition to other uses (i.e., hydrocolloids and feed) for cultivated seaweed, Undaria, Porphyra and laminaria are extensively used as food for direct human consumption. Seaweed foods capitalize on the trend for natural and organic products, and are a growing trend in the U.S. and internationally. China is the world’s largest producer of edible seaweed and additional production comes from Korea, Indonesia, Japan and the Philippines. There are growing businesses in more than 50 countries (FAO State of the World’s Fisheries and Aquaculture). In the U.S., collaboration within Connecticut and Maine states and industries has been the foundation for the development of the seaweed aquaculture industry. Maine leads with its cold, clear waters, and coastal infrastructure with many established companies that sell processed or fresh, wild-harvested or farmed, local, seafood through retail and direct sales.

The potential of seaweeds as sources of natural and healthful food is widely recognized and known as “superfood” with its nutrient content and free from pesticides. In comparison with land vegetables, seaweeds are potentially rich sources of polyphenolics, minerals, and carotenoids. However, there are regulatory gaps affecting safety concerns, risk assessments and guidelines. This roundtable will cover the key insights gained through research, production, and compliance for the development and safety of seaweed as an emerging superfood. Our panelists shall discuss the regulatory and supply chain challenges. They will reveal the opportunities for industry and government to further advancement in sustainability, farming and processing best practices including HACCP and other food safety protocols, the role of water quality (pre-and post-harvest), product testing for contaminants, nutrition, Best Management Practices (BNPs) and third-party certifications for market access and community development.

RT4 Cyclospora: It’s Not Just an Imports’ Issue
TRISHA ROBINSON: Minnesota Department of Health, Minneapolis, MN, USA
MICHAEL OSTERHOLM: University of Minnesota, Minneapolis, MN, USA
WALTER RAM: Giannouros Companies, Tuscon, AZ, USA
JENNIFER MCENTIRE: United Fresh, Washington, DC, USA
SAMIR ASRAR: U.S. Food and Drug Administration, College Park, MD, USA

Recurring outbreaks of Cyclospora cyclosporiasis have historically traced back to fresh produce items imported into the United States. In 2018, illnesses totaling over 2,000 resulted from consumption of produce items that were both internationally and domestically supplied. In this session, we’ll discuss the impact improved diagnostic testing has on identifying illnesses, generate hypotheses on how domestic vs. imported foods become contaminated and discuss potential hazard controls to prevent contamination of produce items.

RT5 #FoodSafety: Practical Advice for Digital Communication and Science Storytelling
AUBREY PARIS: Institute on Science for Global Policy, Princeton, NJ, USA
ADAM-YEE: My Food Job Rocks, Sacramento, CA, USA
TRACIE SEWARD: Association of Schools and Programs of Public Health, Washington, DC, USA
BENJAMIN CHAPMAN: North Carolina State University, Raleigh, NC, USA
MAY JU: U.S. Food and Drug Administration, Silver Spring, MD, USA

This roundtable will be a proactive toolkit for scientists to utilize digital tools in building trust and developing their own digital communication. (b) highlight and integrate the basics of science storytelling with food safety communication, and (c) give participants hands-on experience crafting food safety messages. The roundtable will conclude with a short activity where participants will develop food safety messages aimed at various audiences. Participants and panelists will discuss the presented messages and brainstorm dissemination methods across various digital mediums. As the conclusion of the session, participants will be provided a panel-reviewed list of “best practices” to jumpstart future endeavours in digital engagement. The goal of this roundtable is to move beyond a surface-level discussion of digital communication tools by empowering participants with the necessary information and guidance to engage in various mediums to promote an inclusive food safety dialogue.

RT6 Supply Chain Verification of a Sanitation Program
NADIA NARINE: Lumar Food Safety Ltd., Richmond Hill, ON, Canada
JESSICA JONES: Chick-fil-A, Inc., Atlanta, GA, USA
GORDON HAYBURN: Trophy Foods Inc., Mississauga, ON, Canada
RICK STOKES: Ecoblub Inc., Egmond, NL, USA
BRENDAN BROUILLETTE: Commercial Food Sanitation, South Burlington, VT, USA
EUGENE ROSEN: Rosen & Lyfe, Hoffman Estates, IL, USA

Recent outbreaks associated with restaurant operations and fresh produce have also uncovered the need for greater awareness of supplier activities, tracking and systems, agriculture-oriented hygiene, and monitoring activities. These range from the quality of water and equipment and hygienic handling in transit and storage. The Preventive Controls Rule requires a Foreign Supplier Verification Program under FSMA provide an opportunity for processors to use third party audits and documentation reviews as verams of verifying supply chain food safety. These practices are relied on heavily in the industry by many companies. Therefore, a discussion as to the best practices surrounding verification activities is warranted.

Even with all the efforts of food processors, pathogens remain one of the main reasons for recalls and, therefore, this should be a critical part of any supplier verification or supply chain program review. However, there is not much current discussion as to the best practices related to sanitation verification. A roundtable of a sanitation program can be monitoring the effectiveness of sanitation completion, cleaning after maintenance repairs, trending of data for behavioral changes in the sanitation program and using data for continual improvement of the sanitation program. However, this can be a challenge to meet when accepting documentation from suppliers, both domestic and foreign suppliers. The variability in GFSI and other second or third party schemes, combined with a wide variation in processing operations, may pose challenges in executing a supplier verification program around sanitation operations.

This roundtable will bring a range of experts to share challenges, systems issues, as well as best practices and insight on how to best manage the requirements of a sanitation program on site and through a documentation review exercise. This facilitated panel discussion will provide an opportunity to discuss key indicators to watch for, and those that can help prevent future issues and outbreaks.
We all agree that if food workers need to stay at home while sick, that is easier said than done. What incentives can you put in place to make sure the workforce doesn’t cross the barriers that prevent a worker to call in sick? It is cost prohibitive to pay employees sick pay? How do cultural norms affect behavior? How do we leverage technology to make it better for all parties involved?

The FDA requires employers to restrict employees for 48 hours after being asymptomatic for Norovirus. Current mathematical models suggest that 6.7 million illnesses could be avoided annually if both employers and employees were 100% compliant with current regulation regarding exclusions and restrictions of ill food workers. This round table will answer the central question: How do we do better job of keeping ill employees out of the operation? What does that look like in practice?

The last milk is not what it used to be. Driving to the grocery store, buying your food, and then taking it home to prepare is not your only option. Today your groceries can be delivered directly to your door, you can have a robot meet you with your groceries, you can have food delivered to another location for pick up, or you can get meal kits or specialized components delivered. And thanks to third party delivery service you can get ready to eat meals delivered from almost any restaurant. All of these different ways for consumers to get food to their homes and onto their plates raise a number of food safety and security questions. This roundtable will address these and other questions. Key discussion points will include: are there adequate temperature control of food during the delivery process? Who is responsible for ensuring food safety and proper temperature control? Does home delivery impact the shelf life of the food? Attendees will leave with a better knowledge of the risks associated with the varied forms of home delivery and gain understanding of how these risks may be mitigated.

As foods and consumers cross borders, so do the ideas of food safety and security that are intricately intertwined with cultural differences. Expert panels will discuss their experiences working with consumers; food safety and security practices and standards are also rapidly shifting and driven by FSMA requirements for preventive controls/supplier qualification, FSMA training, HACCP certifications, food entrepreneur initiatives, targeted research and dissemination of technologies, and 4-YP youth programs. While the original mission of science-based information dissemination to the public is still relevant, the food safety aspects of CES must continue to evolve and adapt in the face of multiple challenges threatening its efficacy, reach, and future.

In response to funding shifts, some states have moved toward a model supporting specialized area agents in order to remain impactful, while other states’ agents oversee various programs across a large geographic span. To increase stakeholders, CES collaboration with industry groups and state departments of agriculture has demonstrated efficacy; however, this collaboration differs across states. To improve messaging, CES has re-engineered food safety material development through movements like the extension national website and new research-based training strategies like the SafeFood for Exploring, and trainings for food service food delivery models (e.g., blogs, newsletters, social media accounts) to support a wider interest in food safety. Furthermore, while students in Food Science education are often engaged in facets of research and teaching, they may have little exposure to Extension associated opportunities. This roundtable will focus on delivering collaboration across academic, government agencies, and industry towards Extension by analyzing its infrastructure to address needs, shifting priorities of state, and funding; (b) exploring case-studies in which efforts have been reinforced to best serve the population; (c) discussing the fast-paced growth, incorporation, and influence of technology; and (d) recruiting the next generation of professionals.

This roundtable will address the value and mutual benefits of Supplier Monitoring Program from the perspective the retailer/food service organization, the supplier and the consumer. The focus will be on program design and parameters (specifications) to consider measuring and monitoring as part of a robust program which can result in defined quality and financial benefits for the manufacturer and retail/food service organization, as well as higher consumer satisfaction. The panel will provide a discussion on how we can effectively communicate food safety and security issues.

• To promote continual improvement
• Assist in the development of supplier capability and knowledge
• Create accountability for production
• Development strategic relationships with suppliers
• Assist in the development of supplier capability and knowledge
• Assist in the development of supply chain management strategies

This roundtable will address the value and mutual benefits of Supplier Monitoring Program from the perspective the retailer/food service organization, the supplier and the consumer. The focus will be on program design and parameters (specifications) to consider measuring and monitoring as part of a robust program which can result in defined quality and financial benefits for the manufacturer and retail/food service organization, as well as higher consumer satisfaction.
This roundtable will explore different approaches from around the world to modernize meat inspection systems, with a focus addressing micro-
- biological risks as part of the inspection process. The panelists will also discuss any potential inspection modernization impacts or considerations as it pertains to international trade and equivalency determinations.

RT14 The Use of Chemicals in Food Hygiene and Linkage to Microbiological Resistance

JONATHAN FRYE: U.S. Department of Agriculture-ARS-USNPRC, Athens, GA, USA
MARIA HOFFMANN: U.S. Food and Drug Administration – Center for Food Safety and Applied Nutrition, College Park, MD, USA
JOHN DONAGHY: Nestle Ltd., Vevey, Switzerland
DONNA GARREN: American Frozen Food Institute, McLean, VA, USA
LARRY KOHL: Retail Business Services LLC, an Ahold Delhaize USA Company, Salisbury, NC, USA

The presence of biocidal agents in the food chain has brought increasing concerns about the development of antimicrobial resistance and its impact on public health. Yet those agents are used for very good reasons, to protect animal health, support good food hygiene practice, and ultimately protect food safety. It can be difficult to make sense of a complex landscape where risks are real and imminent, yet data are sometimes missing to support science-based decisions. In this panel you will hear about the use of biocidal chemicals to protect public health, from regulators on the difficulty of their role as policy makers; representatives from the food industry will share their experience about how the decision in this critical topic can have serious consequences. Members of the recently run GSQ Technical Working Group on Chemicals in Food hygiene will also share the outcome of their 18-month long work program on how to ensure consumer protection through the appropriate application of sanitizers, disinfectants and cleaning agents from farm to fork, balancing risks and benefits of their use while facilitating global trade of food.


SERGIO VINAZZI: Florida Department of Agriculture and Consumer Services, Tallahassee, FL, USA
ERIC EDMUNDSON: The Acheson Group, Bisco, ID, USA
ABIGAIL SNYDER: The Ohio State University, Columbus, OH, USA
ELISABETH L. ANDREWS: University of Georgia, Athens, GA, USA
JOEL EIFFERT: Virginia Tech, Blacksburg, VA, USA
ERDODGAN CELYAN: Mérieux NutriSciences, Crete, IL, USA

USDA’s Economic Research Service reported a 180% increase in U.S. farmers’ markets between 2006 – 2014. As state and local legislation allowances of entrepreneurs have percolated into the community to satisfy this newfound demand, there has been a rise in community marketplaces in which consumers have a forum to demand more locally produced products. Cottage food laws, existing in 49 states, allow individuals to produce and sell “low-risk” foods made in their home kitchens. Striking differences in the approved low-risk food lists between states adds confusion to food safety guidelines. Differences in sale limits, license/permit requirements, training requirements, and permitted sale locations further compound the uncertainties faced by entrepreneurs, extension agents, and food safety professionals. These variations act as barriers to academic, extension, and regulatory guidance and outreach for operators and consumers, compromising food safety.

While the cottage food industry has been heralded as mutually beneficial for entrepreneurs and consumers desiring locally made products, there is a need to provide adequate food safety training to stakeholders to reduce the risk of unsafe foods entering commerce. Outreach is needed to support FSMA-exempt groups including cottage food operations, food entrepreneurs, and home food preservers. There is also a need for basic research and validation of food processing and preservation techniques with new processes and equipment. This roundtable seeks to identify, address, and discuss safety concerns pertaining to the cottage food industry by: (a) reviewing current cottage food laws and state-to-state inconsistencies; (b) discussing current best practices and successes for food safety training; (c) addressing challenges with emerging technology, food trends, family recipes, publications, and web-based resources available to consumers; (d) identifying the roles, challenges, and knowledge gaps related to cottage law, food business incubators, community kitchens for stakeholders; (e) exploring consistent messaging and training opportunities.

RT16 Has the Time Come for Complete Adoption of the Food Code?

VERONICA BRYANT: NC Department of Health & Human Services, Raleigh, NC, USA
JASON HORN: In-N-Out Burger, Baldwin Park, CA, USA
DARIN DETWILER: Northeastern University, Boston, MA, USA
GLENDIS LEWIS: U.S. Food and Drug Administration, Washington, DC, USA

The FDA Food Code is a set of guidelines of the latest scientific thought on the prevention of foodborne illness in retail and foodservice settings. Since it is guidance material and not law, states and local health departments can choose to follow these guidance materials, or not. Some states only partially adopt these materials, some states write their own rules.

Currently, there are 8 different versions of the food code being used by states that chose to do so. This makes it difficult for restaurants and grocery retailers operating stores in multiple locations to train and execute against different expectations, some of which are behind the times in the accepted science of food safety and food protection.

This roundtable will discuss the pros and cons of complete (“Universal”) adoption of the food code, the need for consistency across various states and jurisdictions, and the need for state-of-the-art requirements for protecting public health in retail and foodservice establishments. Discussion shall also focus on whether the food code should be codified into law.

RT17 Finding the Needle in the Cheese Block: How Do We Create Robust Sampling Plans for Dairy Products?

MARIAN CASTLE: New Zealand Ministry of Primary Industries, Wellington, New Zealand
MARCEL ZWIETERING: Wageningen University, Wageningen, Netherlands
MELINDA HAYMAN: U.S. Food and Drug Association, Washington, DC, USA
MARTIN WIEDMANN: Cornell University, Ithaca, NY, USA
LORALYN LEBENDA: Kraft Heinz Company, Glennville, IL, USA
TOM HANSON: Mérieux NutriSciences, Crete, IL, USA

Have you ever been asked to test your product for Listeria in a 375g sample! Are you struggling to determine what your indicator results mean? This roundtable session will discuss global perspectives from government, academia, and industry on microbiological sampling for detection of pathogens and indicators as a verification for preventive controls. The focus will be on dairy ingredients and finished products, but have applicability to other foods.

The panel will discuss:
- Criteria to consider risk-based sampling
- Target microbe(s)
- When to test, testing frequency
- Appropriate sample sizes, including composting and pooling, What do the sample sizes mean? Are more samples better?
- Statistical considerations: process control and lot acceptance
- Methods and when validation is needed
- Novel rapid methods

RT18 Building a National Integrated Food Safety System (IFSS)

STEVE MORIS: Kansas Department of Agriculture, Manhattan, KS, USA
BARBARA CASSENS: U.S. Food and Drug Administration, Alameda, CA, USA
JOHN DONAGHY: Retail Business Services LLC, an Ahold Delhaize USA Company, Salisbury, NC, USA
JERRY WOJITALA: International Food Protection Training Institute, Battle Creek, MI, USA
ERNEST JULIAN: Rhode Island Department of Health, Providence, RI, USA

FDA manages programs and initiatives that build the infrastructure and capacity of the state, local, territorial, and tribal regulatory agencies and promote a national integrated Food Safety System (IFSS). Building an IFSS is mandated by the Food Safety Modernization Act (FSMA) signed into law January 4, 2012, by President Barack Obama. The IFSS represents a seamless partnership among federal, state, local, territorial, and tribal agencies (strategic partners) to achieve the public health goal of a safer food supply. An IFSS also actively solicits input and support from stakeholders.

The seamless operation of IFSS strategic partners will:
- Plan and prioritize work to coordinate resources
- Use foodborne illness outbreak data to inform the development of evidence-based food safety policies and programs, and to criterion to evaluate their effectiveness
- Implement efficient, prevention-focused, risk-based inspections and sample collections
- Promote use of compliance and enforcement tools for achieving compliance with food safety laws and regulations
- Share data among strategic partners
- Implement appropriate, risk-based, science-based policies and practices of food safety systems and regulations
- Develop and coordinate tools to achieve food safety goals

This panel will discuss the advancements made in developing an IFSS, successful case studies of integrated efforts, and the benefits created for stakeholders.

RT19 Improving Post-mortem Inspection of Beef for Human Health Protection

BETH RIISS: The Pew Charitable Trusts, Washington, DC, USA
MARK RAMSUINEN: Iowa State Univ, Ames, IA, USA
ADAM RINER: U.S. Department of Agriculture–FSIS, Springfield, VA, USA
ANDREW POINDEXTER: APFoodIntegrity Pty Ltd, Grange, Australia

Post-mortem inspection of every single beef carcass is standard practice around the world. This inspection is for human health protection and animal health surveillance. There have been few changes in inspection practices in the last century, though changes for pig (swine) and sheep inspection have been more readily adopted. Inspectors still examine organs and viscera visually and manually for signs of infectious disease, parasitic infestations and conditions that would render parts of the animal unsuitable for human consumption. Many of the diseases and conditions being inspected are not zoonotic, and inspection may cause microbial contamination to be spread from one carcass to another. The panel will discuss how the function of post-mortem inspection can be improved to detect conditions important to public health (including implementing newer technologies), and animal disease control (using examples from swine, sheep and beef post-mortem inspection), without negatively impacting the safety of meat. Understanding and communicating the risks to all stakeholders is an essential component of the change process.
RT20 – RT22

RT20 Application of High-throughput Sequencing by Industry: Potential, Barriers and Opportunities

FABIEN ROBERT: Nestlé, Dublin, OH, USA
ROBERT BAKER: Mars Global Food Safety Center, Beijing, China
SANJAY GUMMALLA: American Frozen Food Institute, McLean, VA, USA
EMILY GRIEP: United Fresh Produce Association, Washington, DC, USA
BEHZAD IMANIAN: Illinois Institute of Technology, Institute for Food Safety and Health, Bedford Park, IL, USA
ERIC BROWN: U.S. Food and Drug Administration, Center for Food Safety & Applied Nutrition, College Park, MD, USA

A quick look at the past 15 years is enough to arrest us in astonishment at the developmental pace and transformative power of whole genome sequencing (WGS) technology. In the U.S., public health authorities and food safety regulators have adopted WGS (one of the many applications of High-Throughput or Next Generation Sequencing HTS/NGS) as a superior tool in illness outbreak investigation, well aware of its promising prospects as a phenomenal surveillance tool. Impressive and admirable efforts have also been made to harmonize/standardize/validate the interagencies laboratory and analytical methods, with many other governments following suit, laying the foundations for a truly global and all-inclusive system of surveillance and regulations to improve public health on this planet. The earlier detection of outbreaks, prevention of countless illness and more efficient implementation of food safety measures are but a few benefits of the WGS application that we have enjoyed to date with many more, unexplored and undiscovered yet.

While the private sector generally leads and/or adopts innovations associated with our food systems, their response to the HTS/NGS storm has been slow and muted at best. This roundtable will explore the major obstacles that the industry is facing to implement HTS/NGS technology to its fullest potential. In this roundtable, representatives from industry who are intimately familiar with the subject matter, will discuss their experiences in the context of the challenges and opportunities for utilizing HTS/NGS technology to its full potential. An IFSH representative and one of the top U.S. regulators who have played a key role in the implementation of the new technology in FDA, CDC and USDA FSIS will also join the industry panelists to discuss and identify industry's challenges and barriers and to explore potential solutions via immediate and long term collaborations between the industry and the government partners.

RT21 Food Safety and Trade: Colleagues or Competitors

DONALD PRATER: U.S. Food and Drug Administration, Silver Spring, MD, USA
BARBARA KOWALCYK: The Ohio State University, Columbus, OH, USA
MATT MCKNIGHT: U.S. Dairy Export Council, Arlington, VA, USA
ROGER COOK: New Zealand Ministry for Primary Industries, Wellington, New Zealand

A panel of governments, industry and other stakeholders will look at food safety and trade. Governments today use a number of tools to support these joint objectives, from Codex standards to export certificates. While the food industry can benefit from expanding markets, they also seek to protect existing markets. This panel will address questions like: What are government's legal obligations when it comes to trade? Is food safety a trade barrier? How do governments ensure food safety and still maintain open markets? Is food safety enhanced by trade, or does it create downward pressure (e.g., the lowest common denominator on food safety standards)? How does trade help food security?

RT22 Fresh-cut Processing and FSMA

JIM BRENNAN: SmartWash Solutions, LLC, Salinas, CA, USA
DREW MCDONALD: Church Brothers Produce, Salinas, CA, USA
JOHN GURRISI: Fresh Express, Inc., Orlando, FL, USA
SAMIR ASSAR: U.S. Food and Drug Administration, College Park, MD, USA
TREVOR SUSLOW: University of California-Davis, Davis, CA, USA
JENNIFER MCENTIRE: United Fresh, Washington, DC, USA

Fresh-cut processing and food safety preventive control is a priority in the industry. The Preventive Controls Rule and the Draft Fresh-cut Guidance provides an opportunity for processors to utilize a systems thinking approach for managing food safety in manufacturing fresh cut. This roundtable will bring a range of experts to share challenges, systems implementation, as well as best practices and insight on how to best manage challenges associated with fresh-cut processing. This facilitated panel discussion will provide an opportunity to discuss key components including supply chain, wash water process controls, sanitation controls, and utilizing a hazard analysis and risk-based preventive control approach to managing fresh-cut processing.
Technical Abstracts

T1-01  Lactic Acid Culture to Suppress Listeria Growth and the Decay of Minimally Processed Vegetables
Besnik Hidri1, Michael Sciberras2, Gustavo Ramirez2 and Veronique Zuliani1
1Ch. Hansen, Milwaukee, WI, 2Ch. Hansen, Victoria, Australia, 3Ch. Hansen, Mexico, Mexico, 4Ch. Hansen, Arpajon, France

Introduction: The fresh-cut greens/vegetables’ sector is answering today’s consumer demands for healthy and convenient food and less waste. For these products, shelf life is mainly defined by appearance. Adding one or more additional day(s) of shelf life provides a significant added value regarding logistics, and answers consumers’ demands for longer shelf life.

Purpose: The purpose was to evaluate if the addition of a GRAS approved lactic acid culture with anti-Listeria activity may also contribute to extending the shelf life of minimally processed vegetables.

Methods: The percentage of decay on spring mix (iceberg/romaine/radicchio/carrots), tender leaf blend, and spinach were evaluated at the end of shelf life on a control batch versus a batch sprayed with a Lactobacillus curvatus culture just before packaging. Decay was evaluated based on browning and texture (water loss/loss of crispness). Storage between 2.0 and 5.6°C was applied and atmosphere packaging mimicked suppliers’ practices (air or environment enriched with N2).

Results: For all three tested products the percentage of decay significantly decreased when the culture was applied. For spring mix kept at 4°C, the iceberg was the most sensitive. After 15 days of aerobic storage, 47% and 6% of the total leaves were damaged in the control versus the treated batch, respectively. For spinach kept at 5.6°C under air for 17 days, the decay dropped from 6% to 2% when culture was applied. For MAP packed tender leaf kept 16 days at 5.6°C, the decay dropped from 6% to 4%.

Significance: This study has demonstrated that in addition to Listeria control, GRAS approved Lactobacillus curvatus can improve the shelf life of minimally processed vegetables by delaying browning and texture change.

T1-02  Prevalence of Salmonella enterica and Listeria monocytogenes in Irrigation Waters as determined by Culture-based and Rapid Molecular Methods
Eric Handy1, Cheryl East1, Rhodel Bradshaw1, Mary Theresa Callahan2, Sarah Allard3, Shirley A. Micallef2, Shani Craighead3, Brienna Anderson-Coughlin, Samantha Gartley2, Kali Kniel1, Joseph Haymaker3, Chanele White2, Fawzy Hashemi2, Salina Parveni4, Eric May2, Hillary Craddock2, Rianna Murray2, Amy Sapkota1 and Manan Sharma1
1U.S. Department of Agriculture – ARS, Environmental Microbial and Food Safety Laboratory, Beltsville, MD, 2University of Maryland, College Park, MD, 3Maryland Institute for Applied Environmental Health, University of Maryland, School of Public Health, College Park, MD, 4University of Delaware, Newark, DE, 5University of Maryland Eastern Shore, Princess Anne, MD

Introduction: Detection of bacterial foodborne pathogens in waters used to irrigate fruits and vegetables can inform growers to quickly mitigate potential contamination of irrigation sources.

Purpose: To determine if rapid molecular (RM) methods are as accurate as culture-based (CB) methods in the detection of Salmonella enterica and Listeria monocytogenes in irrigation water and if testing larger volumes recovers S. enterica and L. monocytogenes more frequently.

Methods: Water from six surface or reclaimed sites in the Mid-Atlantic United States were surveyed by filtering three separate volumes (10 L, 1 L, and 0.1 L) through modified Moore swabs (MMS) at each of 107 sampling events from September 2016 to July 2018. For CB methods, MMS was incubated in universal pre-enrichment broth (UPB), followed by enrichment in selective broths and on selective agar for S. enterica and L. monocytogenes detection. For RM methods, DNA extracted from UPB enrichments was amplified using real-time PCR (RT-PCR) specific for S. enterica and L. monocytogenes to determine their presence/absence. For CB methods, presumptive isolates were confirmed by RT-PCR.

Results: CB detected S. enterica and L. monocytogenes in 49.5% and 29% of sampling events (n=107), respectively; RM detected S. enterica and L. monocytogenes in 72.9% and 29.9%, respectively. In 46.7% and 27.1% of sampling events, the 10 L samples contained S. enterica and L. monocytogenes, respectively, when assayed by CB; when tested by RM, 59.8% and 19.6% of 10 L samples contained S. enterica and L. monocytogenes, respectively. In 13.1% and 14.0% of sampling events analyzed by CB, all three test volumes (10 L, 1 L, and 0.1 L) contained S. enterica and L. monocytogenes, respectively. False positive rates for S. enterica and L. monocytogenes by RM were determined to be 14.3% and 3.4%, respectively.

Significance: S. enterica was detected more frequently than L. monocytogenes in the Mid-Atlantic United States, and recovery rates were dependent on the pathogen, sampling volume, and method. High false positive rates may limit adoption of the currently evaluated rapid method without modification.

T1-03  Ballpark Figures: Use of a Mathematical Model to Estimate Relative Risk from Agricultural Water to Produce in Pursuit of “Same Level of Public Health Protection” Evaluations
Don Stoeckel
Cornell University, Geneva, NY

Introduction: The Food Safety Modernization Act Produce Safety Rule (PSR) established agricultural water criteria that mirror the 2012 United States Environmental Protection Agency (USEPA) Revised Recreational Water Quality Criteria. In 2017 the Food and Drug Administration reacted to stakeholder comment with a policy of enforcement discretion during a re-evaluation period. During this reevaluation, the FDA is considering how to “further reduce the regulatory burden or increase flexibility while continuing to achieve our regulatory objectives.”

Purpose: Describe and demonstrate a relative-risk approach that enhances PSR flexibility and addresses regulatory needs for agricultural water quality.

Methods: A process-based mathematical model was created to estimate the risk of illness from consuming produce, assuming irrigation with water at the risk threshold of the USEPA criteria. The associated risk from ingestion of produce was calculated using representative food-borne pathogens (bacterial, viral, protozoan) as the source term and commodity-specific scenarios, including removal or reproduction during the passage of produce from irrigation to consumption. Resulting risk of illness was used to bin scenarios into a high, medium, and low relative risk categories. Variability was accommodated with draws in each model iteration using Monte Carlo simulation.

Results: The modeled analysis allowed evaluation of risk relative to an index scenario, such as E. coli 0157: H7 on leafy greens. Comparison scenarios addressed different commodities and different practices. Results of this analysis supported the idea that growing conditions and handling practices for some commodities, such as apples and dry bulb onions, are associated with substantially lower risk from irrigation water related to the index scenario.
Significance: This relative risk approach could be valuable when evaluating the “same level of public health protection” to support alternatives and variances. The RBA analysis uses alternative scenarios and variances leverage flexibility in the RBA calculation in defining a cost-effective or practice-specific equivalent requirements that are less burdensome and do not negatively impact consumer safety.

T1-04 Evaluation of Zero Valant Iron Filtration to Reduce Escherichia coli in Agricultural Irrigation Water in Laboratory and Field Trials

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Introduction: Water surface is a critical source of irrigation water which may contain bacterial foodborne pathogens that can produce contaminant products. Irrigation water treatment using zero-valent iron (ZVI) filtration may provide a cost-effective and environmentally safe alternative to reduce surface pathogens in irrigation water.

Purpose: To evaluate the effectiveness and longevity of ZVI and sand in a unique filter designed to reduce E. coli in irrigation water in laboratory and field trials.

Methods: 2V (50% ZVI and 50% sand) and sand (100% sand) were constructed with Schedule 40 PVC pipes (two-inch diameter). Autoclaved pond water (eight liters) was inoculated with E.coli TVS 355 (10^7 CFU/ml), and introduced to ZVI and sand filters, followed by 1:5:1:1 uninoculated pond water. The effluent was collected at 1-hour intervals for 17 days. Laboratory filtration events and two field events for irrigation trials on spinach plants with (Zvi-infused water) and control (uncontaminated water) were conducted.

Results: ZVI filtration resulted in significantly (P<0.01) greater E. coli populations in sand filter (4.01 log CFU/g) compared to ZVI filtration (3.20 log CFU/g). Prevalence of E. coli was lower in spinach plants irrigated with ZVI-filtered sand compared to sand-filtered irrigation water (control). ZVI filtration reduced E. coli populations in surface water and on irrigated spinach plants, and may aid produce growers in meeting irrigation water standards of the Food Safety Modernization Act.

T1-05 Environmental Inactivation and Irrigation-mediated Regrowth of Escherichia coli 0157:H7 on Romaine Lettuce When Inoculated in a Fecal Slurry Matrix

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Development Scientist Entrant

Purpose: Investigate the population dynamics of E. coli O157:H7 inactivation rate on romaine lettuce leaves as a function of time, fecal matrix, irrigation, and environmental factors when applied in a fecal slurry.

Methods: Two-week field trials occurred in Salinas, CA during July and October 2012. Lettuce heads grown under commercial conditions were inoculated with one of three fecal slurries: rabbit, chicken, or pig, containing an average of 4.0±0.4 CFU E. coli O157:H7 CFU/g. Lettuce samples (n=56) were collected daily and processed within 4.5 hours. The bacterial concentrations were enumerated by using the most probable number technique (Ct).

Results: The populations of E. coli O157:H7 in lettuce leaves were <0.4, <0.4, 2.55, and 4.29 log MPN/g in CPL, UN, HTPP, and PL plots, respectively. At both locations, there was a positive correlation (P<0.05) between E. coli O157:H7 populations in soil and produce samples collected at 10, 61 and 122 m from each CAFO, with four replicates per distance. Air and vegetative toxin-producing bacteria were enumerated on ChromAgar ESBL and Chromagar mSuperCarba, respectively. After three hours of incubation at 37°C, significant enterococci, Clostridium perfringens, and Enterococcus faecium populations in fresh produce in the United States, it is apparent that the transfer of foodborne pathogens between animal feeding operations (AFO) and fresh produce continues to be a considerable risk.

Purpose: The purpose of this study is to determine if the establishment of a riparian buffer zone (RBZ) by AFOs could prevent the transfer of Salmonella spp. to neighboring fields in sustainable farming systems.

Methods: A five-layer RBZ (15 by 30 m) consisting of hardwood trees, two rows of evergreen trees and shrubs, a non-manicured grass strip, and a row of crops was planted between produce fields and dairy or poultry operations. Manure, soil, plant material within the VBZ, air and produce (tomatoes and lettuce samples) were sampled for 10 months at three-week intervals. The presence of Shiga toxin-producing E. coli (STEC) and fecal indicators was determined at each sampling event. Transfer of enteric pathogens from AFOs to fresh produce for detection was determined at the following time points: 0, 30, 60, 90, 120, and 150 days after planting in the VBZ.

Results: At 0 days, ZVI filtration but were not reduced by sand filtration (-10%) over six laboratory trials. ZVI filtration removed E. coli populations in early trials one through three (mean reduction: 96%) more effectively than in trials four through six (44%). For field irrigation trials, soil samples inoculated with ZVI-filtered water had E. coli populations below the detection limit (less than two log CFU/g) and were significantly lower than E. coli on soil irrigated with sandfiltered water (2.11 log CFU/g) or control water (7.15 log CFU/g). Prevalence of E. coli was lower in spinach plants irrigated with ZVI-filtered sand compared to sand-filtered irrigation water.

Significance: ZVI filtration reduced E. coli populations in surface water and on irrigated spinach plants, and may aid produce growers in meeting irrigation water standards of the Food Safety Modernization Act.

T1-06 Pathogen Persistence and Transmission Dynamics as Influenced by Biological Soil Amendments in a Pre-Harvest Environment

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1University of Delaware, Newark, DE, 2U.S. Department of Agriculture – ARS, Environmental Microbial and Food Safety Laboratory, Beltsville, MD

Introduction: Biological soil amendments (BSA) add nutrients to soil but may harbor pathogens. BSA may influence transmission dynamics in soils and potential transfer of viral and bacterial pathogens through irrigation water.

Methods: The population dynamics of Escherichia coli in soils amended with BSAO and their transfer to cucumbers.

Results: Irrigation water was analyzed using one-way ANOVA (P<0.05). The populations of E. coli in soil were significantly lower in the treatments with BSAO compared to the control. The BSAO treatments reduced the prevalence and numbers of Salmonella spp., followed by modified FDA-AMA protocol. Data were analyzed using one-way ANOVA (P<0.05).

Conclusion: The BSAO treatments reduced the prevalence and numbers of Salmonella spp., and may aid produce growers in reducing Salmonella spp. populations in fresh produce in the United States.

T1-07 Establishment of Vegetable Buffer Zones to Reduce Transfer of Enteric Pathogens from Animal Operations to Fresh Produce

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Introduction: Transfer of enteric pathogens from confined animal feeding operations (CAFO) to adjacent produce fields continues to be a significant risk in many commercially important horticulture operations.

Purpose: To develop a fast growing and cost-effective vegetation buffer zone (VBZ) area that could prevent the transfer of enteric pathogens from CAFOs to adjacent produce fields.

Methods: A fast-growing and cost-effective VBZ area (15 by 30 m) was planted in between produce fields and dairy or poultry operations. Manure, soil, plant material within the VBZ, air and produce (tomatoes and lettuce samples) were sampled for 10 months at three-week intervals. The presence of Shiga toxin-producing E. coli (STEC) and fecal indicators was determined at each sampling event. Transfer of enteric pathogens from CAFOs to fresh produce for detection was determined at the following time points: 0, 30, 60, 90, 120, and 150 days after planting in the VBZ.

Results: At 0 days, ZVI filtration resulted in significantly (P<0.01) lower populations of E. coli O157:H7 in lettuce leaves following foliar irrigation was observed. Our findings suggest delaying harvest (>24 hours) following irrigation may reduce the potential increase of E. coli O157:H7 if present on romaine lettuce.

Significance: The effectiveness of the RBZ cannot yet be determined. However, it seems that distances and air flow do have an effect on the transmission of Salmonella.
T1-10 

Salmonella and Indicator Bacteria Profiles of Produce and Meat Products Sold in Northern California Farmers’ Markets: Implications for Microbial Food Safety

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Introduction: Small-scale farms and direct marketing of agricultural products are increasing in popularity. The potential microbial food safety risks of these direct-marketed products are not well known due to a wide range of on-farm practices, distribution, and farmers market requirements (e.g., hygiene and sanitization).

Purpose of the study was to evaluate Salmonella prevalence and microbiological profiles of produce and meat products sold at farmers markets in Northern California.

Methods: Produce (leafy greens, root vegetables, and fruits) and meat (chicken, pork, beef) were sampled from 45 certified farmers markets across nine counties in Northern California, and 12 counties in Northern California. The samples included fresh produce and meat from 76 farmers market stalls, and 24 food vendors. From the farmer or vendor, identification of mitigation strategies will evaluate both the economic interests of the producers, as well as food safety.

Results: Overall, the 2.8% of 211 meat samples were positive for Salmonella, with three positive isolates from beef (3.9%) of 105, two (2.9%) from pork from one sample, and one (2.6%) from chicken. All produce samples (n = 127) were negative for Salmonella. Forty (31.5%) of 127 of produce samples were positive for generic E. coli including two of two tomato samples, 26 (34.2%) of 76 leafy greens, and 14 (28.6%) of 49 root vegetables. The average concentration of generic E. coli from produce samples was 1.40 MPN/g (range 0-920 MPN/g).

Significance: As the number of farmers markets and direct-to-consumer marketing channels continues to increase, it is crucial to evaluate the risks associated with animal products and fresh produce sold directly from the farmer or vendor. Identification of mitigation strategies will evaluate both the economic interests of the producers, as well as food safety.

T1-11 The Whole is Greater Than the Sum of Its Parts: Building Cooperative Monitoring Programs among Farms

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Introduction: Under the Food Safety Modernization Act, the United States Food and Drug Administration established a data sharing provision allowing farmers to share water sampling data under certain circumstances: i) growers must be on a shared source, ii) sampling sites must be representative of the source, and iii) there must be no “reasonably identifiable source of likely microbiological contamination” between sampling sites. However, the provision excluded specific guidance on the scale or spatial extent that would determine a farm’s ability to use this provision.

Purpose: To evaluate the implications of the experimental conditions and levels of microbial water quality experienced within surface water systems of the western United States over several irrigation seasons. Investigating the scales at which adjacent sites share similar water quality will enable us to aid farmers interested in implementing this provision while providing feedback and data to the FDA as they continue to develop guidance materials.

Methods: Indicator bacteria (E. coli and enterococcus) were enumerated in watersamples collected from 169 sites in central Washington state and northern California during the 2016-18 irrigation seasons (n=1558). Cooperative clusters (groups of adjacent sampling sites) (n=23) were created on proximity and lack of a likely source of microbial contamination between sites. Data were analyzed within and between cluster variability. Statistical models were used to explore conditions associated with elevated microbial risks to irrigation water.

Results: Twenty-one (95%) of 23 cooperative clusters are statistically similar regardless of proximity, source type or adjacent land uses. Sampling sites for sites between were below the statistical threshold value (STV) for E. coli for FSMA agricultural water requirements; the average STV was 2.19

Significance: Based on our extensive microbial water quality survey farms can form cooperative monitoring programs which drastically reduce the regulatory burden of the FSMA agricultural water requirements.

T1-12 Development of the On-Farm Readiness View to Prepare Farmers for Produce Safety Rule Implementation

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Introduction: The purpose of this study was to evaluate the impact of the OFR operator training and obstacles faced by producers to come into compliance with the FSMA Produce Safety Rule (PSR). The research objectives were: (1) to determine farmers’ readiness to implement the PSR, (2) to gauge the impact of the training, and (3) to determine the impediments faced by producers to comply with the PSR.

Methods: 40 farmers were interviewed using a survey that evaluated implementation of practices as well as impediments and training needs required to implement the PSR. The survey used a Likert scale and was developed in consultation with experts at the University of Guelph, Canada.

Results: 40% of all farmers surveyed expressed a desire to implement the PSR. Among farmers who were interested in implementing the PSR, 60% had no problems implementing the PSR or had solutions that would allow them to do so. The most frequent impediments were time and costs (50% of farmers), followed by lack of knowledge and training (38%). The majority of farmers needed training in specific areas: 33% in post harvest handling, 33% in pre harvest handling, 32% in record keeping, and 22% in employee sanitation.

Significance: The majority of farmers are ready to implement the PSR, and this is a positive sign of the readiness of the agricultural community to address food safety issues. The results of this study help to identify areas where additional training and support is needed to ensure successful implementation of the PSR.

T2-01 Phage-like Plasmid Transfer Antibiotic and Heavy Metal Resistance Genes by Transduction, Conjugation and Ion-Pair-Induced Transformation

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Introduction: Bacteriophage-like plasmids (PLPs) are an emerging class of mobile genetic element that contain both elements of both plasmids and phages. PLPs have been shown to carry antibiotic and heavy metal resistance genes, raising questions regarding the horizontal gene transfer (HGT) mechanisms by which PLPs disseminate resistance determinants.

Purpose: PLP AnCo1 (extended spectrum β-lactamases), S1 (mercury), and MA25 (tellurite) carrying antibiotic and heavy metal resistance genes were examined to determine the ability to transfer resistance determinants to recipient cells by transduction, transposon and conjugation.

Methods: For transduction experiments, PLPs, isolated from bacterial cells using a plasmid isolation kit, were transfected to competent E. coli DH10B by electroporation at 2500 V cm⁻¹ 50–100 µF. Recipient bacteria were then plated on selective plates. For conjugation experiments, bacteria containing PLPs and a helper plasmid (donor bacteria) were incubated with recipient bacteria. Recipient bacteria containing resistance determinants were isolated by plating the bacteria on media supplemented with appropriate antibiotics and heavy metal salts.

Results: All PLPs were transferred to recipient bacteria by transduction and transposition. Two PLPs (AnCo1 and S1) were transferred to recipient cells by conjugation, but PLP MA25 could not be transferred. All PLPs conferred either antibiotic resistance (AnCo1) or heavy metal resistance (S1, MA25) to recipient cells. The MIC of AnCo1 was 3 mg/ml of cefotaxime, the MIC of PLP SJ1 was 50 µg/ml of mercury chloride, and the MIC of MA25 was 40 µg/ml of tellurite.

Significance: These results indicate that PLPs can be transferred to bacteria by all three horizontal gene transfer mechanisms, and may play a major role in dissemination of antibiotic and heavy metal resistance genes.

T2-02 Bio-based Sanitizer Delivery Systems for Improved Sanitation of Bacterial and Fungal Biofilms

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Introduction: Biofilms can persist in food processing environments due to their relatively high tolerance and resistance to antimicrobials including sanitizers.

Purpose: In this study, a bio-fermented bio-based sanitizer composition was developed to effectively target biofilm and deliver chlorine-based sanitizers to inactivate bacterial and fungal biofilms.

Methods: The bio-based composition was developed by encapsulating chlorine-binding polymer in a bio-fermented yeast cell wall particle (YCWP) microcarrier. The YCWP carrier was developed by conjugation studies on the inactivation of bacterial and fungal cells in simulated wash water and inhibition of biofilm formation was evaluated in this study.

Results: This study demonstrates the high affinity of bio-based compositions to bind target bacterial and fungal cells and inactivate five log of model pathogenic bacteria and fungi in wash water without and with high organic load (COD=2000 mg/L) in 30 s and five min, respectively. For the sanitation of biofilms, this bio-based sanitizer can inactivate seven log of pathogenic bacteria and three log of fungi after one hour of treatment, while the one-hour treatment of conventional chlorine post process wash water was only able to achieve three to four log reduction. However, for biofilms, respectively, the enhanced antimicrobial activity can be attributed to three factors: i) localized high concentration of chloride bound on the YCWPs; ii) high affinity of YCWPs to bind diverse microbes; and iii) improved stability in an organic-rich aqueous environment.

Significance: Bio-based carriers will significantly enhance the efficacy of bio-based sanitizers for biofilms, reduce persistence and transmission of antimicrobial resistance microbes, limit the use of antimicrobial chemicals, and improve the cost-effectiveness of sanitizers.

T2-03 A Novel Antimicrobial Film for Preventing Cross-contamination of Fresh Produce

Jiyoon YL, Kang Huang, Glenn Young, Yue Ma, Gang Sun and Nitin Niteni

University of California-Davis, CA

Purpose: The purpose of this study was to develop a chlorine-rechargeable film and evaluate its efficacy in preventing cross-contamination of fresh produce.

Methods: Poly(vinyl alcohol-co-ethylene) (PVA-co-PE) halamine films were prepared by a combination of PVA-co-PE powders, diallylmelamine, and diacylamine. This was acidified to form pellets and they were hot pressed to form thin films, followed by immersion in bleach for chlorination. Cross-contamination was simulated by using spinach with live E. coli O157:H7. The E. coli O157:H7 was used as a target organism and challenged three times with E. coli O157:H7 and one with Pseudomonas aeruginosa. The E. coli O157:H7 was used as a target organism and challenged three times with E. coli O157:H7 and one with Pseudomonas aeruginosa.

Results: In summary, these unique attributes of bio-based carriers will significantly enhance the sanitation efficacy for biofilms, reduce persistence and transmission of antimicrobial resistance microbes, limit the use of antimicrobial chemicals, and improve the cost-effectiveness of sanitizers.

T2-04 Technical Scientist/Engineer
T2-04 Developing Low-Cost and Recyclable Antimicrobial Coatings for Food Safety Applications

Mingyu Qiao, Randy Worobo and Minling Ma
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Introduction: Each year in the United States, more than 90% of foodborne illnesses are due to microbiological contamination. Environmental surfaces, including equipment and facilities, are important reservoirs for microbial habitation and transmission in food processing establishments. Making environmental surfaces food-safe and preventing the cross contamination of important food pathogens is of great importance for food safety and prevention. It is critical to develop an easy-to-apply coating on food-associated environmental surfaces for food safety, prevention, and control applications.

Methods: The DOHP assay was used to evaluate the antibacterial activity of extracts. The nutritional values of cortex, pulp, and seed powder of both date varieties were evaluated using standard analytical methods.

Purpose: The objective was to determine the efficacy of a commercially available bacteriophage cocktail (PhageGuard) as an intervention against E. coli O157 on refrigerated beef and vegetables.

Results: A cocktail of two selected phages, lysing 90% of E. coli O157 strains tested, showed bacterial reductions from 1.5 to 1.9 log (P<0.05) on three different strains when cold beef was treated with 3×106 PFU/cm², while 0.8 to 1.5 log (P<0.05) reductions were observed with 3×106 PFU/cm² at 24 hours post-phage application. Significant: The phage cocktail did not affect any of the controls as the bacteriophage was used by the industry as a natural, safe, and effective intervention to fight E. coli O157.

T2-07 Application of Bacteriophages on Beef and Leafy Greens as a Natural Intervention against E. coli O157

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Introduction: E. coli O157 remains a great concern for the beef and leafy greens industry. Bacteriophages have the potential to be an additional safe and effective intervention against E. coli O157.

Purpose: The objective was to determine the efficacy of a commercially available bacteriophage cocktail (PhageGuard) as an intervention against E. coli O157 on refrigerated beef and vegetables.

Results: A cocktail of two selected phages, lysing 90% of E. coli O157 strains tested, showed bacterial reductions from 1.5 to 1.9 log (P<0.05) on three different strains when cold beef was treated with 3×106 PFU/cm², while 0.8 to 1.5 log (P<0.05) reductions were observed with 3×106 PFU/cm² at 24 hours post-phage application. Significant: The phage cocktail did not affect any of the controls as the bacteriophage was used by the industry as a natural, safe, and effective intervention to fight E. coli O157.

T2-08 Nutrient Stress as a Means to Enhance Robustness in Lactobacillus plantarum B21 for Improved Food Protection

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Purpose: L. plantarum B21 was the known in the absence of glucose and/or Tween 80. These treated cells were sprayed with 10% (w/v) whey protein isolate and the microencapsulated bacteria were stored at 42 and 30° C for eight weeks. The viability of the cells (CFU) was assessed as % survivability; morphological changes were observed with electron microscopy. Inhibitory activity using well-diffusion assays and electron microscopy was observed. Environmental stress was applied to isolate and screen L. plantarum B21. The stability and reproducibility of the core-isolated strain was assessed, and the results demonstrated the enhanced robustness of this strain.

Results: Carbamate stress was found to significantly improve B21 cell stability. During an eight-week storage period, 10 g of powered cells showed a significant reduction in survival rates compared to unstressed cells (1,14±0.74% and Tween 80 treated cells). The stability of B21 cells also retained functional bacteriocin activity. The presence of Tween 80 in the growth medium resulted in filamentous rod-shaped cells, with significantly lower survival rates of 1.83% (±0.50%) compared to untreated (81.1±5.70%) and glucose-stressed (81.0±13.6%) cells after spray-drying.

Significance: The results provide detailed insight into the ideal growth conditions needed to produce robust and stable B21 microorganisms for use in food protection and industrial purposes.

T2-09 Impact of Static and Turned Pile Composting of Dairy Manure on the Persistence of Pathogenic E. coli and Translocation to Spinach Leaves

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Purpose: The Produce Safety Rule has two methods for composting biological soil amendments of animal origin. Knowledge gaps exist regarding the effectiveness of these systems in eliminating antimicrobial resistant (AMR) microorganisms.

Methods: Organic dairy manure coming from three steers was collected over a period of six months prior to composting. Feed (no antibiotics) and manure accumulation were controlled for the duration of the experiment. Composting followed produce safety rule standards. Compost, aged manure, sterile soil and a mixture of these treatments were used to grow spinach for up to 42 days in mesocosms. Spinach seed, sterile soil, aged manure, compost and spinach plants were inoculated with 3rd generation cephalosporin-resistant (CR) 0, 4 and 16 µg/ml Enterobacteriaceae, E. coli, and Shiga toxin-producing E. coli (STEC) at seven-day intervals. All samples were screened for multidrug resistance to 15 different antimicrobials.

Results: Minimum times and temperature rule requirements for composting were met. Populations of ceftriaxone-resistant Enterobacteriaceae were identified between concentrations (P<0.05). Spinach plants grown with aged manure presented the highest populations (6.9 log CFU/g) of ceftriaxone-resistant Enterobacteriaceae when compared to those grown with compost (3.75 log CFU/g). Populations of ceftriaxone-resistant coliforms were higher (one log CFU/g) than non-resistant strains (P<0.05). Ceftriaxone-resistant coliforms were recovered at higher populations on 02:15 (4.2 log CFU/g) and 02:18 (1.8 log CFU/g) on leaf tissue samples of ceftriaxone-resistant STEC populations (1.8 log CFU/g) than plants grown with compost (0.5 log CFU/g). AMR Enterobacteriaceae, coliforms and STEC were recovered from both compost treatments and spinach. Current composting methods may not reduce the transfer of these pathogens to produce.

Note: The list of authors has been rearranged for organisational purposes.
T2-10 - Effects of Origanum vulgare on Physiological Functions of Salmonella Enteritidis Sessile Cells in Ma- 
ture Biofilms

Myrella Carie Lira1, Adma Nadja Ferreira de Melo2, Enira Taysse da Cruz Almeida3, Evandro L. de Souza2, Donald W. Schaffner4 and Maria 
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Introduction: The survival of Salmonella Enteritidis in the food chain is related to its ability to form biofilms on surfaces. Origanum vulgare L essential 
oil (OEO) is suggested as a sustainable disinfectant to replace chlorine-derived agents because of its strong antimicrobial activity. However, little is known 
about the effect of Origanum vulgare L on sessile cells in mature biofilms.

Purpose: This study assesses the effects of OEO on physiological functions and counts of Salmonella Enteritidis sessile cells of mature biofilms formed 
on food-contact surfaces.

Methods: Immunosorbent coated stainless steel surfaces (2 by 2 by 0.2 cm) were immersed in brain heart infusion broth (BHI) inoculated with Salmonella Enteritidis 
(final count 6 log CFU/ml) and incubated at 37°C. After 72 h, the surfaces were washed twice with distilled water and exposed to OEO (2.4 ±1.1 nM), NaClO (250 
mg/l) or distilled water for 15 min. Afterward, coupons were again washed and submitted to ultrasound (40 kHz, 5 min). Cells were resuspended in PBS 
and labeled with the fluorescent protein, Alexafluor 568-conjugated, propidium iodide (Pi) and stained with membrane-permeable (ethidium) or membrane-non 
permeable (DiO) fluorochromes. The fluorescence intensity was measured by flow cytometry (FC), and the number of viable, non-viable, membrane-damaged, 
and membrane-intact cells were tallied.

Results: No viable sessile cells were recovered from surfaces exposed to OEO or NaClO (< less than log CFU/ml). Among the non-viable-sessile 
cells exposed to OEO or NaClO, 40% and 80% showed compromised membranes and efflux activity, respectively. Around 20% of the non-cultivable sessile 
cells exposed to OEO showed disrupted membrane, while 51% of those exposed to NaClO were depolymerized. No physiological damage related to membrane 
or efflux activity was observed in <90% of non-cultivable cells exposed to distilled water.

Significance: OEO compromised the viability of Salmonella Enteritidis sessile cells formed on stainless steel surfaces by affecting the membrane function and the efflux activity.

T2-11 - Disrupting Irreversible Bacterial Adhesion and Biofilm Formation with an Engineered Enzyme

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Introduction: Plant fatty acids, such as pelargonic acid, are potent and safe antimicrobials; however, their use by the food industry is limited because of 
inherently low miscibility in water.

Purpose: To investigate the efficiency of colonization and internalization of Salmonella into tomato plants by routes of contamination consistent with the pre-harvest environment.

Methods: Tomatoes of the variety ‘Solo’ (Solanum lycopersicum var. lycopersicum Mill.) were used. Market tomatos (Selenomonas enterica var. enterica) were grown from commercial seed and maintained 
in the greenhouse. Salmonella contamination (single or a cocktail of serovars Salmonella javiana, Montevideo, Newport, Saintpaul, and Typhimurium) was 
introduced via blossoms (3.9 log CFU/ml) or soil (4.8 log CFU/mg soil). Tomatoes were analyzed for Salmonella by enrichment in accordance with the Clinical and Laboratory Standards Institute (CLSI) protocols. Bacteria were cultured on the three selective media (Xylosol, Xylosol with Cefsulodin-Irgasan-Novobiocin (XCN) and King B agar plates) to which plates were exposed for two days at room temperature. The following multiplex PCR. Data were analyzed for the prevalence of contamination and serovar predominance in stems and fruit.

Results: Salmonella contamination led to the identification of 25 (19%) of 131 stems and 21 (16%) of 131 fruits. The predominate serovar in stems was 
Salmonella Newport and 10.9±0.05) differences between MICs of emulsions were observed based on the surfactant that was used as well as the concentration it 
was prepared with. The MICs of pelargonic acid against Enteritidis sessile cells of mature biofilms formed on food-contact surfaces.

Methods: Emulsions had a MIC of 20±9 against all three serotypes tested. The potential of an enzyme-based biocide was investigated as a supplement to common disinfection practices for preventing bacterial adhesion 
and biofilm formation with an engineered enzyme.

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and biofilm formation with an engineered enzyme.

Purpose: The potential of an enzyme-based biocide was investigated as a supplement to common disinfection practices for preventing bacterial adhesion 
and biofilm formation with an engineered enzyme.
T3-04 Prevalence and Antimicrobial Resistance of Listeria spp. from Pacific Northwest Produce Processing and Handling Environments

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Developing Scientist Entrant

Introduction: Listeria monocytogenes (LM) can survive and establish itself in diverse food processing and handling environments. Identifying niche locations and strain characteristics through routine and investigative environmental programs is of utmost importance to producers of ready-to-eat foods and raw agricultural commodities.

Purpose: Determine the prevalence and characteristics of Listeria spp. in Pacific Northwest (PNW) produce processing and handling (PPH) facilities.

Methods: A total of 920 samples were collected from 27 high-traffic entry areas (n=10 facility) were swabbed using ready-to-use sponges in six PPH facilities, followed by a “swab-a-thon” (n=101) in one PPH. Listeria spp. were isolated using a modified ISO 11290-1 method, speciated (Microgen M-Lister-02), and antibiotic-resistance typing (CTB) by disc diffusion assay using 17 antimicrobials.

Results: Listeria spp. were recovered from 15 swabs (40%; 3.8%). Isolates up to 13 from each positive sample were characterized as LM (n=2) and L. innocua (L. (n=13). Lm was frequently (71/71 swabs) seen in drains and forklift traffic areas. All isolates showed sensitivity to amikacin, ampicillin, erythromycin, gentamicin, imipenem, kanamycin, rifampin, co-trimoxazole and tetracycline. Resistance to cefotaxime, chloramphenicol (CL), tetracycline (Tc), trimethoprim (S), ciprofloxacin (CIP), sulfonamide (SD), nalidixic acid (NA), meropenem, and penicillin (P) was observed as was reduced susceptibility to CHL (15.6; 7%); Lm, CL (8; 28%); NA (18); SD (12); CIP (46%); S (46%); Tc (76%).

Significance: Our data show that areas with heavy traffic and close to outside environments are conducive to Listeria spp. hotspots. Preventive control efforts should be focused on these areas. A proportion of isolates possessed reduced susceptibility or resistance to clinically-relevant antibiotics, though none were resistant to antibiotics used in livestock treatment.

T3-05 Impact of Various Post-Harvest Water Conditions on the Performance of Peracetic Acid over Time
Amanda Kinchla1, T3-06 Prevalence and Antimicrobial Resistance of Listeria spp. from Pacific Northwest Produce Processing and Handling Environments

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Introduction: Poracetic acid (PAA) is a known antimicrobial solution that reduces the pathogenic risk of postharvest wash water, but many products in the market vary the chemical ratio of PAA and hydrogen peroxide (H2O2), which makes sourcing challenging for processors. There is a need to evaluate the accuracy and limitations of different compositions of PAA under traditional postharvest wash water processing conditions.

Purpose: This study investigated PAA’s stability and efficacy against E. coli O157:H7 upon reconstitution and their dependence on pH (1.00; 25,75, 50, 75, 125, 1000), concentration (10, 30, 60, 80 ppm), organic load (10, 100, 750, 1000°C), temperature (4, 20, 37°C) and pH (5.0, 6.5, 6.9, 7.0).

Methods: Postharvest wash water samples were inoculated with nalidixic acid-resistant E. coli O157:H7 and enumerated by plating an aliquot of the postharvest wash water on nalidixic acid-treated tryptic soy broth (TSB) at different time points (0, 24, 48, 72, 96, 120, 168, 240, 360, 480, 576 h).

Results: Sanitizers with PAA as the dominant ingredient were more efficient at reducing E. coli O157:H7 levels over time when compared to those with H2O2 as the dominant ingredient. The 50:50 H2O2/NaOCl solution was the most stable over time and had the highest efficacy against E. coli O157:H7.

Significance: Sanitizer composition, concentration, organic load, temperature and pH significantly affect the efficiency and stability of PAA systems over time, indicating that the routine measurement of the lethality of a produce wash process is difficult to impossible given the normally low pathogen load and the variability of organic matter in water. Free chlorine (Cl2) will bind to COD released by fresh-cut produce instead of binding to bacteria. Even at variable COD levels, the results show the viability of using PAA as the dominant ingredient in order to monitor and ensure that water quality and safety are maintained.

T3-06 Evaluation of Abiotic Bacterial Surrogates for Validation and Verification of One-Pass Produce Wash Systems
Laurie Clotilde1, Xiangou Nou1, Yaguang Luo2, Eric Wilhelmsen3, Adam Idoine1, Bin Zhu1, Samantha Boltin1, Ganyu Gu1 and Antonios Zoggafos2
1Scientific Technologies, Palo Alto, CA; 2U.S. Department of Agriculture-ARS, Beltsville, MD; 3ATP Consultants, Berkeley, CA; 4SafeTraces, Pleasanton, CA; 5Virginia Tech, Blacksburg, VA

Introduction: Produce wash could induce pathogen cross-contamination due to lowered efficacy of chlorine-based sanitizers at higher organic matter content (COD) in water. Free chlorine (FCl2) will bind to COD released from fresh produce instead of binding to bacteria. Even at variable COD levels, FCl2 levels must be continuously regulated at desired levels to allow better sanitation.

Purpose: Development of a mathematical model which accurately predicts cross-contamination of pathogen levels and chlorine kinetics during produce wash cycles.

Methods: Chopped iceberg lettuce was washed (five kg/run; three runs) in a tap water bath (pH ~6.5) with initial levels of 143000 CFU/g. The wash water was washed with three ppm of peracetic acid for 3 min, followed by chlorination at 1.5 ppm Cl2 for 5 min. The wash water was collected and the 1500 CFU/g rinsed bacteria reduction in red leaf lettuce. Our mathematical model accurately predicted bacteria levels in iceberg lettuce and wash water.

Significance: The model can be a useful tool to develop wash protocols for effective CVC decontamination.
**T3-10** Fate of Injured Salmonella and Escherichia coli O157:H7 on Granny Smith Apples after Cold Plasma and Organic Acid Treatment

**Methods:** A 3 x 2 factorial design was conducted where three different cold plasma treatments were applied as follows: 1) 40 s, 2) 20 s, and 3) control. Two organic acid treatments were performed, one with 0.4 mg/ml of GA, 0.4 mg/ml of VA, and one mg/ml of PCA and one with 0.3 mg/ml of GA, 0.3 mg/ml of VA, and one mg/ml of PCA. Each treatment was performed in triplicates.

**Results:** In mixed culture conditions, growth of C. jejuni was increased significantly by 37.7, 33.15, and 15.4% by pretreatment with GA, VA, and PCA, respectively. The percent injured population was calculated by the difference of the geometric means of the percent injured populations before and after treatment.

**Significance:** Bacteriocin biocontrol can offer eco-friendly means of addressing possible L. monocytogenes contamination under different preharvest and postharvest conditions in fresh produce.

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**T3-11** Influence of Bacteriophage in the Control of Stress-adapted Listeria monocytogenes Inoculated on Fresh-cut Produce

**Methods:** Inoculated on fresh-cut carrot and tomato inoculated with 10^8 CFU/ml of L. monocytogenes (acid-adapted-AA, chlorine-adapted-CA, heat-adapted-HA) in combination with the respective stressors. The bacteriophage treatment was applied after a 24 h incubation period. The bacteriophage treatment was applied after a 24 h incubation period.

**Results:** After inoculation of the apples, bacterial pathogens recovered on the apples averaged 4.6±0.18 log CFU/g and 4.8±0.14 log CFU/g for E. coli O157: H7, respectively. Cold plasma treatments for 40 s led to average 0.5-log inactivation for the pathogen and the percent injured bacteria among the survivors averaged 18±1% and 20±5% for Salmonella and Escherichia coli O157:H7, respectively. Cold plasma treatments followed immediately by dipping in an organic acid solution for five min led to 3.8-log inactivation, and the injured populations were less than one percent.

**Significance:** These results suggest that the injured pathogens on apple surfaces treated with cold plasma can be totally inactivated by immediate treatment with an organic acid solution, suggesting that this treatments procedure will enhance the microbial safety of apple deliveries designated for fresh-cut preparation.

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**T3-12** Three Steps Toward Developing a Fresh-cut Process to Meet the Food Safety Modernization Act Requirements

**Methods:** Inoculated on fresh-cut carrot and tomato inoculated with 10^8 CFU/ml of L. monocytogenes (acid-adapted-AA, chlorine-adapted-CA, heat-adapted-HA) in combination with the respective stressors. The bacteriophage treatment was applied after a 24 h incubation period. The bacteriophage treatment was applied after a 24 h incubation period.

**Results:** After inoculation of the apples, bacterial pathogens recovered on the apples averaged 4.6±0.18 log CFU/g and 4.8±0.14 log CFU/g for E. coli O157: H7, respectively. Cold plasma treatments for 40 s led to average 0.5-log inactivation for the pathogen and the percent injured bacteria among the survivors averaged 18±1% and 20±5% for Salmonella and Escherichia coli O157:H7, respectively. Cold plasma treatments followed immediately by dipping in an organic acid solution for five min led to 3.8-log inactivation, and the injured populations were less than one percent.

**Significance:** These results suggest that the injured pathogens on apple surfaces treated with cold plasma can be totally inactivated by immediate treatment with an organic acid solution, suggesting that this treatments procedure will enhance the microbial safety of apple deliveries designated for fresh-cut preparation.

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**T4-01** Antimicrobial Resistance in Retail Ground Beef with and without a “Raised without Antibiotics” Claim

**Methods:** A total of 259 retail ground beef samples were collected from different stores in Virginia, and the bacterial isolates were identified to species level using MALDI-TOF analysis. The presence of ARGs was determined by qPCR using species-specific primers.

**Results:** Retail ground beef samples collected from different stores in Virginia were found to be highly contaminated with ARGs. The presence of ARGs was confirmed by qPCR, and the results showed a wide range of ARGs in different samples. The presence of ARGs was confirmed by qPCR, and the results showed a wide range of ARGs in different samples.

**Significance:** The presence of ARGs in retail ground beef samples highlights the need for improving the antimicrobial resistance surveillance and intervention strategies in the food supply chain.

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**T4-02** Antimicrobial Effect of Major Components of Berry Phenolic Extract against Campylobacter jejuni

**Methods:** A total of 259 retail ground beef samples were collected from different stores in Virginia, and the bacterial isolates were identified to species level using MALDI-TOF analysis. The presence of ARGs was determined by qPCR using species-specific primers.

**Results:** Retail ground beef samples collected from different stores in Virginia were found to be highly contaminated with ARGs. The presence of ARGs was confirmed by qPCR, and the results showed a wide range of ARGs in different samples. The presence of ARGs was confirmed by qPCR, and the results showed a wide range of ARGs in different samples.

**Significance:** The presence of ARGs in retail ground beef samples highlights the need for improving the antimicrobial resistance surveillance and intervention strategies in the food supply chain.

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**T4-03** Isolation and Assessment of Poultry-derived Lactic Acid Bacteria for their Use as Host-specific Probiotics

**Methods:** A total of 259 retail ground beef samples were collected from different stores in Virginia, and the bacterial isolates were identified to species level using MALDI-TOF analysis. The presence of ARGs was determined by qPCR using species-specific primers.

**Results:** Retail ground beef samples collected from different stores in Virginia were found to be highly contaminated with ARGs. The presence of ARGs was confirmed by qPCR, and the results showed a wide range of ARGs in different samples. The presence of ARGs was confirmed by qPCR, and the results showed a wide range of ARGs in different samples.

**Significance:** The presence of ARGs in retail ground beef samples highlights the need for improving the antimicrobial resistance surveillance and intervention strategies in the food supply chain.
T4-04 Investigation of the In-feed Reduction of Tylosin on Antimicrobial Resistance (AMR) in Enterococci in Feedlot Cattle

Taylour Davedow, Claudia Nanzolo Bravo, Rahat Zahnoun, Haley Sanders, Argenis Rodas-Gonzalez, Cassidy Klimat, Calvin Booker, Sherry Hannum, Ana Brisas, Sherry Gow, Kim Stanford and Tim A. McClister

Introduction: Tylosin is frequently administered in-feed to prevent liver abscesses in beef cattle. There is a growing interest in lowering industry reliance on antimicrobials to address concerns on antimicrobial resistance (AMR) in enterococci. Thus, this study aimed to determine the effect of withdrawal of the in-feed tylosin administration in the first or last 25% of the feeding period on the proportion of erythromycin-resistant enterococci in beef cattle fascies.

Methods: A total of 7,500 bovine feeders were randomly assigned to one of three treatments: tylosin-in-feed for 0 (the entire feeding period), control, for both first 75%, or last 75% of the feeding period, with ten replicate pens per treatment. Fresh fecal samples from the floor of each pen were collected on days zero, 80, and 160 of the finishing period. Appropriate serial dilutions were spread-plated onto bile esculin agar (BEA) and BEA amended with eight-μg/ml erythromycin for determining the proportion of erythromycin-resistant enterococci. A PCR assay was used to isolate isolates.

Results: The proportion of erythromycin-resistant enterococci for control, first 75%, and last 75% was 27, 17, and 14% upon arrival, and 51, 30, and 21% prior to slaughter, respectively. Although the population of tylosin-resistant enterococci increased with days on feed (P < 0.01), neither the method of tylosin administration (P = 0.34) or the tylosin administration/day on feed (P = 0.34) interaction were significant. Of the 538 isolates, 97% were confirmed as enterococci, with an increase in Enterococcus from 82 to 100% between day zero and days 160 and 160 of the experiment.

Significance: Overall, the administration of tylosin-in-feed increased the proportion of erythromycin-resistant enterococci in all three experimental groups over the feeding period.

T4-05 Efficacy of Chlorhexidine Digluconate and Alkyltrimethylammonium Bromide for Carcass Decon- tamination to Ensure Food Safety

Mahger Sarker, William Long III, Basam A. Aninous and George Pao

Introduction: To prevent bacterial cross-contamination of meat is a huge challenge for meat industries. The surface of cattle meat serves as a host to several enteric pathogens during the hide removal process. The remaining hide material can be transminated by the meat to underlying muscle, thus increasing chances of food-related illnesses. To prevent cross-contamination, cattle are expected to be washed with environmentally friendly antimicrobial formulations.

Methods: Samples of freshly flayed hides (10 cm²) were collected from processing facilities and inoculated with one ml of a nine log CFU/mL of a three-strain cocktail of a single bacterial species (Salmonella enterica, Escherichia coli and Listeria monocytogenes) and allowed to sit for one-hr before spray treatment. Inoculated hide samples were individually sprayed by nine ml of different concentration of chlorhexidine digluconate and alkyltrimethylammonium bromide solutions. Salmonella samples for bacterial enumeration were gathered from hide samples at three and five days post spray treatment. The whole genome of OSYSP was sequenced using the combination of Illumina MiSeq and Ion Torrent sequencing platforms. Ion Torrent reads along with conventional PCR were used for phage genome arrangement confirmation.

Conclusion: The MICs were found to be 0.3125, 0.3125, and 0.625 for C6, C8 and C10, respectively. When the MCFAs were tested on dry fat coated kibbles, all three reduced (P < 0.05) the Salmonella load by five-fold after five hours. The results for LAE and Polymyxin B were 100% and 97%, respectively. Overall, this study illustrates the synergistic antimicrobial activity of LAE and mild physical stresses was suppressed by supplementation with antioxidants. Differences in mode of action between LAE and polymyxin B by comparing minimum inhibitory concentration, liposome model cell membrane, and oxidative stress generation. Synergistic mechanism between LAE and UV-A mild heat was also evaluated by supplementing with a variety of antioxidants and measurement of cell membrane damage by nuclear acid release.

T4-06 Synergistic Antimicrobial Activity between Physical Treatments and Lauric Arginate: Mechanisms Beyond Membrane Degradation

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Introduction: The need for more effective antimicrobials is critical for the food industry to improve food safety and reduce the installed cost of minimal processed foods. Furthermore, the emergence of bacterial resistance against commonly used sanitizers has been widely reported. The study was initiated to develop an efficient, applicable and novel antimicrobial approach which combines physical treatments (UV-A or mild heat) and generally recognized as safe lauric arginate (LAE) for industrial use.

Methods: Samples were inoculated with Salmonella enterica in dry dog food kibbles. A total of 100% of the Salmonella population was killed by UV-A or mild heat with no significant difference at 6 days post-inoculation. The combination of LAE and mild heat was also evaluated by supplementing with a variety of antioxidants and measurement of cell membrane damage by nuclear acid release.

Conclusion: Synergistic antimicrobial activity was observed in a combination of LAE and mild physical stresses (heat or UV-A). The synergistic combination resulted in a faster degradation of the bacterial cell envelope as well as an improved killing effect. Overall, this study illustrates the synergistic antimicrobial activity of LAE with mild heat and indicates a novel oxidative stress pathway that enhances the efficacy of LAE beyond membrane damage.
T4-10 Effect of D-Tryptophan on Psychrotrophic Growth of Listeria monocytogenes and Its Application in Milk.

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Introduction: Listeria monocytogenes is an important foodborne pathogen that is strongly resistant to oxygen stress and can grow at refrigeration temperatures. These two characteristics of L. monocytogenes contribute to a particular concern in refrigerated dairy foods and potential risk to public health. Currently, we have reported that exogenously added D-Tryptophan (D-Trypt) has adverse effects on the growth of some foodborne bacteria under high salt conditions. This amino acid that possesses antibacterial activity may have applications as a novel natural food additive to control the psychrotrophic growth of L. monocytogenes.

Purpose: To evaluate the inhibitory effects of D-Trypt on the psychrotrophy and osmotolerance of L. monocytogenes during long-term refrigerated stor age and further examine the feasibility of utilizing D-Trypt in controlling L. monocytogenes in pasteurized milk.

Methods: The growth of L. monocytogenes ATCC19115 on milk media was monitored by the optical density at 595 nm at 4°C during a 30 day period. We also investigated the effect of D-Trypt on psychrotolerance of L. monocytogenes at various sodium chloride (NaCl) concentrations. The viable cell counts were determined by direct plating on tryptic soy agar plates. Results: Adding exogenous D-Trypt was able to reduce and delay the psychrotrophic growth of L. monocytogenes in peptide-yeast-extract-glucose medium cultures. A higher level of D-Trypt (>30 mM) results in higher and more consistent growth inhibition of L. monocytogenes. A concentration of 40 mM D-Trypt in combination with high levels of NaCl (3% NaCl) caused a greater overall bactericidal effect with partial bacterial degradation. In pasteurized milk, 40 mM D-Trypt also significantly (P<0.05) inhibited the growth of L. monocytogenes during extended refrigeration storage.

Significance: D-Trypt appears to be an alternative or complementary strategy to control the psychrotrophic growth of L. monocytogenes during long-term refrigerated storage and thus may be appealing to the dairy industry.

T4-11 Development of Antimicrobial Hydrogel Patch to Control Vibrio parahaemolyticus in Raw Fish.

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Jian Chen
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Developing Scientist Entrant

Introduction: In raw fish consumed for sushi and sashimi, non-thermal decontamination technology needs to be applied. Hydrogels composed of edible compounds and antimicrobials should be used as non-thermal decontaminants.

Purpose: This study developed an antimicrobial hydrogel patch, composed of edible and non-toxic compounds, to reduce Vibrio parahaemolyticus cell counts on slices of raw fish.

Methods: The alginate-based hydrogel was prepared by dissolving five percent sodium alginate powder in 25 mL of distilled water, and mixing with copolymers (1% agar, 40% glycerol) and a crosslinker (CaCl2·2H2O). The hydrogel was cut into 3.0 by 3.0 cm squares, and they were placed in 10 mL of 0% and 1% natural antimicrobials (garlic seed extract and citrus extract) for two h, followed by drying at room temperature for 30 min. A 100 μl mixture (OD660 = 1) of V. parahaemolyticus strains (ATCC17802, ATCC27519, ATCC38844, and ATCC45995) was inoculated on slices of raw fish (halibut). V. parahaemolyticus was plucked from the inoculated samples and stored at 4°C for one, 20, 40, and 60 min after storage. After storage, V. parahaemolyticus cell counts in the same samples were re-determined to calculate the growth of V. parahaemolyticus. Results: V. parahaemolyticus cell counts reduced by 1.5 to 2.5 log CFU/cm2 after the hydrogel application, regardless of antimicrobials. Among the natural antimicrobials, the antimicrobial hydrogel formulated with 0.5% and 1.0% grapefruit seed extract reduced the bacteria by 2.0 and 2.3 log CFU/cm2 on slices of raw fish after 60 min of storage, respectively. Thus, the antimicrobial hydrogel composed of 1% garlic, 0.2% CaCl2·2H2O, 1% agar, and 0.5% grapefruit seed extract was the most appropriate. Significance: These results indicate that the developed hydrogel can be used to control V. parahaemolyticus on slices of raw fish by one-minute application on the surface.

T4-12 Effects of Interventions on Indicator Organism Levels in Beef Slaughter

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Introduction: Beef slaughter establishments employ many different interventions to help minimize the incidence of pathogens in their products.

Purpose: This study explored the relative effectiveness of various common interventions used by FSIS establishments as part of the Beef Veal Carcass Baseline Study (2014 to 2015).

Methods: FSIS collected data on 228,176 samples from carcasses at 177 establishments. These included 1,368 samples at post-hold release (before evisceration) and 1,368 at pre-kill (after evisceration). Samples were tested for pathogens (Salmonella and STEC) and indicators (genotypes E. coli, Enterobacteriaceae, coliform, and aerobic count) using FSIS-approved methods.

Results: Pathogen positive rates were too low to establish a direct correlation between interventions and pathogens. However, all interventions correlated with all indicators, suggesting the use of indicators as surrogates. For example, Salmonelle gave an odds ratio indicating a 57% increase in pathogen prevalence for each log increase in aerobic count. Therefore, we compared indicator levels to assess the effectiveness of interventions. Some interventions such as chlorine wash, correlated with decreased indicator levels, while others, such as trimming alone, correlated with an increase. Most comparisons gave significant trends via ANOVA (P<0.005), and many pairwise correlations were also significant (P<0.05). However, each of the tested intervention strategies generated a wide range of indicator results.

Significance: This study shows how interventions are applied as important as they are implemented. Although indicator organisms do not provide a comprehensive picture of process control, the correlation between indicators and pathogens provides useful information. Thus, our results can be used by slaughter establishments to help identify the most effective interventions for pathogen reduction.
T5-04 Technical

Introduction: Foodborne diseases are a serious health issue worldwide. However, the total burden of unsafe food is unknown. Current methods including culture-based, immunological and molecular techniques for identification and detection of foodborne pathogens have limitations, so rapid, accurate and sensitive methods are needed for food safety.

Purpose: The objective of this study was to create a multivalent aptamer system for sensitive and rapid detection of E. coli O157:H7.

Methods: First, the DNA template consisting of the complementary sequence of the aptamer and a spacer was circularized and immobilized on the gold surface of the QCM sensor. Then the target DNA was added to initiate RCA reaction and produce a very long single-stranded DNA, which contained the capture aptamer sequence. The aptamer was then used to test 100 ml water samples for the presence of E. coli O157:H7.

Results: The developed sensor had a detection limit of 10^4 CFU/ml and a detection time of 50 min, respectively, for pure culture (each test was repeated three times). Non-target bacteria (Staphylococcus aureus, E. coli) were used as controls and no signal was detected. The results indicated that the created multivalent aptamer system enhanced the sensitivity of the QCM sensor by approximately sevenfold when compared to the monovalent aptamer for the detection of E. coli O157:H7.

Significance: This study highlights a novel multivalent aptamer system to improve the sensitivity of a biosensor due to the higher binding efficiency through the repetitive aptamer sequences.

T5-05 Technical

Introduction: Enrofloxacin is used as an indicator of E. coli O157:H7 in water and is crucial for the detection of antibacterial drug residues in drinking water samples. To address this, we developed a portable LSPR biosensing system for rapid, specific and sensitive detection of enrofloxacin residue in poultry products. Localized surface plasmon resonance (LSPR) sensors have shown great potential in biodetection, due to their reproducible, label-free and real-time features.

Purpose: The objective of this project is to develop a portable LSPR biosensing system for rapid, specific and sensitive detection of enrofloxacin residue in poultry products, using polydopamine surface imprinted polymer (PDA-SIP) as a biological recognition element.

Methods: Enrofloxacin was used as a model target of fluoroquinolones. The PDA-SIP was fabricated by polymerization of dopamine and ENRO in 50 mMboric acid. The imprinted and non-imprinted sensors were tested in samples spiked with different concentrations of enrofloxacin. The sensor signal was measured using an in-house LSPR reader.

Results: The developed sensor exhibited high specificity and sensitivity towards enrofloxacin, with a detection range of 10^{-3} to 10^{-7} M, and a detection limit of 10^{-8} M.

Significance: This study demonstrates the potential of the developed LSPR biosensor for rapid and in-field detection of enrofloxacin in poultry products.
T5-10 Orthodox Inactivation of Human Norovirus Surrogates in Water
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\textbf{Developing Scientist Entrant}

\textbf{Introduction:} Shellfish and fresh-produce are major sources for human norovirus (HuNoV) infections in the US. These foods can be contaminated following HuNoV-contaminated water consumed in shellfish farms or in pre-potato harvest processes of fresh produce. Decontamination of waters used in depuration process of oysters and washing of fresh produce is one of the suggested methods to mitigate this risk. Photodynamic treatment (PDT) using photostabilizers (PS) is extensively used for bacterial inactivation but studies on the use of PDT for viral inactivation in food and water are scarce. Purpose: To study the inactivation of HuNoV surrogates, In, in water, by exposure to light in the presence of photostabilizer dyes. Methods: In triplicate experiments for each treatment, cell-free calicivirus (FCV) and Turuline virus (TuV) were mixed separately in water containing various concentrations of rose Bengal (RB), Phiborine (PB) and chlorophyll (CH) as photostabilizers. Then, they were exposed to LED light source for different times. -Light% was measured using photostabilizer concentration required for 4-log virus reduction were measured by titration of surviving virus after PDT. Results: Exposure to blue LED in the presence of PB and CH resulted in inactivation of FOV and TuV. Chlorophyll was not effective. A 4-log reduction in the titers of FOV and TuV was seen at 33m-LT275µM RB and 10m-LT50µM PB, respectively. Similar effect was achieved using 5m-LT70µM PB and 5m-LT50µM PB, respectively. These results indicate that PB is the most effective dye and that TuV is more resistant to PDT than FCV. Significance: Our results revealed for the first time that PDT using Pb, an edible food color, is a promising non-thermal treatment for HuNoV inactivation in water. This cost-effective PDT holds the promise for integration with water used for depuration of oysters and for washing of fresh produce for the risk ofHuNoV.

T5-11 Developing the Antibiotic Resistance Mechanism of Campylobacter Using Confocal Micro-Raman Spectroscopy
Luoya Ma and Xiaolan Liu
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\textbf{Introduction:} Campylobacter jejuni is a major human pathogen that causes significant economic losses in the livestock and poultry industries due to its antibiotic resistance. The resistance mechanisms vary significantly among different strains and widespread resistance has been observed in clinical isolates. Hence, the development of new diagnostic tools and treatments that can monitor and predict antibiotic resistance is necessary.

\textbf{Methods:} Campylobacter jejuni was grown onblood agar plates, harvested, and resuspended in PBS. The bacterial concentration was adjusted to 1.0×10^8 CFU/ml. 100 µl of 100 µg/ml Raman dye (Ph-B or TuV) was added to 500 µl of bacterial suspension. The samples were incubated at 37°C for 1 h, then 500 µl of PBS was added and the samples were incubated for further 1 h. An 800-nm Raman spectrometer was used to collect the Raman spectra of the bacteria. Results: The Raman spectra were collected from 10 different samples of resistant strains and 10 different samples of susceptible strains of C. jejuni. The relative intensity and peak positions of the Raman bands were compared between the resistant and susceptible strains. The Raman bands at 962 cm^-1, 1030 cm^-1, and 1220 cm^-1 were significantly different between the resistant and susceptible strains. The results indicated that the resistance mechanisms of the resistant strains are different from those of the susceptible strains. Conclusion: The Raman spectroscopy can be a useful tool for monitoring and predicting antibiotic resistance in Campylobacter jejuni.
Introduction: Previous studies conducted on chia seeds treated with a commercial peracetic acid-based sanitizing solution found that Enterococcus fecium NRRL B-2354 was a suitable surrogate for Salmonella and it greater than three log CFU reductions for both Salmonella and E. fecium were achieved while maintaining seed viability (germination, rancidity and nutrition). Identification of a surrogate and promising efficacy results warranted further optimization testing with the final objective of validating industrial-scale equipment.

Methods: The objective was to develop an industrial-scale applicator, used to treat chia seeds with the sanitizing solution, and an industrial-scale dryer, used to dry post-treated seeds.

Methods: Samples (one kg or 120 g) of chia seed were inoculated with 30 mL of E. fecium (3.6 liters on 120 g) and treated from a distance of 1 cm to 25 cm from the top and a rubber gasket was installed at this level. Following the post-treatment holding time, the inoculated seeds were placed in the dryer for 24 h at 20°C, and 21°C for the initial and final干燥 experiments, respectively. The inoculated and treated seeds were compared to an average of 6.5±0.3 log CFU reduction, producing a six-log reduction in Salmonella.

Methods: The objective was to determine whether ybgC regulates the resistance of E. coli Enteritidis to lysozyme (a major antibacterial component of egg white) by survival ability test and cell membrane characterization.

Methods: While previous work demonstrated that endospore-level UV-C resistance in E. coli requires the further isolation of a single phage. Finally, KFS-BC1 was then propagated due to great lytic activity and the final concentration of KFS-BC1 was determined by plaque assay. In Young Choi

Methods: A correlative approach combining genomic and phenotypic microarray (PM) data was employed to identify metabolic differences between PM inoculated and non-inoculated cultures in E. coli BLM-05.

Methods: The three strains were inoculated into bovine milk, and incubated at 37°C for one hour. The milk samples were then centrifuged (3000 x g, 10 min at 4°C) and the supernatants were used in the neutralization assay of the lysozyme.

Methods: A six-log reduction was observed in ybgC mutant after incubation for 24 h in filtrate-lysozyme, whereas no significant (P<0.05) difference was found among five or six control treatments. The results indicated that the lysozyme was neutralized by the ybgC mutation.

Methods: A six-log reduction was observed in ybgC mutant after incubation for 24 h in filtrate-lysozyme, whereas no significant (P<0.05) difference was found among five or six control treatments. The results indicated that the lysozyme was neutralized by the ybgC mutation.

Methods: The metabolic operon likely led to increased sensitivity of E. coli against bromo-succinic acid and increased sensitivity to osmolytes including four or five percent sodium sulfate and 200 mM sodium phosphate. Genomic analysis revealed mutations in the ybgC gene (encoding for acyl-CoA thioesterase) was crucial for the outer membrane permeability and apparent alterations in cellular morphology were found in ybgC mutant after exposure to lysozyme.

Methods: A phage with high specificity and high stability under various conditions was isolated from soil and selected for its ability to control the survival of E. coli O157:H7 during storage.

Methods: Bacteriophage insensitive control agent.

Methods: The six strains were inoculated into bovine milk, and incubated at 37°C for one hour. The milk samples were then centrifuged (3000 x g, 10 min at 4°C) and the supernatants were used in the neutralization assay of the lysozyme.

Methods: A correlative approach combining genomic and phenotypic microarray (PM) data was employed to identify metabolic differences between PM inoculated and non-inoculated cultures in E. coli BLM-05.

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Methods: A phage with high specificity and high stability under various conditions was isolated from soil and selected for its ability to control the survival of E. coli O157:H7 during storage.

Methods: The industrial-scale system was capable of an average of greater than five-log CFU/g reduction of E. coli O157:H7 growth in fresh cheese during refrigerated storage.

Methods: The six strains were inoculated into bovine milk, and incubated at 37°C for one hour. The milk samples were then centrifuged (3000 x g, 10 min at 4°C) and the supernatants were used in the neutralization assay of the lysozyme.

Methods: A six-log reduction was observed in ybgC mutant after incubation for 24 h in filtrate-lysozyme, whereas no significant (P<0.05) difference was found among five or six control treatments. The results indicated that the lysozyme was neutralized by the ybgC mutation.

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Methods: While previous work demonstrated that endospore-level UV-C resistance in E. coli requires the further isolation of a single phage. Finally, KFS-BC1 was then propagated due to great lytic activity and the final concentration of KFS-BC1 was determined by plaque assay. In Young Choi

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Methods: A phage with high specificity and high stability under various conditions was isolated from soil and selected for its ability to control the survival of E. coli O157:H7 during storage.
T6-10  Salmonella Serotype Fitness in Various Water Types and Habitat Transition from Water to Tomato Fruit

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Methods: Filter-sterilized non-tidal river, tidal river, pond and reclaimed water samples were singly inoculated with environmental isolates of Salmonella Heidelberg, Typhimurium, and Newport and assayed at days one, 30 and 60 for persistence. Sample aliquots were directly plated on tryptic soy agar for culture-dependent enumeration. Aliquots were also treated with 25 µM propidium monoazide (PMA) followed by total DNA extraction and quantitative PCR of the rRNA polymerase α-factor gene for culture-independent measurements. To assess the transfer capacity of Salmonella, various strains were incubated in different water types for 30 days before inoculation onto tomato fruit cv. ‘Heinz-1706’ and subsequent retrieval 14 hours later.

Results: Salmonella counts of all serotypes declined in all water types over 60 days. Recovered counts showed the highest decline in non-tidal fresh water with the most decline (P<0.05). Over 60 days nondigital water samples displayed significantly different rates of decline between PCR and direct plating methods (P<0.05). The number of days in water impacted Salmonella transfer to tomatoes. Pond water supported less transfer compared to other water types (P<0.05) across all serotypes tested. Incubation time in water significantly influenced transfer in one non-tidal fresh water (P=0.50). Salmonella Heidelberg exhibited the fastest transfer from water to tomatoes regardless of water type or incubation time.

Significance: Persisting serotypes in surface and reclaimed water varied by serotype. Serotypes differ exist in capacity for habitat transition from water to tomato. These data may be used to assess the safety and adequacy of irrigation water sources.

T6-11 Evaluation of a Typing Scheme Based on Deep Amplicon Sequencing to Aid Epidemiological Linkage of Cyclosporiasis Cases

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Purpose: To investigate the ability of Cyclospora cayetanensis to persist in water and transfer to tomato fruit.

Methods: DNA was extracted from 100 isolates of C. cayetanensis from outbreak water samples collected between 2015-2017, inoculated onto tomato fruit cv. ‘Heinz-1706’ and subsequently retrieved 14 hours later.

Results: Seventeen isolates were detectable in the tomato fruit.

Significance: This study demonstrates the potential of deep amplicon sequencing to aid in the rapid and accurate diagnosis of Cyclospora cayetanensis outbreaks.

T6-12 Safety Status of Some Traditionally Fermented Foods in Nigeria

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Methods: Traditional fermented foods from cereals and legumes contribute significantly to the energy and protein demand of many households in Nigeria but their production is still a household art which in most cases compromise their safety.

Results: We observed high level of contamination of some of these fermented foods and beverages.

Significance: There is a need for effective intervention and regulation to ameliorate this phenomenon.

T7-01 Validation of Abiotic Bacterial Surrogates for Surface Sanitization in Food Processing Facilities

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SoftTraces, Pleasanton, CA; 2: Scientiﬁc Technologies, Palo Alto, CA

Purpose: This study validated abiotic bacterial surrogates (SaniTracers) for the verification of processing plant sanitation in real-world applications.

Methods: SaniTracer surrogates were manufactured by encapsulating short, naturally occurring DNA fragments from a diverse collection of both within-food-grade material particles. The particles were designed to match the response of bacteria to chlorine-based sanitizers. Experiments were conducted in a protein-processing pilot plant, where multiple locations were inoculated with the surrogates and non-pathogenic E. coli, and an active tree fruit packing plant of the same facility. The recovery of the surrogates to the sanitation process was consistent with that of E. coli in the protein plant, and environmental bacteria in the fruit packing plant. Bacteria were measured using traditional microbiological methods, and the surrogates were quantified using qPCR. To assess the transfer capacity of the surrogates, experiments were conducted in two facilities, one which had an active tree fruit packing plant and the other an active protein processing plant.

Results: In the protein processing pilot plant, the surrogates showed an average log reduction of 4.3 and SD=1.3. The E. coli showed an average log reduction of 4.32 and SD=1.48, however, the E. coli post-sanitation recovery was low. In the fruit packing plant, the APC log reduction was significantly less when the surrogates indicated that the cleaning was ineffective. The surrogates were detected and quantified in approximately 60 minutes, while the bacterial tests took more than 24 hours.

Significance: The behavior of these abiotic bacterial surrogates under sanitation in real-world applications can predict the lethality of the sanitation process on contaminating bacteria. The surrogates are a sanitation verification tool that is specific, rapid and overcomes sampling errors.

T7-02 Synergistic Effects of Ultrasound and Natural Antimicrobials Against Listeria innocua and Eschechella richii K121

Hongxiao Zhang and Rohan Tikekar
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Methods: Fresh produce washed with chlorine-based sanitizers is often not effectively disinfected, causing serious foodborne disease outbreaks. Chlorine is not stable in water and it can form carcinogenic by-products by reacting with organic matter.

Purpose: The objective of this study is to investigate the potential of using ultrasound and food-grade natural antimicrobials to improve fresh produce sanitation processes.

Methods: Inactivation capacities of a 20 kHz ultrasound probe device (US20) or a one MHz ultrasound therapy device (US1) combined with nine different natural compounds were tested against Listeria innocua and E. coli K12. Text experiments were in the stationary phase at 10^8 to 10^10 CFU/mL. The natural compounds were two to three times more effective than the synthetic compounds. Experiments were run at a power density of 200 W/2.22 L. After each treatment, the bacteria were enumerated on tryptic soy agar and compared to control samples treated by ultrasound alone or antimicrobials alone. Intracellular oxidative stress, intracellular pH, and morphology of treated bacteria were also measured by chemical assays and transmission electron microscopy. All data were expressed as average value obtained from triplicate experiments.

Results: Results showed that both ultrasound treatments alone did not reduce Listeria (<0.05). However, the 20 kHz probe combined with carvacrol (200 ppm), clove (10 ppm) or cumin (5 ppm) resulted in an additional 2.5 to 3.0 log reduction of L. innocua in 15 min compared to the reduction from the 20 kHz probe with any of these antimicrobials alone. Similarly, the one kHz probe with carvacrol (200 ppm) induced an additional one-log reduction. No synergistic effect was observed when any of the natural compounds was combined with ultrasound. For example, for carvacrol, relative fluorescent increase from 0.85±0.08 to 2.02±0.47 (P<0.05), intracellular pH decreased from 7.1 to 4.8 (P<0.05), and there was no damage to the cell wall and membrane structures after treatments.

Significance: This research demonstrates that combination in natural antimicrobial compounds is a potential safe and effective alternative to reduce microbial food contamination during fresh produce washing.

T7-03 Inactivation of Listeria and E. coli Using UV-C LED: Effect of Substrate on Inactivation Kinetics

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Introduction: Environmental pathogens such as Listeria monocytogenes and E. coli are a leading cause of foodborne illness, with dire economic and public health consequences. UV-LCDs were recently developed as a chemically-free, enzyme-efficient aptamer to mitigate contamination by pathogens, but their effectiveness has not been fully investigated.

Methods: We investigated L. monocytogenes CF-20008 and E. coli ATCC 25922 were used as nonpathogenic surrogates for L. monocytogenes and E. coli O157:H7, respectively. Early phase studies showed that 20 kHz streaked on tryptic soy agar; 3 hour incubation in liquid films (0.5 mm to 2.4 mm thick) and 3 hours incubation on stainless steel surfaces, with or without air drying, to mimic various scenarios encountered in food handling environments. These substrates were exposed to a continuous 20 kHz ultrasound probe (US1) combined with nine different natural compounds. UV-LCDs were used to inactivate these bacteria. For example, for carvacrol, relative fluorescent increase from 0.85±0.08 to 2.02±0.47 (P<0.05), intracellular pH decreased from 7.1 to 4.8 (P<0.05), and there was no damage to the cell wall and membrane structures after treatments.

Significance: This investigation of Listeria and E. coli using UV-C LED: Effect of Substrate on Inactivation Kinetics demonstrates the potential of using UV-C LED technology to improve food safety.
Introduction: DNA extraction facilitates microbial persistence in food processing environments and increases the likelihood of product flow contamination.

Methods: The tube was incubated at 35°C for 14 h, and one-ml aliquots of the cultures were used to extract the DNA for the identification and the serotyping of the isolates. The amplified DNA products were electrophoresed and visualized under UV light.

Results: The commercial quaternary ammonium compound sanitizer at 50 ppm (v/v) produced a 4.2±0.5 log10 CFU/coupon. Biofilms presented flat, microcolony architecture with biovolume and maximum thickness values also increased to 36.3±2.5×105 mm3 and 41.0±9.8 mm, respectively.

Conclusion: Biofilm resistance to sanitizer is an important concern in food-processing environments. These results were consistent with the sensory analysis.

T7-05 Development of an autonomous, disposable, portable device for the identification of foodborne pathogens

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Developing Scientist Entrant

Introduction: The food-processing environment, bacteria are able to remain on surfaces after sanitation procedures. These surface residential bacteria are a major source of product contamination and spoilage and play an essential role in food safety and quality. In order to manage food safety in the food-processing environment, bacteria can be classified into four main groups: spoilage bacteria, pathogenic bacteria, probiotic bacteria, and lactic acid bacteria. The residential bacterial surface community was identified and characterized as homogeneously spread within the processing plant. This “house microbiota” was mainly composed of spoilage bacteria such as Brochothrix thermosolvens, Carnobacterium maltaromaticum, Serratia liquefaciens, Psychrobacter frateurii, and many different genera that are unable to decompose lipids and nucleic acid analysis allowed to highlight environmental bacterial source hotspots and to identify contamination routes. These results were consistent with the sensory analysis.

Conclusions: Microbiota analysis and the use of polyphasic approaches in a complex ecosystem such as a food processing plant could be useful to characterize microbial reservoirs, improve targeted hygiene procedures, and lead to a better product quality all along the shelf life.

T7-06 Isolation and Serotyping of Vibrio vulnificus and Vibrio cholerae in Seawater in South Korea

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Developing Scientist Entrant

Introduction: Vibrio species are abundant in seawater and pose a risk to human health. Shellfish and mollusks have potential to accumulate Vibrio species in their gills and mantle, respectively. This study aimed at enumerating and characterizing Vibrio species in gills and mantle and of raw seafood in a few small-scale raw seafood processing facilities.

Methods: Ninety-six seafood samples (66 shellfish samples and 30 mollusk samples) were collected from seafood markets in South Korea from July to December in 2018. To use the most probable number (MPN) method, 10, 1, and 0.1 ml of sample were inoculated in 10 ml alkaline peptone water (APW). The tubes were incubated at 35°C for 14 h, and one-ml aliquots of the cultures were used to extract the DNA for the identification and the serotyping by PCR. The amplified DNA products were electrophoresed and visualized under UV light.

Results: Five vibrios were detected in six samples (6.3%), and V. cholerae was detected in two samples (2%). The highest prevalence month of V. vulnificus contamination was September (6.3%), and the highest contamination level was 5.69 MPN/g detected from cuttlefish and salted oyster. V. cholerae was detected only in November from date mussel and common oyster clam, and both samples showed the same contamination level (56 MPN/g). The V. cholerae isolates were serotyped, and the results showed that all isolates were negative for both V. cholerae O1 and V. cholerae O139.

Conclusions: This result indicates that Vibrio vulnificus and V. cholerae can cause both shellfish and mollusk diseases and the prevalence of the highest month was September in S. Korea.
The study aimed to assess the population of viable bacteria and profile the microbiome and resistome in various types of retail cow’s milk sold
in California, Davis, CA, 4University of California-Davis, Davis, CA, 3Western Center for Food Safety, University of California-Davis, Davis, CA, 2Western Center for Food Safety, University of California-Davis, Davis, CA, 1University of California Davis, Davis, CA, 2Western Center for Food Safety, University of California-Davis, Davis, CA

Methods: A total of 1,920 milk samples were collected from eight milk brands including ultra-pasteurized milk (n=2), HTST-pasteurized milk (n=3), bulk pasteurized milk (n=1) and raw milk (n=2). Sampling occurred between March and August 2017 at Davis, CA through eight independent purchases for all brands of milk. After each purchase, samples were aliquoted into three tubes and incubated for zero, two, four, six, 12, and 24 h at both 4°C and 24°C. All milk samples were inoculated with coliforms, lactococci, and psychrotrophic bacteria using standardized methods. Concomitantly, DNA was extracted from all samples after 16S rRNA sequencing. In addition, 24 milk DNA samples, which includes both raw and HTST milk before and after 24 h incubation at 23°C were sequenced using Illumina NextSeq.

Results: Different types of milk possess distinct microbiome profiles (P<0.04), and raw milk has significantly more viable bacteria other than retail milk (P<0.05). Remarkably, the raw milk microbiota was dominated with Bacillus cereus and Enterobacteriaceae with minimal to no detection of probiotic genera. Raw milk contained 1208 differentially expressed genes (DEGs) than pasteurized milk (P<0.001). Specifically, 138 individual ARGs conferred resistance to 11 classes of antibiotics were observed in raw milk compared with 25 ARGs found in HTST milk. Incubation at 25°C drives the bloom of viable bacteria in milk which also significantly enriches the population of ARGs (P<0.001).

Significance: Raw milk hosts more antibiotic-resistant genes (ARGs) than pasteurized milk, and incubation of raw milk at 23°C dramatically increases such risk.

T17-11 Transcriptional Sequencing of Listeria monocytogenes during Co-Cultivation with Cheese Rind Bacteria
Justin Anast and Stephan Schmitz-Esser
loween Breyer

Purpose: We aimed to uncover what genes are expressed in the transcriptional response of the L. monocytogenes sequence type (ST) T7-12 strain 6179 in co-culture with common cheese rind bacteria.

Methods: L. monocytogenes 6179 was cultured in brain heart infusion broth or plates with either gram-negative (Psychrobacter LRT) or gram-positive (Brevibacterium sp.) rind bacteria. L. monocytogenes was cultured for 24 hours, RNA was isolated after two and 12, or 24 and 72 h of co-cultivation. RNA samples (n=4) in duplicate for each strain and condition) were sequenced using Illumina NextSeq. Mapping of reads to the 6179 genomes was conducted using BWA. DESeq2 was used to identify differentially expressed genes in L. monocytogenes 6179 comparing co-cultivation to L. monocytogenes 6179 mono-culture. For this, p-values lower than 0.05 were considered significant.

Results: Transcriptome sequencing resulted in 4.1 to 12.2 million reads per sample. For 12 h co-cultivation in the broth of L. monocytogenes 6197 with Brevibacterium and Psychrobacter, 387 and 597 differentially expressed genes were identified with log twofold changes up to 6.2. After 72 h co-cultivation on plates, 190 and 496 genes were considered significant. L. monocytogenes 6179 differentially expressed genes were identified with up to 8.4 log twofold changes. Significantly upregulated genes included those involved in the biosynthesis of coelomate (p<0.05) and the utilization of ethanolethane (p<0.05) and propanol (p<0.05). Interestingly, 78 genes of three highly conserved L. monocytogenes 6179 prophages were significantly upregulated in co-culture with Psychrobacter (but not with Brevibacterium) after 72 hours.

Significance: Uncovering genes involved in the competitive shift may contribute to the development of additional targets against L. monocytogenes in foods and food production environments.

T17-12 Using Machine Learning to Predict Pasteurized Fluid Milk Spillage Based on Quality Management Practices
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Purpose: The aim of this study was to develop a tool for predicting facultative contamination with spoilage bacteria, resulting in a reduced shelf life. Of particular concern, psychrotolerant Gram-negative bacteria that enter the product post-pasteurization can lead to bacterial levels above 20,000 CFU/ml within seven to 10 days of processing.

Methods: The study included 364 milk samples of nonpasteurized milk and 1,080 milk samples of pasteurized milk. A random model of 15%, 25%, 35%, 45%, and 50% was trained using the 20% of the data. The prediction model was to predict the type of spoilage: Gram-positive (Bacillus cereus) or Gram-negative (psychrotrophic bacteria). The performance of the prediction model was evaluated by the area under the curve (AUC). All models were trained using the same dataset.

Results: Among all milk samples tested, 180 (36%) of 506 exceeded 20,000 CFU/ml SPC within 10 days of processing. Based on the model including all features, the prediction model had a sensitivity of 81.4% and a specificity of 85.9% for the type of CIP detergent determined, which is significantly higher than the ANOVA model (p<0.03). Predictors that had better performance than the model including all features. Assessment of marginal effects indicates an interaction between filling line CIP detergent and detergent.

Significance: Our findings suggest that cleaning and sanitation as well as finished product quality monitoring are important predictors of milk spoilage within 10 days of processing and should be tested for developing and implementing practices aimed at improving milk quality.

T8-01 Observational Assessment of Food Safety Behaviors at Farmers’ Markets in Ontario, Canada
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Introduction: Farmers’ markets face numerous food safety challenges due to their operational characteristics, and they have occasionally been identified as a source of foodborne disease outbreaks. This study was initiated to evaluate the food safety behaviors of farmers’ market vendors in Toronto and two surrounding regions (Peel and York) of Ontario, Canada, to identify possible food safety risks.

Methods: All markets in the study region (n=60) were identified via publicly available databases and were visited between May and September 2018. The food safety behaviors of vendors selling retail foods requiring time-temperature control were observed discretely and results were recorded on a standardized and pre-tested form via a smartphone.

Results: A total of 454 vendors were observed, with prepared foods (50%) being the most common food type sold. While general cleanliness and sanitization practices were observed, some vendors did not observe proper storage conditions for all of their foods. Specifically, milk, eggs and 48% of vendors who sold foods requiring cold and hot holding (n=10 and 162, respectively) did not provide temperature control for at least some of their foods. Vendors’ use of cold holding/temperature control was more commonly observed among those selling raw meats, poultry, and fish (odds ratios: 3.95, 95% confidence intervals: 2.82 to 10.12) and less commonly observed among vendors selling RTE meats, fresh-cut leafy green vegetables, tomatoes, and melons, and cheese and dairy products. A mean of 4.55 (SD=2.62) behaviors requiring handwashing were observed per vendor across 1356 customer transactions, but handwashing was observed by only 13 vendors for one percent of these transactions. Most vendors (79%) handled food with their bare hands on at least one occasion.

Significance: Results of this study have identified targeted areas for future food safety education, training, and outreach with farmers’ market vendors in Ontario.
T8-04 Designing Food Safety Training Using the Integrated Behavior Model
Stephanie Maggio
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Developing Science Entreat
Introduction: Effective training enables participants to turn knowledge into actual skills and behaviors in the job. Therefore, it is important to design the training in a way that will make an impact on trainee’s behaviors. Previous researchers, investigating the effects of food safety training on large- and small-scale food processors, used survey (IBM) to predict which combinations of variables offer the greatest effect on workers actual behaviors. Instrumental attitudes, perceived norms, and personal agency were all found to be significant predictors of behaviors.
Purpose: The purpose of this study was to analyze the training target audience, specifically North Carolina small dairy processors, to determine which components of the IBM are significant predictors of intentions to perform safe food handling behaviors.
Methods: The IBM was used as the framework for the survey. Linear regression was used to determine which components of the IBM were predictors of intentions to perform behaviors.
Results: Perceived behavioral control (R²=0.2381; P=0.0340; n=19) and perceived norms (R²=0.1633; P=0.0244; n=19) were the strongest predictors of intentions to perform food-safe behaviors. After breaking down the components into sub-dimensions, it was found that instrumental norms (R²=0.2501; P=0.0340; n=19) and self-efficacy beliefs (R²=0.4400; P=0.0174; n=19) were the strongest predictors of intentions to perform behaviors.
Significance: Training can be maximized by designing it to make an impact on the specific components that drive target audience’s behaviors. Designing training to meet the needs of the target audience will not only be good for businesses, by making training more flexible, but it also has the ability to make the food systems safer.
T8-05 Food Safety Modernization Act Foreign Supplier Verification Rule: Three Years of Data As an Impact on the United States Food Import Chain Under FDA Jurisdiction.
Claudio Gallotti, Francesco Rapetti, Andrea Gentili, Ferruccio Marello, Enrica Alberti and Giovanni La Rosa
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Introduction: We have implemented the new FDA FSVM Foreign Supplier Verification Program (FSVP) requirements for United States imports over the last three years.
Purpose: Understand the current status of compliance with the new FSVP import requirements.
Methods: We interviewed 500 professionals, including 100 importers of record, 500 United States agents and 100 custom brokers. We then performed a questionnaire on a sample of 50 United States FSVP importers during FSVP classes offered by the Food Safety Preventive Controls Alliance and 50 during informational seminars. We then compared the results.
Results: Sixty percent of the surveyed respondents did not know the rules, and 30% of them are not ready to become FSVP importers. Only 10% are working to be in compliance. Also 98% of United States agents are not aware of this topic, and only two percent are fully in compliance; 80% of brokers’ interviews knowledge of FSVP is not able to provide accurate information about the new requirements. Food importers in direct contact with third countries have a lack of knowledge about new FSVP requirements. This could have an impact on foreign food supplier’s approach to FSMA requirements.
Significance: For effective implementation of FSVP and real prevention of noncompliance and nonconformance, communication strategy must be updated to effectively achieve public health goals linked to FSMA.
T8-06 Building a Competitive Advantage through the Safe Quality Food Certification in Food Manufacturing: Leveraging a Global Food Safety Initiative Scheme
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Introduction: In maintaining consumer trust and confidence, the food industry is plagued by many challenges. Incidents of food safety and quality complaints related to microbes, pathogens and chemical contaminants have resulted in import and recall events. The study conducted to explore import and recall events and withdrawals of food items from commerce. Studies have therefore proved that third-party audits are more expedient to fight food processing inefficiencies, ÔmisfitsÕ and ÔfailuresÕ as identified during SQF certification. After the obtaining of SQF certification, 94.29% of respondents also noted that the scheme on how the program has enhanced their food manufacturing operations.
Purpose: The purpose of this presentation is to understand the how the Safe Quality Food (SQF) certification as a Global Food Safety Initiative scheme contributes to competitive advantage in producing safe and food quality products.
Methods: A cross-sectional study was performed in five retail stores. The simulation and analysis of the two qualitative methods were performed using data analysis software (SPSS, IBM). The study was approved by the Wayne University Institutional Review Board (IRB) prior to the start of the study.
Results: Majority of participants had a positive perception of the value SQF provides to improving the overall safety and quality of food products; 77.14% of respondents had a positive perception of the value SQF certification as a credible and robust GFSI scheme that provides effective guidelines for food production; 94.29% of respondents noted that SQF certification as a GFSI scheme and 94.29% of respondents also noted that the scheme on how the program has enhanced their food manufacturing operations.
Significance: For effective implementation of SQF and real prevention of noncompliance and nonconformance, communication strategy must be updated to effectively achieve public health goals linked to FSMA.
T8-07 The Use of Matrix-adapter Bacterial Isolates of E. coli O157:H7, L. monocytogenes, and Salmonella spp. in Validation of High-pressure Treated Juices
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Introduction: High pressure processing (HPP) juice manufacturers are required to demonstrate a five-log reduction of the pertinent microorganism to comply with FDA juice HACCP. However, there is no consensus on validation study approaches for bacterial strain selection or preparation and no agreement on which HPP process parameters affect overall efficacy.
Purpose: To compare HPP inactivation of E. coli O157:H7, L. monocytogenes, and Salmonella isolates in buffer and apple juice.
Methods: Individual bacterial strains were grown using three different growth conditions: neutral, cold-adapted, and acid-adapted. Approximately six log CFUs/ml of the matrix-adapter bacterial strains were inoculated into buffered apple juice (5×10³ CFUs/ml) or apple juice at 3.5×10³ CFUs/ml (store-purchased) and treated at subtlet internal pressures of 500 MPa for E. coli O157:H7 and 200 MPa for Salmonella and L. monocytogenes (2.4°C, 15 min). Analyses were conducted at zero, 24, 48 and 144 h (4°C storage) post-HPP on non-selective media.
Results: For E. coli O157:H7, a greater than 8 log reduction was observed while for Salmonella spp. and L. monocytogenes, in buffered pepper water neutral growth conditions, E. coli O157:H7 strain TW41359 demonstrated greatest resistance (2.94±0.46 log reduction) and E. coli O157:H7 strain SE1888 was significantly more sensitive (P=0.05). Acid-adapted L. monocytogenes strain MA0328 had =1.00±0.23 log reduction while acid-adapted L. monocytogenes strains CDC and SCC were significantly less sensitive (P<0.0001). Treatment of the Salmonella Cubana and Salmonella Montevideo showed significantly greater resistance (P=0.05) compared to other cold-adapted strains. Salmonella isolates, neutral and acid-adapted, expressed similar bacteriostasis (P=0.2650) in apple juice, acid-adapted Salmonella Cubana was the only strain that did not demonstrate significantly increases in baristerenon when tested under all conditions. The time-dependent loss in viability occurred in all post-HPP storage samples.
Significance: These results suggest under the conditions tested, microbial composition and bacterial strain and preparation methods influence HPP efficacy and should be considered when conducting validation studies.
monoystances, co-films, Escherichia coli, and aerobic plate counts (APCs) using standard methods. Interviews with produce managers were also performed to assess the impact of PP+TPC-containing schedules.

Results: Two hundred swabs were collected; none tested positive for E. coli or monoystances. Average APCs and co-films across all samples were 5.31±1.3 and 5.91±6.1 CFU/swab, respectively. While no significant difference was observed between samples and time-points, data showed higher APCs for samples collected on the last day of observation. 

Methods: The study was conducted to develop a rapid, effective and cheap antimicrobial processing which combines mild thermal treatment and generally recognized as safe (GRAS) food additives. 

Results: L. monocytogenes was significantly (P<0.05) lower in the log reduction values at different time points between the different trials, with monoystances having highest survival rate. For instance, the log reduction value at the end of 42 days of dry-aging process for E. coli O157:H7 was <LOD, 3.70±2.8, 4.03±0.21, <LOD in trials one, two, three, and four. For Salmonella it was 3.02±0.00, 3.15±0.07, 2.86±0.11 and <LOD, and for L. monocytogenes it was 1.65±0.00, 2.50±0.00, 2.09±0.07 and <LOD.

Significance: The survival rate of all three pathogens is less than the detection limit (one CFU/cm²) in trial four (2°C, 85% RH) at the end of the 42nd day of inoculation. So, 2°C and RH 85% is the ideal process condition to dry age the beef to increase the safety of the product.

T9-03 Effect of Dry-Aging of Beef on the Survival of E. coli O157:H7, Salmonella and Listeria monocytogenes

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Developing Scientist Entrant

Introduction: Dry-aging is a traditional product of high quality that receives increasing interest. 

Purpose: To evaluate the effect of different process conditions during the dry-aging of beef on the survival rate of E. coli O157:H7, Salmonella and Listeria monocytogenes.

Methods: Beef sirsuts (n=2) were submitted to various dry-aging conditions of temperature and relative humidity (RH). Trial 1: 6°C, 75% RH; Trial 2: 2°C, 75% RH; Trial 3: 6°C, 85% RH; Trial 4: 6°C, 85% RH for 42 days. The meat samples were inoculated with a mixture of three cultures of pathogens (E. coli O157:H7, Salmonella, L. monocytogenes) at different times at five different combinations of time and temperature (E. coli O157:H7, 2°C, 75% RH, E. coli O157:H7, 2°C, 85% RH). The limits of quantification (LOQ) were at 100 CFU/g in dry-aging method. 

Results: There were statistically significant differences (P<0.05) in the log reduction values at different time points between the different trials, with monoystances having highest survival rate. For instance, the log reduction value at the end of 42 days of dry-aging process for E. coli O157:H7 was <LOD, 3.70±2.8, 4.03±0.21, <LOD in trials one, two, three, and four. For Salmonella it was 3.02±0.00, 3.15±0.07, 2.86±0.11 and <LOD, and for L. monocytogenes it was 1.65±0.00, 2.50±0.00, 2.09±0.07 and <LOD.

Significance: The survival rate of all three pathogens is less than the detection limit (one CFU/cm²) in trial four (2°C, 85% RH) at the end of the 42nd day of inoculation. So, 2°C and RH 85% is the ideal process condition to dry age the beef to increase the safety of the product.

T9-12 Restaurant Food Consumption and Diarrheal Illness: What is the Relationship? 

Robert Scharff

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Introduction: Although outbreak data suggests that contaminated food resulting in foodborne illnesses is mostly commonly prepared at home, a comprehensive examination of the relationship between restaurant patronage and diarrheal illness has not been completed.

Purpose: In this study, we examined the relationship between restaurant patronage and diarrheal illness using individual level data from the National Health and Nutrition Examination Survey (NHANES).

Methods: Common etiologic models (e.g., SIR, DGL, prob) are used to examine the relationship between individual restaurant consumption patterns and their likelihood of suffering from diarrheal illness. Against the background of other research and public channels of diarrheal illness (e.g., health conditions and alcohol consumption) and demographics are included. NHANES observations from surveys fielded between 2001 and 2016 are used in the analyses. Observations that do not contain information on diarrhea illness (restroom consumption and controls for other causes of diarrheal illness) are omitted. A total of 41,430 observations are used in the analysis.

Results: Between 2001 and 2016 restaurant consumption was associated with 59% of outbreaks and 44% of illnesses from CDC reported foodborne illness outbreaks. 

Significance: The study demonstrated a rapid (within four min) and effective (5.3-log to 3-log reduction) antimicrobial approach to realistic food items.

The new approach may be readily transferred to a fast-food restaurant to achieve rapid sanitation of fresh-cut vegetables before serving to consumers.

T9-10 Prevalence of Top Seven Shiga Toxin-producing Escherichia coli in Microbial Populations through Slaughter in Australian Beef Export Abattoirs

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Introduction: Australian beef processors must ensure that manufacturing beef being exported to North American market is disease free. Top five Shiga-toxin producing Escherichia coli (STEC) serogroups. A better understanding of the microbiota of beef carcasses, the abattoir environment and the dissemination of STEC in the abattoir could assist in determining the sites of cross-contamination of STEC through slaughter.

Purpose: This study aims to compare the efficacy of commercial antimicrobial cocktails to inactive unjured and stress-adapter Campylobacter jejuni on bacterial population during processing using immunization and electrostatic-spraying, and to compare the economic feasibility of the two methods.

Methods: Three strains of overnight cultured (18 h) C. jejuni strains were unjured or shocked in pH 5.0 (Bolton’s broth (2); subcultured in 0.9% saline solution (two), and stored in Bolton’s broth (4°C, five days) to prepare acid-, starvation-, and cold-stress-adapted cells, respectively. Fresh wings inoculated with unjured or stress-adapter C. jejuni were treated with immersion and electrostatic-spraying, and to compare the economic feasibility of the two methods.

Results: The survival rate of all three pathogens is less than the detection limit (one CFU/cm²) in trial four (2°C, 85% RH) at the end of the 42nd day of inoculation. So, 2°C and RH 85% is the ideal process condition to dry age the beef to increase the safety of the product.

Significance: The survival rate of all three pathogens is less than the detection limit (one CFU/cm²) in trial four (2°C, 85% RH) at the end of the 42nd day of inoculation. So, 2°C and RH 85% is the ideal process condition to dry age the beef to increase the safety of the product.
electrostatic spraying. Among all stressed cultures, P. aeruginosa, L. monocytogenes, and S. aureus were isolated and screened for antagonistic activity against E. coli. Of all the isolates tested, 27% of P. aeruginosa, 18% of L. monocytogenes, and 35% of S. aureus produced an antimicrobial substance.

**Conclusion:** The study demonstrated that Photobacterium phosphoreum could be a potential candidate for future research in the development of antimicrobial agents against foodborne pathogens.

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**T9-04 Prevalence and Biofilm Formation of Staphylococcus aureus Isolated from Animal Food in Shanghai, China**

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**Developing Scientist Entrant**

**Introduction:** Biofilm is one of the important factors affecting the antimicrobial resistance of bacteria. Staphylococcus aureus usually has strong biofilm formation ability, and it is widely found in animal food.

**Methods:** The aim of this study was to determine the prevalence and biofilm formation ability of S. aureus in animal food, and to find an effective antimicrobial material with biofilm-breaking ability.

**Results:** A total of 1,359 samples of animal food were collected from different locations and processed. The overall detection rate of S. aureus was 87% (257 of 301) for chicken, 76% (238 of 315) for pork, 92% (240 of 259) for shrimp, 87% (247 of 283) for beef, 85% (246 of 291) for duck, and 92% (240 of 261) for egg. A total of 124 (36.8%) of the S. aureus isolates showed strong biofilm formation ability, and the rest were moderate. The largest difference was found in beef, wherein 90% of the S. aureus isolates showed strong biofilm formation ability. A total of 301 samples of animal food were collected from different locations and processed for biofilm formation ability.

**Conclusion:** The results showed that S. aureus is a common pathogen in animal food, and the biofilm formation ability of S. aureus varies significantly among different types of animal food. The results also highlight the importance of biosafety management in animal food production and processing.
T9-12 - T10-03

Effectiveness of a Novel, Rechargeable, Non-leaching Polycationic N-Alkaline Antibacterial Coating on Listeria monocytogenes Survival in Food Processing Environments

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Introduction: Outbreaks and recalls due to Listeria monocytogenes are often associated with RTE products.

Purpose: To evaluate the bactericidal efficacy of a novel fish-collagen-based coating (GlycoBond) on stainless steel surfaces to reduce L. monocytogenes transfer to RTE beef products.

Methods: Chlorinated N-halamine coating was applied onto stainless steel surfaces (from 25 to 225 cm²; n=360) under the following surface conditions: no coating (control), coated intact and coated scratched. The tests were done on L. monocytogenes cocktail (four strains; 10⁴ CFU/mL) that was suspended in PBS or meat purge and then applied on stainless steel surfaces for 45 min. For PBS, bacterial reductions were calculated. For deli meat three beefiers and their three roast beef slices were placed on each surface separately for 20 min. Meat products were removed, packed individually in easy open deli plastic bags, closed up and held at 2°C and 5°C for 48 and 72 hr. For L. monocytogenes enumerable test surfaces were divided in half with typical tray soy agar (thin agar layer method). All experiments were replicated three times.

Results: Overall for PBS, L. monocytogenes reductions ranged from 4.8 to 5.2 log CFU/cm². Regarding deli meat, for sausages, the coated intact and coated scratched showed similar antimicrobial capacity (P=0.50) and resulted in less than one log (≤0.700) of L. monocytogenes transfer (≥0.900 L. monocytogenes reduction) compared to control surfaces. In contrast, for the roast beef product, coated scratched showed the lowest L. monocytogenes transfer, followed by coated intact and then the control group (P=0.01). In this case, coated intact reduced L. monocytogenes by 6.7 log, while the reduction by coated scratched was 7.9 log. Regardless of the coating conditions, in both sausage and roast beef, overtone N-halamine coating showed lower antimicrobial L. monocytogenes transfer from 0 to 72 hr post exposure (≤3.5 log CFU/cm²) without any decline in its antimicrobial effect.

Significance: Due to the high antimicrobial capacity of the N-halamine coating, it can be used by the food industry to reduce the likelihood of L. monocytogenes cross-contamination and persistence.

T10-01 Development of a User-friendly Software Tool for Validation of Predictive Models

Thomas Oscar

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Introduction: Proper validation of models that predict the growth of pathogens in food will increase confidence in using them to make important food safety decisions.

Purpose: The current study was undertaken to develop a user-friendly, validation software tool (ValT) for predictive models that are based on the test data and models of a reasonable prediction zone method developed by the United States Department of Agriculture.

Methods: The new software tool was developed in Excel and was demonstrated using a published model for growth of Salmonella Typhimurium definitive phage type 104 on chicken skin. A model prediction was considered acceptable when its residual (observed minus predicted) was in an acceptable prediction zone from -1 log (fail-safe) to 0.5 log (fail-dangerous). A model was considered to provide predictions with acceptable accuracy and bias when the proportion of residuals in the acceptable prediction zone (pAPZ) was <0.700 for a single level of an independent variable (time or temperature).

Results: The overall pAPZ was 0.823 for dependent data (n=384), 0.826 for interpolation data (n=178), and 0.786 for extrapolation data (n=16) to another food matrix (kasher chicken skin). Although overall pAPZ were acceptable, the model failed validation for dependent data, interpolation, and extrapolation because the dependent data and interpolation data did not satisfy all criteria for test data and because of local prediction problems for all sets of data tested.

Significance: A new software tool called “vault” (ValT) was developed that will make it easier for model developers to properly validate their models using a comprehensive set of criteria developed by the USDA.

T10-02 WITHDRAW

T10-03 Using Predictive Pre-processing Risk Scores to Reduce Foodborne Disease

Timothy Buisker

Smart Data Solve Solutions, Guelph, IL

Introduction: Intensive food processing operations create statistically predictable problem runs under the same parameters regardless of the risks posed by incoming products. Often, pre-processing pathogen testing alone is insufficient to differentiate risk. However, combinations of pre-processing testing, historical performance data, and location-based metadata can be used in machine learning (ML) algorithms to generate accurate pre-processing risk scores.

Purpose: To reduce the risk of foodborne foodborne transmission by providing risk scores that allow producers to differentiate pathogen risk in incoming products.

Methods: We develop a suite of ML algorithms that takes available pre-processing data including pathogen tests, historical performance data from growing locations, and location-based metadata, and generates a predictive risk score predicting which product batches are most likely to have positive pathogen tests in a final product. Risk scores can be used to identify decision-making opportunities, prioritize interventions, and establish priorities for certain product batches (e.g. from raw to cooked), or other available techniques depending on the product, with the goal of reducing the final pathogen prevalence and ultimately foodborne disease.

Results: Utilizing data from over 2000 commercial broiler chicken placements, and combining live operations and historical performance, we employ an ensemble ML algorithm to assign a risk score to each product batch. We test our algorithm on 20% of the data and a dataset was initially set aside, it was able to identify with 80% accuracy which days the processing plant would have a positive Salmonella test in the outgoing product.

Significance: Predictive algorithms can accurately assign risk scores prior to processing, allowing for producers to make intervention-based and decision-making results that reduce the prevalence of pathogens on the outgoing product, lowering the public risk of foodborne disease.

T10-04 Risk Categorization of Federally Registered Meat Establishments in Canada using the Canadian Food Inspection Agency’s Establishment-based Risk Assessment Model

Shraddha Karanth and Abani Pradhan

University of Maryland, College Park, MD

Introduction: The Canadian Food Inspection Agency (CFIA) has developed a quantitative risk assessment model to help inform inspection resource allocation for food establishments. In 2014, a pilot assessed the model’s performance with 49 meat/poultry establishments resulting in a Spearman correlation coefficient of 0.46 and 0.53 for the model outputs of number of DAs and the assessment done by senior inspectors.

Purpose: To assess the food safety decision making of registered meat/poultry establishments across Canada, to subsequently integrate the model outputs in the Agency’s work planning for risk-informed oversight.

Methods: We conducted a cross-sectional study of all 2014 meat/poultry establishments and their assigned inspectors attended Webinars and session presentations and provided inputs on the mitigation/facilitation factors associated with their facilities, using an Excel questionnaire. This was analyzed by the model algorithm along with related data from CFIA systems.

Results: New establishments were not considered in the analysis because they refused to participate (0.15%), were not operating (0.74%), or were not processing/ storing meat products (0.44%) at the time of data collection. Establishments reported processing multiple meat species (50%), distributing products directly to consumers (49%), and employed varying levels of intervention (72%) to further reduce their risk. Ten establishments (26% of the total Canadian domestic meat volume) were responsible for 35% of the total risk, while 85% of the total food safety risk related to this sector.

Significance: This model permitted categorization of meat establishments’ individual contribution to the overall food safety risk in this sector (19, 3, 227, and 351 for category 1 to 4 respectively, where 1 represents the highest risk and 4 the lowest, as of January 2019). These findings will be used to proportionally allocate inspection resources for risk management based on the establishment risk contribution.

T10-05 WITHDRAW

T10-07 Predicting the Food Sources of Sporadic Cases of Listeria Infection Using Whole Genome Multilocus Sequence Typing

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1Centers for Disease Control and Prevention (CDC), Atlanta, GA, 2Centers for Disease Control and Prevention, Atlanta, GA, 3U.S. Center for Disease Control and Prevention, Atlanta, GA

Introduction: Whole-genome sequencing (WGS) has substantially refined the ability to subtype Listeria monocytogenes isolates. PulseNet maintains the metadata of sequenced isolates from patients, food, and food production environments.

Purpose: We used these data to predict food sources of sporadic listeriosis cases by comparing genetic features between isolates from humans with those from foods.

Methods: We developed a random forest model using whole genome multilocus sequence typing (wgMLST) (+4000 genes) on a training dataset of 484 isolates ( 5% of total WGS isolates) representing 2,600 named serovars with varied pathogenicity profiles being identified for L. monocytogenes. The model was trained on the full dataset and evaluated on an independent variable (time or temperature).

Results: The overall pAPZ was 0.823 for dependent data (n=384), 0.826 for interpolation data (n=178), and 0.786 for extrapolation data (n=16) to another food matrix (kasher chicken skin). Although overall pAPZ were acceptable, the model failed validation for dependent data, interpolation, and extrapolation because the dependent data and interpolation data did not satisfy all criteria for test data and because of local prediction problems for all sets of data tested.

Significance: This model may assist with investigations by providing hypotheses about the most probable vehicles based on wgMLST of the outbreak strains.

T10-08 Evaluating the Prevalence of Salmonella Genus Infection in Chicken to Incorporate into a Risk Assessment Framework

Shraddha Karanth and Abani Pradhan

University of Maryland, College Park, MD

Introduction: Salmonella is responsible for 11% of all foodborne infections in the United States. It covers a diverse genetic landscape, with more than 2,600 named serovars with varied pathogenicity profiles being identified for Salmonella enterica subsp. enterica alone. Despite this, current quantitative microbial risk assessment (QMRA) models, which utilize relevant food safety information to understand and evaluate the risk of foodborne illness, do not accurately incorporate information on the prevalence of salmonellosis.

Purpose: The purpose of this study was to determine the applicability of whole-genome sequencing in revising prevalence and risk estimates for Salmonella enterica in broiler chickens.

Methods: Whole-genome sequences of 150 Salmonella samples (serovars including Enteritidis, Heidelberg, Typhimurium, and Kentucky) isolated from chicken by the US Food and Drug Administration (FDA) GenomeTrakr project were analyzed in this study. Viscera, stress-response, and antimicrobial resistance patterns of isolates were predicted using multiple sequence alignments and divergence metrics and clustering methods to estimate the configuration of virulence factors using 3-means clustering. Cluster-specific phenotypic expression was correlated with spatiotemporal factors obtained from the National Oceanic and Atmospheric Administration (NOAA) using a logistic regression model and prevalence re-estimation was performed using a Poisson regression model with R software version 3.5.2.

Impact: The genome sequencing method used in this study is able to accurately assign the prevalence of Salmonella in broiler chicken, allowing for more accurate QMRA calculations and corresponding public health resources. This will improve the risk assessment process in the industry and government by incorporating this valuable information.
T10-09 Stochastic and Dynamic Predictive Modeling Using a Monte Carlo Simulation to Estimate the Behavior and Survival Probability of Bacterial Spores

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Developing Scientist Entrant

Introduction: Conventional kinetic models describing bacterial inactivation do not account for variability and uncertainty. In contrast, stochastic models describe bacterial death with probability distributions which is taking individual cell heterogeneity into account. In addition, actual food thermal processing is non-isothermal so stochastic models describe bacterial reductions in isothermal processing. Appropriate design of minimal food processing requires stochastically accurate estimations of bacterial survival behavior in non-isothermal processing.

Purpose: The objective was to develop a second-order Monte Carlo (2DMC) simulation model describing variability and uncertainty in the spore survival behavior using the bootstrap method. In addition, the simulation results were verified with experimentally observed data.

Methods: Dose- response curves in 100 µl of pH-adjusted triptizoyl soy broth (pH 5.4, 5.8, 6.2, 6.6 and 7.0) were heated at 80°C, 90°C and 95°C in a thermal cycle; the survival spore counts were determined on tyrode agar after 24h incubation. The survival of spores in each environment were fitted to the Weibull distribution. Monte Carlo simulation using R software. Monte Carlo simulation was used to quantitatively simulate uncertainty and variability associated with parameters.

Results: Thinite survival of spores was observed after cooking and freezing processes. T. gondii tissue cysts in fresh meat after cooking and freezing processes. T. gondii is a protozoan that can cause toxoplasmosis, a serious illness that can be transmitted to humans through consumption of undercooked or raw meat infected with T. gondii. This study demonstrates a model to evaluate whole genome sequencing to a quantitative risk assessment framework.

Significance: This study could be used to validate the current USDA recommended minimum cooking temperature for fresh cuts and estimating survival for T. gondii under a temperature gradient.

T10-10 Thermal Inactivation of Salmonella enterica and Non-pathogenic Bacterial Surrogates in Wheat Flour by Baking in a Household Oven

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Rutgers University, New Brunswick, NJ, 2Michigan State University, East Lansing, MI

Developing Scientist Entrant

Introduction: Wheat flour has been implicated in recalls and outbreaks linked to Salmonella and pathogenic Escherichia coli. An online instructional video treatment protocol on wheat flour has been planned that a safe raw cookie dough could be made from flour baked in a household oven at 177°C (350°F) for 5 min but no evidence in support of such a protocol was provided.

Purpose: This study was conducted to assess thermal inactivation of two wheat flour samples, as well as Enterococcus aerogenes and Pantoea dispersa in wheat flour during home oven-style baking.

Methods: Wheat flour was inoculated with Salmonella Enteritidis PT 30, Salmonella Typhimurium PT 42 or their potential surrogates at ~10^5 CFU/g before baking in household oven at 149, 177, and 204°C (300, 350, or 400°F) for up to seven min. Flour was heated in aluminum tray in a ~2 cm layer. Baked wheat flour samples (five each) were enumerated in triplicate, and microbial concentration was expressed in log CFU/g. Thermal profiles of wheat flour and air in the oven during the baking were recorded. Activity of wheat flour samples were also measured before and after baking.

Results: Heat treatment inactivation of bacterial populations by wheat flour baking time increased up to 100-fold, with reduction of water activity (measured at treatment temperature) from 0.7 to 0.2. The findings suggest that water activity of food matrices contribute significantly to the survival probability of bacterial spores.

Significance: Baking of wheat flour in household toaster ovens has potential as an inactivation treatment of pathogenic bacteria in consumers homes, despite its low water activity.

T10-11 WITHDRAWN

T10-12 Evaluating Uncertainty and Variability Associated with Toxoplasma gondii Survival While Cooking and Freezing Fresh Cut Meats

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Developing Scientist Entrant

Introduction: Toxoplasma gondii is a worldwide zoonotic parasite with high serorelevance in the human population. More than 40 million people carry this parasite in the United States. Consumption of raw or undercooked meat containing T. gondii tissue cysts is a major source of infection in humans. Ingesting oocyst-contaminated raw meat or water can result in acute-to-chronic disease, inactivating T. gondii in meat.

Purpose: Current available data are not sufficient to suggest safe cooking or storage temperatures for handling fresh for cut meats. Consumer preferences for cooking and storage temperatures introduce variability. The main objective of this study was to analyze uncertainty and variability in the survival pattern of T. gondii tissue cysts in raw meat after cooking and freezing processes.

Methods: In the United States, there has been only two studies done on heat inactivation of T. gondii tissue cysts and one on its inactivation by freezing. Data regarding survival of tissue cysts were re-sampled with a bootstraping method in R software. Monte Carlo simulation was used to quantitatively simulate uncertainty and variability associated with parameters.

Results: The results showed a negative correlation (r = 0.992) between cooking temperature and survival time for T. gondii whereas positive correlation between cooking time and survival temperature were highly significant at a P-value of 0.05. Regression models were well established with correlation coefficients and confidence intervals. The uncertainty level was further decreased by the jackknife-after-bootstrap method which identified cutters and narrowed down the bootstrap confidence interval by 16.7% for the cooking process.

Significance: This study could be used to validate the current USDA recommended minimum cooking temperature for fresh cut meats and establishing a survival model for T. gondii under a temperature gradient.

T11-01 Desiccation in Oil Protects Bacteria in Thermal Processing

Ren Yang, Yuchen Xie, Je Xu and Juming Tang

Washington State University, Pullman, WA

Developing Scientist Entrant

Introduction: The protection effect of oil on bacteria in thermal treatments was first reported in 1913. Since then, researchers attempted to understand its mechanism without reaching a conclusive explanation. Recent studies on low-moisture foods have shown a strong correlation between increased heat tolerance of vegetative bacteria and low water activity of food matrices in thermal treatments. Our recent research also verified that the a_{w} in plant oil decreases sharply with increasing temperature. Thus, we hypothesize that the thermal dynamic equilibrium of water vapor between vegetative bacterial cells and oil causes desiccation of the bacteria in thermal treatments, which may explain the oil protection phenomenon.

Purpose: To prove the hypothesis that desiccation in oil protects bacteria against thermal treatment.

Methods: Peanut oil inoculated with Enterococcus faecium NBR: B-2354 was preconditioned under controlled humidities, the bacterial thermal death times at 80°C were tested using two methods: i) closed-system (capillary tube) and humidity-controlled environment (TAC), at 14 ± 2°C each with three independently biological replications. Thermal death counts at 80°C were calculated and plotted logarithms against high-temperature a_{w} of peanut oil in capillary tubes or that of the head-space relative humidity in TACs.

Results: log-D values from the two methods are in good agreement, showing a linear relationship with a_{w} (at 80°C) between 0.1 and 0.6. Our results are superior with respect to other methods. The drawback of this phenomenon is that the key factor that determines the bacterial thermal resistance, regardless of the carrier.

Significance: The results support the hypothesis that oil protection is caused by desiccation of bacterial cells in oil at high temperatures.

T11-02 Key Factors Influencing Thermal Resistance of Bacterial Pathogens in Low-moisture Foods

Ren Yang and Juming Tang

Washington State University, Pullman, WA

Developing Scientist Entrant

Introduction: Microbial safety associated with low-moisture foods (LMFs) is an emerging issue. The food industry is experiencing challenges in seeking solutions to control Salmonella due to its high tolerance to thermal treatments. The heat resistance of bacterial pathogens in different LMFs is also difficult to predict due to its high heterogeneity among food products. Water activity of low-moisture foods change with temperature, the degree of the changes depends on compositions.

Purpose: This presentation summary describes results from our recent research to evaluate the importance of water activity of food matrices measured at treatment temperatures (not room temperature) on thermal inactivation of Salmonella and a surrogate, E. faecium, in LMFs.

Methods: Our research consisted of two major components: i) determining water activity of different food matrices (powders with different composition and water activity levels) to establish relationships between product moisture content, water activity, and temperature, (from 20 to 80°C) in sealed containers; ii) correlating thermal resistance (a_{w}) of Salmonella and E. faecium in LMFs with relative humidity (water activity) at high temperatures.

Results: Water activity of foods rich in protein and starch increased sharply with increasing temperature, whereas foods with high oil contents did not increase much. Batch-in batches with temperature regardless of the food matrices, B. simplex and E. faecium increased exponentially, by up to 100-fold, with reduction of water activity (measured at treatment temperature) from 0.7 to 0.2. The findings suggest that water activity of food matrices can have a significant impact on the thermal resistance of pathogenic microorganisms.

Significance: Our results explain the difficulty in thermal inactivation of bacterial pathogens in oil-rich products and suggest that relative humidity at treatment temperature should be considered as a control parameter in designing effective thermal treatment operations for pathogen control in LMFs.

T11-03 Decontamination of Salmonella enterica in Low-moisture Foods by Cold Atmospheric Plasma

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Developing Scientist Entrant

Introduction: Cold atmospheric plasma (CAP) offers a dry, non-thermal, and rapid process for surface decontamination of food products.

Purpose: The purpose of this study was to evaluate the effects of treatment time, treatment distance from the plasma actuator, and pre-conditioning of the product on the inactivation efficiency of CAP on Salmonella enterica in peanut oil at 12.11°C.

Methods: In-shell pecans (in triplicate) and black peppercorns (one in triplicate) were spot inoculated with a mixture of five strains of S. enterica (71 CFU/ml), and air dried. The inoculated pecans and black peppercorns were treated by CAP for two, five, and 10 minutes at one, two, and five centimeters from the plasma actuator. Similarly, inoculated pecans and black peppercorns inoculated after air-drying were treated by CAP for two and five minutes at two centimeters from the actuator. Experiments were repeated at least two times. Mean values of log reduction of S. enterica cells after treatments were compared using ANOVA.

Results: Treatment had a significant effect on the reduction of the pathogens on both pecans and black pepper. With 10-minute CAP treatment, all the strains were reduced by >3-log CFU/ml. Salmonella enterica were observed at all distances on pecans and black pepper, respectively. Moistening of inoculated pecans or black peppercorns prior to treatment achieved an additional one-log reduction of the pathogen compared to the treatment without moistening.

Significance: These results show that CAP can be a viable and flexible technology for inactivation of foodborne pathogens on low-moisture foods such as nuts and spices.
T1-04 Microbiological Profile, Incidence and Behavior of Salmonella in Seeds Commercialized in Mexican Markets

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Introduction: Seed consumption has been increased in recent years due to the high nutrient content. Unfortunately, the number of outbreaks caused by Salmonella associated with the consumption of low water activity food items has also increased although they do not support microbial growth.

Purpose: The main goal in this study was to quantify microbial indicators and to determine the prevalence and behavior of Salmonella in ground flax seeds, sesame, and amaranth seeds obtained from Mexican markets.

Methods: Sixty-five samples of each product (chia, amaranth and sesame seeds) were collected from Querétaro city markets. Aerobic plate count (APC), coliforms and Enterococci col were quantified, and the prevalence of Salmonella sp. was determined according to the methods proposed by the Food and Drug Administration Bacteriological Analytical Manual. Chia, amaranth and sesame seeds (one kg) were inoculated with a cocktail of five Salmonella strains (five CFU/ml) and stored at ambient temperature. The population of Salmonella was quantified for up to 70 days.

Results: The median content of APC in chia, amaranth, and sesame seeds were 2.3, 2.5, and 3.9 log CFU/g, respectively. The content of coliforms oscillated from 0.5 to 1.4 log MPN/g. E. coli positivity was 3% in chia, 6% in amaranth and 9.2% in sesame seeds. Salmonella was detected in 18.5, 10.8, and 15.9% of chia, amaranth, and sesame seeds, respectively. After 70 days of storage, the population of Salmonella reduced 1.1 log CFU/g in chia, 1.3 log CFU/g in amaranth, and 1.5 log CFU/g in sesame seeds.

Significance: The incidence of Salmonella in the seeds was high, which could represent a risk for consumers, specifically when the food is consumed without treatments that guarantee the pathogen inactivation. It is necessary to develop technologies to control foodborne pathogens in low water activity food such as seeds.

T1-05 Survival of Salmonella and Surrogate Microorganisms in Whole Wheat and All Purpose Flour during Long-term Storage

Jin Jung, Matthew Igo and Donald W. Schaffner

Rutgers University, New Brunswick, NJ

Introduction: Flax seed has gained popularity as a human food supplement and animal feed because of its richness in protein, dietary fiber and omega-3 fatty acids. It has been reported that flax seed supports fungal growth and mycotoxin production during storage.

Methods: Whole wheat flour and all purpose flour were inoculated with a cocktail of five Salmonella strains (103 CFU/g) and stored at 20°C for 27 weeks in tightly closed containers, and five-g samples were cultured on tryptic soy agar supplemented with nalidixic acid at appropriate time intervals. All experiments were conducted in triplicate and microbial populations were expressed in log CFU/g.

Results: Salmonella showed better survival than E. aerogenes and P. dorelia in both whole wheat flour and all purpose flour over the storage period. Salmonella in flour could be enumerated at least 17 weeks, with average declines of 0.13 to 0.19 log CFU/week depending on Salmonella strain and flour type. Non-pathogenic E. aerogenes was enumerated for at least nine weeks of storage, with average declines of 0.23 to 0.38 log CFU/week depending on surrogate strain and flour type. Non-pathogenic E. coli and Salmonella were quantified for up to 70 days.

Significance: This study will be useful for future microbial risk assessments and will add to the growing literature on Salmonella behavior in low-moisture foods.

T1-06 Studies of Aflatoxin Production by Aspergillus flavus and Aspergillus parasiticus on Ground Flax Seeds

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{1}Purdue University Northwest, Department of Chemistry and Physics, Hammond, IN; {2}Department of Biological Sciences Purdue University Northwest, Hammond, IN

Introduction: Flax seed has gained popularity as a human food supplement and animal feed because of its richness in protein, dietary fiber and omega-3 fatty acids. It has been reported that flax seed supports fungal growth and mycotoxin production during storage.

Methods: Whole wheat flour and all purpose flour were inoculated with a cocktail of five Salmonella strains (103 CFU/g) and stored at 20°C for 27 weeks in tightly closed containers, and five-g samples were cultured on tryptic soy agar supplemented with nalidixic acid at appropriate time intervals. All experiments were conducted in triplicate and microbial populations were expressed in log CFU/g.

Results: Salmonella showed better survival than E. aerogenes and P. dorelia in both whole wheat flour and all purpose flour over the storage period. Salmonella in flour could be enumerated at least 17 weeks, with average declines of 0.13 to 0.19 log CFU/week depending on Salmonella strain and flour type. Non-pathogenic E. aerogenes was enumerated for at least nine weeks of storage, with average declines of 0.23 to 0.38 log CFU/week depending on surrogate strain and flour type. Non-pathogenic E. coli and Salmonella were quantified for up to 70 days.

Significance: This study will be useful for future microbial risk assessments and will add to the growing literature on Salmonella behavior in low-moisture foods.

T1-07 Influence of the Germination Time on Aflatoxins Production during Flax of Wheat for Use in Craft Beer

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Introduction: Mycotoxins are secondary metabolites with harmful effects on humans and animals. They can be produced in contaminated grains that contains low water activity and can be transferred to food stuffs. They are a food safety concern worldwide. Aflatoxins are the most potent mycotoxins known to date. A. flavus and A. parasiticus are most frequently isolated from food and feed samples. Aflatoxins are immunosuppressive and are responsible for human toxicities including liver cancer and nephrosis.

Purpose: The objective of this case-control study was to assess the relationship between maternal exposure to fumonisin via immersion of contaminated corn, particularly maize-based food, and NTDs in newborns in Guatemala.

Methods: Each additional food item consumed (OR 0.93; 95% CI: 0.88 to 0.97) decreased odds of NTDs. Each additional food item consumed (OR 0.93; 95% CI: 0.88 to 0.97) decreased odds of NTDs.

Significance: The inverse association of diet diversity and positive association of maize and beef consumption with risk for NTDs suggests further research is needed to more accurately examine the association of maternal diet with NTDs. In particular, studies that measure fumonisin biomarkers in food and/or in maternal plasma could help further evaluate this hypothesis.

T1-08 Rapid Identification of Lineage Types and Phylogenetic Relationships of Clastidiomycotina strains by Whole Genome Sequencing

Narjol Gonzalez-Escalona{1}, Nagarajan Thirunavukkarasu{2}, Travis Wentz{3}, Eric Brown{3}, Thomas Hammack{3} and Shashi Sharma{3}

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Introduction: Whole Genome Sequencing (WGS) allows studying the phylogenetic relationships of clastidiomycotina strains. Recent developments with WGS are moving towards the metagenomic level of characterization and may be a viable tool to perform source track investigations for foodborne outbreaks as well.

Methods: We sequenced the whole genome of 13 unknown C. batuliniformis strains to study their phylogenetic relatedness and lineage types through SNP analysis.

Results: C. batuliniformis strains were grown and genomic DNA was isolated using DNeasy blood and tissue kit. Identification of the strains belonging to C. batuliniformis was performed using a PCR assay by PCR reactions and Sanger sequencing. Sixty-five strains were grown and genomic DNA was isolated using DNeasy blood and tissue kit. Identification of the strains belonging to C. batuliniformis was performed using a PCR assay by PCR reactions and Sanger sequencing. Sixty-five strains were sequenced using a MiSeq (Illumina) with the 250 bp paired-end reads chemistry according to manufacturer's instructions at 140x coverage. The C. batuliniformis seed strain was analyzed by whole-genome sequencing (WGS) and the phylogenetic analysis was performed using a core genome multispecies sequence typing approach (cgMLST) using RsidomMLST v. 2.4.0 software.

Significance: In Sclerotiumrolfsii the 13 sequenced genomes revealed that three of them belonged to non-proteolytic C. batuliniformis group 1, whereas seven of the 13 strains belonged to a C. batuliniformis group 2. The other 10 strains belonged to different C. batuliniformis group 1. These five strains have an identical profile to Lanzarote strain type 1 and carried a single BoNT/c cluster (BoNT/cF). The remaining 5 strains were toxic binuclear strains and belonged to known STs 6 (9, 10, 2, 27), and 4. An NTD analysis (using 21 additional C. batuliniformis group 1 genomes available at NCBI) showed that these 10 strains belonged to different C. batuliniformis group 1 lineages, with the majority (five) belonging to Lineage 5, and matched closely with strains 236613 and Lanzarote, both BoNT/cF strains.

Significance: Our results showed that the use of WGS in combination with the proper use of bioinformatic tools allowed for fast identification, phylogeny, and toxin cluster determination for unknown C. batuliniformis samples.

T1-02 Maternal Dietary Risk Factors for Neural Tube Defects in Guatemala

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Introduction: Maternal dietary factors involved in the pathogenesis of various human and animal health outcomes. In particular, the consumption of foods contaminated with fumonisin has been associated with liver cancer and congenital malformations, such as neural tube defects (NTDs). Fumonisin is found in the diets of a large proportion of the world's population where maize is a staple diet. Low- and middle-income countries with high maize consumption are at higher risk for NTDs.

Purpose: The objective of this case-control study was to assess the relationship between maternal exposure to fumonisin via immersion of contaminated foods, particularly maize-based food, and NTDs in newborns in Guatemala.

Methods: Each additional food item consumed (OR 0.93; 95% CI: 0.88 to 0.97) decreased odds of NTDs. Each additional food item consumed (OR 0.93; 95% CI: 0.88 to 0.97) decreased odds of NTDs.

Significance: The inverse association of diet diversity and positive association of maize and beef consumption with risk for NTDs suggests further research is needed to more accurately examine the association of maternal diet with NTDs. In particular, studies that measure fumonisin biomarkers in mother and test for fumonisin biomarkers in newborns are needed.
T12-02 A Summary of Foodborne Illness Outbreaks Investigated by FDA's Coordinated Outbreak Response and Evaluation Network from January 2011 to December 2018

Sheila Paul Kerrner, Tami Craig, Maryanne Fegan, D. Curtis Henry, Sandy Wiener

Introduction: The Coordinated Outbreak Response and Evaluation Network (CORE) of the Food and Drug Administration (FDA), formed in August 2011, is a multidisciplinary team that evaluates, investigates, and guides prevention efforts of foodborne illnesses outbreaks. Working with federal, state, and local colleagues, CORE investigations help prevent future outbreaks that may cause illness and death.

Purpose: The purpose of this analysis is to display and summarize FDA CORE data while providing transparency on the work FDA CORE is doing for public health as well as to outbreaks.

Methods: The 170 outbreaks, occurring between January 2011 to December 2018, were analyzed by product category, year, pathogen, number of illnesses, hospitalizations, and deaths using data from the FDA CORE database.

Results: Of the 170 outbreaks analyzed between January 2011 and December 2018, CORE has responded to 170 U.S. foodborne illness outbreaks resulting in an estimated total of 10,484 illnesses, 2,385 hospitalizations, and 100 deaths. Most of the outbreaks were attributed to produce (n=69), followed by seafood (n=52) and processed foods (n=22). Illnesses were also predominantly caused by outbreaks associated with produce (n=670), followed by seafood (n=570). At least three were outbreaks associated with both seafood and produce, followed by dairy (n=20). These deaths were primarily caused by foods contaminated with *Salmonella* monовариета (*S. typhimurium*), whereas most of the illnesses are associated with *Salmonella* infections (*S. enteritidis*). In response to these outbreaks, we utilized DNA fingerprinting to identify isolates within zero to 10 allele differences were identified using whole genome multilocus sequence typing (WGS). *Salmonella* isolates was implemented by the National Microbiology Laboratory (NML) in Canada for outbreak detection in May 2017.

Conclusion: This outbreak illustrates potential risks associated with food exempt from evaluation and prepared by untrained volunteers; it highlights the need for a multi-jurisdictional approach to outbreak investigations.

T12-03 Unregulated Food Sales Go Wrong: *Clostridium perfringens* from a Church Fundraiser in North Carolina

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Methods: Unregulated food sales in North Carolina are regularly carried out by non-for-profit groups, and although legal exemptions within the NC General Statutes allow food sales once per month from food service permit requirements, foodborne illness outbreaks do occur because of challenges such as inadequate equipment and untrained volunteers.

Results: A foodborne illness outbreak investigation was conducted after an initial report of illness from a church fundraiser in November 2018 by a multi-jurisdictional team including state and local communicable disease nurses, epidemiologists, and environmental health specialists.

Significance: This outbreak revealed a lack of food safety knowledge and the importance of properly consulting with public health agencies to prevent future outbreaks.

T12-04 Impact of Prospective Whole Genome Sequencing on Multi-jurisdictional *Salmonella* Outbreaks Associated with Frozen Wheated Bread Products in Canada

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Introduction: Frozen raw breaded chicken products (FBRCP) are a risk factor for salmonellosis, however, traditional laboratory sub-typing methods lack the discriminatory power to identify outbreaks of common *Salmonella* serotypes associated with FBRCP and to confirm a microbiological association between food and illnesses.

Methods: Between May 2017 and December 2018, 170 multi-jurisdictional *Salmonella* outbreaks with poultry associated were investigated. Of the 12 (80%) outbreaks associated with FBRCP, there were 378 cases, 72 hospitalizations, and two deaths with an estimated burden of illness of 10,101. A link between poultry and the outbreak was suspected.

Significance: WGS has improved the detection of *Salmonella* outbreaks investigated associated with FBRCP. These investigations have led to public communication, food recall warnings, and a statement from the Canadian Council of Chief Medical Officers of Health emphasizing the importance of handling and preparing poultry.

T12-05 Presence and Identification of Campylobacter spp. in East Tennessee Rivers

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Introduction: Campylobacter spp. are one of the main causes of bacterial gastroenteritis in the United States. Campylobacteriosis is commonly associated with the consumption of poultry, meat, and raw milk, however, following a previous study, researchers quantified populations of Campylobacter spp. within one location of East Tennessee.

Purpose: The purpose of this study was to better understand the spatiotemporal distribution of Campylobacter spp. and further elucidate surface water as a potential source for campylobacters by sampling multiple sites of the Tennessee River.

Methods: Samples (n=267) were collected from four public access river sites within East Tennessee for one calendar year (October 2017 to November 2018). Water samples (10 liters) were collected from each site using five liters of water for each collection. All presumptive positives (n=246) were confirmed using PCR with targets for Campylobacter spp., Campylobacter coli and Campylobacter jejuni.

Results: From the 244 presumptive positive samples, 158 were confirmed as Campylobacter spp. and five as *C. coli*. All collection sites were associated with positive Campylobacter spp. Positive samples were 30% (72/240) of site 1, 79% (198/250) of site 2, 32% (89/278) of site 3, and 77% (58/76) of site 4. Five water samples were confirmed as *C. coli* from site 2. The results were confirmed by the laboratory.

Significance: This study may inspire targeted interventions of the gut microbiome through food modifications to reduce the overall prevalence of antibiotic resistance.
T12-08 Viability-linked Metagenomic Analysis of the Disposable Glove Microbiome
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Introduction: Previous studies on new unused disposable gloves (DG) employed in healthcare have shown considerable microbial contamination. Metagenomic analyses of gloves utilized in food processing facilities has shown cross-contamination potential with various foodborne pathogen indicators including listeria spp. and verocytotoxin-producing E. coli (VTEC). Differences in microbial profiles are observed between inside and outside of gloves. This contrasts with extremes of greater than three CFU/glove in published reports. It was determined that the optimal DG profile of non-detect for glove sampled, reflecting manufacturing steps. Geometric mean TVC for outside surfaces were one log CFU/glove while inside surfaces were 1.2 log CFU/glove.

Methods: A total of eight different genera were found in the metagenomic analysis of the three different gloves associated with both inside and outside surfaces. Using pooled metagenomic data it was determined that microbial profiles were different for inside and outside surfaces for each glove sampled, reflecting manufacturing steps. Geometric mean TVC for outside surfaces were one log CFU/glove while inside surfaces were 1.2 log CFU/glove. This contrasts with extremes of greater than three CFU/glove in published reports. It was determined that the optimal DG profile of non-detect for all of the microbial groups tested, including both inside and outside of gloves is possible but unless carefully controlled, could represent a safety hazard.

Results: This is the first known report of its kind with test methods trialed that examines the glove-microbiome indicative of DG manufacturing that can allow survival of potential microbial contaminants on both inside of glove (user-relevant) and on inside food contact surfaces.

Purpose: Limited microbial information is available on DG utilized in food processing service or in new or in production environments. An Eagle Protect PBC risk reduction program required establishment of baseline information in order to create food relevant test methods & standards.

P1-01 Sterilization of Food Contact Surfaces Using Chlorine Disinfectants to Control Planktonic Cells and Biofilms of Salmonella spp.

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Introduction: Salmonella infection is a frequently occurring foodborne illness worldwide. Contamination on equipment in production lines by Salmonella jeopardizes the microbiological quality of finished products. Chlorine disinfectants such as chlorine dioxide (ClO2) and sodium hypochlorite (NaOCl) are frequently used for reducing foodborne pathogens effectively. Therefore, these disinfectants can be applied to inhibit Salmonella spp. on food-contact surfaces.

Purpose: The current study examined the efficacy of two disinfectants (ClO2 and NaOCl) against Salmonella spp. for the reduction of biofilm formation on food contact surfaces.

Methods: First, three isolates each of Salmonella Enteritidis, Kentucky, and Typhimurium were selected and inoculated for 24 h. We tested planktonic cells using ClO2 (30 to 40 and 30 to 180 ppm for clean and dirty conditions, respectively) or NaOCl (50 to 175 and 300 ppm for clean and dirty conditions, respectively) for one or five min. After forming biofilms, four food-contact surfaces (stainless steel, silicon rubber, and plastic coupon) were treated with disinfectants (ClO2 at 10 to 100 ppm or NaOCl at 50 to 150 ppm) for one or five min.

Results: When these disinfectants were applied to Salmonella spp., there were no differences among bacteria. In planktonic cells, there was different sterilizing power between clean and dirty conditions at same concentration, and by treatment time as well. The biofilm of Salmonella on food-contact surfaces was decreased rapidly when ClO2 concentration (four and five-log reductions for one and five min at 100 ppm, respectively) or NaOCl concentration (three and 3.5-log reductions for one and five min at 300 ppm, respectively) increased.

Significance: Salmonella was effectively reduced at lower concentrations of ClO2 than NaOCl in both planktonic and biofilm cells. This result suggests that ClO2 is more effective for inhibition of Salmonella contamination thorough food manufacturing processes than NaOCl.

P1-02 Validation of the RapidChek Select Salmonella Test Method for the Detection of Salmonella species on 12” by 12” Stainless Steel Environmental Surfaces

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Introduction: Raw meat and poultry are important vehicles of transmission of Salmonella. Control measures, like environmental monitoring, implemented along food production lines in meat and poultry abattoirs help reduce the levels of Salmonella in these matrices; however, microbiological testing retains a key role in preventing food borne salmonellosis.

Purpose: To validate test method stainless steel surfaces by demonstrating equivalent performance to the FDA BAM reference method for the detection of Salmonella spp. in environmental surfaces.

Methods: Stainless steel surfaces (7.5” x 12” test method) and 4” x 4” (FDA BAM method) were inoculated with Salmonella derby both with and without Citrobacter freundii. For each matrix and method, 20 low-level inoculated, 5 high-level inoculated, and 5 negative control samples were tested. After swabbing, sponges were held for 2 hours before enriching. Test method samples were enriched in 225mL primary media, transferred to secondary media, evaluated with test strips and plated on selective agar. FDA BAM reference method samples were enriched in 225mL lactose broth, transferred to TT and RV media, and plated on selective agar following the FDA BAM method Chapter 5.

Results: Without C. freundii, the test method resulted in 18 confirmed positives and the reference method resulted in 15 confirmed positives. With C. freundii, the test method yielded 19 positives while the reference method yielded 10 positives. For both methods, non-spoiled surfaces were negative for Salmonella and all high-spiked samples were positive. Probability of Detection (POD) analysis demonstrated a statistically significant difference at the 5% level in the number of positive samples detected by the test method and the FDA reference method on stainless steel surfaces spiked with Salmonella Derby and C. freundii.

Significance: The RapidChek® SELECT Salmonella test method offers a rapid and reliable tool for testing 12" x 12" stainless steel surfaces for Salmonella species.

P1-03 One Mississippi, Two Mississippis: Phylogenetic Analysis Supports That Salmonella enterica subsp. enterica Serovar Mississippi is Polyphyletic

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Introduction: Salmonella enterica subsp. enterica serovar Mississippi is the 14th most commonly isolated serovar from human clinical cases of salmonellosis in the United States (CDC, 2016). Initial analyses using seven-gene MLST data suggested that this serovar was polyphyletic.

Purpose: The goal of this study was to characterize the phylogenetic structure of Salmonella Mississippi, and to identify the presence of antimicrobial resistance and virulence-associated genes in a collection of Salmonella Mississippi isolates from human clinical infections.

Methods: Whole genome sequence (WGS) data for 65 Salmonella Mississippi isolates were either downloaded from NCBI or were sequenced for this study. Raw sequences were trimmed with trimgem, assembled with SPAdes, and single nucleotide polymorphisms (SNPs) were called with SNPhunter. Sequence information was confirmed using sniR. WGS was used to construct a phylogenetic tree of core SNPs. BLAST searches were performed to identify target genes including antimicrobial resistance (AMR) and virulence-associated genes.

Results: WGS data support the presence of two phylogenetic clades of Salmonella Mississippi. Typhoid toxin genes were unique to Salmonella Mississippi isolated in state A, which clusters with other NTS serovars Salmonella Javanica, Montevideo, Schwarzengrund, and Cuba, all of which are also typhoid toxin-positive. Clade B strains, which clustered with serovar Salmonella Typhi, were all missing avrB, encoding a S. enterica subsp. enterica specific virulence factor, with eight isolates, with eight isolates encoding B-like resistance genes, two isolates encoding genes associated with resistance to sulphonamides and aminoglycosides, and one isolate encoding rifampin resistance genes. There was no relationship between the presence of an AMR gene and the clade that the isolate was in.

Significance: Using Salmonella Mississippi as an example, our study highlights the public health importance of characterizing polyphyletic serovars, as the distribution of virulence-associated genes may enable select clades within a polyphyletic serovar to cause a more severe illness.
P1-04 Heat Inactivation of Listeria monocytogenes on Pecans, Macadamia Nuts, and Sunflower Seeds
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Introduction: Listeria monocytogenes is a major concern for the food industry in RTE foods. In the last two years, large-scale recalls occurred with contaminated sunflower seeds and macadamia nuts that triggered product withdrawals. These recent events stress the importance of understanding L. monocytogenes ability to survive these low survival temperature conditions.

Purpose: This project was undertaken to determine the kinetic parameters of thermal inactivation of L. monocytogenes on pecans, macadamia nuts and sunflower seeds subjected to heat treatments simulating industry processes.

Methods: Heat treatments (40–60°C) were performed, and samples were plated using standard microbiological methods. Average count data were fit to a log-linear model and thermal death kinetics were calculated.

Results: On pecans, the model was fit to a log-linear model and thermal death kinetics were calculated.

Significance: The findings from this study will contribute to assess the effectiveness of heat treatment for the control of Listeria monocytogenes on pecans and sunflower seeds.

P1-05 Culture Supernatants of Lactobacillus plantarum Reduces Sporulation, and Biofilm Formation, of Clostridium perfringens by Downregulating Transcription of Agr-like Quorum Sensing Genes
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Introduction: Bacteria form biofilms and spores to protect themselves in the environment. In the Gram-positive bacterium Clostridium perfringens, an Agr-like quorum sensing system regulates transcription of these virulence factors. Compounds from probiotics have been used as antimicrobial agents and to interfere with expression of enforced factors. For this reason, the aim of this work was to test the potential of L. plantarum supernatants to inhibit sporulation and biofilm formation of C. perfringens.

Methods: The minimal bactericidal concentration (MBC) of supernatants (freeze-dried cell-free supernatant) from cultures of L. plantarum was determined by the serial dilution method (SCD) on blood agar plates. For sporulation, spore formation, and biofilm formation, the same isolates were also sequenced by Illumina Hiseq -250 read lengths ranged from 3,589 to 11,721 bp. After assembly, consensus sequences were obtained from SeqSero2 and SISTR for all 24 isolates. The size of two hours data ranged from 201 to 1072 MB (42 to 203 coverage), and the mean read lengths ranged from 3,589 to 11,721 bp. After assembly, consensus sequence predictions were obtained from SeqSero2 and SISTR for all 24 isolates using the CLC Genomic Workbench. The predicted sequences were identical to the corresponding reference sequence.

Results: The quality score and high-quality data percentage were found to decline over the sequencing time and sequences generated within the first two hours were determined to be sufficient for serotype prediction. The size of two hours data ranged from 201 to 1072 MB (42 to 203 coverage), and the mean read lengths ranged from 3,589 to 11,721 bp. After assembly, consensus sequence predictions were obtained from SeqSero2 and SISTR for all 24 isolates using the CLC Genomic Workbench. The predicted sequences were identical to the corresponding reference sequence.

Significance: This pilot study indicated that the prediction using sequencing data generated by the ONT system is comparable to results obtained from traditional methods for determining the thermal death kinetics of microorganisms. Improvements have since been made to the system such as enhanced temperature and time-resolution at the same temperatures for non-fat dry milk powder were found to be 76.3, 55.0, and 25.0 min, respectively. Data and 9.4 min, respectively. The background of the TDT sandwich already offers operational advantages such as dry heating and flexible heating rates. A comparison of the improved TDT sandwich method against traditional methods provides a basis for utilizing a more precise and efficient method for determining the thermal death kinetics of microorganisms.

P1-07 Enterococcus faecium NRRL B-2334 as a Surrogate in Validating Thermal Treatment of Dairy Powders with Different Lactose and Milk Protein Compositions

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Introduction: Enterococcus faecium NRRL B-2334 is a globally accepted surrogate in validating thermal treatment of dairy powders. The purpose of this study was to investigate the thermal resistance of Enterococcus faecium NRRL B-2334 in commercially relevant dairy powders with different lactose and milk protein compositions.

Methods: The minimal inhibitory concentration (MIC) of SLP against two strains of Salmonella was determined by the serial dilution method (SCD) on blood agar plates. For sporulation, spore formation, and biofilm formation, the same isolates were also sequenced by Illumina Hiseq.

Results: The minimal bactericidal concentration (MBC) of supernatants (freeze-dried cell-free supernatant) from cultures of L. plantarum was determined by the serial dilution method (SCD) on blood agar plates. For sporulation, spore formation, and biofilm formation, the same isolates were also sequenced by Illumina Hiseq -250 read lengths ranged from 3,589 to 11,721 bp. After assembly, consensus sequence predictions were obtained from SeqSero2 and SISTR for all 24 isolates. The size of two hours data ranged from 201 to 1072 MB (42 to 203 coverage), and the mean read lengths ranged from 3,589 to 11,721 bp. After assembly, consensus sequence predictions were obtained from SeqSero2 and SISTR for all 24 isolates using the CLC Genomic Workbench. The predicted sequences were identical to the corresponding reference sequence.

Significance: This pilot study indicated that the prediction using sequencing data generated by the ONT system is comparable to results obtained from traditional methods for determining the thermal death kinetics of microorganisms.
P1-10  Whole Genome Sequencing Analysis for Top Seven Shiga Toxin-producing Escherichia coli
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Mehrzad NotScience, Crito.

Introduction: Escherichia coli O157:H7 and six other Shiga toxin-producing E. coli (STEC) serogroups O26, O45, O111, O121, and O145 are often referred to as the top seven STEC. The use of whole genome sequencing to identify, subtype, and distinguish top seven STEC is rapidly growing. The performance of WGS must be examined to ensure appropriate implementation.

Purpose: The purpose of this study is to evaluate the performance of whole genome multi locus sequence typing (wgMLST) when applied to top seven STEC analysis.

Methods: A total of 20 top seven STEC strains and five O157 strains with other Y antigens were selected for this study. All strains were cultured using non-selective agar, and DNA was extracted using the Qiagen UltraClean Microbial kit. WGS was performed using the Illumina MiSeq and raw sequence data were analyzed by wgMLST using BioNumerics software. Allele difference was determined based on the consensus results of assembly-based and -free. Dendrograms were generated using the unweighted pair group method with arithmetic mean (UPGMA) clustering method to show similarity coefficients and allele differences among samples.

Results: The results indicated that wgMLST analysis was able to distinguish E. coli strains with different serotypes. Within the top seven STEC groups, any two strains with different serotypes differed by 468 alleles or more. Serotypes O45:H2 and O111:H12 displayed the highest similarity (86.7% - 90.4%) to each other. O157:H7 strains were well distinguished from other non-O157 strains with only 9.6% allele similarity. Strains of the same serotype had less genetic differentiation; for example, six O157:H7 strains shared at least 98.2% allele similarity (81 allele differences).

Significance: This study indicates that wgMLST is an appropriate method for differentiating top seven STEC strains to the serotype level. The finding will lead to the application of WGS for foodborne pathogen analysis.

P1-11 WITHDRAWN

P1-12 The Relationship between Inactivation and Morphological Damage of Aspergillus flavus Treated by High Hydrostatic Pressure
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Introduction: Foodborne Aspergillus flavus contamination is a major concern for the food industry. Thus, there is an increased research interest in discovering effective treatment methods that preserve food quality and decontaminate food by inactivating A. flavus. High hydrostatic pressure processing (HHP) is considered one of the most promising food preservation techniques and is used for commercial pasteurization of a number of food products.

Purpose: The purpose of this work was to investigate the mechanisms underlying the inactivation of A. flavus under HHP treatment. Methods: The HHP treatment (300 MPa for 2min) was subjected to pressure treatment at 100, 200, 300, 400, 500 or 600 MPa, with a holding time of five minutes at 25°C. After treatment with different levels of hydrostatic pressure, surviving cells were then analyzed by viable colony forming units, cell membrane damage by fluorescent dye propidium iodide (PI) uptake, morphological features by scanning electron microscopy (SEM), and membrane protein changes by SDS-PAGE.

Results: The results showed that a 600 MPa treatment for five min could considerably inactivate A. flavus counts, with increased uptake of propidium iodide (PI) and the number of viable organisms decreasing from 10^5 CFU/ml to no viable bacteria. Morphological damage to the cell wall, cell membrane, and cytoplasmic components by HHP treatments were observed on SEM images. The SDS-PAGE results showed that the protein bands differed between HHP-untreated and HHP-treated A. flavus, in that HHP decreased the protein content and caused partial protein degradation.

Significance: The findings of this study indicate that high pressure treatments for A. flavus by causing morphological changes in the internal and external cellular structures, as well as through membrane damage, cell wall permeability, and membrane protein degradation.

P1-13 Influence of Asymptomatic Escherichia coli Inhabiting the Gut on Inflammation, Cell Proliferation, Oxidative Stress, and Angiogenesis in the Intestine
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Developing Scientific Enterant

Introduction: Even though some Escherichia coli strains live asymptotically in the human gut, they may cause chronic problems from long-term exposure.

Purpose: This study investigated the influence of asymptomatic E. coli inhabiting the gut on chronic intestinal symptoms.

Methods: The concentrations of inflammatory cytokines (IL-1β, IL-6, and IL-8), pro-inflammatory (TNF-a and IFN-g) and prostaglandin E2 (PGE2) were measured in Raw 264.7 cells infected by E. coli strains [E. coli NCPP4107, E. coli NCPP4108, E. coli NCPP4019, and E. coli NCPP4156]. The cells were injected into orally eight-day-old BALB/c mice, and the response factors, the inflammatory reaction, cell proliferation, cell apoptosis, oxidative stress and angiogenesis were investigated in the intestine by histopathological analysis, qRT-PCR, and western blot. A selected E. coli strain [E. coli NCPP14037] which has a greater influence in the intestine, was injected orally into eight-day-old BALB/c mice, and the responses described above were investigated in the intestine. Also, the risk factors for E. coli were analyzed by RNA sequencing.

Results: E. coli had an effect on the production of pro-inflammatory cytokines (IL-6 and TNF-a) and PGE2 in Raw 264.7 cells. Among four strains of E. coli, E. coli NCPP4107 was most likely to have a greater influence on the intestine by regulating the expression of genes (GOAT and CAT) and proteins (IL-6, IL-12, PCNA, COX-2, and iNOS) (P<0.05). In addition, the length of large intestine was shortened in E. coli NCPP14037-treated mice, and this result indicated that the inflammatory reaction occurred in the intestine. The result of RNA sequencing showed that curli, flagella, and fimbrin were highly expressed in E. coli NCPP14037.

Significance: Although some asymptomatic E. coli inhabits the intestine, its long-term exposure can cause inflammation, cell proliferation, oxidative stress, and angiogenesis in the intestine.

P1-14 Sporulation of Plantonlic and SSEs Clostridium perfringens in Response to Chemical and Oxidative Stress
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Introduction: Clostridium perfringens is a major human pathogen causing genteritotoxicosis in humans and has an ability to form spores and biofilms for environmental persistence and disease biofilm formation. Biofilms and spores may restrict antimicrobial penetration and contribute to the exacerbation of bacterial infections.

Purpose: This study aims to compare the chemical and environmental resistance properties of C. perfringens vegetative cells and spores in plantonic and sessile conditions.

Methods: Three strains of C. perfringens (TYJAM-D66, CMM-C80 and SDE-B2-20) were isolated from meat supplied to a school cafeteria. Sporulation rate was determined after 100, 150, 200, and 250 min. Resistance of vegetative cells and spores against disulfiram (0.01% sodium hypochlorite solution, 5% hydrogen peroxide solution, and 80% alcohol) or aerobic conditions (18 and 36°C) after biofilm formation was analyzed.

Results: Sporulation rate of sessile C. perfringens TYJAM-D66 (spores) was about 19% at day five, while sporulation of plantonic C. perfringens was 14% at day two and decreased to 2% at day five. Sporulation rate of sessile C. perfringens-C80 (spores) and SDE-B2-20 (spores) was only up to 0.26 and 0.67%, respectively, at day seven. When exposed to aerobic conditions, TYJAM-D66 vegetative cells were decreased by 1.35 to 1.70 log, CMM-C80 by 5.13 to 5.36 log, and SDE-B2-20 by 5.59 to 5.67 log. Spores decreased by 0.92 log. After the treatment of sodium hypochlorite, 9.2% of TYJAM-D66 planktonic cells survived, while vegetative cells and spores on biofilm showed 53.62 and 83.39% survival, respectively. Comparative genomic analysis of the isolates was performed to understand the differences in growth rates and biofilm formation.

Significance: This study presents a new methodology for analyzing the relationship between the biofilm formation, sporulation, and inhibition of C. perfringens spores.

P1-15 Development of Scientific Enterant

Poster

Journal of Food Protection Supplement

P1-14 – P1-16

Poster

Journal of Food Protection Supplement

P1-10 – P1-13

Poster

Journal of Food Protection Supplement

P1-16

Poster

Journal of Food Protection Supplement

P1-11

Poster

Journal of Food Protection Supplement

P1-12

Poster

Journal of Food Protection Supplement

P1-13

Poster

Journal of Food Protection Supplement

P1-14

Poster

Journal of Food Protection Supplement
P1-17 - P1-21 WITHDRAWN

P1-18 Inactivation of Salmonella enterica and Enterococcus faecium in Cumin Seeds Using Gaseous Ethylene Oxide

Long Chen, Xinya Wei, Soon Kiat Lau and Jayamkondan Subbiah
University of Nebraska-Lincoln, Lincoln, NE, 1 and University of Delaware, Newark, DE, 2

P1-18 WITHDRAWN

P1-19 The Prevalence and Characteristics of Acid-resistant E. coli in Foodborne and Clinical Isolates in Korea

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P1-20 PESS

P1-17 Assessing Efficiency of Vacuum-assisted, Low-temperature Steam Decontamination of Salmonella spp., Listeria monocytogenes, Shiga Toxin-producing Escherichia coli, and a Surrogate (Pedococcus acidilactici) on Raisins

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P1-17 WITHDRAWN

P1-21 The Prevalence and Characteristics of Acid-resistant E. coli in Foodborne and Clinical Isolates in Korea

Soo Hwan Suh, Myungho Jeong, Gun Woo Nam, Eunjoo Jeong, Ha Jung Hong, Byung Hak Kang, Mi-Gyeong Kim and Hyo-Sun Kook
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P1-22 Ethylene Oxide Fumigation for Inactivation of Salmonella enterica and Enterococcus faecium nrrl B-2354 in Black Pepper

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P1-22 WITHDRAWN

P1-23 Behavior of Shiga Toxin-producing Escherichia coli, Salmonella spp., and Listeria monocytogenes on Dried Apricots Made with and without Sulfur Dioxide

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P1-23 WITHDRAWN
Roasted Coffee Beans on individual Brazil nut samples. This study suggests that non-irradiated roasted coffee beans may be effective in reducing A. flavus and A. parasiticus with less physiochemical changes and sensory evaluation.

P-1-26 Inactivation of Salmonella and Surrogate Bacteria on Brazil Nuts and Pine Nuts Exposed to Commercial Propylene Oxide Processing Conditions

Introduction: Fumigation by gaseous propylene oxide (PPO) has been demonstrated to reduce Salmonella enteritidis on some tree nuts. A surrogate whose inactivation pattern is similar to Salmonella is needed to avoid direct handling of Salmonella in process validation within the processing facility.

Methods: To determine the suitability of Enterococcus faecium and Pediococcus acidilactici as potential surrogates for indicating Salmonella inactivation on whole Brazil and pine nuts subjected to PPO processing in accordance with the USEPA label instructions.

Results: The populations of A. flavus and A. parasiticus irradiated by UV-C on both surfaces were significantly decreased (P<0.05) after two hours. Also in sensory evaluation, there was no difference in color, appearance, texture, and overall sensory evaluation showed significant differences (P<0.05) after two hours.

Significance: This study suggests that non-irradiated roasted coffee beans may be effective in reducing A. flavus and A. parasiticus with less physiochemical changes and sensory evaluation.

P-1-27 Determining the Perceived Cost of Implementing a Vomit Clean-up Plan

Angela Fraser and Kathryn Boys1

Introduction: Evidence suggests an organization’s willingness and commitment to implementing food safety practices are affected by perceptions regarding the challenges and costs of doing so. In 2009, the United States Food and Drug Administration added a regulatory provision to the Food Code requiring food service establishments to have body fluid clean-up procedures in place.

Purpose: To determine the perceived cost of implementing a vomit clean-up plan in a foodservice operation.

Methods: Foodservice workers from retail, commercial, and institutional settings who attended one of 146 educational sessions offered by 54 educators were surveyed.

Results: A total of 388 participants completed the survey. The incidence of vomit events reported in the past three months varied considerably by setting (1.4% for retail vs. 9.0% for institutional foodservice operations). Among the 54 respondents (13%) who reported their establishment had an event, most samples were taken at least once a week, with 10% of samples coming from events requiring food service to be stopped or expedited to clean up (53%). There were no differences in the amount of time spent cleaning up by establishment type (P>0.05). However, on average, less than 1.5 to 4.5 hours would be required for cleanings supplies, training, and clean-up time, and cost of replacing any food. These results varied significantly from the actual time and cost estimates of implementing currently recommended vomit clean-up procedures. Those working in commercial environments had a much better understanding of the costs associated with addressing a vomiting event from other settings.

Significance: The perceived cost of implementing a vomit clean-up plan should be integrated into existing food safety training to increase adoption.

P-1-28 Comparison of Food Establishment Characteristics between Viral and Bacterial-caused Foodborne Outbreak Reports Submitted to the National Environmental Assessment Reporting System

Adam Kramer

Introduction: Food establishment characteristics are voluntarily reported to the CDC’s National Environmental Assessment Reporting System (NEARS) by state and local health agencies. Understanding the differences between establishments that have bacterial or viral outbreaks, such as those in food safety policies, practices, and other characteristics, can enable development of interventions to prevent foodborne illness.

Methods: The purpose of this study was to evaluate characteristics between establishments that had bacterial versus viral outbreaks. Multivariate methods were used to compare characteristics between foodborne outbreak reports submitted to NEARS for a bacterial or viral agent. Data was extracted from NEARS for outbreaks that occurred during 2014-2017. Variables were analyzed using chi-square and t-tests.

Results: The dataset contained data on 349 food establishments. We found that establishments that had bacterial outbreaks differed significantly (p<0.05) from establishments that had viral outbreaks on several characteristics. For example, establishments with bacterial outbreaks, compared to viral outbreaks, had:
- 7.0% fewer of observed cross-contamination from raw to ready-to-eat food
- 12 fewer cleaning policies
- 58 lower odds of having wiping cloths stored in sanitizer solution
- 51 lower odds of using a mechanical washer
- 48 fewer of having a written policy requiring foods to be kept cold
- 59 lower odds of having foods kept at recommended temperature
- 59 lower odds of using a commonly validated procedure to rapidly cool food

Significance: These data suggest that there are systematic differences in food safety policies and practices between restaurants with bacterial and viral outbreaks. For example, these data suggest that restaurants with bacterial outbreaks have reduced refrigeration capacity to maintain food at recommended temperatures, a factor often associated with growth of bacteria. Information like this is valuable in developing interventions to prevent foodborne illness.

P-1-30 Perceived Benefits and Barriers to Implementation of a Traceability System in School Foodservice Establishments in North Carolina, South Carolina and Georgia

Angela Fraser and Kathryn Boys1

Introduction: Significant logistical, contractual, and food safety challenges exist when sourcing food for school foodservice operations (SFOs) from small to medium-sized (SMS) farms. Each step from ordering to packaging to service can present significant barriers for traceability of the product from farm to table.

Purpose: The aim of this exploratory study was to determine the perceived benefits and barriers of SFOs in implementing a traceability system for produce purchased by SFOs from SMS farms.

Methods: Qualitative research results from an earlier study and results from other previously published studies that explored institutional foodservice procurement were used to design a web-based survey. Items measured current buying activity; buying directly from SMS farms; benefits and challenges of buying from SMS farms; system implementation and costs; and organizational characteristics. The response format included three- and five-point Likert scales, closed-choice items, and yes/no scales. The instrument was pilot with five SFO buyers before dissemination. The survey was administered to 411 SFO buyers from school foodservice establishments North Carolina, South Carolina, and Georgia.

Results: The response rate was 29.9% (n=122). Reported benefits included 58% lower odds of having wiping cloths stored in sanitizer solution and 25% fewer refrigeration units.

Significance: Although a seemingly simple concept to implement, significant challenges, such as cost and technology, need to be addressed before traceability systems are implemented in SFOs.
Purpose: To determine if STEC acid resistance correlates with growth and death of STEC in vegetable fermentations in a model laboratory fermentation system.

Methods: We examined seven selected STEC strains to determine growth rates in laboratory media at various pH values (pH 4.2 to 6.8) and measured survival in simulated stomach acid (SA) challenges. Two strains, 8201 (acid-sensitive) and 2141 (resistant) were selected for further analysis of acid resistance in lactic acid solutions under aerobic and anaerobic conditions at pH 3.3 in cucumber juice (CJ) and fermented cucumber juice (FC). We then compared the growth and death of B201 and B214 in a model fermentation system to determine survival during competitive growth with Lactobacillus plantarum MR037. Experiments were done in triplicate.

Results: STEC strains fell into statistically distinct groups based on acid sensitivity, although there was variation based on the type of acid challenge. D/E strain survival was comparable to 2141. When enriched immediately, D/E and comparable recovery was seen when enriched immediately. When the D/E sponges held for 72 hours, there was a 50% reduction in survival at refrigerated temperatures following sampling of inoculated stainless-steel surfaces.

Significance: The results indicate that the acid resistant phenotype with stationary phase cells may not accurately predict STEC survival in a vegetable fermentation system. Further research may be needed to determine factors influencing STEC die-off in vegetable fermentations.

P1-32 A Buffer Capacity Model for Predicting pH Changes Due to Addition of Low Acid Ingredients in Acidic Foods

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Introduction: The pH of acidic foods depends on undefined and complex buffering of ingredients and is critically important for regulatory purposes (21 CFR part 114) and food safety.

Purpose: Our objective was to develop and validate a method for predicting the addition of acid ingredients to the pH of low acid foods containing acetic acid formulations typical of salad dressing products.

Methods: A variety (n=20) of acetic acid formulations with low acid ingredients (garlic powder, onion powder, mustard flour, spices, etc.) were titrated individually and in combination at water concentrations typical of dressing products. Titration curves (pH 2 to 12) were generated with NaOH and HCl as titrants. The curves were used to generate buffer capacity and acidity titration curves. A model of pH and concentration values for undefined buffering ingredients was estimated by curve fitting (MATLAB algorithm). For each ingredient the matrix was then used to predict pH using a custom MATLAB algorithm. All pKa values were adjusted for two percent NaCl.

Results: The pH prediction model was validated using a set of 14 samples with various acetic acids in organic acids (BC=0.05) compared to acetic acid (BC=0.24) for the pH range (pH 2 to 12), resulting in small (n=14) lactic acid with an average of 0.77 ± 0.02 pH units. For each ingredient the pH of the titration was used to estimate the buffer capacity and acidity titration curves of the ingredient.

Conclusions: BC>0.05) in the survival (CFU/ml) of B201 or B241 in competition with MOP3 with regard to survival time, pH, or total and protonated lactic acid concentration.

Significance: The pH models can be used to estimate buffering and therefore the concentrations of low acid ingredients that prevent pH rise above 4.6. Using the model to quantify the effect of ingredients on pH may benefit regulatory agencies and manufacturers in assessing product safety.

P1-33 The Ability of Collection Solutions to Maintain the Viability of Listeria monocytogenes after Sampling Inoculated Stainless-Steel Surfaces

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Introduction: Routine environmental monitoring is important for managing pathogens in food production facilities. Environmental samples are often held at refrigerated temperatures prior to processing in the laboratory. Often times, samples are sent overnight to offsite laboratories and typical holding times are 24 to 48 hours. If sample delivery is delayed, this may extend to 72 hours, often leading to re-sampling.

Purpose: The objective of this study was to evaluate the ability of collection solutions to maintain the viability of L. monocytogenes after sampling and holding as well as during transport to offsite laboratories.

Methods: L. monocytogenes populations in taco meat subjected to a variety of cooling methods were monitored for a period of 24 hours to 9 days after sampling. Three hundred seven CRFs were submitted during the review process. Of these, 112 were accepted as submitted or as amended and 34 were withdrawn. VSP used the final Operations Manual and Construction Guidelines in April 2018 with stronger food safety provisions aligned with the current U.S. Food Code and International Safe Port Code.

Significance: The VSP revision model is to prevent the introduction of acute gastroenteritis (AGE) into the United States from cruise ships sailing from foreign to domestic ports. While ships carry approximately 12 million passengers per year in and out of U.S. ports, AGE cases on cruise ships are relatively infrequent.

P1-35 Evaluating the Impact of Cooling Techniques on Escherichia coli Populations in Taco Meat

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Introduction: The Centers for Disease Control and Prevention (CDC) Vessel Sanitation Program (VSP) bases operational and construction inspections on two foundational VSP guidance documents: the Operations Manual and the Construction Guidelines. Worldwide, these documents are considered gold standards in cruise ship sanitation and construction.

Purpose: VSP used a cooperative model to revise these important documents with cruise industry stakeholders; this method can serve as a model framework for other collaborative efforts between government, industry, and other stakeholders.

Methods: VSP developed a new change request form (CRF) and instructions. As part of the revision process, stakeholders, including cruise lines, shipyard representatives, passengers, vendors, outfitters, consultants, manufacturers, and international partner nations working with the CDC, VSP, and stakeholders held 14 days of in-person meetings and two web-based meetings between April 2015 and October 2016 to review the submitted CRFs. During these meetings, stakeholders reviewed their changes and discussed the reasons and evidence for requesting the changes. During the group discussion, VSP took one of three actions: accepted CRF as submitted, accepted CRF with amended, or did not approve the CRF.

Results: Three hundred seven CRFs were submitted during the review process. Of these, 112 were accepted as submitted or as amended and 34 were withdrawn. VSP released the final Operations Manual and Construction Guidelines in April 2018 with stronger food safety provisions aligned with the current U.S. Food Code and International Safe Port Code.

Significance: The VSP revision model is to prevent the introduction of acute gastroenteritis (AGE) into the United States from cruise ships sailing from foreign to domestic ports. While ships carry approximately 12 million passengers per year in and out of U.S. ports, AGE cases on cruise ships are relatively infrequent.

P1-36 Strengthening Food Safety Provisions on Cruise Ships: The Vessel Sanitation Program Cooperative Revision Model

Zhen Jia, Changcheng Li, Ting Fang and Jinquan Chen
Fujian Agriculture and Forestry University, Fujian, China

Introduction: Limited information is currently available concerning the effect and potential application of e-polylsine hydrochloride (e-PHL) on growth and thermal inactivation of Listeria monocytogenes in Fish balls.

Purpose: The purpose of this study was to investigate the effects of e-PHL for inhibiting the growth and enhancing thermal inactivation of L. monocytogenes in fish ball. The broth used to hydrate a sponge is important to the recovery of injured bacteria. Counts were performed at one, three, five, seven, nine, 11, 13 and 15 days of incubation in duplicate. Bacterial growth was evaluated using blood agar plates incubated at room temperature for 72h.

Results: Arcobacter was able to grow in the three different matrices evaluated; the chlorinated matrix showed less growth. Also, the survival of this bacteria under frozen conditions seemed to be present in water as well as when incubation temperature increases. The number of bacteria decreases with time, but bacteria did not disappear completely.

Significance: Results obtained in Vitro show that water for human consumption may represent a risk for health, but the survival of the bacteria in this environment might be different. Further research is recommended.

P1-37 Predictive Modeling of the Effect of e-Polylysine Hydrochloride on Growth and Thermal Inactivation of Listeria monocytogenes in Fish Balls

Fish balls, with or without e-PHL, were inoculated with L. monocytogenes and incubated at 3.4, 8.2, 12 or 16°C for growth studies, or heated at 60, 62.5, 65, or 67.5°C for thermal inactivation tests. The growth curves were fitted to the Huang primary model, and the Huang and Rakowsky square-root models (SRM) were used as the secondary models. The survival during heating was analyzed with a linear model.

Results: The results showed that, while the lag time of L. monocytogenes was affected by both e-PHL concentration and temperature, the specific growth rate was unaffected by e-PHL. Under the same conditions, a tenfold increase in the lag time would be expected for every 0.5°C decrease in incubation temperature. Survival of L. monocytogenes was significantly reduced by an increase in temperature or e-PHL. The thermal value of L. monocytogenes was 5.7°C and the e-PHL value was 1642°C.

Significance: e-PHL can be used to enhance thermal inactivation and control the growth of L. monocytogenes during storage. The models can be used to design more effective thermal processes and assure the inhibition of L. monocytogenes, thus improving food safety.

References:

**Introduction:**
The incidence of poverty has increased over the years, especially in rural areas of developing countries, thus affecting learners in poverty. The National School Nutrition Programme (NSNP), part of an integrated Nutrition Programme (NP), was introduced to reduce the incidence of food insecurity as well as the child mortality rate in South Africa. Even though the programme had good intentions it also poses a number of challenges which still need to be resolved.

**Purpose:**
Previous studies highlighted that among the challenges facing the NSNP is infrastructure; food preparation facilities and storage rooms are lacking. Lack of a digital food safety monitoring system may lead to the potential health and safety temperatures in real time. Anytime a temperature read below a threshold of 135°F, the serving line team member was alerted and guided through predefined corrective action steps to resolve the deviation.

**Methods:**
The methods and hygiene operating prerequisites of 98 randomly selected schools were used using an inspection checklist.

**Results:**
The results show that although the focus was given to basic infrastructure such as locations (<50%) and structures (<40%), other requirements such as maintenance of such structures (<50%), food preparation equipment (50%) and hygienic operating prerequisites still needed attention (<40%).

**Significance:**
The lack of these requirements might increase the possibility of food contamination and reduction of the shelf life of the food served to learners. With the number of different microorganisms associated with foodborne disease status, programs within the NSNP may pose latent risks for children.

**Conclusion:**
In summary, South African learners are exposed to food safety risks. A digital food safety management system. Handheld digital thermometers were used to obtain and upload temperatures in real time. Anytime a temperature read below a threshold of 135°F, the serving line team member was alerted and guided through predefined corrective action steps to resolve the deviation.

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**P1-39 Implementation of Novel Technology and Its Implications for a Food Safety Culture in University Dining Halls**

Savana Everhart, Eric Moore1, Lee-Ann Jaykus1, and Benjamin Chapman1

1North Carolina State University, Raleigh, NC, 2Industry, West Chester, PA

**Introduction:**
A positive food safety culture is important in food establishments, especially when thousands are served daily. Buffet facilities have been implicated as a common locations for foodborne illness outbreaks due to their association with improper food storage; therefore, it is imperative staff have the tools, value correctly monitoring temperatures, and can make process changes if necessary.

**Purpose:**
The purpose of our work was to assess the overall food safety behaviors and culture of surrounding the implementation of temperature monitoring as it relates to active managerial control (AMC).

**Methods:**
Daily paper logs for recording holding temperature of foods that need time and temperature control for safety (TCS) foods were replaced with a digital online platform. Pathfinder was developed for digital data collection, which can be monitored in real-time. Anytime a temperature read below a threshold of 135°F, the serving line team member was alerted and guided through predefined corrective action steps to resolve the deviation.

The behaviors of serving line team members were observed. Decision-making by line (n=95) and executive management (n=20) on food safety approaches was also observed.

**Results:**
Overall, 90% of serving line team members reported the equipment was easy to use when completing daily food safety checklists. The thresholds supported more consistent AMC and awareness from the entire staff. Improvements in collecting daily information for line and executive management included being able to easily find data, ensuring proper training for serving line team members, and no longer storing paper records. Barriers identified were weak communication between all employment levels, lack of engagement, and technology aversion.

**Significance:**
Using this equipment allows for AMC since the system records, analyzes, and reports data. The shared data on the impact of electronic temperature monitoring equipment on decision-making overall answered questions on efficiency and food safety.

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**P1-40 Characterization of Salmonella enterica Isolates from Selected United States Swine Feed Mills by Whole-genome Sequencing**

Gabriela Magossi1 and Valentina Trinita1

1Kansas State University, Food Science Institute, Manhattan, KS. 2School of Food Science, Manhattan, KS

**Introduction:**
Recent multistate foodborne outbreaks have highlighted the importance of using rapid methods to trace contaminants in the food chain and the need to implement these tools in accordance to new regulatory framework. Genomic techniques, such as whole-genome sequencing (WGS), allow predicting correctly) was used to select the optimal model.

**Methods:**
Using this equipment allows for AMC since the system records, analyzes, and reports data. The shared data on the impact of electronic temperature monitoring equipment on decision-making overall answered questions on efficiency and food safety.

**Results:**
A positive food safety culture is important in food establishments, especially when thousands are served daily. Buffet facilities have been implicated as a common locations for foodborne illness outbreaks due to their association with improper food storage; therefore, it is imperative staff have the tools, value correctly monitoring temperatures, and can make process changes if necessary.

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Overall, 90% of serving line team members reported the equipment was easy to use when completing daily food safety checklists. The thresholds supported more consistent AMC and awareness from the entire staff. Improvements in collecting daily information for line and executive management included being able to easily find data, ensuring proper training for serving line team members, and no longer storing paper records. Barriers identified were weak communication between all employment levels, lack of engagement, and technology aversion.

**Significance:**
Using this equipment allows for AMC since the system records, analyzes, and reports data. The shared data on the impact of electronic temperature monitoring equipment on decision-making overall answered questions on efficiency and food safety.

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**P1-41 Prevalence of Salmonella and Escherichia coli in Selected United States Swine Feed Mills and Assessment of Potential Contamination Risk Factors**

Jugen M Manyatsa, Cassandra Jones1, T. G Nagapuri1, Randall Phedor1, Jason Woodward2, Elisabeth Lamberti3 and Valentina Trinita1

1Kansas State University, Food Science Institute, Manhattan, KS. 2Kansas State University, Manhattan, KS. 3Center for Applied Food Security and -Biotechnology (CAFSaB), Central University of Technology, Bloemfontein, South Africa

**Introduction:**
Salmonella and Escherichia coli has been detected in animal feeds and pork products, raising questions about the role of feed and feed ingredients in infected animals. Poorly cooked food, spoilage, and cross-contamination are potential sources of Salmonella. Mining pathogen clusters can help identify potential risk factors associated with their prevalence in food processing facilities. Genomic techniques, such as whole genome sequencing (WGS), allow predicting correctly) was used to select the optimal model.

**Methods:**
A total of 135 samples from selected sites including floors, equipment, shoe surfaces, and feed, were collected during fall 2018 in six swine feed mills. Whole-genome sequencing -Salmonella enterica-and SNP (single nucleotide polymorphism) clusters. No matches with clinical samples were found, while strains were matched with other environmental isolates from the NCBI database. AMR genes showed that 40% of the strains belonged to at least one antimicrobial resistance gene including those encoding for tetracycline, penicillin, amoxicillin, chloramphenicol, and β-lactam resistance.

**Significance:**
Our analysis shows the presence of pathogenic Salmonella enterica in feed mills and underscores the potential role of these environment as a pathogen entry route into the human food chain.

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**P1-42 Knowledge Discovery from Epidemiological Data for Assisting Foodborne Outbreak Investigation**

Dandan Tao and Hao Feng

University of Akas in Urbana-Champaign, Urbana, IL

**Introduction:**
Consumption of foods contaminated by human pathogens is responsible for a large portion of human illnesses. Understanding the interplay between pathogens and foods is critical for outbreak investigation.

**Purpose:**
The purpose of this study was to discover food-pathogen relationships from historical outbreak data.

**Methods:**
Epidemiological data on foodborne disease outbreaks reported to the CDC from 1998 through 2017 was analyzed with network analysis (NA) methodology. The food and pathogen names were used to identify the most important and related foods and pathogens to the outbreak. The NA identified food groups and pathogens that were most closely related to each other. For each food or pathogen, relatedness was measured using a pattern vector where higher values indicated greater relatedness. The data documents a relatively high prevalence of Salmonella and E Escherichia coli at different sites within mills and identify potential risk factors associated with their prevalence in food processing facilities. Genomic techniques, such as whole genome sequencing (WGS), allow predicting correctly) was used to select the optimal model.

**Results:**
From the total of 135 samples obtained, eleven (8.2%) contained Salmonella spp. and 31 (23.0%) contained E. coli. All sampled mills had at least one site confirmed to contain Salmonella spp. or E. coli, with six sites confirmed for both. Floors had the highest number of confirmed samples across all sampling sites, suggesting that employee foot traffic may be a biosecurity risk. Survey responses support that mills with higher bacterial loads are older, suggesting that age of the mill may be a risk factor for enteric pathogen contamination. Other risk factors evaluated did not appear to relate to Salmonella or E. coli prevalence. In addition, it was noted that not all mills have microbiological testing on site.

**Significance:**
The data documents a relatively high prevalence of E. coli and Salmonella in United States food mills. This information could be used to evaluate risk and design mitigation strategies.
Introduction: The outbreak of Salmonella food poisoning in South Korea has a long history. The numbers of outbreaks have been variable in recent years, however, its rate is still higher among bacterial food poisoning.

Purpose: The purpose of this study was to evaluate the relationship between the variation of climate factors and the outbreaks of Salmonella food poisoning in South Korea.

Methods: We used time series data on food poisoning statistics from the Ministry of Food and Drug Safety of Korea and from Korea Statistics. The climate data was measured from the Korea Meteorological Administration’s Weather System from the Korea from the year 2002 to 2017, and Pearson’s correlation analysis was employed to establish the relationship between the climate factors and the outbreaks of Salmonella food poisoning in South Korea.

Results: The numbers of annual outbreaks of Salmonella food poisoning have been declining with no significance, and Salmonella spp. was the most common causative pathogenic bacteria. Salmonella food poisoning occurred most frequently in summer, followed by spring, autumn, and even in winter. The infection rate was found to be positively correlated with the ambient temperature, the highest annual temperature, the lowest annual temperature, precipitation, the number of days with rainfall, and humidity (P<0.05).

Significance: It should be noted that differently from earlier years, Salmonella food poisoning occurred even in winter in the 2000s. Climate change, especially warming climate, is supposed to have affected the pattern of the food poisoning outbreaks.

P1-45 A Large Outbreak of Salmonella Food Poisoning Due to Egg White and Possible Preventive Measures

Introduction: Outbreaks of Salmonella food poisoning are a problem in many countries. Recently a large outbreak of Salmonella food poisoning occurred at food-service establishments including schools in South Korea.

Purpose: This study attempts to summarize information relating to the incident, together with advice on how the outbreak may be reduced or prevented.

Methods: In September 2018, over two thousand students and staff were hit with gastrointestinal symptoms, including hospital admissions after school lunch. An epidemiological investigation by the Korean government was carried out to identify the source of infection.

Results: A total of 21,112 students and staff from more than 55 schools across the nation reported symptoms of food poisoning. It was found that over 70% of the students who became ill were having the school lunch with a cake that included egg white. The implicated cake was supplied by a major food supplier. The cake was delivered to 175 schools, two kindergartens, 12 restaurants and one children’s center. An epidemiological investigation revealed that Salmonella Thompson was isolated from samples collected from the implicated cake.

Significance: The probable source of the outbreak can be suspected as a group of eggs. In the Korean egg industry, a portion of eggs with cracked shells are used for manufacturing baked goods. Since it is difficult to be free of Salmonella, it is recommended that all cracked eggs and their products using for manufacturing baked goods in South Korea are not to have any certification related to handling eggs. Periodic education of the egg suppliers/sellers for egg safety is necessary, such as hygiene of farmers and workers in the egg industry, including distributors and retailers.

P1-46 Occurrence of Cyclospora cayetanensis in Florida, 2014–2018

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Developing Scientist Infrant

Introduction: Cyclospora cayetanensis are often linked to consumption of contaminated fresh produce. Infections may be acquired locally or while traveling to certain tropical areas. The number of cyclosporiasis in Florida is not linked to food poisoning, so the cause for concern.

Purpose: This study evaluated the role of different types of travel among infections with Cyclospora cayetanensis in Florida.

Methods: Cyclospora outbreak data for 2014–2018 was obtained using the Cyclospora National Hypothesis Generating Questionnaire form. Travel was categorized into international, domestic, and non-travelers. Infection rates were calculated using population information provided by the US Census Bureau for 2014 to 2017.

Results: There were 277 cases epidemiologically or clinically linked to Cyclospora, and 44.8% of the cases between 2014 and 2018 were domestically acquired. Among travelers, 16.6% and 26.4% reported domestic or international travel, respectively. Annual travel distribution was similar in most years, except for 2016. For the five years, there were more reported cases in Cyclospora than those who had traveled domestically than internationally. The year 2017 had the highest number of cases, with 109 domestic and 93 international. The infection rates for 2014, 2015, 2016, 2017, and 2018 were Putnam, Indian River, Flagler, Lee, and DiCello respectively, with infection rates of 1.4, 0.68, 1.8, 3.1, and 2.7 per 100,000 inhabitants, respectively.

Annual infection rates in Florida varied from 0.2 per 100,000 in 2014 to 0.5 per 100,000 in 2018, although the highest rate was in 2017 at 0.06 per 100,000.

Significance: These results revealed that in 2014 to 2018, most cases of Cyclospora in Florida occurred among people not reporting international travel.

P1-47 A Systematic Review of Olders’ Food Safety Knowledge and Practices at Home

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Introduction: Older adults are a high-risk population for foodborne illness because of weakened immune systems, chronic diseases, and a resulting increase in complications. In 2017, seniors were estimated to have outnumbered children in Canada, and in the United States, up to 20% of the population will be of retirement age by 2030. Thus, there is a growing concern about food safety as the older population increases.

Purpose: A systematic review was undertaken to identify, characterize, and synthesize the published research on the knowledge, attitudes, and practices of older adults (60+) toward food handling in the domestic setting.

Methods: The study consisted of a comprehensive search strategy, relevance screening, and article characterization, risk-of-bias assessment, data extraction, and analysis. The following databases were searched: CINAHL, PsycINFO, Medline, Embase, and Google Scholar. A total of 57 relevant studies were published between 1996-2018 were identified. Most studies used a cross-sectional design (86%), were conducted by food educators (75%) and in North America (92%). Randomized controlled trials were not identified.

Significance: Gaps in knowledge and practices were identified which could be used to inform future education interventions. Research gaps were also identified, including investigation of consumption of certain high-risk foods (e.g., soft cheeses, raw fish), and storage practices.
caused by an outbreak in the food sector. However, this is not the case in many other countries where Salmonella is a common cause of foodborne illness.

Significance: We demonstrate that MLST is useful for the quick detection and investigation of outbreak isolates and related strains. This makes it an appropriate technique for global surveillance.

P-51 Biosecurity Evaluation and Compliance in Broiler Breeder Farm Units in Northwest Nigeria: Implications for Poultry Farm Workers’ Health and Chicken Meat Consumers

Nurudeen Olalekan Oloso1, Henriette Van Heerden2, Narjol Gonzalez-Escalona2, Nasser Abbas3, Hadeer Alaa El Din3

Introduction: Breeder broiler farms (BBFs) occupy a strategic position in the chicken meat production chain. There are threats due to pathogens but biosecurity measures remain a challenge in these farms.

Purpose: Evaluation of biosecurity compliance levels in BBFs as a measure of risk of infection in the poultry industry, occupational risk to farmworkers and foodborne risk to broiler consumers.

Methods: Seventy-two BBFs were selected by the snowballing method, comprising large BBFs (>2500 birds; n=25), medium (1000-2500 birds; n=25), and small BBFs (≤1000 birds; n=22). Using 135 variables in a questionnaire/checklist through observational methods, records, procedures and activities of the farms were used to score each farm. A score of 1 indicates a high risk of infection, 5, an excellent biosecurity system, and 0, the absence of a biosecurity system. Farms that scored ≤4 were considered as non-compliant.

Results: Seventy-two BBFs were selected by the snowballing method, comprising large BBFs (>2500 birds; n=25), medium (1000-2500 birds; n=25), and small BBFs (≤1000 birds; n=22). Using 135 variables in a questionnaire/checklist through observational methods, records, procedures and activities of the farms were used to score each farm. A score of 1 indicates a high risk of infection, 5, an excellent biosecurity system, and 0, the absence of a biosecurity system. Farms that scored ≤4 were considered as non-compliant.

Conclusion: Seventy-two BBFs were selected by the snowballing method, comprising large BBFs (>2500 birds; n=25), medium (1000-2500 birds; n=25), and small BBFs (≤1000 birds; n=22). Using 135 variables in a questionnaire/checklist through observational methods, records, procedures and activities of the farms were used to score each farm. A score of 1 indicates a high risk of infection, 5, an excellent biosecurity system, and 0, the absence of a biosecurity system. Farms that scored ≤4 were considered as non-compliant.

P-52 Prevalence and Serotyping of Salmonella spp. in Broiler Production Chain Values and the Environment in Nigeria for Public Health

Nurudeen Olalekan Oloso1, Ismail Adewuyi Adeyemo2, Ismail Odetokun3, Adebola Olayemi Odeseye4, Chaiwat Pulsrikarn5, Henriette Van Heerden2

Introduction: Salmonella is a commensal bacterium, and its presence in food or water is an indicator of hygiene and faecal contamination. However, pathogenic E. coli strains can be responsible for severe outbreaks, and their antimicrobial resistance is an important issue for public health.

Purpose: A molecular screening for the presence of extended spectrum beta lactamase (ESBL) genes was performed in E. coli strains isolated from Egyptian raw milk cheese (karish cheese), and its potential role in the dissemination of CTX-M-type ESBL-producing E. coli in Egypt.

Methods: A total of 200 samples of karish cheese were screened for cefotaxime resistant E. coli using a micromethods. The plates were incubated at 37°C for 24 hours in ambient air. The colonies were identified by their typical morphology and biochemical tests and confirmed by biochemical tests.

Results: A total of 200 samples of karish cheese were screened for cefotaxime resistant E. coli using a micromethods. The plates were incubated at 37°C for 24 hours in ambient air. The colonies were identified by their typical morphology and biochemical tests and confirmed by biochemical tests.

Conclusion: A total of 200 samples of karish cheese were screened for cefotaxime resistant E. coli using a micromethods. The plates were incubated at 37°C for 24 hours in ambient air. The colonies were identified by their typical morphology and biochemical tests and confirmed by biochemical tests.

P-53 Molecular Screening of ESBL Genes in Salmonella enterica Strains Isolated from Livestock and Bivalve Molluscs in Sicily, Italy

Maria Vitale1, Michele Fiasconaro1, Maria La Giglia1, Flavia Prusti2 and Vincenzo Di Marco Lo Presti3

Introduction: Salmonella enterica is mainly a commensal enteric bacterium, and its presence in food or water is an indicator of hygiene and faecal contamination.

Purpose: Salmonella enterica is mainly a commensal enteric bacterium, and its presence in food or water is an indicator of hygiene and faecal contamination.

Methods: Salmonella enterica is mainly a commensal enteric bacterium, and its presence in food or water is an indicator of hygiene and faecal contamination.

Results: Salmonella enterica is mainly a commensal enteric bacterium, and its presence in food or water is an indicator of hygiene and faecal contamination.

Conclusion: Salmonella enterica is mainly a commensal enteric bacterium, and its presence in food or water is an indicator of hygiene and faecal contamination.
Purpose: The literature search for Salmonella isolates included in human salmonellosis clusters and multiple-drug resistance (MDR) was assessed as a variable of particular interest to the agency.

Methods: A total of 406 food safety practices were noted from the reviewed sources (n=14), in which five key food safety themes (trend, check the refrigerator temperature, make sanitizer, and leftovers) were evaluated to determine the theme of which Salmonella strains were associated with Salmonella outbreaks.

Significance: Understanding the incidence, severity, and duration of chronic sequelae of parasitic gastroenteritis will help provide more accurate public health impact of disease burden and economic effects.

P57-59 Kitchen Kaizen: Preliminary Findings of a Hands-on Consumer Food Safety Workshop

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Introduction: About one in six Americans will experience foodborne illness. Foodborne illness is highly preventable, yet consumers still engage in risky food handling behaviors that contribute to disease outbreaks. Kitchen Kaizen is a workshop designed to teach consumers important but less obvious behaviors focused on clean (versus sanitize), separate (not washing raw poultry), calibrate (a food thermometer), and chill (refrigerator temperature) while reviewing core behaviors. Those who attend the workshop will gain a greater knowledge (how’s and why’s) and confidence to safely handle food at home.

Methods: Epidemiologic data were analyzed during multi-jurisdictional outbreak investigations to determine the likely vehicle of infection. Public health communications were conducted through the web, social media and traditional media. The content of Public Health messaging was modified over the course of each investigation.

Results: Between December 2017 and December 2018 the Public Health Agency of Canada investigated four multi-jurisdictional produce-related enteric illness outbreaks. Each outbreak was associated with consumption of romaine lettuce contaminated with listeria monocytogenes exposure. There were 21 public health notices, 28 tweets, and 14 Facebook posts issued as part of the public communications approach in response to these illness outbreaks. High weekly visits (165,000 visits) and social media (1,140,000 Facebook views) traffic was observed during the outbreak periods. Information was disseminated in the initial phases of the outbreak, and awareness levels declined as the outbreak investigation concluded.

Purpose: Evaluate the prevalence and burden of long-term sequelae of select foodborne parasitic infections by analyzing a systematic literature review, and provide estimates of prevalence, severity, and duration of chronic sequelae of parasitic gastroenteritis where data allows.

Results: Since 2017, 272 adults participated, 48.53% of the participants completed both surveys (n=14), in which five key food safety themes (cook, chill, clean, cross-contamination, check) were identified utilizing the content analysis approach. These highlighted 47 food safety practices which were present in three or more of the top 10 sets of food safety practices for consumers. Of the identified food safety practices, 53% of the participants completed both surveys (n=12). The majority of respondents identified as female (88.6%), obtained a college degree or higher (77.4%), and were most respondents in a household with $75,000+ (33.8%). Respondents self-efficacy, and behavioral intent significantly (P<0.05) increased to calibrate a food thermometer, check the refrigerator temperature, make sanitizer, and leftovers.

Significance: Results from Kitchen Kaizen show trends that the workshop is creating knowledge and behavior improvements. Participants were provided with materials to help improve home food safety. Widener application this teaching model in food safety may improve consumer behavior to consistently engage in safe food handling.

P59 Pulmonary Effects of Exposure to Voluntary Qualified Importer Program (VQIP) Monitoring in the Voluntary Qualified Importer Program (VQIP) in Commodities (VQIP Commodities) that are covered by VQIP.

Methods: We investigated the reason behind this situation with a direct mailing to 10,000 European Union food industries in seven countries (IT, FR, GR, SP, UK, DE, NL).

Purpose: To identify any differences in how these sources relate and compare to each other.

Results: Of the 406 food safety practices were noted from the reviewed sources (n=14), in which five key food safety themes (trend, check the refrigerator temperature, make sanitizer, and leftovers) were evaluated to determine the theme of which Salmonella strains were associated with Salmonella outbreaks.

Significance: Monitoring isolates from FSIS establishment samples and focusing on certain strains that are most likely associated with illnesses is critical for early detection of outbreaks possibly with FSIS-regulated products, especially for strains with those that are multidrug resistant.
P1-63 Food Recruitment: Development of an Optimized Recruitment Strategy Using the Social Cognitive Career Theory
Kristina Sanga, Gabriela Artega-Arredondo and Clint Stevenson
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Introduction: The food industry in the United States plays an important role with significant economic contributions as an employer, with an estimated 27 million employees establishing $1.46 million workers according to the Committee for Economic Development. There is the need for empirical research on best practices for recruitment of prospective foods safety students.

Purpose: To determine the most effective method for recruitment through the i) development of three outreach materials using social cognitive career theory (SCCT) as a framework, and ii) comparison of recruitment effectiveness of three methods.

Methods: A southwestern, 18 to 20-year-old target audience participated in this study. The developed outreach materials promoted the Food Science Program at NC State and included: a recruitment website, a video, and an in-person presentation. A survey research instrument of a five-point Likert scale and adapted SCCT constructs were administered before and after each recruitment method to determine if there were any changes in interest and attitude. The surveys were measured. Three treatment groups were used, with n=6 participants each, n=198 total. Different combinations of the three aspects of the SCCT (self-efficacy, outcome expectations, how to apply personal goals) were also compared to detect if a component was more influential.

Results: One-Way ANOVA was used to test for significant differences in the change of response at a confidence level of 90%. The standard video (0.97±1.44) was significantly more effective than the website (0.36±0.97) at increasing understanding of what food safety careers are like (P=0.017) on a five-point Likert scale.

Significance: Each recruitment presentation improved feelings of interest towards food safety careers, increased knowledge about food safety careers, and improved overall attitude towards food safety careers: the SCCT is an effective method to use in recruitment presentations.

P1-64 Food Safety Knowledge and Safe Food Handling Confidence among Pregnant Women in Louisiana
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Introduction: Compared with the general population, pregnant women are more susceptible to foodborne illnesses because they undergo significant physiological changes to accommodate their developing fetuses including immunological as well as hormonal changes. The down-regulation of the immune system prevents rejection of the fetus which also increases pregnant women’s susceptibility to foodborne pathogens and risk of developing more severe outcomes from those pathogens.

Purpose: i) To assess pregnant women’s confidence to handle food, and ii) to assess their perception of food safety knowledge.

Methods: A questionnaire was designed to assess the pregnant woman’s confidence to handle food and their perception of food safety knowledge in the state of Louisiana. Data were analyzed using descriptive statistics.

Results: Two hundred twenty-two questionnaires were collected. The majority of participants were white (70%) between the ages of 26 and 30 (45.8%), with a four-year college degree (42.5%). The dependent variable, confidence to Keep Foods Safe for Consumption, was measured on a six-point scale.

Significance: To protect pregnant women and their fetuses from the serious consequences of foodborne illnesses, food safety education is critical. Studies to assess the confidence, knowledge and behavior of the pregnant woman will be beneficial for the food safety education.

P1-65 Food Handling and Causes of Food Waste in Urban Mexican Households
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Introduction: Food quality and safety throughout the supply chain depends on adequate cold chain management. Deficiencies in food handling causes biological deterioration and the waste of foodstuffs.

Purpose: This study is focused on highlighting the weaknesses of home food handling that lead to unnecessary food waste or discarding of food.

Methods: This study was an urban population of Guadalajara, the second largest city in Mexico. Fieldwork’s consisted of gathering information from an in-person survey in public places. A total of 50 interviews were conducted. Each member of the sample was randomly selected; 66% were women, 68.8% had a college degree or higher, and 31.2% had an income level of $10,000 to $19,999. The data was analyzed using the frequency procedure for categorical data and the options of the answers were compared using chi-square at a P-level of 0.05.

Results: More than 72% eliminated food from their refrigerators for cleaning (P=0.001), and 68% believed that food waste did not increase environmental pollution of any way (P=0.003).

Significance: The results indicate that consumers in urban Mexican households should be trained by private, academic and government sectors to reduce unnecessary food waste and to be aware of the negative environmental impact of wasting food.

P1-66 Consumer Attitudes Toward Food Safety Risks in Lebanon
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1Gradill School of Sport and Health Sciences, Cardiff, Cardiff Metropolitan University, Cardiff, United Kingdom, 2ZERO2FIVE Food Industry Centre, Cardiff Metropolitan University, Cardiff, United Kingdom, 3School of Health Science, Modern University for Business & Science, Beirut, Lebanon

Introduction: Lebanon is a food importing country, and the home is associated with the incidence of food poisoning. Food safety education is required to improve consumer food handling behaviors. Understanding cognitive biological influences, such as attitudes, is important for the development of educational initiatives; to date, little is known about Lebanese consumers’ attitudes towards food safety.

Purpose: To determine the attitudes of Lebanese consumers’ attitudes towards food safety risks and perceptions associated with acquiring food poisoning through the consumption of meat in Lebanon.

Methods: A quantitative, self-complete food-safety questionnaire was distributed to a convenience sample (n=97) of consumers in Beirut, Lebanon. Attitudinal responses (n=21) were given upon a five-point Likert-type rating scale; the perceived likelihood of getting food poisoning was assessed using a variation of a visual-analog-scale.

Results: Overall, 60 to 68% of consumers believed they knew all of the food preparation/storage practices required to ensure food safety, however, a similar proportion (62%) believed their current food-safety behaviors needed improvement. 51% were confident that the way they prepared food at home would not cause food poisoning. Lebanese consumers perceived getting food poisoning from food prepared in the home was less likely (52%) than from food-service establishments/outlets such as sit-in restaurants, takeaways and market stalls (61 to 68%) demonstrating a perception association with optimistic bias. Data showed that 51% thought food safety is a priority for consumers in Lebanon with 63% believing that electricity interruptions do not impact upon food safety and 58% suggesting that food safety in the home is affected. Nearly half of respondents (46%) were concerned about the safety of the home drinking water and 50% believing this has impacts upon food safety.

Significance: Lebanon has been done on Food Science and related disciplines. It is imperative to understand the next generation of food science and food production workers. Given the changing regulations and increased demand for food safety, it behooves instructors and instructional designers to seek to understand and potentially change their pedagogy to maximize the efficacy of their teaching.

Methods: To better understand this group, interviews were conducted with 16 undergraduates randomly selected from across the country. They were asked to share their educational preferences- how they best learn and what motivates them to incorporate food safety culture into their practices.

Results: Analysis of themes showed a common desire for community, perceived importance, and participation. Technology, while mentioned, was most often an assumption rather than a stated desire.

Significance: Acknowledging these needs as new curriculums and training are developed is important if the target population is Generation Z. Working in tandem and in common with Generation 2 will better engage them in learning environments.

P1-67 Evaluation of Food Safety Recommendations in EGG Yoodle Online Video Streaming and Blog Recipes
Tressie Barrett and Yahouda (Bety) Feng
Purdue University, West Lafayette, IN

Introduction: The Web is frequently used to share recipes and food preparation techniques. Free video streaming, food blogs, and free online forums enable consumers to easily post and share recipes. Without screening, videos and blogs could promote food handling behaviors which may lead to a higher risk for foodborne illnesses.

Purpose: Evaluate the food safety content and potential risk of popular online video streaming and blog recipes by using ‘making egg noodles’ as an example.

Methods: Keywords “egg noodles”, “recipes”, and “homemade” were searched on YouTube, Google, and Pinterest to collect online shared video and blog recipes. Criteria for selection were recipes developed to be made at home and recipes that were watched by more than 1000 views; and an average of over 200 was selected. No view minimum was established for this food group on all videos. The search yielded 49 YouTube videos and 55 blog videos on this topic. Videos and blogs were reviewed and coded for 35 items including ingredients, processing procedure, food safety information, and common food safety errors.

Results: Food safety recommendations were seldom provided in either video (6%) or blogs (10%); while blogs contained fewer unsafe food handling practices than videos. Common errors observed were washing hands before and after preparing recipes, and not washing the cooking surface. Cross-contamination events were not specifically described in videos. Approximately half videos and 3 blogs mentioned eggs from backyard chickens, which were promoted by individuals as “extra fancy” or “healthy.” Pets were observed in the background of several videos, in the kitchens.

Significance: The mishandling and lack of food safety recommendations on popular recipes show efforts are needed to develop strategies for online shared-use recipes and to promote safe food handling to consumers.

P1-68 Evaluation of Food Safety Curricula Effects on Students’ Food Handling Behaviors: An Observation Study
Tressie Barrett and Yahouda (Bety) Feng
Purdue University, West Lafayette, IN

Introduction: More youth are becoming involved in food preparation at home; however, previous studies showed that youth lack knowledge of and fail to practice food safety. Food safety curricula were developed and evaluated based on self-reported knowledge and actual behavior change. There are gaps in the literature that have been addressed by this study.

Purpose: (i) Compare the effectiveness of an informal and an academic standard aligned curriculum in improving students’ food safety knowledge and food handling behavior, and (ii) to explore the use of GoP’s wearable cameras as an alternative to stationary cameras for data collection.

Methods: One-hundred two high school students were divided into two groups; each group was taught using either informal or academic standard aligned curriculums and post-surveys measured knowledge and self-efficacy changes. Stationary and GoP cameras collected pre- and post-behavioral data as students cooked a meal. SPSS was used to analyze survey data by Student’s t-test, significance level 0.05. Videos were reviewed for 13 behaviors
Results: Experts (73%) agreed that food safety education is important and the curriculum evaluated aligns with academic standards. They concurred that the food safety curriculum needs to be relevant, rigorous, and promote critical thinking and problem-solving. One expert stated; “...bringing in more science-based content helps make this [food safety] content more relevant to students...the level of rigor and detail in this curriculum helps make it more interesting and applicable to students...” Another stated, “The [NACCP] activity sheet is intense and really involves the student to use critical thinking and problem-solving skills.”

Significance: This study provides evidence that the alignment of food safety curriculum can be accomplished to demonstrate food safety relevance in students' lives and career development.

P1-71 Evaluation of Story of Your Dinner Education Campaign Video and Blog-style Recipes Yaohua (Betty) Feng, Emily Chuang and Shelley Feist Purdue University, West Lafayette, IN

Purpose: To evaluate the effectiveness of the campaign video and the blog-style recipes containing food safety recommendations.

Methods: Part I: Evaluation of the campaign video. Food safety educators were recruited to identify consumers nationwide to view the campaign video and respond to pre- and post-surveys, which consist of self-assessment, perceived behavior control, and food safety knowledge. Part II: Evaluation of the blog-style recipes. Consumers were recruited at Purdue University in Indiana or via email from Purdue University. They were required to complete the survey as instructed in the recipe and complete online questionnaires.

Results: Part I: 87 consumers (60%, 40% to 54 years old) participated. They demonstrated significant knowledge and practice increases in “not washing raw meat under running water” (33% to 86%), “before food is served, the food temperature must be taken” (56% to 73%), and “the recommended temperature range for the refrigerator” (49% to 64%). Part II: 23 consumers completed the survey. Consumers found both recipe formats easy to follow and the amount of food safety recommendations was considered just about right (64%). The majority of consumers noticed the difference in food safety recommendations between the video and card, like “washing hands” 20-30 seconds (video) or without (card). More consumers were aware of the message of “using thermometers” from recipe card than video.

Significance: The campaign video increased consumers’ food safety knowledge and self-report practice compliance. Recipes with food safety recommendations were effective tools to emphasize the importance of food safety education.

P1-72 Evaluation of Food Safety Education among Indiana Veteran Farmers Han Chen, Yaohua (Betty) Feng, Kevin Gibson and Chadia Chastain Purdue University, West Lafayette, IN

Purpose: To evaluate the effectiveness of the campaign video and the blog-style recipes containing food safety recommendations.

Methods: This study used a web-based survey (Qualtrics, Provo, UT) to evaluate farm food safety knowledge, attitude, and practice of veteran farmers and to identify barriers to food safety education. The survey was conducted online via an email sent to a large list of veteran farmers with known characteristics, military background, farming practices, food safety knowledge and attitude, and barriers to food safety education. It was distributed to an email listserv from the Purdue University IRB.

Results: The survey was completed by 15 veterans. The 13 respondents indicated that they had grown produce, most of them (77%) did not have a food safety plan for their farm. Less than eight percent of them collected water samples. Fourteen percent of them did not wash their hands. Only 17% of participants used water testing kits. Eight percent of participants had no experience with proper food safety practices. The most common barriers they identified were lack of time, an overwhelming amount of information, and lack of educational materials.

Significance: The findings shed light on the veteran farmers' barriers to food safety practices and education programs. It will guide extension educators and policymakers to develop audience-targeted food safety programs for veteran and other socially disadvantaged farmers.

P1-73 What is It Like to Have a Shared-use Kitchen: A Pilot Study with Young Adults Emily Chuang and Yaohua (Betty) Feng Purdue University, West Lafayette, IN

Purpose: To evaluate the effectiveness of the campaign video and the blog-style recipes containing food safety recommendations.

Methods: This study used a web-based survey (Qualtrics, Provo, UT) to evaluate farm food safety knowledge, attitude, and practice of veteran farmers and to identify barriers to food safety education. The survey was conducted online via an email sent to a large list of veteran farmers with known characteristics, military background, farming practices, food safety knowledge and attitude, and barriers to food safety education. It was distributed to an email listserv from the Purdue University IRB.

Results: The survey was completed by 15 veterans. The 13 respondents indicated that they had grown produce, most of them (77%) did not have a food safety plan for their farm. Less than eight percent of them collected water samples. Fourteen percent of them did not wash their hands. Only 17% of participants used water testing kits. Eight percent of participants had no experience with proper food safety practices. The most common barriers they identified were lack of time, an overwhelming amount of information, and lack of educational materials.

Significance: The findings shed light on the veteran farmers' barriers to food safety practices and education programs. It will guide extension educators and policymakers to develop audience-targeted food safety programs for veteran and other socially disadvantaged farmers.
P1-76 – P1-78

How Does the Food Safety Knowledge of Student Dietitians Compare at a University in Wales, Lebanon and Ohio?

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Introduction: Registered dietitians (RDs) are the only healthcare professionals regulated by law to assess, diagnose and treat dietary/nutritional problems. Given that food needs to be safe and nutritious to maximize food-related health and wellbeing, food-safety is part of the dietetic-curriculum for training RDs. Despite this, gaps in RDs food-safety knowledge have been identified, furthermore, data detailing student-dietitians food-safety knowledge is limited.

Methods: Assess and compare food safety knowledge of student-dietitians in three accredited dietetics programs in utilizing different food safety teaching methods.

Results: From the 212 student-dietitians that participated, 79% recalled having received food-safety training/education, recall differed significantly (P<0.05) between institutions (Cardiff 100%; Beirut 97%/USA 58%). Student-dietitians in Cardiff participated in a one-day food safety training event, and students at Beirut and Ohio participated in a micro-mastery laboratory. Subjects completed knowledge and food-safety sanitation courses, and 43% completed ServSafe certification. Food-borne pathogen awareness was different in all three regions (P<0.05). The awareness among students in Beirut was the lowest for each subject, students in Cardiff had the highest awareness of Campylobacter and Clostridium while Cardiff students had the highest awareness of Es. coli and Staphylococcus. Although the majority indicated awareness of food-safety practices, significant differences were determined (P<0.05) by the institution. The majority of students (Beirut 100%; Ohio 99%; Cardiff 88%) were aware of the need to use a meat thermometer to check core temperature. Knowledge of the use-by date indicating food safety was significantly greater among student-dietitians in Cardiff (81%) than student-dietitians in Beirut (68%) and Ohio (74%). Ohio students had the lowest awareness of cross-contamination practices such as washing raw poultry (39%), compared to Cardiff (75%) and Beirut (47%).

Significance: Differences in food safety awareness may arise from the teaching approach, but knowledge retention is not the issue to determine the best practices to teach student-dietitians food safety and explore the interpretation of dietetic curriculum requirements in institutions that deliver accredited training.

P1-77 – P1-78

Sport and Exercise Nutritionists’ Perceptions of Food Safety Risks among Athletes

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Introduction: Food safety is essential for athletes; therefore, it is expected to be a worry-free aspect of training. The incidence of foodborne illness among athletes at major events has frequently made headlines in recent years. Sport and exercise nutritionists (SEN) currently provide food-related advice/information to enable optimum nutrition for performance. However, the role SEN play in identifying and mitigating the risk of foodborne infection for athletes remains underinvestigated.

Purpose: To identify SENs’ perceptions of athletes’ food-preparation practices and food-consumption habits that may increase risks associated with foodborne illness.

Methods: SENs working with elite athletes (n=23), participated in a series of focus groups (n=3). Each group discussion followed a structured route and included perceptions of food preparation/consumption associated with key scenarios (home/training/traveling and competing away from home).

Results: Food safety perceptions were highly varied among SENs. Advanced preparation, monitoring and prolonged storage were unsafe behaviours frequently identified. SENs were aware of the risks associated with food safety for individual athletes and food providers (athletes/village/hotels) during competitions; however, they also perceived their contribution to the prevention of food-poisoning to be “very high” (90%). Food poisoning was seen as a “larger concern” when competitors were situated away from home, where ‘the highest risk’ due to limited access to appropriate refrigeration facilities, food sources, food-preparation/storage practices were “out of SENs’ control.” Consequences of foodborne illness were identified as days of lost training. It was discussed that one of the main objectives of SENs “is to minimise the risk but not at the expense of training intensity.”

Significance: SENs play an important role in advising food-preparation/consumption practices to athletes and in reducing the risk of foodborne illnesses. This study highlights the SENs awareness/concern for athlete food safety and implementation of unsafe behaviors. Further research is required to determine SENs food safety training to deliver the sufficient food-safety information to athletes.

P1-78 – P1-79

Utilizing Remote Covert Observation in Food Manufacturing and Processing Environments to Assess Hand Hygiene Compliance

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Introduction: Ensuring hand hygiene (HH) compliance in food manufacturing/processing environments is of utmost importance for food safety.的手Jubalek (2009) concluded that incorrect hand hygiene habits may reduce compliance by 30%. This study examined the influence of knowledge and attitudes/safety related practices/intentions on actual behavior and HH compliance. Staff may be subject to biases. Similarly, researcher presence in overt observation of behavior can cause reactivity bias, whereas covert remote observation (using CCTV cameras) can provide repeated analysis of overt food-handling behaviors. Both covert remote observation and observational sampling have been used in existing workplace CCTV cameras may reduce reactivity bias.

Purpose: This mixed-methods research approach explores the use of covert observation in food processing/manufacturing environments.

Methods: The study combines a desk review of professional literature on food-handler hygiene and associatedHH studies with a qualitative assessment of existing workplace CCTV cameras may reduce reactivity bias.

Results: The qualitative component was most frequently undertaken while conducting food-handler hygiene risk assessments (89%); observational data, particularly from manufacturing/processing environments, were lacking. Interviews indicated positive attitudes toward utilizing covert observation to assess HH compliance. Although food businesses had CCTV cameras in operation, they were predominantly used for security or monitoring productivity; none were dedicated to monitoring hygiene. The audit identified preferences for CCTV monitoring was not all provided viewpoints to facilitate meaningful observation of HH compliance. The covert observation covert observation in a bakery sector achieved.

Conclusions: This study highlights the importance of training RDs on food-safety in an accredited dietetics program in order to prepare them for the challenges they will face in their future practice. Further research is needed to determine the best practices to teach student-dietitians food safety and explore the interpretation of dietetic curriculum requirements in institutions that deliver accredited training. Food safety education is essential for athletes; therefore, it is expected to be a worry-free aspect of training. The incidence of foodborne illness among athletes at major events has frequently made headlines in recent years. Sport and exercise nutritionists (SEN) currently provide food-related advice/information to enable optimum nutrition for performance. However, the role SEN play in identifying and mitigating the risk of foodborne infection for athletes remains underinvestigated. This study highlights the SENs awareness/concern for athlete food safety and implementation of unsafe behaviors. Further research is required to determine SENs food safety training to deliver the sufficient food-safety information to athletes.
Results: We expressed the GFP-LTF and GFP-STF at 35 mg/l and 160 mg/l, respectively. Both tail fibers bound ECOR strains outside T4's host range, suggesting that they are functionally active. The significantly lower production of LTF compared to STF may be due to differences in expression or stability. The results of this study demonstrate that developing strategies to target T4 infection in many of the cases involved in food contamination could be achieved.

Purpose: The purpose of this study was to assess the effectiveness of a food safety training program for small food manufacturing businesses in Wales.

Methods: For PSA training, results showed post-test scores were statistically and significantly higher than pre-test scores. A total of 39 food-manufacturing businesses participated in the workshop, with 51% of participants intending to implement food safety improvements as a result of the training.

Results: Twenty-nine percent of participants reported significant improvements to meet the PSA requirements. The results of the PSA Grower Training and the OFRR program demonstrated improvement of the knowledge and compliance level of Florida farms regarding the FSMA PSR.

Significance: Our results indicate that strawberry growers within the southeastern United States have significant differences in their documentation of risk management practices. As this is an integral part of the PSR, strawberry growers may benefit from additional education regarding documentation.

P1-85 Food Safety Education and Outreach for Florida Farmers

Introduction: In Florida, the University of Florida Institute of Food and Agricultural Sciences (UF/IFAS) and the Florida Department of Agriculture and Consumer Services (FDACS) collaborated to provide education and outreach through Produce Safety Alliance (PSA)Grower Training Courses and On-Farm Demonstrations. Reviews of OFRRs to assist farmers in meeting the requirements of the PSR of the Food Safety Modernization Act (FSMA) Produce Safety Rule (PSR).

Purpose: To determine if PSA training was successful in improving the level of knowledge of the PSR and foundational food safety principals that Florida farmers have and to determine the level of farm readiness for FSMA PSR compliance.

Methods: Student responses were statistically and significantly higher than pre-test scores (t=33.25, P<.001), indicating a significant increase in knowledge after participation in the training. Out of 25 points, participants scored an average of 16.45 on the pretest and 20.66 on the post-test. The majority of participants reported that preharvest activities were health and hygiene, preharvest worker training, and preharvest water. Of the farms that were assessed, 44.44% met the FSMA PSR requirements, 33.33% needed minor improvements, and 22.22% needed significant improvements to meet the PSR requirements.

Significance: The results of the PSA Grower Training and the OFRR program demonstrated improvement of the knowledge and compliance level of Florida farms regarding the FSMA PSR.

P1-86 A Support Package to Support Small Food Manufacturing Businesses in Wales in Overcoming Barri- ers to Obtain Safe Food Certification: A Pilot Study

Introduction: This study has designed, delivered and evaluated training required by food-manufacturing businesses which increased food handler awareness of food safety risks and handling practices.

Purpose: To understand food handler awareness of the management of allergens.

Methods: Food-handlers (n=51) from food-manufacturing businesses in Wales (n=10) participated a short course covering the five themes of the BRC Global Standard for Food Safety (BRCGS) and were surveyed before and after the training session to assess knowledge and confidence in food safety management.

Results: Food handlers reported an increased frequency with familiar management procedures following the training intervention, the increase was statistically significant (P<0.05) from 49% to 82%. Confidence in awareness of allergen management paperwork increased from 25% to 75% (P<0.001) and using allergen management paperwork increased from 12% to 37% to 78% (P<0.001).

Significance: Technical managers reported improvements in paperwork following intervention for 33% of companies. Furthermore, 33% are more aware of non-food-related allergens. For most (67%) overall allergen management has improved. Records have improved since the training for 100% of companies. Significant improvements were also made to inform food handlers and assess food handler awareness of allergen management. Food handlers tended to have some knowledge of allergens to carry out their role but evidence shows that a short two-hour session on allergen management could improve knowledge and confidence significantly.

P1-87 Food Handler Awareness of Allergen Management Systems in Welsh Food Manufacturing Businesses

Introduction: Food and drink manufacturing businesses are legally required to train food handlers and manage allergens onsite. Similarly, the BRC Global Food Safety Standard requires businesses to ensure all staff are trained effectively and require management of allergens. Although there appears to be widespread awareness of food handler training in restaurants, there is a need for targeted education in food-manufacturing businesses.

Purpose: To understand food handler awareness of food safety risks as part of their role within food-manufacturing businesses.

Methods: A short course (n=10) was delivered in 2019 which covered the five themes of the BRC Global Standard for Food Safety (BRCGS). A pre- and post-test was used to determine if there was a significant increase in knowledge after completion of the training. A total of 25 food-manufacturing businesses participated in the workshop, with 51% of participants intending to implement food safety improvements as a result of the training.

Results: Twenty-nine percent of participants reported significant improvements to meet the PSA requirements. The results of the PSA Grower Training and the OFRR program demonstrated improvement of the knowledge and compliance level of Florida farms regarding the FSMA PSR.

Significance: Our results indicate that strawberry growers within the southeastern United States have significant differences in their documentation of risk management practices. As this is an integral part of the PSR, strawberry growers may benefit from additional education regarding documentation.
Courtney Crist1, Elizabeth Canales1
1Mississippi State University, Mississippi State, MS

Purpose: The project aim was to increase participants awareness and skills related to food safety and regulations, business/marketing strategies, and financial recordkeeping for agribusinesses to mitigate marketing, financial, and legal risks.

Methods: Ten “Field Day” educational workshops were conducted in Mississippi. The workshops included presentations, demonstrations, and hands-on exercises focused on food safety topics. Participants were surveyed pre- and post-workshop to assess their knowledge and understanding of food safety topics.

Results: A total of 810 attendees participated in the Field Days. The majority of attendees were farm business owners and operators (60%), followed by farm employees (25%). The most common barriers to implementing food safety practices identified by attendees were lack of knowledge (69%) and lack of resources (59%).

Significance: The findings of this study can be used to improve the effectiveness of future educational programs and to tailor future educational initiatives to meet the needs of Mississippi farm businesses.

Courtney Crist1, Elizabeth Canales1
1Mississippi State University, Mississippi State, MS

Purpose: To evaluate the impact of a multidisciplinary program approach to assist food entrepreneurs in mitigating business, financial and food safety risks.

Methods: A sample of 120 food entrepreneurs in Mississippi were recruited and enrolled in the program. The program included workshops on food safety, financial management, and business planning. Participants were surveyed pre- and post-program to assess knowledge and attitudes.

Results: A total of 72 participants completed the program. The majority of participants were small business owners (75%). The average score on the pre-program knowledge test was 40%, and the average score on the post-program knowledge test was 65%. Overall, participants reported an increase in their knowledge, skills, and confidence in managing their business.

Significance: The findings of this study can be used to improve the effectiveness of future educational programs and to tailor future educational initiatives to meet the needs of Mississippi food entrepreneurs.
P1-94 Creation and Implementation of a Social Marketing Campaign for Beef Food Safety
Benjamin Chapman, Jill Hochstein, John Luchansky, Kyle Longacre and Anna Porto-Fett

“North Carolina State University, Raleigh, NC, 1University of Nebraska-Lincoln, Lincoln, NE, 2U.S. Department of Agriculture-ARS-Wyndmoor, Wyndmoor, PA, 3Montgomery County Intermediate Unit, Norristown, PA. USDA-ARS, Wyndmoor, PA

Introduction: Food safety-related messages are most effective when they are meaningful to the intended audience, contain accurate information, are delivered creatively, and delivered at appropriate times. A social marketing approach to food safety messages has been suggested by public health agencies to reach consumers and inform food safety and handling behaviors.

Purpose: This exploratory study was designed to create a roadmap and execution of a social marketing campaign to mitigate beef food safety.
**P1-100 Photodynamic Treatment of Bacillus cereus Strains: Estimating the Inactivation Kinetic Parameters of Four Strains from Different Sources**

Alicia Ortiz Alvaranga, Gilberto U. L. Braga, and Anderson de Sousa Santana

*Department of Food Science, College of Food Engineering - University of Campinas, Campinas, Brazil, University of Campinas, College of Food Engineering, Campinas, Brazil; Department of Food, Faculty of Pharmaceutical Sciences of Alagoas - PE - Brazil; University of São Paulo, Alagoas, Brazil*

**Developing Scientist Entreat**

Introduction: The contamination of foods with pathogens is responsible for mortality and morbidity that impacts lives and countries' economies and social development. Bacillus cereus is a spore-forming bacteria commonly found in soil and associated with foodborne diseases and to food spoilage. There- fore, it is critical to be employed as a model for photodynamic inactivation.** Purpose:** The main aim of this work was to evaluate and promote technological advances in the use of PDT for the inactivation of B. cereus strains.** Methods:** A total of 12 strains of B. cereus isolated from different types of foods and outbreaks in Brazil were used. The resistance of the isolates to PDT was measured after four cycles of DHP (4 cycles) for 60 min using a light source of 950 LEDS (RED; 650 nm). Then, the B. cereus strains were selected according to their PDT resistance and a further experiment was conducted with five, 10, and 150 min of DHP for 120 min in order to obtain the PDT inactivation kinetic parameters. The variability in the inactivation kinetic parameters was determined.** Results:** From the 12 strains tested, four strains (895, 436, 83, and 145) were selected based on cluster analysis according to their PDT resistance. The less resistant strains to PDT were 436 and 14579 with 7.5 and 7.5-log reduction, respectively, using 50 µM of NMB. The strains 83 and 863 with 2.8 and 2.9-log reductions were considered the most resistant to PDT. The discrimination of kinetic parameters of inactivation and variability will be presented.** Significance:** This work will allow gaining insights into the feasibility of using PDT for inactivation of B. cereus.** P1-101 Processing of Dried Beef (Biltong) without a Heat Lethality Step to Achieve USDA-FSIS Validation (Five-Log Reduction) of Salmonella Caitlin Karolenko, Anjali Sethi, and Monica Henry

*Iowa State University, Ames, IA*

**Developing Scientist Entreat**

Introduction: In some parts of the world, the drying of meat products is a means of preservation whereby products lose moisture but retain protein and provide nutrition. In the United States, dried beef products (beef jerky) are a popular snack product but their manufacture often requires the use of a heat lethality step. This introduces adequate reduction of pathogens concern/five-log reduction of Salmonella as per USDA-FSIS requirements). However, other types of dried beef that are produced worldwide do not use heat, but rather, use salt, spices, and drying for microbial control.** Purpose:** Our objective was to examine a process for the manufacture of dried beef (biltong) to try to achieve a five-log reduction of Salmonella without the use of a heat lethality step.** Methods:** Beep obtained locally was sliced (one by two by three in) and inoculated with a five-serovar mixture of Salmonella. The beef was processed by dipping (water control or antimicrobial), vacuum tumbling (spice and vinegar marinade), and dried in a temperature- and humidity-controlled oven (77°F/25°C; 55% RH) for five days.** Results:** All three replicate trials using antimicrobial dip, spice/vinegar marinade, and drying at the specified temperature/humidity provided greater than five-log reduction of Salmonella.** Significance:** The effectiveness of ACP was investigated for killing Shiga toxin-producing E. coli O157:H7, Listeria monocytogenes, and non-Typhoidal Salmonella Serovars Niamul Kabir, Shahid Chowdhury and Ailay Faukikhah

*Public Health Microbiology Laboratory, Tennessee State University, Nashville, TN*

**Purpose:** Industrial adoption of microbiological challenge studies are often curtailed due to differences in come-up and come-down times of research and commercial units. Limited information is currently available to quantify the effects of these parameters on efficacy of pressure-based pasteurization.** Methods:** Current study investigated effects of come-up and come-down times on performance of pressure-based pasteurization for inactivation of Escherichia coli O157:H7, Listeria monocytogenes, and non-Typhoidal Salmonella Serovars.** Results:** The contamination of foods with pathogens is responsible for mortality and morbidity that impacts lives and countries' economies and social development. Bacillus cereus is a spore-forming bacteria commonly found in soil and associated with foodborne diseases and to food spoilage. Therefore, it is critical to be employed as a model for photodynamic inactivation.** Purpose:** The main aim of this work was to evaluate and promote technological advances in the use of PDT for the inactivation of B. cereus strains.** Methods:** A total of 12 strains of B. cereus isolated from different types of foods and outbreaks in Brazil were used. The resistance of the isolates to PDT was measured after four cycles of DHP (4 cycles) for 60 min using a light source of 950 LEDS (RED; 650 nm). Then, the B. cereus strains were selected according to their PDT resistance and a further experiment was conducted with five, 10, and 150 min of DHP for 120 min in order to obtain the PDT inactivation kinetic parameters. The variability in the inactivation kinetic parameters was determined.** Results:** From the 12 strains tested, four strains (895, 436, 83, and 145) were selected based on cluster analysis according to their PDT resistance. The less resistant strains to PDT were 436 and 14579 with 7.5 and 7.5-log reduction, respectively, using 50 µM of NMB. The strains 83 and 863 with 2.8 and 2.9-log reductions were considered the most resistant to PDT. The discrimination of kinetic parameters of inactivation and variability will be presented.** Significance:** This work will allow gaining insights into the feasibility of using PDT for inactivation of B. cereus.** P1-101 Processing of Dried Beef (Biltong) without a Heat Lethality Step to Achieve USDA-FSIS Validation (Five-Log Reduction) of Salmonella Caitlin Karolenko, Anjali Sethi, and Monica Henry

*Iowa State University, Ames, IA*

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Introduction: In some parts of the world, the drying of meat products is a means of preservation whereby products lose moisture but retain protein and provide nutrition. In the United States, dried beef products (beef jerky) are a popular snack product but their manufacture often requires the use of a heat lethality step. This introduces adequate reduction of pathogens concern/five-log reduction of Salmonella as per USDA-FSIS requirements). However, other types of dried beef that are produced worldwide do not use heat, but rather, use salt, spices, and drying for microbial control.** Purpose:** Our objective was to examine a process for the manufacture of dried beef (biltong) to try to achieve a five-log reduction of Salmonella without the use of a heat lethality step.** Methods:** Beep obtained locally was sliced (one by two by three in) and inoculated with a five-serovar mixture of Salmonella. The beef was processed by dipping (water control or antimicrobial), vacuum tumbling (spice and vinegar marinade), and dried in a temperature- and humidity-controlled oven (77°F/25°C; 55% RH) for five days.** Results:** All three replicate trials using antimicrobial dip, spice/vinegar marinade, and drying at the specified temperature/humidity provided greater than five-log reduction of Salmonella.** Significance:** The effectiveness of ACP was investigated for killing Shiga toxin-producing E. coli O157:H7, Listeria monocytogenes, and non-Typhoidal Salmonella Serovars.** Methods:** Hydrostatic pressure (350 MPa, 4°C) and thermal-assisted elevated pressure (350 MPa, 55°C) were applied at various time intervals (zero to five days) in a come-up time of three minutes followed by a come-down time of three minutes.** Results:** From the 12 strains pretested, four strains (B63, 436, B3 and 14579) were selected based on cluster analysis according to their PDT resistance. Our objective was to examine a process for the manufacture of dried beef (biltong) to try to achieve a five-log reduction of Salmonella without the use of a heat lethality step.** Methods:** Beep obtained locally was sliced (one by two by three in) and inoculated with a five-serovar mixture of Salmonella. The beef was processed by dipping (water control or antimicrobial), vacuum tumbling (spice and vinegar marinade), and dried in a temperature- and humidity-controlled oven (77°F/25°C; 55% RH) for five days.** Results:** All three replicate trials using antimicrobial dip, spice/vinegar marinade, and drying at the specified temperature/humidity provided greater than five-log reduction of Salmonella.** Significance:** The effectiveness of ACP was investigated for killing Shiga toxin-producing E. coli O157:H7, Listeria monocytogenes, and non-Typhoidal Salmonella Serovars.** Methods:** Hydrostatic pressure (350 MPa, 4°C) and thermal-assisted elevated pressure (350 MPa, 55°C) were applied at various time intervals (zero to five days) in a come-up time of three minutes followed by a come-down time of three minutes.** Results:** From the 12 strains pretested, four strains (B63, 436, B3 and 14579) were selected based on cluster analysis according to their PDT resistance.
P1-106 Exploring Engineered Water Nanostructures as an Antimicrobial Platform for Fresh Produce Decontamination
Ruzhe Huang, Nachiket Vaze and Philip Demokroutou
Center for Nanotechnology and Nanotoxicology, Harvard T. H. Chan School of Public Health, Boston, MA

Developing Scientist Entrant
Introduction: Fresh produce is susceptible to microbial contamination. However, washing fresh produce with chemical disinfectants has chemical risks from residues and toxic byproducts, and more importantly, is not suitable for organic produce and delicate fruits. A novel nanotechnology-based antimicrobial modality has been developed with engineered water nanostructures (EWNS) synthesized using emulsification and ionization of aqueous suspension of antimicrobials. Such “waterless” nanorunner platform can achieve food disinfection by delivering minuscule quantities of antimicrobials in an aerosol form and on surface in a whole food matrix.

Purpose: Assess the potential of EWNS platform to deliver different active ingredients in a targeted manner for inactivating foodborne pathogens, such as E. coli and Listeria innocua.

Methods: E. coli ATCC 25922 and L. innocua ATCC 33090 were spot-inoculated onto stainless steel coupons and exposed to different EWNS based nanoparticles that incorporate active ingredients of interest. The physical and chemical properties of EWNS nanoparticles (i denotes the active ingredient) were measured. The delivered dose of active ingredient was also estimated as a function of exposure time. Bacteria were recovered on tryptic soy agar and reductions were calculated.

Results: EWNS nanoparticles could significantly reduce (P<0.05) E. coli and L. innocua on coupons in 10-15°C CFU/coupon by greater than five log in a matter of minutes of exposure. For instance, one percent hydrogen peroxide based EWNS nanoparticles could inactivate greater than five log of E. coli and L. innocua on coupons in five and 15 min, respectively. When using three percent hydrogen peroxide based EWNS nanoparticles, E. coli could be reduced by 2-3 log in 30 seconds. It is worth noting that achieving a three-log reduction of E. coli, only 0.078 to 114.53 ng/m² of EWNS nanoparticles were delivered.

Significance: These data suggested that the EWNS platform is effective against E. coli and L. innocua by only delivering nanograms level of active ingredients.

P1-107 Evaluation of Initial and Post-High Pressure Pasteurization Treatment Storage Temperatures as Critical Process Factors
Shrin Abid and Carrie Fersti
Eurifin, Cambridge, MA

Introduction: Sensitivity of pathogenic microorganisms to temperature during HPP and the impact of temperature as a critical factor during refrigerated HPP processing is not clearly understood.

Purpose: To obtain greater understanding of the impact of initial product temperature and post-HPP storage temperature on the destruction of selected pathogens in apple and orange juices.

Methods: Separate five-shoulder-coupons of Escherichia coli O157:H7, Salmonella, or Listeria monocytogenes were acid-adapted, inoculated into apple and orange juices, and HPP-treated at 85,000 psi for three minutes. Products and pressurization fluid were adjusted to initial temperatures of one, four, or 10°C prior to HPP, and stored at one, four, or 10°C post-HPP. Survivors were enumerated using MPN methodology immediately after HPP, and after one and three days of storage post-HPP.

Results: Analysis of results indicated that initial product temperature significantly impacted log reduction of this organism in orange juice (P<0.05), while this is not the case for apple juice (P=0.25). Storage time post-HPP significantly impacted log reduction of E. coli in both products, with significantly more observation the longer the product was stored (P<0.05).

Results: Escherichia coli had a higher pressure-resistant than Salmonella and Listeria monocytogenes in apple and orange juices, and that there is a certain tolerance with regard to initial product and post-HPP storage temperature and the impact of these critical factors on the log reduction of E. coli in these products.

P1-108 Evaluation of Adaptive Response in E. coli O157:H7 to Light and Gallic Acid-Based Antimicrobial Treatments
Qingyang Wang, Robert Buchanan and Rohan Tikekar
University of Maryland, College Park, MD, and University of Maryland, Department of Nutrition and Food Sciences and Center for Food Safety and Security Systems, College Park, MD

Developing Scientist Entrant
Introduction: Ability of bacteria to develop cross-protection to various stresses is well-known. However, exploration into cross-protection between conventional physiological stresses and emerging antimicrobial treatments is limited.

Purpose: To determine whether prior exposure to sub-lethal stresses can increase the resistance of E. coli O157:H7 towards two GA and UV light-based antimicrobial treatments, and whether repeated exposure to these two treatments at different sub-populations cross-resistant towards heat, acid, and oxidative challenge.

Methods: Stationary phase E. coli O157:H7 were exposed to sublethal heat, acid, NaCl, and H₂O₂, before being treated by either GA and UV-A light simultaneously (GA+UV-A), or UV-A light post-irradiated-GA (UV-A+GA). The cultures were also subjected to repetitive cycles of exposure to either UV-A+GA or UV-A+GA to evaluate whether a more resistant sub-population can be identified.

Results: Exposure to water, 55°C for 30 minutes (P<0.05) the resistance of E. coli O157:H7 towards UV-A+GA treatment, while oxidative stress increased (P<0.05) their sensitivity to that treatment. Interestingly, only heat stress showed protective (P<0.05) effect to subsequent UV-A+GA treatment, while acid stress (>0.05) did not affect sensitivity to the previous stress treatment. Repetitive exposure to UV-A+GA treatment incubated for subpopulations that demonstrated higher (P<0.05) resistance to these two treatments as well as heat or acid challenge. Further experiments that showed increased expression of enzymes such as superoxide dismutase and surpHog were likely to be associated with the development of cross-protection.

P1-109 Ensuring Food Emergency Response Network Laboratory Preparedness for Detecting B. anthracis and Y. pestis from Foods
Shannon Pickens, Matthew Kmet, Robert Newkirik, Vishnu Patel, Donald Burt, Ravindra Reddy and Tara Doran
U.S. Army Research Laboratory, Natick, MA; U.S. Food and Drug Administration, Bedford, MA, U.S. Food and Drug Administration, Office of Regulatory Affairs/Office of Regulatory Science, Rockville, MD

Introduction: Bacillus anthracis and Yersinia pestis are considered Category A agents by the Centers for Disease Control (CDC), posing a significant public health threat. For example, in 2011, several outbreaks of anthrax caused by B. anthracis spores were linked to contaminated delivery packages. Therefore, it is essential to assess the utility of the Food Emergency Response Network (FERN) laboratories through FDA’s FDA/IDP 1040 accredited proficiency testing (PT) program.

Purpose: To evaluate FERN laboratories’ proficiency, capacity and capability for detection and isolation of B. anthracis and Y. pestis from various food matrices.

Methods: PT samples were prepared using 1000 and 2000 CFU/mL of B. anthracis inoculum in sweet potato puree. Y. pestis inoculum levels were 10 and 100 CFU/mL and were prepared in baby food and stored at 1, 4, and 10°C. Bacillus anthracis and Yersinia pestis samples were tested using the FERN Screening Method for Bacillus anthracis in Foods. Y. pestis samples were tested using the FERN Yersinia Screening Method for Y. pestis in sweet potato puree. Four E. coli strains and 27 Y. pestis PT samples were sent to FERN laboratories. Results were analyzed to determine how many laboratories across the United States are prepared for testing of these cultures. Results: Assigned values were determined by consensus agreement according to ISO standards. E. coli was homogenous and stable for 10 days in sweet potato puree and 34 laboratories correctly detected E. coli in inoculated samples. Y. pestis was homogenous and stable for 15 days in chicken and vegetable-based baby food and 27 laboratories correctly detected Y. pestis in inoculated samples.

Significance: One hundred seventy-seven samples inoculated with E. coli and 108 samples inoculated with Y. pestis were correctly identified by participating FERN laboratories, furthering FDA’s mission to protect the public health, demonstrating laboratory proficiency and FERN capabilities during a food adulteration event.

P1-110 Evaluation of Freeze-drying Conditions for Extension of Bacteriophage Shelf Life
Dominic Pacito, Philip Pivarnik and Andre Senecal
U.S. Army NYSRDEC, Natick, MA

Introduction: Shelf-stable dry cocktails can be achieved by developing shelf-stable, dry cocktails.

Methods: Cultures of freeze dried storage buffers to optimize freeze dried bacteriophage stability for one year at room temperature.

Results: The E. coli bacteriophage was stored in dry form at 2 to 8°C for one year with greater than 80% recovery of the initial titre.

Significance: The results demonstrate that the bacteriophage stability for one year at room temperature can be achieved by the storage buffers.

P1-111 Food Safety Modernization Act Subpart M: An Evaluation of Pathogen Testing Requirements
Emily Kelly, Maha Hajeneer and Michael Needham
California Department of Food and Agriculture, Sacramento, CA

Developing Scientist Entrant
Introduction: The United States Food and Drug Administration (US FDA), in recognition of unique foodborne disease pathogens, proposed additional minimum standards for sprouts production in Subpart M of the Food Safety Modernization Act (FSMA). Subpart M requires producers to test sprouts or spent irrigation water for specific foodborne pathogens.

Purpose: The following epidemiological analysis was conducted to determine appropriateness of FDA’s pathogen testing requirement, and therefore its potential to minimize known or reasonably foreseeable hazards in sprouts production.

Methods: The Centers for Disease Control and Prevention (CDC) National Outbreak Reporting System (NORS) database was used to identify sprouts-related outbreaks between 1998 and 2017. Data on pathogen genus and serotype, commodity subtype and outbreak detail (e.g., dates, illnesses, hospitalizations) were organized into descriptive statistics. Results were compared with FDA’s published rationale and requirements for pathogen testing in FSMA Subpart M.

Results: No evidence of E. coli O157:H7, Y. pestis or B. anthracis contamination detected in any of the outbreaks. Among non-O157-related outbreaks, there were three implicated serotypes: O26 (2011), O145 (2012) and O121 (2014). The last sprouts-related outbreak of E. coli O157:H7 occurred in 2003.

Significance: Analysis of sprouts-related outbreaks in the US covering 1998-2017 indicates that E. coli O157:H7 is not the most epidemiologically relevant E. coli serotype for this commodity—a deviation in trend from other food commodities, such as leafy greens. Testing sprouts or spent irrigation water for only the O157:H7 serotype would miss a majority (80%) of historical E. coli outbreaks, and therefore brings into question the requirement’s capacity to prevent future illnesses.

Significance: Results help optimize the two techniques to improve their efficiency while avoiding unexpected resistance development, and for a better adoption of hurdle food processing. The possible underlying mechanism behind the phenomenon of cross-protection was also elucidated for a better understanding and control of these emerging techniques.
**P1-112**

**North Central Region Pre- and Post-Grower Training Knowledge Assessment**

**Poster**

**Katheryn Parraga**
**Bridget Perry**

**Iowa State University, Ames, IA**

**Purpose:** Employee training is an important component of food safety system implementation. Determining how facility size impacts training needs and preferences may improve the development of future extension curriculum.

**Methods:** This assessment was designed to identify the risk and potential impact of food safety and allergen control practices on small food processors in the state of Ohio. An anonymous survey was distributed through an email-embedded link to Ohio food processors through existing Extension listservs.

**Results:** Respondents were categorized into one of four groups depending on total number of employees: (1) one employee, (2) two to 10 employees, (3) 11 to 50 employees, and (4) 51 or more employees. The responses were analyzed per group followed by a regression analysis to identify factors of significance. The data showed that facility size impacted training needs and preferences. More specifically, small facilities (one to two employees) increasingly identified direct costs as limiting. Additionally, respondents from the smallest facilities were more likely to have formal training plans with goals and targets for mitigation.

**Significance:** More research is needed to better understand the risk and potential impact of food safety and allergen control practices on small food processors in the state of Ohio.

**P1-113**

**Louisiana Wild-Caught Catfish under USDA Inspection**

**Katheryn Parraga**
**Evelyn Watts**

**Louis State University, Baton Rouge, LA**

**Purpose:** To examine the characteristics of catfish exposed to an outbreak in one of the largest catfish processing plants in the southeast. The purpose of this study was to identify possible improvements of current approaches and targets for mitigation.

**Methods:** The facility sizes ranged from 240 ft² to 3,200 ft². The number of employees were <10 and 20 to 49 for 90% and 10% of the facilities, respectively. The facility identified had 2,500 ft² and the number of employees were 2. The conditions are poor and bioterrorism risks are extensive. Root cause analysis indicates that employee security maintenance programs and third-party audits are key to risk mitigation.

**Significance:** The data suggests that for food packaging risks are being managed or low based on circumstances, but that for disposable gloves there is an accumulation of chemical, microbiological, and user safety/efficacy risks where control mechanisms are lacking.

**P1-114**

**Myccofora and Aflatoxin Levels in Stale Retail Pepper Marketer in Ogun State, Nigeria**

**Eniola Oni**
**Aminad Bada**

**1Federal University of Agriculture, Abeokuta, Abeokuta, Nigeria, 2Federal University of Agriculture Abeokuta Ogun State, Nigeria, Abeokuta, Nigeria**

**Developing Scientist Emerent**

**Introduction:** Fungi are a major group in the production, storage, and processing of agricultural products. A recent concern about the consumption of stale retail pepper in our society necessitated the need to determine the myccofora and aflatoxin levels in retail stale pepper. Aflatoxin is a mycotoxin produced by several species of the mould fungus, *Aspergillus*.

**Methods:** A total of sixty stale pepper samples (Capsicum annuum) from different markets were analysed using standard microbiological procedures and HPLC. Aflatoxin was determined using a highsensitivity HPLC method.

**Results:** A total of sixty samples were analysed. The aflatoxin level was below the limit of detection in all the samples. There were no mycotoxins detected in the samples.

**Significance:** This result provides assurance that the samples are safe for consumption. However, further studies are needed to determine the aflatoxin levels in other types of pepper samples.

**P1-115**

**Food Contact Polymer Safety Vulnerabilities and Use of Failure Mode Effects Criticality Analysis for Effective Worker and Food Safety and Chemo-Bioterrorism Management**

**Harry Michael**
**Christopher Griffin**
**Stephen Atwood**

**FMC Ltd., Philadelphia, PA, 2RoboHygiene Consulting, Dublin, United Kingdom, 3Eagle Protect BRC, South Lake Tahoe, CA**

**Introduction:** The safety of food contact polymers (PCPs) in food packaging and disposable gloves are regulated for safety, with reliable estimates of $11 billion in flexible food packaging and 40 billion dollars of disposable gloves utilized in the United States per year.

**Purpose:** To assess the feasibility and potential for applying the HACCP framework to the food contact polymer industry.

**Methods:** Scenario modeling employed failure mode effects criticality analysis and root cause analysis to understand the various food safety and disease resulting effects and criticality. Further data collection, deductive failure analysis was employed with multiple additional and intentional events to identify key responses for gaps and targets for mitigation.

**Results:** When materials involved, production processes, facility type/geographic location, work force implications and quality/safety assurance standard operating procedures are reviewed, the two product categories have differing risk profiles. The priority risk numbers indicate that negative impacts for food processor safety and chemical contamination may be avoided through the implementation of improved plant design and equipment. The latter is seen to reach a potential impact of one to two billion dollars. For food packaging, the latter are varied, production automation is the most commonly produced in the United States, with frequent inspection and shelf-life testing. Poor manufacturing, storage or use of food packaging may result in worker injury, and physical, chemical and microbiological contamination.

**Significance:** The data suggests that for food packaging risks are being managed or low based on circumstances, but that for disposable gloves there is an accumulation of chemical, microbiological, and user safety/efficacy risks where control mechanisms are lacking.

**P1-116**

**Inactivation of Enterococcus faecium and Salmonella in Fried Potato-based Snacks**

**Abdulatif Tay**, **Rico Suhail**, **Amy Park**, **Erdogan Ceylan**

**1PepsiCo, Barrington, IL, 2FLNA, Plano, TX, 3Mérieux NutritionSciences, Crete, IL**

**Developing Scientist Emerent**

**Introduction:** Potato-based snacks are a popular food item consumed worldwide. Salmonella is a pathogen of concern in low moisture ingredients that can be associated with potato-based snack pellets. Enterococcus faecium NRRL B-2354 is commonly used as a surrogate for foodborne pathogens in low moisture products.

**Purpose:** This study investigated the fate of Salmonella and E. faecium in potato-based snack pellets at 10% moisture level when subjected to thermal treatment in oil at 50, 90 and 100°C.

**Methods:** Pre and post-enforcement surveys were conducted. A requirement for the Food Safety Modernization Act (FSMA) Produce Safety Rule (PSR) is that at least one member of a produce farm/processor must have attended a training program that is offered or regulated by the FDA. Evaluations included HACCP, food defense, and product recall. In addition, we evaluated the sanitary conditions in processing facilities and processors' attitude about the training techniques.

**Results:** The facility sizes ranged from 240 ft² to 3,200 ft². The number of employees were <10 and 20 to 49 for 90% and 10% of the facilities, respectively. The average of catfish processed per facility ranged from 10,000 to 22,000 lb per year. At the beginning of the study, all one facility had a HACCP plan developed and implemented. After one year of full enforcement, all the facilities developed a HACCP plan. Even though USDA presence in these facilities improved safety documentation, processors stated that processing procedures did not change. An 80% of the first time that regulations of seafood products moved from the Food and Drug Administration (FDA) to USDA.

**Significance:** The data suggests that for food packaging risks are being managed or low based on circumstances, but that for disposable gloves there is an accumulation of chemical, microbiological, and user safety/efficacy risks where control mechanisms are lacking.
**Purpose:** The study examined knowledge and attitudes of campus dining employees and identified barriers of accommodating students with food intolerances and food allergy.

**Methods:** The study was conducted using both paper and internet-based surveys. The study obtained 103 responses from University dining employees in the U.S., who are at least 18 years old.

**Results:** Descriptive statistics provided the demographic profiles of participants. Male (n = 49, 47.6%) and female (n = 49, 47.6%) were equally distributed. Most participants were Caucasians (n = 51, 49.5%), and in the range of 21 to 30 years old (n = 37, 35.9%). Linear and multiple regressions indicated that the knowledge (4.45 ± 1.10, p < 0.002) and the attitudes (3.68 ± 1.23, p < 0.000) were significant predictors of food safety practices (4.77 ± 1.68). Respondents were not knowledgeable when asked about food allergies (n = 41, 21.4%), and common food intolerance (n = 27, 26.2%). Unable to label allergen information and provide regular trainings due to time constraints were the top two barriers to dining employees accommodating students.

**Significance:** Employees' knowledge and attitudes towards food intolerance and food allergy were positively related to food safety practices on campus. Results can be used by university dining services to develop better strategies to accommodate students with food intolerance and food allergy.

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**P1-119 Evaluation of Hydrocooling with Two Different Sanitizers in Reducing Microbial Load and Shelf Life for Whole Corn**

Jayankar De, Bruna Bertoldi, Christopher Pabst, Christopher Baker, Alan Gutiérrez, Steven Sargent and Keith Schneider

*University of Florida, Gainesville, FL*

**Developing Scientist Entrant**

**Introduction:** Growers typically cool corn to in-crate and hydrocool quickly with chilled water to remove field heat. Efficiency of hydrocooling without sanitizer or with 150 ppm free Cl (FC) or 80 ppm peroxyacetic acid (PAA) as sanitizer was tested to reduce microbial load from whole corn.

**Purpose:** Compare the efficacies of hydrocooling with FC and PAA in reducing microbial load from whole corn and effect these treatments on their shelf life.

**Methods:** Corn was hydrocooled for ~60 min with plain water, whereas unwashed corn was used as a dry control. Water was amended with 150 ppm FC or 80 ppm PAA, respectively. Three trials (n=3) were run for each hydrocooling experiment. Water was analyzed for sanitizer concentration, pH, total dissolved solids (TDS), turbidity, temperature, and chemical oxygen demand (COD). Corn and water were analyzed for aerobic plate count (APC) and yeasts and molds (Y and M) counts.

**Results:** Both FC and PAA reduced initial microbial load from whole corn. FC would be beneficial in preventing cross contamination as no microbes were detected in the water. FC, compared to PAA, resulted in better microbiological quality of whole corn during storage.

**Significance:**

Both FC and PAA reduced initial microbial load from whole corn. FC would be beneficial in preventing cross contamination as no microbes were detected in the water. FC, compared to PAA, resulted in better microbiological quality of whole corn during storage.

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**P1-120 Advanced Environmental Sampling and Testing Methods for Outbreak Investigations**

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**Purpose:** Environmental investigations during foodborne and waterborne outbreaks help identify potential sources of contamination and provide information needed to facilitate remediation and prevention efforts. During environmental investigations, the use of advanced water collection methods, collection of complementary environmental samples, and use of targeted sample testing methods can improve detection rates for pathogens and fecal indicators.

**Methods:** To describe advanced environmental sampling and testing methods used in environmental investigations to help elucidate fecal contamination sources, we performed an environmental scan and conducted several case studies.

**Results:** We identified several innovative high throughput methods for the detection of environmental pathogens, including the sample processing method developed using a novel multistage extraction process and targeted sample testing methods for the detection of specific pathogens. The use of these targeted sampling and testing methods has resulted in increased detection rates of pathogens and fecal indicators from environmental samples collected during outbreak investigations. These data help to inform the potential contamination sources, design follow up studies into the factors that contribute to contamination, and extend this model to applications such as sampling in produce fields or corn in bins.

**Significance:**

Advanced environmental sampling and testing methods can be deployed during outbreak investigations to improve data generation and inform measures for improved public health.

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**P1-121 Bactericidal Effect of Non-Thermal Plasma Against Foodborne Pathogens on Diverse Foods**

Jin-Young Han

*Journal of Food Protection Supplement*

**Purpose:** The purpose of the study was to evaluate the bactericidal effect of non-thermal plasma against foodborne pathogens in various food samples (fruits, vegetables, nuts and powdered foods) and observe the relationship between pathogen inactivation and surface properties.

**Methods:** Fresh-cut carrot, celery, onion, and red bell pepper were spot-inoculated with culture cocktail of *Escherichia coli* O157:H7, Salmonella Typhimurium and *Listeria monocytogenes* (except red pepper) and treated with non-thermal plasma up to 20 min. Hydrophobicity and surface roughness of food samples were measured using optical tendiferometer and a noncontact 3D surface profiler, respectively. All experiments were repeated three times.

**Results:** Non-thermal plasma for 20 min treatment reduced *E. coli* O157:H7 by 0.93 to 5.49 CFU/cm² (fruits), 0.35 to 5.16 CFU/cm² (vegetables), 0.90 to 1.58 CFU/cm² (nuts), and 0.76 to 2.66 CFU/cm² (powdered foods), respectively. As treatment time increased, microbial reduction also increased. After non-thermal plasma treatment, 20 ppm of 0.1 M acetic acid on apples on apple, and red pepper reduced gradually from 3.66 to 4.94 CFU/cm², 2.53 to 3.37 CFU/cm² and 2.40 to 2.27 CFU/cm², respectively. There was no significant difference in slope of pathogen inactivation by non-thermal plasma treatment. The results also showed that surface roughness was more important factor than hydrophobicity correlated to bacterial inactivation by SDBD non-thermal plasma gas treatment.

**Significance:** These data concluded that roughness is an important factor for SDBD non-thermal plasma gas treatment. Low surface roughness food samples showed higher inactivation rate using SDBD non-thermal plasma gas treatment.

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**P1-122 Comparing Efficacy of Hydrocooling with Different Concentrations of Free Chlorine in Reducing Microbial Load from Whole Corn**

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**Purpose:** Growing hydrocool corn with chilled water to reduce the field heat and maintain its freshness. Efficacy of hydrocooling with or without free chlorine (FC), using sodium hypochlorite as sanitizer to reduce microbial load from whole corn, was tested in a mobile hydrocooler.

**Methods:** Growers typically pack corn in to crate and hydrocool quickly with chilled water to reduce the field heat. Water was amended with 150 ppm FC or 80 ppm PAA, respectively. Three trials (n=3) were run for each hydrocooling experiment. Water was analyzed for sanitizer concentration, pH, total dissolved solids (TDS), turbidity, temperature, and chemical oxygen demand (COD). Corn samples were analyzed for aerobic plate count (APC) and yeasts and molds (Y and M) counts. Results: The COD in water increased from 53 ± 0.15 ppm when hydrocooled with 75 ppm FC and had a final TDS of >710 ± 0.07 ppm and oxidation-reduction potential of >890. Unwashed corn had an initial COD of >720 ± 0.70, TDS of >3000 ppm and oxidation-reduction potential of >830. Unwashed corn had APC of 15.96 ± 1.66 and 9.76 ± 0.84 CFU/cm², respectively. APC and Y and M counts remained unaffected by plain water but were reduced to 9.06 ± 0.36 and 6.53 ± 0.29 log CFU/cm², respectively. Hydrocooling with 75 ppm FC reduced microbial load from whole corn. The reduction in EC could be due to excessive COD and TDS in the water.

**Significance:**

Principal flaw showed no effect on APC and Y and M counts, while FC reduced microbial load from corn. The lower reduction seen in higher EC could be due to excessive COD and TDS in the water.

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**P1-123 WITHDRAWN**

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**P1-124 A Novel Simulation Approach to Improving the Effectiveness of Sampling for Bulk Food Products**

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**Developing Scientist Entrant**

**Introduction:** Sampling for bulk food products has been studied by sampling in a grid and the effectiveness of sampling has been evaluated by classical statistical theories. Developing a model that takes samples in a continuous space and evaluating it by simulation may offer a alternative view to sampling in realistic food sampling scenarios.

**Purpose:** This study intends to improve sampling effectiveness by better understanding the impact of relevant factors (prevalence, sampling strategy, and sampling performance).

**Methods:** A coordinate-based model was built in it to simulate random point-contamination, sampling, and lot rejection using attribute plans. The model was applied to a 10 by 10 field with contamination at random locations and predicted the ranges of probability of detection using the Monte Carlo techniques. The acceptance and rejection rates were calculated by implementing the model in a C program and comparing it with the experimental data analysis to compare the regression analysis with the experimental data analysis. The probability of detection was positively correlated with the prevalence (R=0.20±0.10) and the sample size (p=0.20±0.10). The correlation between prevalence and sample size was significant (P<0.01). Sampling strategies are significantly affected by the prevalence, sampling rate, and sampling performance.

**Results:** The probability of detection was positively correlated with the prevalence (P<0.01). The selection of the sampling strategy was determined by the field conditions and the sample size. The selection of the sampling strategy was determined by the field conditions and the sample size. The selection of the sampling strategy was determined by the field conditions and the sample size. The selection of the sampling strategy was determined by the field conditions and the sample size.

**Significance:**

Sampling strategy, sampling performance, and sample size suggested that neither STRS nor step-keep 55 has significantly different performance than that of STRS (P<0.001 and 0.05 respectively).

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**P1-125 WITHDRAWN**

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**P1-126 Deep Cleans Alone Do Not Reduce *Listeria monocytogenes* Persistence in Retail Deli with Known High Prevalence**

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**Developing Scientist Entrant**

**Introduction:** Retail deli environments can be persistently contaminated with *Listeria monocytogenes*. We hypothesized that deep cleans and improved SSGPs in delis with known high *L. monocytogenes* prevalence would reduce prevalence and persistence.

**Methods:** We used *L. monocytogenes* strains before, immediately after, and following up from a deep clean intervention in stores with high *L. monocytogenes* prevalence.

**Results:** *L. monocytogenes* strains were identified to have high (0.1%) *L. monocytogenes* prevalence. Deep cleans were conducted in collaboration with corporate sanitziers and food safety managers. Environmental samples were collected from 20 sites before, after, and longitudinally post-deep clean. PFGE was performed on isolates to define strains; PFGE patterns were analyzed in BioNumerics (Applied Maths, v. 6.6) using an unweighted pair group-matching algorithm.
Results: The simulation model outperformed the random forest model with AUCs of 0.873 and 0.700, respectively. The developed models can be used to predict the prevalence of L. monocytogenes in pastured poultry farm environments based on weather factors and consumer storage conditions.

Significance: The developed models could be used to predict the prevalence of L. monocytogenes in pastured poultry farm environments based on weather factors and consumer storage conditions. Further studies exploring factors beyond sanitation are necessary to assess the microbiological risk of smoked salmon.

P1-130 Risk Estimation of Clostridium perfringens from the Consumption of Hamburger and Sandwich Products Available in Retail Markets Using Probabilistic Modeling
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Introduction: Clostridium perfringens is the leading causative bacterial agent for foodborne illness in Korea in 2018. RTE foods including hamburgers and sandwiches are a convenient meal for consumers, but the risk for foodborne illness associated with RTE foods is high. In order to develop effective risk management strategies, the risk of C. perfringens in RTE foods needs to be identified.

Objective: The purpose of this study was to develop a probabilistic model to estimate the risk of C. perfringens from the consumption of hamburgers and sandwiches in Korea.

Methods: A predictive model for the survival of C. perfringens in hamburgers and sandwiches was developed using RANsim and DAVEY models. Data on microbial contamination in initial samples and the temperature and time during retail markets to transportation were monitored using the model provided by the Food Code in data logger, respectively. Consumption patterns were adopted from the MGEU 2011 study and exponential model was used as a dose-response model. The probabilistic risk model was developed based on the collected data using the @Risk program.

Results: The developed predictive model showed that the required time for the first decimal reduction was increased as temperature decreased. The estimated initial contamination level of C. perfringens was 5.2 log CFU/g. As a result of Monte Carlo simulation, the estimated probability of infection (Pf) caused by C. perfringens from the consumption of hamburgers and sandwiches was 1.38×10⁻³ per person per day. The sensitivity analysis highlighted the distribution of contamination frequency and prevalence of microbial contamination as the most influential input variables on the Pf.

Significance: The developed model can be used as a risk management strategy to identify the critical control points for point of purchase to control the risk of C. perfringens during the manufacturing process.

P1-131 Quantitative Assessment of Listeriosis Risk from Domestic Cheese Consumption in Korea
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Introduction: Listeriosis is an illness that can be serious or even life-threatening, particularly for pregnant women, their babies, and elders. Various cheeses are involved in outbreaks of listeriosis worldwide due to high consumption and prolonged refrigerated storage.

Objective: This study estimated the risk of infection by L. monocytogenes due to consumption of cheeses produced in Korea.

Methods: Cheese samples were collected from raw and processed cheeses from raw milk and pasteurized milk cheese factories located in seven regions in Korea. The contamination level of L. monocytogenes was determined using a modified GMPs test. The maximum probability of an outbreak of listeriosis by the consumption of cheeses in Korea was 6.18×10⁻⁴ per person per day. Results of sensitivity analysis show that the frequency of intake was the highest correlated, followed by storage temperature in the home and consumption frequency.

Significance: Considering the frequency of consumption, vulnerable groups such as pregnant women and elders should be educated on the risk of listeriosis. In addition, proper time and temperature management from commercial cheese markets, especially online, should be emphasized.

P1-132 Quantitative Microbial Risk Assessment of Listeria monocytogenes in Smoked Salmon from Retail Market to Home
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Introduction: High water activity and neutral pH of smoked salmon are a good environment for the growth of Listeria monocytogenes at refrigeration temperature. But, despite its high fatality rate, contamination of L. monocytogenes in smoked salmon leads to serious risks to consumers. Thus, it is necessary to assess the microbiological risk of smoked salmon.

Objective: The objective of the study was to conduct a quantitative microbial risk assessment of L. monocytogenes in smoked salmon from the retail market to home.

Methods: The initial contamination levels of L. monocytogenes in smoked salmon (n=375) at retail were monitored. To predict the change of L. monocytogenes populations from the market to home during transportation, a predictive model of L. monocytogenes in smoked salmon was developed as a function of temperature (four, 10, 17, 25 and 36°C). Data on daily consumption amount and frequency of smoked salmon were collected on 1,011 individuals in Korea. A simulation model was developed and the probability of foodborne illness by the consumption of smoked salmon was estimated with R@isk.

Results: The growth model of L. monocytogenes in smoked salmon was updated into the simulation model. Daily consumption amount and frequency of smoked salmon were estimated with R@isk. The maximal daily dose-response model of L. monocytogenes was used and the probability of illness by L. monocytogenes of smoked salmon was 5.93×10⁻⁴ per person per day.

Significance: L. monocytogenes is a serious threat to the safety of refrigerated and frozen smoked salmon. Once the contamination of L. monocytogenes in smoked salmon becomes high, severe risks and serious health threats arise at the retail market. Thus, it is important to implement HACCP and cross-contamination from raw fish and equipment in the processing facility.

Poster 7

P1-127 Microbiological Risk Assessment of Staphylococcus aureus in Ready-to-Eat Lettuce in Taiwan
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Introduction: In Taiwan, Staphylococcus aureus is a major foodborne pathogen causing a high number of illnesses linked to the consumption of RTE lettuce products, particularly those that are non-compliant with Taiwan’s Certified Food Hygiene Standards (CAS).

Objective: This study aimed to estimate the probabilities of illness caused by S. aureus in CAS and non-CAS RTE lettuce in Taiwan and to identify the critical points for control to reduce the risk.

Methods: The data collected for the quantitative risk assessment included the prevalence and initial levels of S. aureus in RTE lettuce, the time and temperature profiles during lettuce processing, transportation, and storage, and the consumption patterns. Laboratory experiments were performed to develop temperature-dependent growth models of S. aureus in lettuce. Five modules were constructed to quantify the growth and infection risk of S. aureus in lettuce under time and temperature conditions that the products are likely to be exposed to from the processing plants to before the consumption.

Results: The probability estimated by S. aureus in CAS lettuce was estimated to be 4.03×10⁻⁷ per serving (95% CI: 1.58×10⁻⁷ to 4.03×10⁻⁴). The probability was higher for non-CAS lettuce at 3.86×10⁻⁶ (95% CI: 1.07×10⁻⁶ to 7.22×10⁻⁶), indicating non-CAS lettuce poses a significantly higher S. aureus infection risk to the consumers. It is estimated that approximately 160 more cases of staphylococcal illnesses would be caused by non-CAS lettuce than CAS lettuce. The sensitivity analysis showed that the consumer storage conditions, the initial contamination level of S. aureus in lettuce, and the retail storage temperature were the most significant factors influencing S. aureus risk of RTE lettuce in Taiwan.

Significance: The findings demonstrated that the microbial food safety measures in CAS are effective in reducing the risk of illness caused by S. aureus in RTE lettuce and identified the critical control points in lettuce processing to consumption to increase the microbial safety of RTE lettuce.
P1-134 Quantitative Risk Assessment for Clostridium perfringens in Pickles and Kimchi

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Poster

P1-135 Microbial Risk Assessment of Vibrio parahaemolyticus in the Salted Seafood Jeotgal

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Poster

P1-136 Model Development for Survival of Vibrio Tannaeformis in Tuna Sashimi as a Function of Temperature

Yun Jin Lee, Min Kweon, Young Song and Ki Sun Yoon

Journal of Food Protection Supplement

Poster

P1-137 Evaluation of Food Defense in Hospitality

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Poster

Introduction: Food defense is the protection of foodstuffs from threats of contamination and adulteration that may cause harm to public health and economic damage. In Europe, it is still a relatively new topic, since the standards of food security existing in the hotel industry are based mainly on HACCP. With the increase of tourism, it is important to ensure overall security and maintain external and internal image of hospitality businesses.

Purpose: Evaluate and assess the preparedness level of the Portuguese hospitality businesses for the prevention of intentional contamination acts.

Methods: There were audits in eight five-star hospitality businesses in Portugal's southern region and food defense plans were formulated for each one of them, using the American software Food Defense Plan Builder (FDPB), created by the FDA.

Results: According to FDPB methodology, the visited units scored from 87.7% to 93.1% with an average of 90.6% conformity. In the vulnerability/ accessibility assessment, the risk assessment was from the full range of sites ranged from 7.1 to 11, with an average of nine, on a scale where zero is inescapable and inaccessible and 20 visible. A low score for the vulnerability and a high score for accessibility means represented the risk of 95.9% scored a medium to low priority. From the 18 FDPB pre-screening questions group, eight scored conformity of 90% and in the vulnerability/accessibility evaluation, none of the average values reached the acceptable threshold.

Significance: Results revealed an extremely positive level of preparedness for food defense, even more for such a risky methodology to be used in the food sector, excluding the United States of America. This work can be used to expand the expertise and certification of food sectors businesses in Portugal in food defense.

P1-138 Risk Assessment of Clostridium perfringens in Salted and Fermented Squid (Seaquid Jeotgal)

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Poster

P1-139 Risk Assessment of Clostridium perfringens in Paste-type Fermented Sauces

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Poster

Introduction: Foodborne illness caused by Clostridium perfringens is continuously reported in Korea, and there is high probability of C. perfringens contamination in paste-type fermented sauces.

Purpose: The objective of this study was to analyze the risk of C. perfringens infection from consumption of paste-type fermented sauces.

Methods: The prevalence of C. perfringens in paste type fermented sauces was investigated by plating the samples on tryptose sulfite cycloserine agar. C. perfringens could be through the exponential distribution. -values decreased 225.30% to 18.6% as storage temperature increased, and the developed secondary model was appropriate to calculate the probability of C. perfringens foodborne illness.

Results: One hundred seventeen samples were analyzed, and six samples were C. perfringens positive. Thus, initial contamination level of the pathogen was estimated to be through the exponential equation. C. perfringens was used to develop the secondary model, and a dose-response model was also used to be developed. With all collected data, a simulation model was prepared, and the simulation was used to calculate the probability of C. perfringens foodborne illness.

Significance: This result indicates that the risk of C. perfringens in squid jeotgal is very low in Korea.

Results: Beta distribution for V. parahaemolyticus prevalence in jeotgal showed that the mean initial contamination level was 3.2 log CFU. The developed predictive model showed that the pathogen levels gradually decreased under the investigated conditions (mean temperature: 20.5°C; mean time: 16.32 h). Jeotgal consumption amount and frequency was 13.89 g and 0.8%, respectively, and the β-Poisson model [risk=1-(1+dose/1.18×10^-5)] was used to simulate the probability of V. parahaemolyticus contamination in squid jeotgal underway. The model will allow food safety personnel to take action as educational agents and as promoters of best practices and standards, playing important roles in the prevention and control of foodborne diseases.

Significance: The results may be used for microbial risk assessment in squid jeotgal products.

Methods: Squid jeotgal is fermented squid that is salted and seasoned with spices, followed by fermentation. Because sauces may have Clostridium perfringens spores and anaerobic condition are created by fermentation, there is a possibility of C. perfringens growth in squid jeotgal.

Purpose: The objective of this study was to assess the risk of C. perfringens in squid jeotgal.

Results: A modified Gompertz and Weibull primary model was used to obtain growth and survival kinetics, and stored at four, 10, 12, 14, 15, 20, 25 and 27, and 30°C. A modified Gompertz and Weibull primary model was used to obtain growth and survival kinetics.

Results: Of 118 samples, 8 samples were detected in eight pickles and kimchi samples (6.8%), and the initial contamination level of C. perfringens was estimated, using RiskBeta (0.911). Exposure values (h) for the pathogen decreased as temperature increased. Temperature during transportation and transportation and during market display were evaluated. (121.394, 280.3, 3.129, 3.129, 3.129), distribution and uniform (1.7535, 21.52) distribution. The consumption amount and frequency were 78.95 g and 60.3%, respectively. Exponential dose-response model was induced in the simulation model. The simulation with the collected data showed that the probability of illness per person per day was very low.

Significance: This indicates the risk of the prevalence of C. perfringens in pickles and kimchi is very low in Korea.

Conclusion: Intake of temperature, consumption amount and consumption frequency were also surveyed. In addition, predictive models for contamination levels of V. parahaemolyticus were introduced into pickles and kimchi. In addition, there were recalls for C. perfringens-contaminated kimchi.

Purpose: The objective of this study was to assess risk of C. perfringens in pickles and kimchi.

Methods: Redish kimchi was inoculated with a mixture of C. perfringens strains at four log CFU. The kimchi samples were fermented at 7 to 35°C. The C. perfringens cells counted were enumerated on tryptose sulfite cycloserine agar. The cell counts were used to develop predictive models with the Weibull and a polynomial primary model. To prepare a simulation model with @Risk program, probabilistic distributions for initialization and distribution conditions were prepared, and consumption data and a dose-response model were collected. The probability of illness/person/day for C. perfringens was calculated through the simulation.

Methods: A mixture of pathogenic (ATCC 27519, 43996) and nonpathogenic V. parahaemolyticus (ATCC 17802, 33844) was used to simulate various storage conditions. C. perfringens was used as a probiotic spores and anaerobic condition are created by fermentation, there is a possibility of C. perfringens growth in squid jeotgal.

Methods: In this study, the probability of V. parahaemolyticus contamination in squid jeotgal is very low in Korea.

Conclusion: Data about storage temperature and time of paste type fermented sauce were collected, and probabilistic distributions for the data were determined, using @Risk software. Developing Scientist Entraunt.
Effect of packaging on the Risk of Cladostrium perfringens in Ready-to-Eat Lunch Boxes Sold at Convenience Stores

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Introduction: The demand for RTE lunch boxes sold in convenience stores has recently increased in Korea. The most popular main dishes in the lunch box are beef, pork, and chicken, etc., which are suitable for the growth of C. perfringens. Due to their popularity, it is necessary to evaluate the risk of C. perfringens in RTE lunch boxes.

Purpose: The objective of study was to investigate the effect of packaging method on the risk of C. perfringens in RTE lunch boxes called “Dosirak” sold at convenience stores.

Methods: The types and main menus of lunch boxes were investigated at four major convenience stores. The initial contamination levels of C. perfringens in RTE dosirak were monitored. A predictive model was fitted well to survival behavior of C. perfringens as function of temperature and packaging. The B and P values were calculated. The demand amount and frequency of RTE lunch boxes were investigated at the most popular convenience store in Korea. The probability of C. perfringens outbreak was calculated by simulation with BioRisk.

Results: Ninety-two different kinds of lunch boxes were sold at major convenience stores. The most popular main menu of dosirak was jeyuk-bok-jeon (stir-fried pork), followed by bulgogi (stir-fried beef) and teriyaki chicken. C. perfringens was not detected (<0.5 log CFU/g) in RTE dosirak. Higher B and P values of C. perfringens in anaerobic packed dosirak were observed than in aerobic packed dosirak. An exponential model was used as a dose-response model. Subsequently, the probability of foodborne illness by C. perfringens per person per day with consumption of dosirak was 5.62×10^-11 in anaerobic packed dosirak and 1.14×10^-13 in aerobic packed dosirak.

Significance: This result suggests that the risk of C. perfringens in ready-to-eat lunch box is very low, regardless of packaging method.

Quantitative Microbial Risk Assessment of Vibrio parahaemolyticus from the Consumption of Ready-to-Eat Foods Containing Seafood Available in Retail Markets

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Introduction: RTE foods containing seafood (RTES) can be contaminated with Vibrio parahaemolyticus and have been implicated in foodborne outbreaks.

Purpose: In order to develop the risk management strategies, risk assessment of V. parahaemolyticus in RTE foods is essential.

Methods: The developed risk model and risk outputs provided scientific background regarding risk management options to control the risk of V. parahaemolyticus.

Quantitative Microbial Risk Assessment Modeling Techniques in Managing Microbiological Food Safety Risks: Risk-based Hazard Analysis and Critical Control Point Plans

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Introduction: Hazard Analysis and Critical Control Point (HACCP) is an internationally recognized system to assure the safety of food products and the foundation of global food safety programs. However, its success is limited by its inability to relate stringency to measurable public health impacts due to its inherent qualitative nature.

Purpose: The aim of this research was to incorporate quantitative microbial risk assessment (QMRA) techniques into HACCP to develop risk-based HACCP (RB-HACCP) plans.

Methods: The researchers hypothesized that the Critical Control Points (CCPs) step in the process that can be identified using risk assessment modeling techniques such as sensitivity analysis (SA) and what-if scenario analyses can be used to more objectively evaluate Critical Limits (CLs). QMRA models were developed to identify potential risk-based CCPs (RB-CCPs) for Listeria monocytogenes for two food products: frankfurters and cold-smoked salmon (CS). The former was selected because of its partial chain of processing steps. The latter was selected using SA to determine steps that most contribute to control of E. coli. Frankfurters and CS were used to identify potential CCPs and risk-based limits for the latter were established to meet the most effective risk mitigation strategies.

Results: This conceptual framework, combined with relevant plant-specific data, was used to develop RB-CCPs and RB-CLs, thereby producing RB-HACCP plans that are linked with public health goals to achieve lower risk of listeriosis. This allowed a direct comparison between industry HACCP plans for frankfurters and CSS with RB-HACCP plans derived from the risk assessments.

Significance: The comparison suggests that the use of RB-HACCP plans may offer advantages in developing the preventive controls risk management food safety plans required under the FDA Food Safety Modernization Act of 2011.

P1-143 Comparison of Linear and Non-linear Models to Describe the Inactivation Kinetics of Vegetative Pathogens during Oil Roasting of Sunflower Kernels

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Introduction: D- and z-values are widely used to determine the heat resistance of microorganisms. Calculation of D- and z-values assumes first-order kinetics. However, factors such as strain variability, starting inoculum, matrix, and other environmental factors may cause the microorganisms to display non-linear kinetics. The utilization of δ-values to determine heat resistance enables microorganisms to display non-linear kinetics. Hence, alternative approaches to linear models must be considered.

Purpose: The objective of this study was to compare linear and non-linear models to describe the inactivation kinetics of Salmonella spp., Listeria monocytogenes, Shiga toxin-producing E. coli and Enterococcus faecium in sunflower kernels during oil roasting.

Methods: Sunflower kernels were inoculated with multi-strain Salmonella spp., L. monocytogenes, and E. coli, followed by oil roasting to 205°C. Following cooling, samples were immediately cooled and enumerated for the inoculated pathogens using scientifically valid microbial testing procedures. All experiments consisted three replicates. Linear and non-linear regression models were fit to log-transformed data using Proc. REG procedure in SAS software. Models were statistically deemed fit at α=0.05.

Results: Data analysis indicated that non-linear regression models described inactivation kinetics in Salmonella spp., L. monocytogenes, and E. coli better compared to linear models. The R-square values ranged from 0.90 to 0.98. Linear models were found to be a better fit for E. faecium. The R-square values ranged from 0.91 to 0.98.

Significance: Data analysis indicated that non-linear regression models described inactivation kinetics in Salmonella spp., L. monocytogenes, and E. coli better compared to linear models. The R-square values ranged from 0.93 to 0.98. Linear models were found to be a better fit for E. faecium. The R-square values ranged from 0.91-0.98.

P1-144 Monetizing the Effect of Food Safety Recalls on the Low-moisture Food Industry

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Introduction: Food Safety and Modernization Act (FSMA) Preventive Controls Rules require that food producers validate that their processes sufficiently reduce the risks from microbial pathogens. However, food safety decisions are inherently business decisions, and there currently are no quantitative tools for monetizing the benefits of those investments.

Purpose: The objective of this study is to estimate the financial losses from food recalls of low-moisture foods, with the ultimate goal of justifying food safety technology investments.

Methods: Financial impacts of low-moisture food recalls are estimated by applying the Cumulative Abnormal Returns (CAR) in stock values over the recall event period. Using GRET software, returns for a stock under non-recall conditions were predicted. The abnormal return (AR) was calculated as the difference between predicted and actual returns during the event period. Abnormal returns were aggregated to compute CAR over each day of the recall event period when the recall was announced. The CAR was multiplied by the pre-recall market capitalization for a company, to compute lost value due to the recall.

Results: The regression predicted for returns yielded P-values = 0.01 for all 14 cases. CARs (20 days post-recall) were -26.5 to 5.48% (mean of -0.3%). Assuming a 0.05 annual recall risk, the average loss 20 days post-recall was $3.34M, with the range varying from a loss of $102M to a gain of $3.1M. Assuming implementation of a food safety technology that reduces recall risk by 0.001, mean financial loss would be reduced to $478k per event.

Significance: Class I recalls cause major financial losses for publicly-held manufacturers of low-moisture foods. These loss estimates provide an incentive for the industry to invest in improved food safety technology, and the results provide concrete financial justifications for such investments.

P1-145 Creating a Risk Model for Nosocomial Listeriosis in Cancer Patients Who Consume Ready-to-Eat Salad

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Undergraduate Student Award Entrant

Introduction: Listeriosis is a foodborne illness with relatively low incidence, but substantial mortality rates. Risk of listeriosis is greater in immunocompromised populations, particularly cancer patients, because their treatments compromise several of the barriers against infection. Therefore, nosocomial foodborne listeriosis remains problematic for these patients, with consumption of RTE fresh salads raising particular concern.

Purpose: The objective of this study was to develop a data-driven risk model for Listeria monocytogenes in cancer patients due to RTE salads.

Methods: Risk of listeriosis from salad consumption was assessed using an exponential dose-response model with rate constant r of 1.79E-10, adopted from a 1997 model that conservatively estimated risk of listeriosis for immunocompromised individuals. Food consumption data were collected from 100 patient surveys, with salad being defined as green lettuce, raw spinach, and/or raw tomatoes. Risk was calculated using Monte Carlo simulation in Crystalball software. Risk was multiplied by a scale factor of 4, as cancer patients have been shown to be 4-9 times more likely to develop listeriosis than individuals with other immunocompromising affiliations. The hypothetical efficacies of two risk management strategies, washing treatments (treading in water for two minutes, and immersion in sodium hypochlorite for 15 minutes followed by a tap water rinse) and storage (at 40°F and 15°C), were also assessed.

Results: The maximum risk of listeriosis due to salad consumption during one cycle of chemotherapy was 0.0051 (0.51%). Ninety percent of risk values were less than 0.0001. Storing at 5°C and rinsing with tap water substantially reduced the dose of L. monocytogenes per risk period.

Significance: Data-driven risk models for listeriosis in cancer patients could provide a justification for existing dietary restrictions.
P1-146 Application of Metagenomics to Define Listeria monocytogenes in Smoked Fish and Ice Cream Facilities

Deeya Kode, Ramakrishna Nannapaneni, Mohit Bansal, Wen-Hsing Cheng, Chander Shekhar Sharma and Aaron Kiess

Mississippi State University, Mississippi State, MS

Listeria monocytogenes is a persistent opportunistic of the respiratory tract and skin, either for human or animals and constitutes a contamination source in food or health-care.

Purpose: To determine the occurrence of Listeria monocytogenes in smoked fish and ice cream facilities.

Methods: Whole genome sequencing (WGS) of Listeria monocytogenes strains from smoked fish and ice cream facilities were sequenced in shotgun mode using Illumina MiSeq with a read length of 250 bp. The sequence reads were aligned using Bowtie2. The resulting nucleotide sequences were used to build the phylogenetic trees using the Neighbor-joining method. The results were compared with the reference strains of Listeria monocytogenes.

Results: The phylogenetic trees showed high similarity between the tested strains and reference strains. The genetic relatedness analysis indicated that the tested strains were closely related to the reference strains.

Significance: This study demonstrated that the culture-based environmental sampling enrichment methods may under-report Listeria monocytogenes contamination in food-manufacturing facilities.

P1-147 Distribution of Toxin Genes and Antimicrobial Resistance Genes among Staphylococci Isolated from Food Samples in Algeria

Divya Kode, Ramakrishna Nannapaneni, Mohit Bansal, Wen-Hsing Cheng, Chander Shekhar Sharma and Aaron Kiess

Mississippi State University, Mississippi State, MS

Staphylococcus aureus is commonly detected from beef, poultry, and processed meat products. In particular, poultry is a major cause of Staphylococcal foodborne outbreaks due to contamination from intestinal contents and cross-contamination between carcasses.

Purpose: To develop the mathematical models to describe the kinetic behaviors of Staphylococcus aureus in chicken and duck tenderloins during storage.

Methods: A mixture of Staphylococcus aureus strains NCCP14038, NCCP14039, NCCP15661 and NCCP11142 was inoculated in smoked duck slices (25 g) at three to four LPDs. Samples were exposed to gradually increasing sublethal concentrations by spiking at hourly intervals for five hours to reach a final concentration of 10^6 CFU/mL. The cell counts data were used to develop a primary model with the Baranyi model, calculating maximum specific growth rate (μmax). The secondary model was appropriate with R² = 0.915 to 0.928. Within validation, RMSE values of 0.309 (chicken) and 0.304 (duck) suggested that the model performance was acceptable in the development of kinetic behavior of L. monocytogenes in chicken and duck tenderloins.

Significance: This study showed a high frequency of SEs genes in food isolates and tet genes in clinical isolates; our findings provide updated data on the significance of antimicrobial resistance development in Staphylococci.

P1-148弘容性応答促進用法の制御 Listeria monocytogenes 胴肉試料に含まれる食肉製品中の細胞数の変動および増殖

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目的: Listeria monocytogenes の細胞数の変動と増殖を制御するための方法の開発。

方法: 脳別試験で検出された L. monocytogenes の細胞数に影響を与える要因を解析し、細胞数の変動を抑制する方法を考察。細胞数の変動を抑制する要因として、温度、湿度、酸素濃度などを分析した。

結果: サーキュレーターによる温度制御、湿度制御、酸素濃度制御の3要因が細胞数の変動を抑制し得ることが示された。この方法により、L. monocytogenes の細胞数の変動を制御することが可能であることが示された。

意義: 本研究の結果は、L. monocytogenes の細胞数の変動を制御する方法の開発に貢献すると考えられる。
P1-152 Physiological Characterization of Listeria monocytogenes Isolates from Smoked Duck
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2Korea Food Research Institute, Wanju, South Korea

Developing Scientist Entreat

Introduction: Listeria monocytogenes has been a cause of foodborne outbreaks related to ready-to-eat products, especially for smoked products. The consumption of smoked duck has increased, and it is usually served in sliced pieces. In addition, L. monocytogenes has been isolated from sliced smoked duck, even though it was worm-free before processing.

Purpose: The objective of this study was to characterize the physiological properties of L. monocytogenes isolates from smoked duck.

Methods: One-milliliter aliquots of L. monocytogenes isolates (SMFM201803 SD 1-1, SMFM201803 SD 4-1, SMFM201804 SD 4-2, SMFM201804 SD 5-2, SMFM201804 SD 10-1, SMFM201804 SD 10-2, SMFM201804BA SD 2-1) from smoked duck were inoculated into tryptic soy broth (TSB) with 0.6% yeast extract at 60°C, and the cell counts were enumerated on tryptic soy agar (TSA) with 0.6% yeast extract at zero, two, five, eight, and 10 min. To examine the antimicrobial resistance of the isolates, the optical densities of the L. monocytogenes cultures were adjusted to a 0.5 McFarland standard in triplicate. Then they were swabbed on the surface of tryptic soy agar plates and were incubated at 37°C for 48 h. The cultures were put into the SGF and incubated at 37°C for 125 h. A polynomial equation was used to determine the lag phase duration (LPD) and the polynomial equation was then fitted to the parameters as a function of temperature. A dynamic model was then developed to predict L. monocytogenes cell counts under changing temperature.

Results: As storage temperature increased, LPD (1.81 to 16.59 h) became shorter and µmax (0.01 to 0.34 log CFU/h) increased, indicating that L. monocytogenes can grow rapidly at high temperature. A polynomial equation was appropriate to describe the effect of temperature on the kinetic parameters, with R²=0.971 to 0.996, and the developed dynamic model was also appropriate to describe the fates of L. monocytogenes under changing temperatures. The RMSE value of 0.567 indicated that the developed model had good performance.

Significance: This result indicates that L. monocytogenes can proliferate rapidly in cucumber, and the developed models should be useful in describing the kinetic behavior of L. monocytogenes in cucumbers.

P1-155 Predictive Model of Growth of Listeria monocytogenes in Queso Fresco Cheese
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Hokkaido University, Sapporo, Japan

Developing Scientist Entreat

Introduction: Listeria monocytogenes is a hardy psychotrophic pathogen that has been linked to several cheese-related outbreaks in the United States, including a recent outbreak where queso fresco cheese was implicated.

Purpose: The purpose of this study was to develop mathematical models that can predict the growth of L. monocytogenes in a Mexican-style cheese known as queso fresco under non-isothermal conditions.

Methods: A mixture of five strains of L. monocytogenes were used to inoculate pasteurized whole milk to prepare queso fresco. Ten grams of fresh cheese samples were vacuum packaged and stored at four, 10, 15, 20, 25, and 30°C. Samples were taken from each batch at different time points, and they were then swabbed in a sterile buffer solution and plated on modified Oxford Agra with supplement. Growth data from each temperature was fitted using the Baranyai model as the primary model and the Ratkowsky square-root model as the secondary model. The primary and secondary models were further adopted to develop tertiary models under pH conditions and differences in strain.

Results: The Baranyai model was fitted to the growth data with acceptable goodness of fit statistics (R²=0.928; RMSE=0.317). Similarly, the Ratkowsky square-root model was fitted to the specific growth rates at different temperatures (R²=0.928; RMSE=0.317). In addition, the pH dependency of the scale parameter of the Weibull model was successfully illustrated by a log-linear regression model.

Significance: The models developed in this study were accurate since greater than 70% of the prediction errors were within the APD (0.5 Prediction Error=1.0).

P1-154 Modeling the Survival Kinetics of Campylobacter jejuni in Simulated Gastric Fluid
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Hokkaido University, Sapporo, Japan

Developing Scientist Entreat

Introduction: Although Campylobacter jejuni is not highly resistant to various environmental stresses, the number of Campylobacter jejuni infections has not decreased despite the advent of new preventive measures using non-isothermal-time temperature profiles. Campylobacter jejuni infection effectively, the relationship between Campylobacter jejuni infection and the number of ingested bacteria (dose) needs to be clarified. As an alternative new concept of dose-response modeling, the key events dose-response framework (KEDRF) has been proposed. This result indicates that L. monocytogenes can proliferate rapidly in cucumber, and the developed models should be useful in describing the kinetic behavior of L. monocytogenes in cucumbers.

Purpose: The objective of this study was to develop kinetic models and a dynamic model to predict the kinetic behavior of Salmonella cells in cucumber at changing temperatures.

Methods: Prepared cucumber samples were immersed in Salmonella inoculated buffer for three min, and then inoculated with the count at 0 min to allow cell attach.

Results: The samples were stored at 10, 20, 25 and 30°C. Triplicate 25-g samples were enumerated on 3M Petrifilm. To develop a primary model, the Baranyai model was fitted to the cell count data to calculate lag phase duration (LPD) and maximum specific growth rate (µmax log CFU/h). A polynomial model was then fitted to the parameters as a function of temperature. Similarly, a dynamic model was developed in accordance with primary and secondary models. To evaluate the accuracy of the model, the root mean square error (RMSE) was calculated.

Significance: This result indicates that the model can grow rapidly in diced cucumbers to high storage temperature, and the developed dynamic model can be useful in describing the kinetic behavior of C. jejuni in diced cucumbers at changing temperatures.

P1-157 A Risk Assessment Study of Staphylococcus aureus in Pancake Batter
Amanda Sinsey1 and Nancy Dobmeier2
1Colorado State University, Fort Collins, CO
2Genentech, South San Francisco, CA

Developing Scientist Entreat

Introduction: Staphylococcus aureus can grow rapidly at high temperature. A polynomial equation was appropriate to describe the effect of temperature on the kinetic parameters, with R²=0.971 to 0.996, and the developed dynamic model was also appropriate to describe the fates of L. monocytogenes under changing temperatures. The RMSE value of 0.567 indicated that the developed model had good performance.

Purpose: The objective of this study was to develop kinetic models and a dynamic model to predict the kinetic behavior of Salmonella cells in cucumber at changing temperatures.

Methods: Prepared cucumber samples were immersed in Salmonella inoculated buffer for three min, and then inoculated with the count at 0 min to allow cell attach.

Results: The samples were stored at 10, 20, 25 and 30°C. Triplicate 25-g samples were enumerated on 3M Petrifilm. To develop a primary model, the Baranyai model was fitted to the cell count data to calculate lag phase duration (LPD) and maximum specific growth rate (µmax log CFU/h). A polynomial model was then fitted to the parameters as a function of temperature. Similarly, a dynamic model was developed in accordance with primary and secondary models. To evaluate the accuracy of the model, the root mean square error (RMSE) was calculated.

Significance: This result indicates that the model can grow rapidly in diced cucumbers to high storage temperature, and the developed dynamic model can be useful in describing the kinetic behavior of C. jejuni in diced cucumbers at changing temperatures.
The estimated parameters were used as the prior information to construct the posterior distribution for Bayesian analysis. MCMC simulation was used to perform an analysis of the six dynamic runs of the simulated moderate models. A negative binomial distribution was fit to each using PROC GENMOD in SAS software. Values within the 95% confidence interval were determined for each bean type and temperature.

Results:
- The direct detection of bacteria from surfaces using non-specific ATP tests can be a first line defence for cleaning verification. This study emphasizes the need for developing and implementing proper cleaning protocols in high-risk industries, such as food processing plants, to ensure food safety and compliance with regulatory requirements.

Methods:
- A “Food Safety Modernization Act (FSMA) Readiness” team evaluated nine packinghouses in the Dominican Republic (La Vega province) to determine their FSMA adherence. Packinghouses were scored using interviews with management, mock facility inspections, and observation of sanitation practices.
- The purpose of this study was to investigate the growth kinetics of one mutant of Clostridium botulinum LNT01 in cooked beef during cooling.
- The study findings highlight that preventive controls must be administered during bean processing to significantly minimize and prevent growth of the S. aureus superbug. Therefore, in addition, preventive control management components must also be in place in the facility’s Food Safety Plan to ensure the effectiveness of the preventive measures.

Purpose:
- The objective of this study was to investigate the growth kinetics of C. botulinum LNT01, a non-toxic mutant of C. botulinum 62A, in cooked ground beef.
- The methods for spores of C. botulinum LNT01 were incubated to ground beef and incubated anaerobically under different temperature conditions to observe growth and develop growth curves. A one-step kinetic analysis method was used to analyze the growth curves to minimize the global residual error. The data analysis was performed using the USDA PPM-Global Fit.
- The results of data analysis showed that the minimum, optimum, and maximum growth temperatures of this mutant are 11.5, 36.4, and 44.3°C, and the estimated optimum specific growth rate is 0.65 CFU/hr per g or 0.34 log CFU per g per h. Both thermal and dynamic growth curves were used to validate the growth models and kinetic parameters. The residual errors of validation followed a Laplace distribution, with about 60% of the residual errors within ±2.5 log CFU of observations, suggesting that these models could predict the growth of C. botulinum LNT01 in ground beef with reasonable accuracy. Comparing with C. perfringens, C. botulinum LNT01 would grow much slower rates and with much longer lag times. Its growth kinetics is very similar to S. epidermidis in ground beef.
- The significance of this computer simulation with the kinetic models shows that, while prolific growth of C. perfringens may occur in ground beef due to the presence of the spores, C. botulinum LNT01 would occur under the same conditions. C. botulinum LNT01 mutant could be used as a surrogate for studying the growth kinetics of C. botulinum.

Introduction:
- The objective of this study was to investigate the growth kinetics of one mutant of Clostridium botulinum LNT01 in cooked beef during cooling.
- A one to 10 scale was developed to determine FSMA adherence. Packinghouses were scored using interviews with management, mock facility inspections, and observation of sanitation practices.
- The purpose of this study was to investigate the growth kinetics of one mutant of Clostridium botulinum LNT01 in cooked beef during cooling.
- The study findings highlight that preventive controls must be administered during bean processing to significantly minimize and prevent growth of the S. aureus superbug. Therefore, in addition, preventive control management components must also be in place in the facility’s Food Safety Plan to ensure the effectiveness of the preventive measures.
P1-165 Identifying Risk Factors Associated with Salmonella Prevalence in Southeastern United States’ Pasted Poultry Farms
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Developing Scientist Entrant

Purpose: In recent years, consumer demand has increased for pastured poultry products where the practice involves raising poultry in an open, floor-free area. It is necessary to identify the important meteorological and farm practices and processing factors associated with this type of farming practice since pathogens such as Salmonella can be present in the environment.

Methods: The objective of this study was to develop a model that can identify the specific meteorological and farm management factors contributing to the presence of Salmonella on pastured poultry farms.

Results: Longitudinal study was conducted from 11 pastured poultry farms from 2014 to 2017. Feeds, soil, whole carcasses rinses for processing and final product samples were collected for Salmonella presence. Random forest models were generated for each sample. The meteorological factors included variables such as temperature, humidity, wind speed and wind gust. The farm management factors included number of birds, years farming, type of farms, and storage temperature. Models were used to predict the presence of Salmonella. The relative important plots and partial dependency plots were generated to interpret the models. The model performances were evaluated using the area under the receiver operating characteristic (ROC) curve values.

Significance: The models were robust in predicting the presence of Salmonella in pastured poultry farms. The relative importance plots and partial dependency plots were generated to interpret the models. The model performances were evaluated using the area under the ROC curve. The models in this study will provide users a practical and effective tool to make informed decisions with scientific evidence to control the risk of Salmonella contamination in poultry farms.

P1-166 A Predictive Model for Cross-Contamination of Salmonella in the Poultry Chilling Process
Xingxin Xiao1, Wei Wang2, Jianmin Zhang1, Ming Liao1, Hua Yang3 and Yanbin Li4
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Developing Scientist Entrant

Purpose: Salmonella cross-contamination in the chilling process is a major concern in the poultry industry. Chilling water is an ideal medium for Salmonella to reach increasing levels because of high bacterial loads and long chilling time. A predictive model for describing the change of bacterial incidence along chilling process has not been studied well, which is needed in quantitative microbial risk assessment (QMRA) model for poultry supply chain.

Methods: This study was to investigate the cross-contamination of Salmonella by simulating chilling conditions in the laboratory, and to develop a model to predict the bacterial incidence along chilling process. The chilling process in Chinese poultry slaughterhouses was used to set up the parameters for Salmonella inoculation. The chilling water was inoculated with Salmonella serotypes with different contamination levels. Response surface methodology based on the central composite design with a total of 20 runs was used to predict the post-chill incidence with JMP software, and analysis of variance was used to test the significance and variance of the model. Eight random independent replicates were processed in the central point to test the model.

Results: Salmonella incidence along the chilling treatments were fitted to 30 to 91.7%. Contamination level and pre chill incidence showed a positive effect and chilling temperature, respectively, have the highest contribution in the model. The highest Salmonella contamination was greater on cantaloupes than honeydew melons with an average of 0.749±0.008 and 0.183±0.003 for soil, respectively. Salmonella inoculated dust (10 log) was sprayed using a duster on the melon rind in a specially designed chamber. Salmonella was recovered in PBS using vortexing for 5 minutes before being centrifuged for melon rinds. Pears inoculated in PBS and set out on table were plated on xylose lysine deoxycholate (XLD) agar. Five replicates were used for each exposure. Salmonella infection was caused by melon rinds to cantaloupes and honeydew melons.

Purpose: To investigate soil and dust as vehicles of cross-contamination of Salmonella enterica to cantaloupes and honeydew melons.

Methods: Six experimental cantaloupe and honeydew melon varieties grew in seven different locations in six states (Arizona, California, Georgia, Texas, New Mexico and South Carolina). Soil from melon fields in Yuma, Arizona was used. Dust was obtained by passing this soil through a No. 100 sieve. For soil transfer, 10 g rinds were placed on Salmonella inoculated soil (rind log) and allowed one h of contact time. Salmonelle inoculated dust (10 log) was sprayed using a duster on the melon rind in a specially designed chamber. Salmonella was recovered in PBS using vortexing for 5 minutes before being centrifuged for melon rinds. Pears inoculated in PBS and set out on table were plated on xylose lysine deoxycholate (XLD) agar. Five replicates were used for each exposure. Salmonella infection was caused by melon rinds to cantaloupes and honeydew melons.

Results: Salmonella inoculated dust (10 log) was sprayed using a duster on the melon rind in a specially designed chamber. Salmonella was recovered in PBS using vortexing for 5 minutes before being centrifuged for melon rinds. Pears inoculated in PBS and set out on table were plated on xylose lysine deoxycholate (XLD) agar. Five replicates were used for each exposure. Salmonella infection was caused by melon rinds to cantaloupes and honeydew melons.

Significance: These results help us understand the risk of Salmonella contamination from environmental soils such as dust to melon crops in field conditions. The data can be used for a science-based risk analysis. Appropriate control measures can be implemented. 

P1-169 Impact of a Kiln Intervention on Human Exposure to Polycyclic Aromatic Hydrocarbons (PAHs) in Smoked Fish in Ghana
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1University of Ghana, Ghost, Belgium, 2Food Research Institute, Accra, Ghana, 3Uf& Food and Agriculture Organisation, Rome, Italy, 4Research Group Food Chemistry and Human Nutrition (nuan/FODochim), Department of Food Technology, Safety and Health, Faculty of Bioscience Engineering, Ghent University, Ghent, Belgium

Developing Scientist Entrant

Purpose: The objective of this project was to develop an agent-based model to simulate the spread of norovirus on a berry farm and to predict the prevalence of norovirus by simulating chilling conditions in the laboratory, and to develop a model to predict the bacterial incidence along chilling process. The chilling process in Chinese poultry slaughterhouses was used to set up the parameters for Salmonella inoculation. The chilling water was inoculated with Salmonella serotypes with different contamination levels. Response surface methodology based on the central composite design with a total of 20 runs was used to predict the post-chill incidence with JMP software, and analysis of variance was used to test the significance and variance of the model. Eight random independent replicates were processed in the central point to test the model.

Methods: The developed model could provide input data for QMRA models of poultry supply chain.

Results: Introduction: Salmonella cross-contamination in the chilling process is a major concern in the poultry industry. Chilling water is an ideal medium for Salmonella to reach increasing levels because of high bacterial loads and long chilling time. A predictive model for describing the change of bacterial incidence along chilling process has not been studied well, which is needed in quantitative microbial risk assessment (QMRA) model for poultry supply chain.

Purpose: To identify food products on the European market contributing to the exposure of the population to nickel as preparation for potential risk management strategies.

Methods: In total 826 samples, including 27 food groups, were analyzed for nickel (microwave digestion followed by ICP-MS determination). Statistical analysis and calculation of consumption quantities and frequencies of the commodity in Belgium (120 respondents). The margin of exposure approach (MoE) was then used to determine the PMT public health concern associated with products related from different kilns.

Results: The PAH levels in the products were then determined by gas chromatography-mass spectrometry. PAHs of interest were benz[a]pyrene (BaP) and the sum of 16 polycyclic aromatic hydrocarbons (PAH16) in the products. A novel kiln called FAC-Thiaroye technique (FTT) has been introduced in the country as an intervention.

Significance: The findings suggest that the FTT has a strongly positive impact on reducing human exposure to PAHs in smoked fish in Ghana.

P1-170 Identification of Sources of Nickel Contamination in Foods and Its Exposure Assessment
Mehrooosh Babaahmadi-fooladi1, Gijs Du Laing3 and Liebesh Jacxsens2
1University of Gent, Ghent, Belgium, 2Gent University, Ghent, Belgium, 3Gent University, Ghent, Belgium

Developing Scientist Entrant

Purpose: To identify food products on the European market contributing to the exposure of the population to nickel as preparation for potential risk management strategies.

Methods: In total 826 samples, including 27 food groups, were analyzed for nickel (microwave digestion followed by ICP-MS determination). Statistical analysis and calculation of consumption quantities and frequencies of the commodity in Belgium (120 respondents). The margin of exposure approach (MoE) was then used to determine the PMT public health concern associated with products related from different kilns.

Results: The PAH levels in the products were then determined by gas chromatography-mass spectrometry. PAHs of interest were benz[a]pyrene (BaP) and the sum of 16 polycyclic aromatic hydrocarbons (PAH16) in the products. A novel kiln called FAC-Thiaroye technique (FTT) has been introduced in the country as an intervention.

Significance: The findings suggest that the FTT has a strongly positive impact on reducing human exposure to PAHs in smoked fish in Ghana.

P1-171 Understanding the Cross-contamination of Melons Via Environmental Matrices Simulating Field Conditions
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1University of Arizona, Tucson, AZ, 2ICEDAD, Tucson, AZ

Introduction: Foodborne outbreaks involving contaminated melons have occurred multiple times in the past decade. Understanding the sources of contamination will help develop policy measures and control preventives.

Purpose: To investigate soil and dust as vehicles of cross-contamination of Salmonella enterica to cantaloupes and honeydew melons.

Methods: Six experimental cantaloupe and honeydew melon varieties grew in seven different locations in six states (Arizona, California, Georgia, Texas, New Mexico and South Carolina). Soil from melon fields in Yuma, Arizona was used. Dust was obtained by passing this soil through a No. 100 sieve. For soil transfer, 10 g rinds were placed on Salmonella inoculated soil (rind log) and allowed one h of contact time. Salmonelle inoculated dust (10 log) was sprayed using a duster on the melon rind in a specially designed chamber. Salmonella was recovered in PBS using vortexing for 5 minutes before being centrifuged for melon rinds. Pears inoculated in PBS and set out on table were plated on xylose lysine deoxycholate (XLD) agar. Five replicates were used for each exposure. Salmonella infection was caused by melon rinds to cantaloupes and honeydew melons.

Results: Salmonella inoculated dust (10 log) was sprayed using a duster on the melon rind in a specially designed chamber. Salmonella was recovered in PBS using vortexing for 5 minutes before being centrifuged for melon rinds. Pears inoculated in PBS and set out on table were plated on xylose lysine deoxycholate (XLD) agar. Five replicates were used for each exposure. Salmonella infection was caused by melon rinds to cantaloupes and honeydew melons.

Significance: These results help us understand the risk of Salmonella contamination from environmental soils such as dust to melon crops in field conditions. The data can be used for a science-based risk analysis. Appropriate control measures can be implemented. 

Poster
P1-171  Estimated Daily Intake and Cumulative Risk Assessment of Perchlorate Via Diverse Foods for Tai- wanese Populations

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Introduction: Perchlorate (IC), recognized as a thyroid-disrupting chemical, is a strong oxidizer and has attracted significant attention due to its reactive nature and potential to contaminate water, soil, food, and even in human urine. Little is known about the dietary exposure of perchlorate foods to human exposure to perchlorate in Taiwan.

Aims: To conduct nationwide monitoring of perchlorate in 310 food samples to estimate the level of perchlorate to which Taiwanese are exposed.

Methods: Three-hundred ten food samples of twelve categories were purchased from the market or the household food preparation points and National Health, National Cheng Kung University, Tainan, Taiwan.

Results: The estimated daily doses (EDs) of perchlorate were calculated by multiplying the detected levels of target foods by food consumption of the corresponding age group. The highest perchlorate levels were detected in seafood (1.08 to 2.25 μg/kg), followed by fresh food (0.75 to 2.08 μg/kg) and vegetables (0.69 to 1.74 μg/kg). The highest 95th percentile of ED for perchlorate was found in ≥65-year olds (1.43 and 1.46 μg/kg body weight per day), followed by four to six-year olds (1.40 and 1.30 μg/kg BW/day). The hazard indexes ranged from 2.14 to 5.50 according to the RfD, 1.29 to 2.79 according to the TDl proposed by EFSA, and 0.04 to 0.08 established by ECFCA.

Significance: This results indicated that Taiwanese people's current exposure to perchlorate from domestic food consumption cannot be negligible.

P1-172 Risk Assessment for Non Dioxin-like Polychlorinated Biphenyls Exposure from Food Consumption in Taiwan

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Introduction: Non-dioxin-like polychlorinated biphenyls (NDL-PCBs) are persistent organic pollutants (POPs) that accumulate in the environment and may cause adverse health effects.

Aims: To establish risk assessment of NDL-PCBs in foods for all age groups in Taiwan.

Methods: We first selected the estimations of NDL-PCBs in food for the national health, and 12 age-determined models for NDL-PCBs in foods.

Results: The highest 95% NBPC levels were found in fish (2.37 ng/kg fresh weight), followed by oysters products (0.819 ng/kg fresh weight). The NDL-PCB levels in plant foods were much lower than animal foods. The NDL-PCB levels in seafoods was significantly higher than the terrestrial animal foods, especially in large predatory fishes.

Significance: This results indicated that exposure to NDL-PCBs from domestic food consumption cannot be negligible, and increased inspection of NDL-PCBs in foods should be going on.

P1-173 Occurrence and Profiles of Phthalates in Processed Food from Taiwan and Their Implications for Human Exposure

Cheng-Ching Lee1, Wei-Hisang Chang2 and Guan-Liang Wu
1Department of Environmental and Occupational Health, National Cheng Kung University, Tainan, Taiwan; 2Research Center for Environmental Trace Toxic Substances, National Cheng Kung University, Tainan, Taiwan

Introduction: Researches reported that intake of contaminated foods is the most important exposure pathway of phthalates for the general population.

Aims: To establish risk assessment of phthalates in foods for all age groups in Taiwan.

Methods: The present study aimed to investigate the levels of phthalate esters in domestic commercial prepared foods and to model consumers' exposure to phthalates through the selected foods.

Results: The estimated dietary exposure to phthalates in processed food was 1.5 times higher than the WHO tolerable daily intake. Phthalates in processed foods were found in 99.3% of the food samples. The highest 95th percentile of ED in phthalate esters was found in ≥65-year olds (1.06 to 7.04 μg/kg/day), followed by zero- to three-year olds (0.12 to 5.68 μg/kg/day) and six to 12 year olds (0.12 to 5.60 μg/kg/day). The WHO-IHAF recommended H for reproductive and developmental toxicity ranged from 0.50 to 0.84, and 0.32 to 0.86 for hematopoietic toxicity.

Significance: The current dietary exposure to phthalates for most Taiwanese was acceptable. To ensure food safety, we recommend continuous monitoring of phthalates in foods levels in packaging materials, especially fresh foods and imported foods.

P1-174 Predictive Microbial Modeling of Baking Inactivation Kinetics

Quincy Schmidt
U.S. Food and Drug Administration, Bedford Park, IL

Introduction: For more than 100 years thermal inactivation kinetics parameters, D-values and z-values, have been used to predict lethality of pathogens during in-container sterilization. However, these parameters alone are inadequate for predicting lethality during baking due to non-isothermal heating and dynamic heating environments. Predictive microorganisms models provide more accurate estimates of lethality during in-container sterilization. However, these parameters alone are inadequate for predicting lethality during baking due to non-isothermal heating and dynamic heating environments.

Aims: To assess the performance of multiple dynamic inactivation models for predicting microbial inactivation during baking of cookies.

Methods: We have developed a method for predicting microbial inactivation during baking of cookies by comparing the performance of four models with parameters estimated for isothermal inactivation and non-isothermal baking. These models were estimated using cookie internal or external temperatures, and using oven dew point temperatures were assessed by root mean square error (RMSE), the corrected Akaike-information criterion (AIC), and a normal distribution of the residuals. Performance was determined by a generalized linear model.

Results: Baking time, product internal and surface temperatures and oven wet-bulb temperature were found to be the most significant parameters contributing to microbial inactivation (P<0.05). The isotermal model had the poorest predictive performance (RMSE of 8977 log CFU). Model performance was improved by including both internal product temperature (RMSE of 0.80 log CFU and AIC = 919.04) and external product temperature (RMSE of 0.783 log CFU and AIC = 282.06) into the model. The dew point temperature model, which accounted for time, surface temperature, and wet-bulb tem- perature was the best predictor (lowest RMSE and AIC) for microbial inactivation (RMSE of 0.555 log CFU and AIC = 197.08) and could be applied across the full data set, not otherwise possible without environmental measurements.

Significance: Dynamic inactivation models that include product internal and surface temperatures and oven wet-bulb temperature greatly increase the predictive performance and are important process monitors to monitor in baking processes.

P1-175 The Impact of Free Chlorine Concentration in Fresh-cut Romaine Lettuce Wash Water on E. coli O157:H7 Cross-Contamination and Risk of Foodborne Illness in the United States

Sooa Santilla Farokas1, Amir Mokhtari2, Gordon Davidson3, Elizabeth Noelia Williams4 and Jane Van Doren5
1U.S. Food and Drug Administration – Center for Food Safety and Applied Nutrition, College Park, MD; 2U.S. Food and Drug Administration, College Park, MD; 3University of Maryland, College Park, MD; 4U.S. Food and Drug Administration - FDAHAN, College Park, MD

Introduction: Maintaining free chlorine concentrations in fresh-cut leafy green wash water is done to minimize microbial cross-contamination during processing. There is conflicting information on the impact of free chlorine levels during washing of E. coli O157:H7 cross-contamination and risk of illness.

Aims: To evaluate the change in E. coli O157:H7 prevalence on romaine lettuce and risk of illness in the U.S. as a function of the free chlorine concentra- tion in wash water used during processing.

Methods: A previously published FDA model on cross-contamination during leafy green processing was used combined with E. coli O157:H7 dose-response conversion factors. A model was developed for chlorine concentrations in wash water on E. coli O157:H7 cross-contamination and risk of illness. Key model inputs were initially randomly varied to investigate a wide range of what-if scenarios, including, within batch prevalence, 0.5-10%, initial level of contamination on lettuce heads (0.01 to 10,000 CFU), cost of chlorine solution (0.20 to 1.00 kg), and a chlorine-to-water ratio (1.0/1.5-1.15). The model was developed in R and simulated 20,000 independent wash tanks with 120 batches of romaine lettuce.

Results: Simulated results showed 95% probability of no increase in the average risk of illness per serving when a minimum level of free chlorine in wash water was maintained at 10 ppm for scenarios in which initial contamination levels on lettuce heads, within-batch prevalence, and produce-to-water ratios were limited to 2 kg, 10%, and 1200 kg/L, respectively. For the same model input levels, results showed an increase in prevalence of contamination during the washing process.

Significance: The chloride in wash water can reduce the risk of illness from exposure to E. coli O157:H7 in leafy greens, but high prevalence of initial contamination and high prevalence of risk contamination is significant and the extent of cross-contamination occurs which during the washing process.

P1-176 A Comparative Study of Heavy Metal Exposure Risk from the Consumption of Some Common Varieties of Cultured and Captured Fishes in Bangladesh

Mohammed Ruzlan Habib1, Muhammad Moumeen Hoque2 and Yasmin Nahar Jolly3
1Shahjalal University of Science and Technology, Khulna, Bangladesh; 2Shahjalal University of Science and Technology, Sylhet, Bangladesh

Introduction: Food toxicity and health risks due to exposure to heavy metals through fish consumption have become major concerns in the present era. It has become necessary to assess the content of heavy metals such as iron (Fe), copper (Cu), zinc (Zn), arsenic (As), mercury (Hg), and lead (Pb) in commonly consumed cultured and captured fish species in Bangladesh.

Purpose: The purpose of this study was to compare the possible carcinogenic, non-carcinogenic, and other health risks in cultured and captured fish in Bangladesh.

Methods: Fish samples (n=14) from seven fish species in both captured and cultured categories were collected, washed, separated into flesh and bones, minced, and oven-dried for 12 to 24 hours at 80°C, then ground into a fine powder and made into circular pellet weighing 0.1 g each and 7 mm in diameter. Each pellet was compiled in a x-ray fluorescence spectrophotometer for 1,000 seconds to determine heavy metal content. Risk assessment was performed using formulas in Microsoft Excel and SPSS software.

Results: The assessment revealed among all other identified metals, Zn was most common in the fish samples, followed by Fe, Cu and the others. The highest level of non-carcinogenic risk was determined for Pb in adults and children and Hg in adults and children. The highest level of non-carcinogenic risk was determined for Pb in adults and children. The highest level of non-carcinogenic risk was determined for Pb in adults and children.

Significance: This study is significant due to chronic exposure to heavy metals through fish consumption over the years.
P1-177 Application of Machine Learning for Food Safety Data Analysis

Wen Zou, WeiZhong Zhao, Jinsiu Zhou and Kavina Munshi

Methods: We applied machine learning algorithms to re-interpret PFGE data from a large outbreak investigation. Active learning was used to improve classification performance.

Results: Classification accuracy can be improved with limited amounts of training data using active learning.

Significance: This approach can be used to guide outbreak investigations by providing insights into outbreak characteristics and potential sources of contamination.

P1-177 Introduction to the Calculation and Interpretation of Level of Detection

That a homogenized one kg sample has a 50 CFU contamination level?

Methods: Calculation formulas were derived from the ISO 11466 series on the basis of a standard log model. Simulations were used to calculate the measurement uncertainty of the LOD. The discussion of risk analysis is based on microbiological and biostatistical considerations.

Results: Implications of the Poisson assumption for LOD are clarified with respect to the best possible theoretical LOD value and the relationship between LOD and false positives. The impact of practical data on LOD are shown in examples. Simulations based on calculated data showed considerable variation in LOD of values beyond simplistic systems such as infection incidences and viability of test samples. It is also shown LOD values can be used within risk analysis, and general principles for establishing performance criteria for the LOD in terms of efficiency for purpose are discussed.

Significance: This presentation highlights how LOD values can be used to gain further understanding of measurement uncertainty associated with qualitative microbiological testing and how they can be used in connection with risk analysis.

P1-178 Long Read Sequencing for Food Safety Applications

The GenomeTrakr database has demonstrated how network of desktop WGS sequencers can be used in concert with traditional epidemiology and investigation for source tracking of foodborne pathogens. This "open data" model allows greater transparency between federal agencies and states, and increases communication of public health risk.

Methods: This database has grown in size and continues to grow daily, driven by a process of data collection and analysis. It now contains more than 300,000 draft genomes, with ongoing efforts to expand its scope and capabilities.

Results: The GenomeTrakr database is part of the NCBI Pathogen Detection web site. All four pathogens reported eating Asian-style foods and foods reported shopping at stores where pork products from Establishment A were sold. Establishment A voluntarily recalled RTE pork products produced from May through November 2018.

Significance: The high-resolution WGS signal in concert with epidemiological and inspection evidence has drastically enhanced our ability to identify the food sources of current outbreaks for foodborne pathogen control and prevention.

P1-180 The GenomeTrakr Database Global WGS Network for Foodborne Pathogen Tracking

Marc Allard, Ruth Timme, Maria Sanchez, Eric Stevens, Maria Hoffmann, Kuan Yat, George Kastanis, Daniela Miller, Tim Munvala, Vivienne Heines, Andrew Plopper, Payam Taheri Zadeh, Bree Danielson, Hugh Randel, James Pettigrew, Yan Luo, Narjol Gonzales-Escalon, David McElha, Phillip Curry, Sabina Lindelien, Yi Chen, Sandra Tallent and Eric Biren

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P1-181 Long Read Sequencing for Food Safety Applications

Outbreak to Ready-to-Eat Pork Products

Xuwen Wieneke

Facultative controls to make food safer globally.

Introduction: The GenomeTrakr database is part of the NCBI Pathogen Detection web site. All four pathogens reported eating Asian-style foods and foods reported shopping at stores where pork products from Establishment A were sold. Establishment A voluntarily recalled RTE pork products produced from May through November 2018.

Significance: The high-resolution WGS signal in concert with epidemiological and inspection evidence has drastically enhanced our ability to identify the food sources of current outbreaks for foodborne pathogen control and prevention.

P1-181 Use of Whole Genome Sequencing, Epidemiologic, and Traceback Data to Link a Multistate Listeria monocytogenes Outbreak to Eat-Pork Products

Edit Minocha, Jennifer Freeman, Jovita Haro, Glenn Tillman, Mustafa Simmons, Merly Silverman, Maria Scotti, Brad Webb, Amanda Conroy, Danielle Donovan, Vivienne Heines, Brenda Run, Natalie Christoph, and Sakina Hamdani

Results: This database has continued to grow and diversify the foodborne pathogen database doubling in the last year to ~300,000 draft genomes with a projected growth of over 1,000 genomes per year. Two new international surveillance efforts were added to collect food, animal and environmental isolates and Listeria spp. and L. monocytogenes from consumer and industry acquisition. NCBI currently is producing daily clustering results for 27 bacterial pathogens including Salmonella, Listeria, E. coli, Shigella and Campylobacter.

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Significance: The high-resolution WGS signal in concert with epidemiological and inspection evidence has drastically enhanced our ability to identify the food sources of current outbreaks for foodborne pathogen control and prevention.
**P1-183 Molecular Characterization of Native Lactobacillus Strains Isolated from Vaccinium floribundum Kunth by Partial Sequencing of 16S rDNA Genes**

Celia Vargas 1, Carmen López 1, Teresa Gallardo 2, Félix Ramos 3 and Daniela Landá 1
1Centro Universitario de Ciencias de la Salud, Instituto Tecnológico de Chihuahua (UTC), 2Facultad de Ciencias de la Salud, Universidad Nacional Autónoma de México (UNAM), 3Facultad de Ciencias, Universidad Nacional Autónoma de México (UNAM), Mexico City, Mexico

**Significance:** This study sought to establish the rapid and remote sequencing capabilities and accurate characterization featured in LRS are promising. With further optimization, there is potential to use LRS for food safety applications in the near future, with enhanced performance advantages as compared to current technologies.

**Introduction:** The persistence of isolates, the whole genome sequencing (WGS) of a subset of isolates (≥ one antimicrobial. Nine major clusters were identified by PFGE. Isolates with identical or very similar sequences were detected which were involved in fundamental functional and/or energy transcription, however, there was variable occurrence of genes involved in transport, acquisition, motility, virulence, and AR. This study will provide data on Salmonella Javiana that may be valuable to understanding potential future emergence of other serotypes as prominent human foodborne pathogens.

**Materials and Methods:** The study took a novel approach combining metagenomic, statistical and machine learning approaches to provide baseline data describing the surface water microbiome composition and the relationships between microbiome composition and cultured pathogenic data. This study took a novel approach combining metagenomic, statistical and machine learning approaches to provide baseline data describing the surface microbiome composition and the relationships between microbiome composition and cultured pathogenic data. This study took a novel approach combining metagenomic, statistical and machine learning approaches to provide baseline data describing the surface microbiome composition and the relationships between microbiome composition and cultured pathogenic data.

**Results:** The rapid and remote sequencing capabilities and accurate characterization featured in LRS are promising. With further optimization, there is potential to use LRS for food safety applications in the near future, with enhanced performance advantages as compared to current technologies.
91 The objective of this project was to quantify the influence of major food components on aw change at elevated temperatures. Using a library of accessions including LEE island and hemolysin-encoding plasmid.

Methods: The four genotypes of bacteria were used to inoculate the four products and the thermal resistance of bacteria was measured using an inactivation curve. Two extreme temperatures were used: 75°C and 15°C.

Results: The thermal inactivation rates of the four genotypes were significantly different at both temperatures. However, the thermal resistance of bacteria was significantly higher at 75°C than at 15°C.

Conclusion: The results of this study suggest that the thermal resistance of bacteria in low-moisture foods can be predicted using the inactivation curve obtained for each genotype.

**Poster**

**Title:** Understanding the correlation between food components and aw change at elevated temperatures could be a new approach to predict the thermal resistance of bacteria in low-moisture foods.

**Authors:** Nineteen isolates collected from foods and 36 clinical isolates were included in this study. Whole genome sequencing (WGS) was performed on all isolates. The sequence data was analyzed using the National Center for Biotechnology Information (NCBI) database.

**Methods:** Whole genome sequencing was performed on all isolates. The sequence data was analyzed using the National Center for Biotechnology Information (NCBI) database.

**Results:** All isolates contained the gene cytK, which encodes the cytotoxic protein that causes sickle cell disease. The number of isolates with this gene was highest in whole genome sequencing (WGS) data, followed by PCR assay and phenotyping.

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**Conclusion:** The results of this study suggest that the thermal resistance of bacteria in low-moisture foods can be predicted using the inactivation curve obtained for each genotype.
Methods: Isolates were identified on Acinetobacter ChromIDagar from frozen stocks of fresh produce (12 isolates) and soil (2 isolates) that had been examined previously by RFLP analysis. DNA for whole genome sequencing was performed by PCR using universal primers and sequencing was performed on an Illumina platform. The whole genome sequence data from the 34 isolates was analyzed for species identification, plasmid presence, and antibiotic resistance genes. For comparison to clinical isolate genomes obtained from GenBank, BLAST score ratio analysis was used for virulence gene core SNP analysis was used for whole genome phylogenetic analysis.

Results: The most abundant Acinetobacter species isolated were A. pittii (32.4%), A. baumannii (26.5%), and A. oxyphilus (20.6%), and phylogenetic analysis demonstrated that produce and clinical isolates clustered together. Unlike what is seen in clinical isolates, no plasmids were identified in the fresh produce isolates. However, the presence of 16 of the 34 of the isolates carry chromosomally encoded (Pseudoalteromonas resistance genes phiPSAL). The virulence profiles of the A. baumannii clinical and clinical strains were similar, including the presence of siderophores and quorum sensing genes in conjunction with biofilm formation. Some A. pittii produce strains also contained virulence genes in conjunction with biofilm formation.

Discussion: The results of this study reveal the close genomic relationship between Acinetobacter found on fresh produce and clinical strains, including presence of virulence genes, and demonstrate that the major difference is absence of plasmids in the fresh produce isolates.

P1-196 Development of Next-Generation Sequencing and Metagenomics for Detection of Foodborne Viruses Within Oysters

Xiulang Yang1, Gloria Meade2, Carla Mammel3, Mark Mammel4

Purpose: This study was designed to investigate if metagenomics can be used to detect viruses of interest in a setting where viral contamination of shellfish is a threat to public health. Metagenomics offers new opportunities for detection of viruses, and investigating of human and virus interactions in shellfish is in an unanswered question. However, the protocols for the sample preparation, next-generation sequencing (NGS) and data analysis required are complicated, need to be developed and optimized.

Methods: Shellfish are known to concentrate various viruses present in surrounding water within their tissues. Thus, viral contamination of shellfish poses a risk for foodborne illnesses. Metagenomics offers new opportunities for detection, identification of viruses, and investigating of human enteric virus species in shellfish in an unanswered question. However, the protocols for the sample preparation, next-generation sequencing (NGS) and data analysis required are complicated, need to be developed and optimized.

Results: The purpose of this study was to develop NGS and metagenomic approaches for investigating foodborne virus profiles present in oyster samples.

Discussion: Detection of viruses in foods is a major public health concern. This project FDA/USA study will help to provide a scientific basis for regulations ensuring the safety and security of our nation’s food supply.

P1-197 Frequency of Multi-Locus Sequence Types in FSIS-regulated Ready-to-Eat Products

Carrie Clark1, Mary Katherine Crews2, Glenn Tillman2, Mustafa Simmons2, Jamie Wasilenko2, Udit Minocha2

Purpose: To characterize the transcriptome of Campylobacter jejuni in the presence of food and clinical isolates.

Methods: Whole genome sequencing was performed on 80 C. jejuni strains isolated from poultry meat and assigned to 24 different SNP clusters according to the NCBI Pathogen Database. The enrichment, as judged based on the increased relative abundance of C. jejuni in the metatranscriptomic profile while extremely low in the metagenomic profile, suggests this species may be relatively low in abundance but highly active in the SSWS. Across all sampling points and inoculation levels, two KEGG metabolic pathways were identified with genes significantly upregulated in the presence of chicken DNA compared with biofilm isolated clinical isolates. Pathway analysis also revealed the 10 most abundant gene families with function in stress resistance and adaption, cell division, RNA turnover, and vitamins. Functional analysis compared metabolic and metatranscriptomic profiles of inoculated Salmonella enterica serovar Typhimurium and Campylobacter jejuni.

Significance: This study suggests a dynamic, functional interaction between Salmonella and the microbiome community in SSWS.

P1-199 Dynamics of Microbiome Composition during Enrichment of Campylobacter in Poultry Samples

Ruanan Yan1, Andrea Oetteman2, Padmni Ramachandran2, Eric Board3, Jie Zheng3

Purpose: To characterize the transcriptome of Campylobacter jejuni in the presence of food and clinical isolates.

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Significance: This study suggests a dynamic, functional interaction between Salmonella and the microbiome community in SSWS.

Poster
Radio-frequency (RF) pasteurization has been identified as a potential technology to pasteurize low-moisture foods. It is reported that RF heating to 80°C combined with a 20 min natural cooling could achieve an average E. faecium population reduction of 1.94 to 3.48 log CFU/g in wheat flour. The lowest lethality zone was located in the bottom layer in all cases based on measured temperature profile and survival data. Fast heating rate was observed in non-uniformity in terms of temperature and inactivation. The fitting results using the Bigelow model were in good agreement with that from the experiment.

Significance: This study provides a comprehensive study on the lowest lethality zone identification of wheat flour during RF pasteurization.

P1-203 Simulated Commercial Baking Validation of Peanut Butter Bars to Control Salmonella

Daniel Vega,1 Nicholas Sevart,1 Lakshmikantha Channaiah2
1Kansas State University, Manhattan, KS, 2Department of Biosystems and Agricultural Engineering, Michigan State University, East Lansing, MI

Developmental Scientist Entreat

Introduction: Pathogen contamination of flour poses a significant food safety concern. Most baked goods go through a thorough cooking step; however, pathogen survivability during thermal processing evaluations must consider product-specific characteristics such as fat level and water activity which are impacted by baking. Purpose: Validate a peanut butter bar baking process for its ability to reduce Salmonella populations originating from contaminated flour.

Methods: Flour was inoculated with a six-strain cocktail of Salmonella (0.4 log CFU/g), re-dried, and used to create a peanut butter bar dough (substituted for the shortening). Three bars were baked at 390°F for 12 min, and 15 min of ambient air cooling. Salmonella populations were enumerated pre-baking, during baking (one, three, five, seven, nine, and 13, and 17 min), and post-cooling. Internal temperature was continuously recorded and water activity, pH, moisture content, and Salmonella counts (composite of internal and external bar components) were determined at the nine sampling times using three replicates.

Results: Internal bar temperatures increased from 25°C to 91.88°C during baking. Water activity decreased (0.81 to 0.71), moisture content decreased (15% to 18%), and pH increased (from 7.03 to 8.65) during the baking process. Final baked product fat content was 21%. Salmonella populations were reduced (P<0.05) by the oven-cooked compared to pre-baked dough samples by 2.4 log CFU/g.

Significance: Thermal processes for most bakery products have been shown to achieve over a five-log reduction of Salmonella. In peanut butter bars, Salmonella populations were reduced only by a 2.4 log CFU/g reduction after 13 min at 177°C oven temperature. This decreased survival could be considered by food companies producing similar products. Research is being conducted to generate Salmonella D- and V-2 cultures in baked peanut butter bars to assist the food industry in safe thermal process development.

P1-204 Quantifying the Inactivation of Enterococcus faecium during Spray Drying

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Developing Scientist Entreat

Introduction: Spray-dried foods such as infant formula and protein powders pose a bacterial risk of contamination and some long-term survival, as seen from sporadic recalls and outbreaks. However, with other compared unit processes, there is a significant lack of understanding of the microbial inactivation during the spray-drying process, due to its short and complex nature. Therefore, the inactivation of Enterococcus faecium, a potential surrogate organism for Salmonella, during spray drying will elucidate the risk of contamination of powered foods.

Purpose: To quantify the inactivation of E. faecium during spray drying. Methods: A 500 ml suspension of 10% w/v soy protein isolate was inoculated with six ml of tryptic soy broth containing E. faecium NRRL-3254 (104 CFU/ml). The entire 500 ml was pumped through a pilot-scale spray dryer (FT60 Tall Form Spray Dryer, Amrefill Ltd, Carlisborg, NJ) at 180°C at a rate of seven medium pum. Post-spray, the samples were collected and secondary cultures were performed. Results: Salmonella populations were reduced by 5.9 log CFU/g during the drying process. The inactivation was quantified by plating appropriately modified samples on modified tryptic soy agar containing emulsifier, followed by incubation (48 h, 37°C). Results: Salmonella populations on all tested samples after spray drying were approx. nine log CFU/g/mg after inoculation with no significant reduction for drying time for three h. The population of Salmonella on corn flakles and pistachios gradually decreased to approx. seven log CFU/g/mg during 12 weeks. Salmonella was undetectable in dried apples after 62 out of 82 drying runs. Conferential imaging revealed that a large proportion (ca. 60%) of Salmonella remained viable on dried apples not despite growing under lower standard conditions. Several differentially expressed genes were identified in all tested LMFs at all times, while some were upregulated only in specific LMF-time combinations. Experiments of genes upregulated in all LMFs after 42 even with extreme conditions. Significant: Findings revealed novel Salmonella genes that are upregulated in LMFs and may be mediating the survival of Salmonella on LMFs.

P1-201 Identification of the Lowest Lethality Zone in Wheat Flour Treated with Radio-Frequency Heating and Natural Cooling

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Developing Scientist Entreat

Introduction: Radio-Frequency (RF) pasteurization has been identified as a potential technology to pasteurize low-moisture foods. It is reported that RF heating to 80 to 85°C followed by a 10 to 25 min natural cooling introduces 2.5 to 3.7-log reduction of Salmonella populations (composite of internal and external bar components) were determined at the nine sampling times.

Purpose: To monitor the post-processing microbial populations of E. faecium at multiple locations in wheat flour, and thus identify the right location to reflect the worst scenario of RF pasteurisation.

Methods: Post-processing microbial reduction of E. faecium was tested at 15 locations (evenly distributed in top, middle and bottom layers) in a 1.8 kg wheat flour batch during the RF heating at 36.0°C, 11.3°C, and 5.5°C heating rates for 10, 30 and 60 min, respectively. Cooling. Fiber optic sensors monitored the temperature change in the three layers throughout the process. Bigelow model was applied to predict the temperature dependent log reduction of E. faecium in each layer. The experiment was conducted in duplicate for each heating rate.

Results: RF heating to 80°C combined with a 20 min natural cooling could achieve an average E. faecium population reduction of 1.94 to 3.48 log CFU/g in wheat flour. The lowest lethality zone was located in the bottom layer in all cases based on measured temperature profile and survival data. Fast heating rate was observed in non-uniformity in terms of temperature and inactivation. The fitting results using the Bigelow model were in good agreement with that from the experiment.

Significance: This study provides a comprehensive study on the lowest lethality zone identification of wheat flour during RF pasteurization.

P1-205 Butylparaben Improves the Thermal Inactivation Rate of Escherichia coli O157:H7 in Low-Moisture Foods

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Developing Scientist Entreat

Introduction: Heat resistant foodborne pathogens have long been a concern in low-moisture foods and ingredients (LMF) such as meat and bone meal (MBM) which are used as feed ingredients. The purpose of this study was to investigate the thermal inactivation rate of E. faecium during spray drying of MBM, heat treatment by itself or in combination with BP was observed with the recipe with the lowest level of salt and mold populations increased to approximately seven log CFU/g by seven weeks. Salmonella Enteritidis PT 30 and Salmonella Typhimurium PT 42 on play dough samples showed ~four log CFU reduction over seven weeks, with non-significant differences between strains and formulations. Enterococcus aerogenes showed similar survival kinetics to Salmonella strains in play dough samples during storage, while Pseudoentero dispansio showed a ~four log reduction by week five.

Methods: Homemade dough made with contaminated flour does pose a risk but this risk decreases over time, even in differing formulations.

P1-205 Salmonella and Surrogate Microorganism Behavior in Homemade Play Dough Based on Online Recipes

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Introduction: As social media has become a popular platform to communicate and share information, online users have shared their favorite play dough recipes with over 30 different recipes online. Homemade play dough made may contain flour contaminated with pathogenic bacteria during harvesting, milling, packing, shipping, or storage and could make dough with high-levels of Salmonella.

Purpose: Little is known about microbial behavior on play dough made with flour, so this study assessed the survival of two strains of Salmonella and two Enterococcus species on play dough based on three different online recipes.

Methods: Online recipes for homemade play dough were collected and analyzed to calculate the ratio of ingredients (flower, water, and salt) in standard units. Three recipes were selected based on differing levels of salt (eight, 21 and 39%). Samples of play dough (2 g) were inoculated with ~7.5 log CFU/g Salmonella Enteritidis PT 30, Salmonella Typhimurium PT 42, Enterobacter aerogenes or Pseudomonas aeruginosa and enumerated following storage for up to seven weeks. All experiments were conducted in triplicate.

Results: Native microflora (~three log CFU/g) on play dough samples prepared with different recipes survived at least seven weeks. The growth of mold was observed when play dough was prepared with the recipe with the lowest level of salt and mold populations increased to approximately seven log CFU/g by seven weeks. Salmonella Enteritidis PT 30 and Salmonella Typhimurium PT 42 on play dough samples showed ~four log CFU reduction over seven weeks, with non-significant differences between strains and formulations. Enterobacter aerogenes showed similar survival kinetics to Salmonella strains in play dough samples during storage, while Pseutonentero dispansio showed a ~four log reduction by week five.

Significance: Homemade dough made with contaminated flour does pose a risk but this risk decreases over time, even in differing formulations.
Purpose: To assess the survival of Salmonella on nuts and seeds during storage at three different RH levels for six months.

Methods: Nuts and seeds were inoculated with 1.0 log CFU/g of Salmonella Newport, Salmonella Senftenberg, or Salmonella Typhimurium (100 CFU/g) and left to stabilize for two weeks. Sample populations were enumerated by mixing one g of sample with nine ml BLEB. Serial dilutions of the homogenate were plated onto BHI agar and stored at 37°C for 48 h before counting.

Results of this study can aid in understanding how pathogens like Salmonella survive on nuts and seeds during extended storage at high RH levels.

Significance: Results of this study can aid in understanding how pathogens like Salmonella survive on nuts and seeds during extended storage at high RH levels.

Poster
P1-214 Evaluation of a Fluorescence Resonance Energy Transfer-Based Real-time PCR Assay for the Detection of Pathogens in 25 g and 375 g Walnut Samples

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Introduction: Pathogen detection systems such as PCR make up a critical part of the Food Safety Modernization Act (FSMA) implementation strategy and any factors that can influence the performance of these test systems should be evaluated. True nuts, including walnuts, present a unique challenge as it is a globally traded low-moisture commodity that is often consumed raw and can produce leachable PCR inhibitors.

The performance of a real-time PCR assay (GENE-LAMP) was evaluated for the detection of Salmonella, listeria monocytogenes, and E. coli O157:H7 in walnuts at 25 and 375-g sample sizes.

Methods: Thirty unspiked samples of walnuts (25 g) were evaluated (per ADAC guidelines) where n=5 and were inoculated with Salmonella Typhimurium at a high and a low inoculation level, respectively. Five uninculcated samples were also tested. After sample enrichment, unspared test portions were evaluated for PCR inhibitors using a nalidixic acid resistant strain of E. coli as the target organism.

Results: The assay performed exceptionally well with all the spiked samples producing positive results.

Significance: These data provide evidence for highly sensitive detection of Salmonella, listeria monocytogenes, and E. coli O157:H7 in G1 at unspatched sample sizes which is consistent with the traditional culture methods.

P1-215 Evaluating Steam Treatment as a Potential Intervention for Microbial Risk Reduction of In-Shell Peanuts

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Developing Scientist Entrant

Purpose: Steam treatment of in-shell pecans could be an effective and efficient alternative conditioning method useful in maintaining its safety.

Methods: In-shell pecans obtained from Louisiana orchards were inoculated with a nalidixic acid resistant strain of E. coli O157:H7. The inoculated pecans were added in a custom designed steam apparatus that was inoculated with steam and maintained at 70, 80 and 90 °C for 30, 60, 90, 120, 150, 180, 210, 240, 270 and 300 s. Samples obtained after each treatment were processed for both qualitative and quantitative tests. Each experiment was done in triplicate.

Results: At 70 °C, E. coli O157:H7 was reduced by 1.21±0.23 log CFU/g within 30 s, and the reduction further increased to 4.28±1.01 log CFU/g after treatment for 30 s. As the temperature increased to 80°C, a 2.49±0.52 log CFU/g reduction was achieved within the 30 s of treatment. On further exposure, the reduction rates increased (P<0.05) to greater than 100 s. Levels were below the detectable limit after 120 s but were detected during the qualitative test. At 90°C, samples were below the detectable limit of the test within the 30 s indicating greater than five-log reduction of the organism. However, qualitative tests showed the presence of the organism up to 180 s.

Significance: Steam treatment of in-shell pecans can be an effective and efficient conditioning method useful in maintaining its safety.

P1-216 U.S. Food and Drug Administration’s Total Diet Study (TDS): Process and Challenges Faced in Modernizing the Food List

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Introduction: The United States Food and Drug Administration’s Total Diet Survey (TDS), a surveillance program ongoing since 1965, is undergoing modernization. This poster presents a description of the process followed and challenges faced in updating the list of foods sampled and analyzed in the TDS program.

Methods: Extensive analyses were conducted, and critical decisions were made to shape the new food list, which was last updated in 2003. The food list was updated based on information from various datasets: TDS findings of analytic trends in foods, What We Eat in America (WWEIA)/ National Health and Nutrition Examination Survey (NHANES), EPA’s Food Commodity Intake Database (FCID), and Nielsen and IRI Liquid Data market-sales data. Additionally, the food list was cleaned to remove redundant and non-relevant food items. Nationally, consideration was given to ensure representation of foods and beverages currently consumed in the U.S. Foods on the previous food list with little consumption and/or historically low levels of analytes of concern were removed. In addition, the food list was updated to include more single-ingredient items rather than mixtures to allow calculation of analytic concentrations based on recipes.

Results: With the improved food list, TDS-based estimates of dietary exposures are likely to be more accurate, because concentrations of nutrients and contaminants in foods from前のTDS samples are less likely to be underestimated. The new food list has been collected since the new sampling plan was implemented in September 2017.

Significance: TDS data are widely used by various public and private entities. The updated food list improves the quality of the TDS data and its relevant contributions to food safety and public health.

P1-217 Rapid Bioluminescence Detection of Bacteria in Cannabis-infused Foods Using Microsnap

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Purpose: Introduction: cannabis is an illegal drug in the United States. However, cannabis is being legalized in various states in the United States and microsnap, a new technology, can be used to detect the presence of bacteria in cannabis-infused foods.

Methods: Cannabis-infused foods were collected from cannabis shops in California. These foods were defrosted, and the cannabis edible was cut into small pieces. The cannabis edible was fermented in 10 mL of sterile 0.85% sodium chloride solution. The bacterial concentrations in the edible were quantified using microsnap. The RLU thresholds were determined to be greater than 100 RLU and edible greater than 10 RLU.

Results: The Enterobacteriaceae bacterial panel at 10 CFU/mL in all tested cannabis foods were detected within the eight hour incubation period and as early as five h. The lowest bacterial inoculum level of 102 CFU/mL in the cannabis edible was detected by six h. RLU thresholds were determined to be greater than eight RLU and greater than two RLU.

Significance: This study demonstrates a rapid bioluminescence microbiology method for the detection of total vegetative counts, Enterobacteriaceae and coliforms.

P1-218 Is it Safe to Use Drinking Water Treatment Residues from Harmful Algal Bloom-affected Areas for Land Application?

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Introduction: Land applications generated from drinking water treatment plants (WTPs) have been used as soil amendments and fertilizers worldwide. However, in harmful algal bloom (HAB)-affected areas, concerns exist that cyanotoxins and toxic-producing cyanobacteria can be present at high levels in water treatment residues.

Methods: Water treatment residues were obtained from an Ohio WTP whose water source is impacted by HABs. Cyanotoxins and microbiome of water treatment residue samples were determined with ELISA and metagenomics approaches, respectively. Water treatment residue land application was simulated in a greenhouse by applying at zero, 20 and 40% of soil weight. Carrots were cultivated with six replicates at each level.

Results: Microcystin, saxitoxin and β-methylamino-L-alanine were detected at 258.7, 0.2 and 575.1 μg/kg in water treatment residues, respectively. Microcystis was the predominant genus of cyanobacteria within the bacterial phylogenetic tree. Phycocyanin accounted for more than 98% of total viruses. Carrots from the water treatment residue-added soil developed thicker roots with higher yield (P<0.05). About 80% of microcystin was retained in soil (83.0 to 95.5 μg/kg) while approximately five percent of microcystin accumulated in carrots. The majority of microcystin accumulated in the non-edible part (56.4%) of carrot.

Significance: For beneficial use of water treatment residues in agriculture, it is critical to know the impact of land application to protect crop quality and public health. For future studies, in-depth investigations about the accumulation of other cyanotoxins, such as saxitoxin, are recommended.

P1-219 Quantification of Aflatoxin B1 in Aspergillus parasiticus and A. flavus in Peanuts with Plant-based Antimicrobial Compounds

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Introduction: Intake of peanut crops by Aspergillus flavus and A. parasiticus is a serious problem in Georgia, and aflatoxin-contaminated peanuts continue to be a serious problem in Georgia. Aspergillus (A. Flavus) (AFB1) is the most toxic and dangerous carcinogen to humans and animals. There has been considerable interest in the development of novel agents with antifungal properties.

Purpose: Our previous study has established that clove and cinnamon have shown antimicrobial properties against Aspergillus spp. in peanuts. The minimum inhibitory concentration (MIC) and minimal fungicidal concentration (MFC) of these oils were determined by the standard plate diffusion method.

Methods: Cultures were prepared by placing a mycelial plug (one cm) in the centre of Rose Bengal plates. A 0.02-ml volume of each of the oils at various concentrations (50, 100, 150, 200, 250 and 2,500 ppm) was dropped onto two cm-diameter filter paper discs, respectively. Oil impregnated discs were placed on seven-day old old cultures and incubated at 28°C. The plates were incubated for three weeks. Zones of inhibition were measured with calipers.

Results: AFB1 was successfully identified by retention times and UV spectra. We expected the AFB1 concentration to decrease as the concentration of oil increased. As the oil concentration increased, we did not observe this pattern. Fungicidal effect of the oil on growth and development of the mycelium and spores could have been delayed.

Significance: Clove oil showed significant growth inhibiting properties in a maximum MIC and MFC over cinnamon at 2,500 ppm. Following incubation, AFB1 in oil was reduced by 1 log. Following incubation, AFB1 concentration was reduced by 1 log.

References: AFB1 concentration to decrease as the concentration of oil increased, however, we did not observe this pattern. Fungal inhibitory effect of oil on growth and development of the mycelium and spores could have been delayed.

Significance: Clove oil showed significant growth inhibiting properties. Hence, this EO may offer potential as a biological control agent against A. flavus and A. parasiticus in integrated pest management program of peanuts in Georgia or other peanut-growing states.
null
P1-226 Wide Range Host Range of the Genus Felixivirus are Potential Candidates for Salmonella Infantis Biocontrol

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Introduction: Salmonella Infantis is a globally emerging serovar of Salmonella causing human illness. Bacteriophages offer a potential biocontrol solution, however, it is not understood what type of resistance in Salmonella Infantis is selected for by phage biocontrol.

Purpose: We compared the ability of two phages with different host ranges (wide and narrow) to select resistant host mutant bacteria (RMB) and to reduce the bacterial load.

Methods: We performed 12-hour independent experiments using two models of phages of the Felixivirus07 genus infective against Salmonella Infantis: a phage with wide host range (WHR) and a phage with narrow host range (NHR). Challenges were conducted at a MOI of 0.01 in tryptic soy broth at 37°C and enumerated by plaque assay. After challenge, surviving RMB isolation was attempted by using a 0.2-micron filter.

Results: WHR phage decreased the bacterial load by 5.2±0.3 log CFU/ml, whereas NHR phage only decreased the bacterial load by 0.8±0.2 log CFU/ml. WHR phages showed higher selectivity in medium containing 4.8±1.5% water activity when compared to NHR phages.

Significance: This study shows that WHR phages may be more effective against Salmonella Infantis than NHR phages, and provides insight into the emergence of phage resistance. This knowledge will aid in the development of long-term biocontrol solutions for Salmonella Infantis.

P1-227 Determinants of Specificity of the Escherichia coli O157:H7 Bacteriophage PhiV10

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Developing Scientific Enterant

Purpose: The aim of this study was to determine the role of the O157 and H7 antigens in the infection of E. coli O157:H7 by PhiV10.

Methods: The thirteen O157 non-H7 and ten non-O157, H7 colonies were targeted to both binding and lysogenic assays. For binding assays, strains were incubated with 10 PFU PhiV10 per ml at 20 to 21°C. After ten minutes, unbound phages were isolated from bound phage using a 0.2-micron filter and enumerated by plaque assay with E. coli O157:H7, plaque counts similar indicating lack of binding. For lysogenic assays, strains were incubated with PhiV10 nuclease containing a kanamycin resistance marker for twenty minutes at 20:21°C. Lysogens were enumerated on kanamycin plates (50 µg/ml) and confirmed with luminescence assays.

Results: The binding assays showed all strains expressing the O157 antigen successfully bound PhiV10 as they displayed similar PFU as the control. However, all ten non-O157 isotypes expressing the H7 antigen showed no binding of the phiV10. The results of the lysogen assays indicated zero percent infection of non-O157:H7 strains, zero percent infection of E. coli O157:H7 strains.

Significance: The results of this research thus far support the hypothesis that PhiV10 binds to E. coli O157:H7 via the use of the O157 antigen and infects by using the H7 antigen, which will help in optimizing PhiV10 as a biocontrol for E. coli O157:H7.

P1-228 Population Dynamics of Listeria monocytogenes during Rehydration of Dehydrated Potato

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Introduction: Dehydrated plant food products have low water activities and do not support the growth of pathogenic bacteria. During rehydration, the increasing water activity and relatively neutral pH of these foods may require a product assessment to determine the extent to which they support the growth of L. monocytogenes, LPS, and other pathogens.

Methods: The experiments were conducted and a one-year study was conducted by calculating the reduction in weights of duplicate 10-g potato samples after heating at 100°C for 24 h. The initial water content of the dehydrated potatoes after drying 24 h was 4.8±1.5%. After only 2.5 min of rehydration, the water content of the final product increased to 49±6%.

Results: There was a significantly higher (P<0.05) reduction in weights of potato samples after heating at 100°C for 24 h. The experiments showed that WHR phages may be more effective against L. monocytogenes than NHR phages, and provides insight into the emergence of phage resistance. This knowledge will aid in the development of long-term biocontrol solutions for Salmonella Infantis. A significantly higher (P<0.05) reduction in weight was observed in both models in one hour. The RMB frequency was variable in time, with values between 1×10³ and 9×10⁷. The EOP with WH phages showed higher selectivity in media containing 4.8±1.5% water activity when compared to NHR phages.

Significance: This study shows that WHR phages may be more effective against Salmonella Infantis than NHR phages, and provides insight into the emergence of phage resistance. This knowledge will aid in the development of long-term biocontrol solutions for Salmonella Infantis.
Posters

**P1-234 Inactivation of Klebsiella pneumoniae in Hot Water by High Pressure Processing, Thermal Radiation, and Axia Xu1, Shownhuh Sheen1 and Christopher Simmons2**

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**Introduction:** Klebsiella pneumoniae is a bacterial pathogen associated with hospital-acquired respiratory infections (HAI) and urinary tract infections (UTI). The microbiome of recycled bedding sand was evaluated at various stages in the recycling process. As a separate but related project we also determined the inactivation of K. pneumoniae-associated UTI affects ≤50,000 annually. These conditions disproportionately affect women. Consumption or mishandling of contaminated meat and poultry could affect the health of these at-risk populations. The cost of sepsis, UTI, and UTI is estimated to be ≥$2 billion annually.

**Results:** This study was to determine the inactivation kinetics of K. pneumoniae in chicken meat.

**Methods:**

- **K. pneumoniae** multi-isolate cocktail containing clinical and food isolates was inoculated into ground chicken meat (GCM) that was 95% lean.
- **Thermo physical:**
  - D-values were 3.69 and 2.95 at 50°C and 60°C, respectively. Two isolates were selected as the most promising for the next study.
- **Thermo biological:**
  - The temperature was tested at 20°C, 50°C, and 60°C. TP values were 0.53 and 0.35 kGy at 4°C, respectively.
- **Significance:**
  - The advantage of the next-generation sequencing method is the unmatched capacity to identify species without the need to specifically target only a limited set of species. The method is capable of detecting species from a variety of sample types.

**Purpose:** The microbiome of recycled bedding sand is a complex ecosystem with thousands of species with more than a hundred samples simultaneously analyzed.

**Developing Scientist Entrant**

**P1-235 Characterizing the Microbiome of Recycled Bedding, the Environmental Persistence of Salmonella enterica serotype Dublin, and for Preventive Bovine Hepatitis\(^1\)**

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**Introduction:** Recycling bedding sand is a common practice in barns to reduce the cost of bedding. Little attention has been paid to the diverse microbial community in bedding sand and how it might impact dairy cattle health and pre-harvest food safety.

**Methods:** The microbiome of recycled bedding sand was evaluated at various stages in the recycling process. As a separate but related project we also developed a method to detect and enumerate entero serotype Salmonella. Dubin to persist in recycled dairy bedding sand with the native microbial communities.

**Results:** DNA was extracted from recycled sand using phenol-chloroform. The variable regions of 16S rRNA were amplified by PCR, and the products sequenced using Illumina sequencing technology. Genes were identified using Mutter software and analyzed with Ribosomal.

- To assess Salmonella survival, Salmonella Dubin was inoculated into bedding sand at 10⁵ CFU g⁻¹. At days zero, three, and seven the samples were tested for Salmonella.
- The conditions needed to inactivate K. pneumoniae in this study were similar to uninfected E. coli, and less than those typically needed to inactivate S. Typhimurium or S. Typhi.
- **Significance:**
  - Foods treated with intervention technologies are recommended for people with pre-existing medical conditions such as UTI and recurrent UTI, which can have a human genetic component. People with these conditions may benefit from the consumption of meat and poultry treated using these interventions to lessen their risk of infection. Consumer and women’s health goals will benefit from this research, as well as food companies which can provide safer foods for those target populations.

**Developing Scientist Entrant**

**P1-236 Antibiotic Resistance of Lactic Acid Bacteria Isolated from Dairy Products in Tianjin, China**

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**Introduction:** Antibiotic resistance is an increasingly significant problem worldwide. Lactic acid bacteria (LAB) are commonly involved in the manufacture and preservation of a wide range of food products. In recent years, the resistance of LAB has received more attention recently, because they may act as the reservoirs for the antibiotic resistance genes and transfer them to other microorganisms, including human pathogens, through the food chain.

**Methods:** We conduct a preliminary study to investigate the antibiotic resistance profiles of some LAB obtained from fermented dairy products in Tianjin, China.

**Purpose:** Eight different dairy samples were obtained from local markets in Tianjin, China. LAB was isolated and identified to species level using conventional biochemical methods. We used the chromogenic antibiotic resistance assay of all isolates to four antibiotics was analyzed by using disc diffusion method, and the corresponding resistance genes were determined by PCR and sequencing.

**Results:** A total of nine strains (three Lactobacillus bulgaricus and six Staphylococcus thermophilus) were isolated from commercial yogurt and cheese products. The resistance gene profiles of the isolates were as follows: 7 isolated with resistance to ampicillin, 4 isolated with resistance to tetracycline, 3 isolated with resistance to kanamycin, and 1 isolated with resistance to streptomycin. The resistance species of ampicillin were validated to be identical to the gene of tRNA adenine deaminases (AAD) in Staphylococcus epidermidis with a similarity of 99%.

**Significance:** This study revealed the widespread antibiotic resistance in LAB used in dairy products in China, thus the use of a starter in fermented meats should be closely monitored to ensure food microbiological safety.

**Poster**

**P1-237 Salmonella Detection from Large Milk Powder Samples Using the Thermo Scientific SureTect Salmonella Species PCR Assay**

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**Introduction:** Infant formula, milk powders and their ingredients are susceptible to Salmonella and Cronobacter contamination. Separate international standard reference methodologies for Salmonella and Cronobacter detection mean that different sample sizes and separate enrichments must be performed for each method. A harmonized enrichment strategy would be advantageous for microbiologists to minimize the amount of cost and effort spent during testing for these pathogens.

**Purpose:** The purpose of this study was to evaluate the Salmonella detection capabilities for large sample sizes of milk powders and ingredients using the Thermo Scientific SureTect Salmonella spp. PCR Assay in comparison with ISO 6579-1:2017. The same enrichment conditions were applied to the alternative method as the SureFect Cronobacter spp. PCR Assay.

**Methods:** Twelve 350-gram probiotic powdered infant formula (PIF) samples were spiked with Salmonella species and enriched alongside four (unspiked) samples for each species using the Thermo Scientific IAP enrichment formula, before processing according to the alternative method. The ISO method was performed in parallel to assess the accuracy of the method. Thirty-six 375-gram PIF, milk powder and ingredient samples were spiked with Salmonella Typhimurium or Salmonella infantis infants by desiccation and tested alongside seven unspiked samples according to the same study design, as a second unspared study.

**Results:** From the 300-gram PIF-study the alternative method confirmed the presence of Salmonella in 12 (100%) spiked samples after 16 hours incubation while the ISO method confirmed only 11 (91.7%). From the 375-gram samples study, comparable Salmonella detection was also achieved for alternative and ISO methods.

**Significance:** The data demonstrate the comparable performance of the alternative method for Salmonella detection compared to ISO 6579-1:2017 using large sample sizes and enabling one enrichment to be tested for both Salmonella and Cronobacter using PCR assays.

**Poster**

**P1-238 Food Authenticity Testing with Next-Generation Sequencing**

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**Introduction:** The Thermo Scientific NGS Food Authenticity Workflow identifies meat, fish and plant species from foods and feeds. The semi-automated workflow combines DNA extraction, library preparation, Thermo Fisher Scientific Ion Torrent technology and extensive database enabling identification of thousands of species with more than a hundred samples simultaneously analyzed.

**Purpose:** A complete process was developed to study the species authenticity of food products. The method for a streamlined workflow producing quality results.

**Methods:** Foods from different categories were tested to challenge the method including heavily processed foods, fresh and frozen foods, ready-to-eats, liquid foods and dairy products using the Next-Generation Sequencing method. The purpose of this study was to evaluate the accuracy of the method. All species from 19 meat samples, 18 fish samples and 10 plant samples were correctly identified. All plant species listed in the ingredients were detected except 1 in which a couple of species was accidentally analyzed.

**Significance:** The advantage of the next-generation sequencing method is the unmatched capacity to identify species without the need to specifically target only a limited set of species. The method is capable of detecting species from a variety of sample types.

**Poster**

**P1-239 Isolation and Genome Analysis of Lactococcus lactis Strains Characterized for the Potential Utilization of Allulose**

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**Introduction:** Allulose ([D]-Pisco) is a monosaccharide sugar known as a low-calorie high sweetness sweetener. Recently, researchers have interested in its beneficial food and physiological properties corresponding to high-quality foods. To our knowledge, few studies have been reported on the screening of lactic acid bacteria for potential usability of allulose.

**Purpose:** The purpose of this study was to develop and characterize the novel probiotic source of potential lactic acid bacteria to maximize the functional role and application of allulose as a prebiotic in food.

**Methods:** Modified MRS-allulose (1%) was prepared to screen allulose-utilizing lactic acid bacteria. The purpose of this study was to evaluate the accuracy of the method. All species from 19 meat samples, 18 fish samples and 10 plant samples were correctly identified. All plant species listed in the ingredients were detected except 1 in which a couple of species was accidentally analyzed.

**Significance:** The advantage of the next-generation sequencing method is the unmatched capacity to identify species without the need to specifically target only a limited set of species. The method is capable of detecting species from a variety of sample types.
genome sequence and the isolated Lactobacillus strains confirmed for their ability to produce probiotics in a simulated gastrointestinal environment.

Significance: We expect that the isolated Lactobacillus strains could be applied to the food industry as a functional probiotics to enhance health-promoting and gut health.

P1-240 - P1-242

Modulation of Gut Intestinal Microbiota during Prevention of Salmonella with Lactobacillus in BALB/c Mice

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University of Maryland, College Park, MD

Introduction: Gut intestinal microflora comprises an interactive ecosystem possessing considerable microbial diversity. The homeostatic status of this microbiome is the key gut in intestinal defense. Enteric pathogens can compromise beneficial microorganisms by dysbiosis with a loss of intestinal flora and metabolic functions. Several probiotics such as lactobacilli and bifidobacteria are generally known as potential modulators of gut integrity and health.

Methods: In Experiment 1, 30 mice were randomly divided into 5 groups (n=6/group). L. plantarum was used to treat mice orally at a dose of 10^9 CFU/g body weight per day for 1 week. In Experiment 2, 30 mice were randomly divided into 5 groups (n=6/group). L. rhamnosus was used to treat mice orally at a dose of 10^9 CFU/g body weight per day for 1 week. In Experiment 3, 30 mice were randomly divided into 5 groups (n=6/group). L. acidophilus was used to treat mice orally at a dose of 10^9 CFU/g body weight per day for 1 week.

Results: In Experiment 1, the relative abundance of Firmicutes was significantly increased by the treatment of L. plantarum compared to the control group (p<0.05). The abundance of Bacteroidetes was significantly decreased by the treatment of L. plantarum compared to the control group (p<0.05). In Experiment 2, the relative abundance of Firmicutes was significantly increased by the treatment of L. rhamnosus compared to the control group (p<0.05). The abundance of Bacteroidetes was significantly decreased by the treatment of L. rhamnosus compared to the control group (p<0.05). In Experiment 3, the relative abundance of Firmicutes was significantly increased by the treatment of L. acidophilus compared to the control group (p<0.05). The abundance of Bacteroidetes was significantly decreased by the treatment of L. acidophilus compared to the control group (p<0.05).

Significance: The results of this study showed that different probiotic bacteria have different effects on the gut microbiome, which can be used to develop new probiotics for the treatment of gut diseases.

P1-243 - P1-245

Protective Effects of β-Glucan Extracted from Spent Brewer’s Yeast during Freeze-drying and Storage of Probiotics

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Introduction: Yeast β-glucans are glycosyl polymers well-known for their bioprotective properties, such as antioxidant and anti-inflammatory activities. Probiotic cultures for food applications are generally supplied as freeze-dried powders. To avoid cell inactivation during freeze-drying and warrant stability during storage, polysaccharides are used as protective compounds. However, the role of β-glucan forms yeasts as cryoprotective of probiotics is still under investigation.

Purpose: This study evaluated the protective effects of spent brewer’s yeast β-glucan (YBβG) during freeze-drying and refrigerated storage of probiotic cultures.

Methods: The yeast β-glucan was obtained from the cell wall of brewer’s yeast (Saccharomyces cerevisiae) discarded as brewery slurry, using sonication and enzymatic treatments. Fresh biomass of Lactobacillus plantarum 49 (YBβG) and L. plantarum 201 (17% CFG) obtained from culture in MRS, Rogosa and Sorillo broth was used for freeze-drying at 24°C. The freeze-dried suspensions were dried at 55°C, 2.3% (w/v) sucrose-freeze-dried at 24°C, and 55°C, 2.3% (w/v) sucrose-freeze-dried at 24°C. The freeze-dried suspensions were stored at 4°C and 20°C for 24 days. The freeze-dried suspensions were stored at 4°C and 20°C for 24 days. The freeze-dried suspensions were stored at 4°C and 20°C for 24 days. The freeze-dried suspensions were stored at 4°C and 20°C for 24 days. The freeze-dried suspensions were stored at 4°C and 20°C for 24 days. The freeze-dried suspensions were stored at 4°C and 20°C for 24 days. The freeze-dried suspensions were stored at 4°C and 20°C for 24 days. The freeze-dried suspensions were stored at 4°C and 20°C for 24 days. The freeze-dried suspensions were stored at 4°C and 20°C for 24 days.

Significance: This study will provide insight into the production of β-glucan-extracted yeast during freeze-drying and storage, which is a relevant parameter for the development of Cryoprotective β-glucans for use in food applications.
Introduction: Due to a more attentional diet with foods of non-animal origin, a wide variety of vegetable delights is being introduced on the market. In some countries, flatfish have been associated with pests, salsa or guacamole or taboule.

Purpose: To collect information on the scope of these vegetable delights available on the Flemish retail market and to evaluate microbiological quality and safety of these types of products. Vegetarian spreads with major component plant-based proteins (e.g., chickpeas) were excluded from this study.

Methods: Local supermarkets were visited to establish the type of vegetable delights available at retail and information on storage conditions and ingredients were collected. A total of 40 and eight ambient-stable products were subjected to microbial analyses both at time of purchase and (enforced) end of shelf life for both overall quality and hygiene indicators and Listeria monocytogenes. Also ph and a was measured.

Results: In total 172 different food items were found, mainly fruits, and in 1.6% of all tested items, L. monocytogenes was isolated. The pH varied between 3.6 and 5.2, the a ranged between 0.91 and 0.99. No Listeria monocytogenes counts of >10 CFU/g were found. In all tested products B. cereus, sulphite reducing Clostridium, E. coli and coliform were occasionally found but never exceeded 3.65 log CFU/g, aerobic psychrotrophic count and psychrotrophic lactic acid bacteria ranged between less than two to 9.44 log CFU/g and yeast and fungi ranged between less than 1.67 log CFU/g.

The commercial stability was confirmed for the ambient-stable products.

Significance: Large differences were found between the overall microbial quality parameters, indicating variation in production conditions. Most products were at end of shelf life still of very good microbial quality.

P247 Thermal Reduction of Salmonella Inoculated Gelatin in Marshmallow Jennifer Todd-Searle, Danielle Voss, Bartosz Kielczewski, Polly Potier-Germain and Nancy Bontempo

Purpose: To determine internalized salmonella in marshmallow using real-time PCR for clarifying the worst case scenarios.

Methods: A 2.5 log reduction was achieved using a 2.5 minute treatment at 120°C.

Results: Salmonella was not detected in 6 out of 7 samples analyzed.

Significance: This study highlights the need for real-time PCR in verifying the thermal reduction of salmonella in ambient-stable products.

P248 Thermal Reduction of Salmonella spp., Escherichia coli, and Listeria monocytogenes during BBQ Sauce Processing Maurina Mansaray, Ashley Cunningham, Stephanie Nguyen, Christopher Showalter and Deann Alkins-Lewenton

Purpose: To determine the effect of thermal reduction of salmonella in BBQ sauce.

Methods: Three trials were conducted to evaluate the thermal processes.

Results: Salmonella was not detected in any of the samples from the three trials.

Significance: This study highlights the need for real-time PCR in verifying the thermal reduction of salmonella in ambient-stable products.

P249 Quantitative Microbial Risk Assessment of Vibrio cholerae and Vibrio vulnificus by Consumption of Flatfish Sushi and Sashimi Sejong Kim, Yoonejung Yoo, Young-Mog Kim, Kwon-Sam Park, Il Shik Shin* and Yoohan Yoon

Purpose: The objective of this study was to estimate the risk of V. cholerae and V. vulnificus by flatfish sushi and sashimi in Korea.

Methods: Vibrio cholerae and V. vulnificus were isolated from flatfish sushi and sashimi in Korea. The consumption rate and time data for flat-fish distribution were also collected, and appropriate proportionate distributions were then determined. Predictive models that can describe the fate of Vibrio species were cited and the bacterial cell counts were simulated under the distribution condition. Consumption data for flatfish were collected. Data on the consumption of flatfish was collected.

Results: Estimated intervention variables were ≤2.4 log CFU/G for V. cholerae and V. vulnificus, respectively. For intervention conditions, part dermophil was set at 0.1% and 1.5% for Vibrio cholerae (VC) and Vibrio vulnificus (VV) respectively. The distribution of Vibrio vulnificus was set at 77°C and a duration of 15 minutes. Time was set at 0.001 s (minimum time) to 100°C (saturated thermometer injected in the sushi).

Significance: V. vulnificus in flatfish was detected in Korea.

P250 Determination of a Thermal Reduction of Foodborne Pathogenic Bacteria at Mild Heating Temperatures with Inclusion of Parabens Zhujun Gao1, Qi Dong1, Chongao Ge2, Rohan Thilekar3 and Robert Buchanan4

Purpose: This study examined the potential enhancement of inactivation of four foodborne pathogens, Cronobacter sakazakii, Listeria Typhimurium, attenuated Escherichia coli O157:H7 and Listeria monocytogenes, during mild heating by inclusion of parabens in a model matrix.

Methods: A 6-log reduction of L. monocytogenes in Gelatine, a traditional Japanese and Korean fresh brain heart infu- sion broth with selected concentrations of butyl paraben (<250 ppm) just prior to thermal treatment (54°C to 58°C) for 15 min. Heating was conducted using a submerged coil apparatus (Shanxi Technologies), with samples collected at designated intervals. Samples were plated on trypticase soy agar for recovery and enumeration.

Significance: This study demonstrates the potential to reduce the risk of foodborne parabens to allow the use of mild heating temperatures for pasteurization.

P251 Validation of a Drum Roaster for Peanut Roasting in a Jhajharia, Gujarat (India) Peanut Butter Factory

Nancy Dobmeier and Balasubramanyam Kottapalli

Purpose: The purpose of this study was to validate a roasting process for peanuts.

Methods: A 6-log reduction of Salmonella was achieved by roasting peanuts at 185°C for 10 minutes.

Significance: This study validates the roasting process for peanuts in a peanut butter factory.

P252 Validation of Baking as a Kill-Step for Controlling Salmonella in Fruit Filled Pastry Minta Michael1, Daniel Vega2, Lakshminathamma Channaiyah3, George Milliken4, Harshvardhan Thippareddi5, Nicholas Sevart6 and Randall Phelps7

Purpose: This paper validates baking as a final step to control Salmonella in fruit-filled pastries.

Methods: Salmonella was inoculated into apple puree and baked at 570°F (192°C) for 1.5 hours.

Significance: Baking at 570°F (192°C) for 1.5 hours will reduce the risk of Salmonella contamination in fruit-filled pastries.

P253 Validation of Baking as a Kill-Step for Controlling Salmonella in Fruit Filled Pastry

Minta Michael1, Daniel Vega2, Lakshminathamma Channaiyah3, George Milliken4, Harshvardhan Thippareddi5, Nicholas Sevart6 and Randall Phelps7

Purpose: The goal was to determine the lethality of the baking process on Salmonella in fruit-filled pastries.

Methods: Salmonella was inoculated into apple puree and baked at 570°F (192°C) for 1.5 hours.
to determine Salmonella reductions at defined intervals. D-values in pastry dough were determined at 55, 58, and 61°C using thermal-death-time disks and not underdosing or insufficient. However, little is known about the stress response of Biofilm-forming ability despite low cellulose production.

**Methods:** A six-strain mixture of WT and PS

**Results:** Counts of WT and PS Enterococcal colony at 7°C were 2.40±0.7 and 3.29±0.2 prior to treatment, respectively. These counts on day zero were not significantly different from those which were enumerated immediately after an injury-recovery period. The treatment of counts of two-week mature biofilm of WT Enterococcal colony at 15°C were 5.22±0.1 and 4.56±0.1 before and after treatment, respectively, exhibiting low efficiency (P<0.05) of the sanitizer against two-week mature biofilm. Significance: The tested serogroups and phenotypes of pathogen exhibited similar biofilm formation capability and sensitivity to QAC. The sanitizer test at 2.4°C was used to confirm the viability of the isolated strains. The manufacturer appears to be efficacious only against planktonic cells while exhibiting inability for the complete removal of one- and two-week mature biofilm.

**Phenotypic characterization of Biofilm-Forming B. subtilis sp. Identified in the Irish Artisan Bakery Environment

Sakshi Lamba1, MM Dechamama1, Semius Fanning1 and Amalia G. Scannell1

**Methods:** Phenotypic characterization of five selected isolates and B. subtilis NCTC3610 as a control was performed using Congo Red-Calcofluor assay for the co-expression of extracellular matrix components (namely curli fimbriae and cellulose), crystal violet biomass assay, and pellicle formation at the air–broth interface at 30, 37 and 45°C from 18 to 96 h.

**Results:** All the isolates had the ability to form weak, moderate and strong biofilms at different test time and temperature. The isolates exhibited a red, dry (rough) (RDAR) morphology on Congo Red-Coliagar agar after 72 h at 30, 37, and 45°C, indicating the presence of curli fimbriae, while two isolates showed weak (W) and moderate (M) curli fimbriae development. No fluorescence was detected in E. faecalis in QAC-exposed static and in QAC-sprayed dynamic growth in MM medium assessed using crystal violet decrease over time at 30 and 37°C with weak to moderate attachment in all but one isolate which consistently formed strong films at both the temperatures. At 45°C, the biomass production appeared to decrease from 18 to 24 h and then increased for three out of five isolates, however attachment to the surface was strong for all the isolates. Fragile pellicles at the air–broth interface at 37 and 45°C suggested biofilm forming ability despite low cellulose production.

**Significance:** The increased recognition of the nature of B. subtilis biofilms will contribute significantly to the development of strategies for the control of relevant bakery food products.
**P1-259** Prevalence of Methicillin-resistant *Staphylococcus aureus* in the Isidro Ayora General Hospital in the City of Loja, Ecuador

Elisa Baculima, Diana Hualpa, Andrea Cabrera and Fernando Serrano

*Universidad Técnica Particular de Loja, Loja, Ecuador*

**Introduction:** Methicillin-resistant *Staphylococcus aureus* (MRSA) causes various infections of both community and hospital origin. The interest in this study is due to insufficient information about this pathogen in Ecuador.

**Methods:** To determine the prevalence of MRSA in Isidro Ayora Hospital in the city of Loja, Ecuador, 331 nasal and blood samples and 92 food samples were collected. The food samples were seeded in Baird Parker agar by inoculating one ml in three boxes. Fifty g of the product was weighed and mixed with 450 ml ofButterfield’s phosphate-buffered saline and incubated 45 to 48 h at 35°C. The identification of S. aureus and MRSA in all samples was carried out by catalase, coagulase, and mannitol tests. For the confirmation of MRSA, colonies were inoculated on Mueller Hinton agar withcefotaxin and incubated under aerobic conditions for 18 hours at 37°C. The samples are considered MRSA when the inhibition zone is ≥ 21 mm.

**Results:** From 10 food samples, it was determined that 12 (1%) of them were positive for S. aureus, of which 1 (0.7%) was confirmed as MRSA and sensitive to linezolid. In clinical samples a greater prevalence of this microorganism was reported in nurses (50%), doctors (14%) and internal medical (28%). The results suggest a need for application and implementation of good hygiene and sanitation practices.

**Significance:** The existence of S. aureus in prepared foods suggests a potential risk to the health of patients and hospital staff.

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**P1-260** A Comprehensive Study of bioMérieux VIDAS SET II and the r-Biopharm Risdecern SET Total to Detect the Presence of Staphylococcal Enterotoxins Using Matrix Dependent Extractions from a Variety of Foods

Ashley Aurand-Cravens, Beth Johnson, Vaneet Arora, Patricia Rule and Stan Bailey

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**Introduction:** Herein, the performance of a secondary confirmation assay, the Use the VIDAS SET Total assay for use in detection and confirmation of SETs in foods. The VIDAS SET assay uses an automated ELISA (Enzyme Linked Fluorescent Assay) based technology and the Risdecern SET Total is a manual ELISA method. Both kits screen for staphylococcal enterotoxins and use different extraction protocols. Box C2 used 0.1 ml in the VIDAS SET assay for use in detection and confirmation of SETs in foods. Box C2 used 0.1 ml in the Risdecern SET assay for use in detection and confirmation of SETs in foods. The VIDAS SET assay uses an automated ELISA (Enzyme Linked Fluorescent Assay) based technology and the Risdecern SET Total is a manual ELISA method. Both kits screen for staphylococcal enterotoxins and use different extraction protocols. Box C2 used 0.1 ml in the VIDAS SET assay for use in detection and confirmation of SETs in foods. Box C2 used 0.1 ml in the Risdecern SET assay for use in detection and confirmation of SETs in foods.

**Purpose:** The purpose of this study was to compare the two assays for toxin detection in a variety of foods and to evaluate the performance of the different extraction methods.

**Methods:** Twenty-three different food matrices and six proficiency samples were evaluated for a total of 60 VIDAS SET II and 47 Risdecern SET analysis. Each matrix was spiked with SET A or B at one log of CFU/g. Once detected via VIDAS SET II, the same extraction material was used for confirmation with the Risdecern SET Total assay.

**Results:** Both tests correctly reported all the spiked samples as positive and the control samples as negative. The VIDAS SET II was able to correctly detect the toxin when present, following five different matrices’ unique extraction protocols. The Risdecrern also correctly detected toxin presence/absence in each of the six proficiency samples.

**Conclusion:** We concluded that both assays can be relied upon to use the presence of SETs using the recommended sample extraction methods but that the initial and the secondary testing could be carried out using the same extraction material. Lessons learned and challenges with the different extraction methods will be shared.

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**P1-261** Hurdle Enhancement of Antimicrobial Efficacy of Acidic Electrolyzed Water on Bacillus cereus Spp

Using Ultrasoundization

Ruling Liu, Donghong Liu and Xiaonan Lu

*Zhejiang University, Hangzhou, China, Zhejiang University, Hangzhou, AP, China, Food, Nutrition and Health Program, Faculty of Land and Food Systems, The University of British Columbia, Vancouver, BC, Canada*

**Developing Scientist Entrant**

**Introduction:** Bacillus cereus spp have tough and metabolically inert structures whose formation is a strategy for this bacterium to survive in unfavorable conditions in the food industry. It is of significant concern to the food industry because it produces two thermostable toxins and causes massive foodborne disease. Thus, it is critical to develop effective technology for inactivation of bacterial spores.

**Purpose:** This study evaluated the mitigation effects of ultrasonic treatment combined with acidic electrolyzed water (AEW) on B. cereus spp.

**Methods:** B. cereus spp were treated with ultrasound and AEW separately and combined, followed by investigation of the antimicrobial effects. Flow cytometry and electron microscopy were used to investigate changes in the physiological status and ultrastructure of spores. The trials were replicated 3 times in each experiment.

**Results:** AEW treatment induced 1.05 ± 1.37 log CFU/ml reduction of B. cereus spp while the sporidal effect of ultrasound was minor. In comparison, combined treatment showed a 2.29 log reduction of spores and to have a synergistic effect. Moreover, simultaneous antimicrobial treatment was more effective to motivate spores than successive antimicrobial treatments. Flow cytometry combined with SYTO 16/P7 staining analysis revealed that ultrasound hydrotized the cortex and AEW partially damaged the integrity of the inner membrane of spores. We also identified that treatment of AEW with ultrasound destroyed spores’ structure (e.g., cortex) that subsequently decreased the resistance of spores. Electron density of spores appeared to be heterogeneous after AEW treatment.

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**P1-262** Isolation and Characterization of Vibrio parahaemolyticus Protected from Laboratory Validation Foodborne Pathogens by PAEs Released into Olive Oil

Hua-Ru Su, Cheng-Wei and Tai-Yuan Chen

*National Taiwan Ocean University, Keelung, Taiwan, 2University of Maryland, College Park, MD*

**Purpose:** To determine the prevalence of MRSA in Isidro Ayora Hospital in the city of Loja, Ecuador

**Introduction:** Methicillin-resistant *Staphylococcus aureus* (MRSA) causes various infections of both community and hospital origin. The interest in this study is due to insufficient information about this pathogen in Ecuador.

**Methods:** A standardized agar dilution method was used to establish risk analysis for PAEs released into olive oil.

**Results:** From 92 food samples, it was determined that 12 (13%) of 92 were positive for *S. aureus*, of which 1 (0.7%) was confirmed as MRSA and sensitive to linezolid. In clinical samples a greater prevalence of this microorganism was reported in nurses (50%), doctors (14%) and internal medical (28%). The results suggest a need for application and implementation of good hygiene and sanitation practices.

**Significance:** The existence of S. aureus in prepared foods suggests a potential risk to the health of patients and hospital staff.

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**P1-263** The Migration of Phthalate Esters from Packaging Materials to Olive Oil under High Temperature Storage

Hua-Ru Su, Cheng-Wei and Tai-Yuan Chen

*National Taiwan Ocean University, Keelung, Taiwan, 2University of Maryland, College Park, MD*

**Purpose:** The presence of methicillin-resistant *Staphylococcus aureus* (MRSA) is a major concern to the food industry because it produces two thermostable toxins and causes massive foodborne disease. Thus, it is critical to develop effective technology for inactivation of bacterial spores.

**Methods:** The standards included DMP (dimethyl phthalate), DEP (diethyl phthalate), BBP (benzyl butyl phthalate), DBP (di-butyl phthalate), DEHP (di-2-ethylhexyl phthalate) and DINP (di-n-butyl phthalate) were chosen as target compounds.

**Results:** The highest released level for DMP (PET bottle) was 16.84±2.66 (µg/ml), and the predicted consumption levels were 0.0092 and 0.0089 mg/kg body weight/day for male (64 kg body weight) and female (52 kg body weight) respectively.

**Conclusion:** The predictions finish such that the lowest range of DEHP can reach to five percent (relative standard deviation, 95% CI) the DMP and DEP migrated out of containers, including PET, HDPE and HDPE. Sealed and unsealed were within 4.77 to 4.84 mmHg/ml and 9.1 to 9.11 mmHg/ml, respectively. At other PET bottles, the migration was increased 6.56 folds that at 30 days sealed PET bottles.

**Significance:** The highest released level for DMP (PET bottle) was 16.84±2.66 (µg/ml), and the predicted consumption levels were 0.0092 and 0.0089 mg/kg body weight/day for male (64 kg body weight) and female (52 kg body weight) respectively.

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**P1-264** WITHDRAWN

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**P1-265** Insight into bacterial communities present in commercial chopped romaine lettuce processed in early and late seasons

Chao Liao and Luxin Wang

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**Developing Scientist Entrant**

**Introduction:** Due to recent outbreaks associated with chopped romaine lettuce, the abundance and diversity of bacterial communities present in these products needs to be better evaluated.

**Purpose:** The aim of this study was to evaluate the changes of bacterial communities present in three brands of chopped romaine lettuce products harvested in California and one brand harvested in Florida.

**Methods:** Commercial romaine lettuce products (a total of 72 bags) were purchased from the local grocery market in early (September to October) and late (March to April) seasons of 2018. Three brands of commercial products were chosen with two brands from California and one brand from Florida. DNA was extracted from these samples on the “use-by” dates and the dynamic changes of microbial communities were evaluated by plating sample homogenate onto plate count agar (PCA) and anaerobic agar (APC) and by conducting 16S rRNA sequencing.

**Results:** The abundance of *Vibrio parahaemolyticus* was observed in all three brands of chopped romaine lettuce products (0.5 x 10^6 CFU/g) respectively.

**Conclusion:** The highest released level for DMP (PET bottle) was 16.84±2.66 (µg/ml), and the predicted consumption levels were 0.0092 and 0.0089 mg/kg body weight/day for male (64 kg body weight) and female (52 kg body weight) respectively as recommended by the Scientific Committee on Food of the European Commission (EC-SC).
Proteobacteria with relative abundance from 15.09 to 59.46% and Firmicutes with relative abundance from zero to 84.48% dominating across all samples. Early season samples had significantly higher Firmicutes than late season samples (P<0.05). At the genus level, Paenibacillus, Lactococcus, and Rhodococcus were the top three genera in all samples.

Significance: This study provided important insight into microbial communities present in commercial lameau rice products. Results will directly benefit the risk assessment of products when they approach "use by" dates.

P2-266 Analyzing Microbial Community Change of Turkey Litter Compost Due to Heat Exposure Using 16S High Throughput Sequencing

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Objective: Turkey litter compost samples were collected from farms in South Carolina and Georgia. The compost was heat treated using different temperatures and holding times in an anaerobic chamber. The microbial community composition was compared in the compost before and after heat treatment, and the effectiveness of the compost treatment for removing DNA from dead cells was confirmed. Further, the identification of indigenous genes in poultry litter surviving the physical heat treatment may lead to future studies on biological control of pathogens in soil amendmen.t

Significance: The microbial community analysis was optimised for turkey litter compost, and the effectiveness of PMA treatment for compost on removing DNA from dead cells was confirmed. Further, the identification of indigenous genes in poultry litter surviving the physical heat treatment may lead to future studies on biological control of pathogens in soil amendment.
Complications on the cost of foreign materials for food are reported to the health centers in Japan. Each health center has archived data on Miyagi from telephone population surveys conducted for the whole of Japan and for Miyagi prefecture. We merged the telephone survey data conducted in 2016 surveillance is strongly suggested, in order to identify and prioritize food safety measures more precisely and to monitor the effectiveness of risk management.

We estimated the burden of foodborne illness for Campylobacter, Salmonella and Vibrio parahaemolyticus in Japan from 2006 to 2016. The most frequently reported food items were bugs followed by hairs, metals, and plastics. Prepared dishes, confectionary and farm products were the most reported food items. The highest number of complaints occurred in food production/processing/cooking business sectors, followed by retail and food service sectors. Significant: These data enable us to recognize the situation of contamination by foreign materials in food in Japan, and to guide food business operators in preventing foreign food contamination by serious.

There were 14,379 complaints reported on contamination by foreign materials during the data collection period (approximately three years). Respondents (age: 36.7±9.3 years) were mostly meat traders (106, 52.2%) and butchers (56, 27.6%) with 178 (87.7%) reporting work-associated food safety risks. Of these respondents, 102 (52.5%) had some formal education and 146 (71.8%) had some form of training on food safety.

A cross-sectional survey involving random sampling of 203 workers from five slaughterhouses were assessed using a pretested structured questionnaire. Information on food safety risks was obtained using a numeric scoring system. Data were analyzed with descriptive statistics, and univariate analysis was performed.

- **Results:**
  - The mean score for the control group was 65.2 and for the intervention group it was 77.3. The intervention group had a significantly higher score than the control group (p < 0.001)
  - The proportion of workers reporting some knowledge of food safety risks in the intervention group was significantly higher than the control group (p < 0.001)
  - The proportion of workers who reported practicing some risk management practices in the intervention group was significantly higher than the control group (p < 0.001)

- **Significance:**
  - The study provided empirical basis for the development of suitable policies and interventions to mitigate food safety risks in Nigerian slaughterhouses.
  - The study has important implications for the improvement of food safety management in Nigeria, particularly in the context of the extensive tourism industry.

- **Purpose:**
  - The objective of this study was to perform a transcriptomic analysis of strain 62A grown in two media, toxin production medium (TPM) and non-toxin production medium (NPM). The study aimed to elucidate the mechanisms underlying their genetic regulation.

**Research Question:**
- *What is the genetic basis for toxin production in strain 62A of *Clostridium botulinum*?*

**Materials and Methods:**
- *Strain 62A in Chilled Poultry Carcasses in Algeria* was used in the study.
- The study was conducted at the Laboratory of Food Hygiene and Quality Insurance System (LHAS), National Veterinary School, Algeria.

- **Results:**
  - The study found that the toxin production is controlled by a complex regulatory network involving multiple genes.
  - The study identified a putative toxin protein (botC) which is encoded by the gene BotC.

- **Significance:**
  - This study has implications for the development of food safety control measures in the poultry industry in Algeria.

- **Purpose:**
  - The objective of this study was to evaluate the implementation of HACCP (Hazard Analysis and Critical Control Point) system in food manufacturing companies in the Emirates of Dubai.

**Research Question:**
- *How effective is the implementation of HACCP system in food manufacturing companies in the Emirates of Dubai?*

**Materials and Methods:**
- A cross-sectional survey involving random sampling of 203 workers from five slaughterhouses were assessed using a pretested structured questionnaire. Information on food safety risks was obtained using a numeric scoring system. Data were analyzed with descriptive statistics, and univariate analysis was performed.

- **Results:**
  - The mean score for the control group was 65.2 and for the intervention group it was 77.3. The intervention group had a significantly higher score than the control group (p < 0.001)
  - The proportion of workers reporting some knowledge of food safety risks in the intervention group was significantly higher than the control group (p < 0.001)
  - The proportion of workers who reported practicing some risk management practices in the intervention group was significantly higher than the control group (p < 0.001)

- **Significance:**
  - The study provided empirical basis for the development of suitable policies and interventions to mitigate food safety risks in Nigerian slaughterhouses.
  - The study has important implications for the improvement of food safety management in Nigeria, particularly in the context of the extensive tourism industry.

**Purpose:**
- The objective of this study was to perform a transcriptomic analysis of strain 62A grown in two media, toxin production medium (TPM) and non-toxin production medium (NPM). The study aimed to elucidate the mechanisms underlying their genetic regulation.

**Research Question:**
- *What is the genetic basis for toxin production in strain 62A of *Clostridium botulinum*?*

**Materials and Methods:**
- *Strain 62A in Chilled Poultry Carcasses in Algeria* was used in the study.
- The study was conducted at the Laboratory of Food Hygiene and Quality Insurance System (LHAS), National Veterinary School, Algeria.

- **Results:**
  - The study found that the toxin production is controlled by a complex regulatory network involving multiple genes.
  - The study identified a putative toxin protein (botC) which is encoded by the gene BotC.

- **Significance:**
  - This study has implications for the development of food safety control measures in the poultry industry in Algeria.
Growth method should be considered when testing desiccation resistance or survival in food systems. Samples of fresh, whole, hulled strawberries were contaminated with different levels of *L. monocytogenes* and then aged for 48 to 72 hours at 4°C. Half of the samples were placed in -30°C for four to six days after aging, while the remaining were analyzed via direct plating and MPN. The frozen disks were then thawed and analyzed in the same manner as the fresh samples. Overall, there was a significant reduction in *L. monocytogenes* concentration after aging freezing, but some growth was still present. The enumeration method that showed to be more sensitive in this scenario was MPN.

Purpose: The purpose of this study was to determine the fate of *L. monocytogenes* in frozen strawberries and to evaluate the best enumeration method for this matrix and environmental condition.

Methods: Samples of fresh, whole, hulled strawberries were contaminated with different levels of *L. monocytogenes* and then aged for 48 to 72 hours at 4°C. Half of the samples were placed in -30°C for four to six days after aging, while the remaining were analyzed via direct plating and MPN. The frozen disks were then thawed and analyzed in the same manner as the fresh samples. Overall, there was a significant reduction in *L. monocytogenes* concentration after aging freezing, but some growth was still present. The enumeration method that showed to be more sensitive in this scenario was MPN.

Significance: Our study provides evidence that we must be concerned about *L. monocytogenes* in not only fresh but also frozen produce.
P2-04 - Dried Spices and Their Role in Final Product Quality – A Case Study

Jack Mouradian, Shabnam Mohammad and Kevin Hsiao
Promega Corp., Madison, WI

Introduction: Antimicrobial activity of dried spices against E. coli O157:H7 is a well-known phenomenon. This study examined the feasibility of adding dried spices after irradiation to reduce the contamination levels of this pathogen in meat products.

Methods: Irradiated and non-irradiated beef (ground and whole), ham, and turkey were used in Smoked Sausage, Pork, and Turkey Fajitas, respectively. Dried spices, including dried onion, garlic, and oregano, were added at 1% and 2% of the final product weight, followed by cooking under standard conditions. Samples were collected at two time points: before and after cooking.

Results: A significant reduction in the number of E. coli O157:H7 was observed in both irradiated and non-irradiated samples when dried spices were added. The highest reduction was observed in the Smoked Sausage samples with the 2% spice addition, with a decrease of over 7 log units.

Significance: This study suggests that the addition of dried spices to irradiated meat products can significantly reduce the number of E. coli O157:H7, making it a feasible and effective intervention strategy for food safety improvement.

P2-07 - Assessing Bacterial Viability by Monitoring Adenine Nucleotides and Adenylate Charge in Response to Biocide Treatment

Saleh Iqbal, Subbahul Modyonjal and Kevin Hsiao
Promega Corp., Madison, WI

Introduction: Adenine nucleotides provide a valuable tool for assessing the effect of biocide treatments such as chlorine (0.5 and 1.5 ppm), chloramine T (two ppm), and gluclaraldehyde (0.1%) in a time-dependent manner up to 20 min. We also tested these biocides for a period of up to 120 min to determine the log kill for each biocide concentration. These studies were carried out to determine the time required for each biocide (0.5 and 150 ppm) to achieve complete inactivation of E. coli O157:H7.

Methods: E. coli O157:H7 was grown in LB broth culture by the plate count method. The minimum inhibitory concentration (MIC) was determined using the disc diffusion method. The MIC was then used to determine the time required for complete inactivation of the bacteria.

Results: At the MIC, the bacteria were killed within 30 min for both chlorine and chloramine T. The results suggest that the combination of chlorine and chloramine T is effective in completely inactivating E. coli O157:H7.

Significance: This study highlights the potential of using adenine nucleotides to monitor bacterial viability in response to biocide treatment, which can provide important insights into the effectiveness of these treatments.

Poster presentations
P2-10 Evaluation of Bactericidal Effects of Phenylisothiocyanate on Shiga Toxin-producing E. coli in Beef Products
Eunyoung Cho, Jin-Soo Kim, Young-Tae Park, and June-Mi Kim
Department of畜肉学, Nong-Agricultural Technology University, Gyeongsan, Korea

Purpose: The purpose of this study was to compare the inhibitory properties of a chemical and a natural sodium-free preservative on the growth of Shiga toxin-producing E. coli.

Methods: A Danish CSS processor in collaboration with DTU Food used a combination of salting with injection brining followed by treatments on the surface of the fillets to determine if there was an optimal distribution of additives. The main product characteristics of CSS were three percent water-phase NaCl and pH of 6.1. A new study was done to demonstrate the synergistic antimicrobial activity of crude extracts from Brassica rapa var. fenn.

Results: The purpose of our study was to screen cultures in our collections, or those occurring naturally, that can reduce vegetable nitrate to nitrite. New sources of nitrate reducing organisms may provide more efficient methods of generating nitrite.

Significance: Our results showed that WC and RC extracts which proved to be potentially effective can be used as natural alternative preventives to conventional nitrite.
P2-17 Physiological Damage Caused to Cells of Salmonella Enteritidis PT4 by Continuous Exposure to Mint (Mentha piperita L.) Essential Oil

Adina Nadja Ferreira de Melo, Geany Targino de Souza Pedrosa, Erika Tayse da Cruz Almeida, Evandro L. de Souza, Donald W. Schaffner¹ and Marciane Magnani²

¹Federal University of Pará, João Pessoa, Brazil; ²Federal University of Pará, João Pessoa, Brazil; ³Universidade de Zango, Zanzibar, Tanzania; ⁴Federal University of Pará, João Pessoa, Brazil

Introduction: The Mentha piperita L. essential oil (MEO) is a “green” antimicrobial. The activity of MEO against Salmonella has been primarily associated with changes to cell membrane permeability, which may cause leakage of vital intracellular components. Little is known about the effects of continuous exposure of Salmonella to MEO.

Methods: This study evaluated the effects of continuous exposure to MEO on the physiology of a strain of Salmonella Enteritidis PT4 isolated from chicken meat associated with foodborne disease outbreaks.

Results: Salmonella Enteritidis PT4 grown in LB broth were exposed to 2.5 µL/mL of MEO in brain heart infusion (BHI) broth or to BHI broth on 12 consecutive days. At the end of the exposure, cells were centrifuged (450g, 10 min, 4°C), washed twice and resuspended in PBS after every 24 h of exposure. Cells were labelled with propidium iodide for membrane integrity, ethidium homodimer for intercellular activity, and 5- and 5.5-didodecylaminotetrazolium chloride for respiratory activity. Flow cytometry data for acquisition was set on 10,000 FSC and CDG were gated for analysis. In each acquisition, 10,000 events were analyzed. Density plots were generated with flowJo software and dot plot analysis of FL1 vs. FL2 was used to establish fluorescence of each population. Cytograms were analyzed using BD Accuri C6 software.

Results: After exposure to MEO, approximately 93% of lactobacilli remained intact, while 5% demonstrated efflux changes and 2% exhibited decreased respiratory activity. In contrast, the percentage of Salmonella Enteritidis PT4 cells demonstrating efflux changes increased to 47%, while 7% showed decreased respiratory activity. Cells not exposed to MEO remained largely polarized with normal efflux pump activity and intact membranes.

Significance: These results show that MEO exerts inhibitory effects through a multi-target mechanism in Salmonella Enteritidis PT4 cells, but continuous exposure appears to create a sub-population of cells able to repair the injuries.

P2-18 Cell Damage Caused by Mandarin Essential Oil to Autochthonous Sporadic Lactic Acid Bacteria in Orange Juice

Geany Targino de Souza Pedrosa, Adina Nadja Ferreira de Melo, Erika Tayse da Cruz Almeida, Evandro L. de Souza, Rafael Pagan and Marciane Magnani

¹Federal University of Pará, João Pessoa, Brazil; ²Federal University of Pará, João Pessoa, Brazil; ³Universidade de Zango, Zanzibar, Tanzania; ⁴Federal University of Pará, João Pessoa, Brazil

Introduction: Lactic acid bacteria comprise the largest group of raw fruit microbiota and can easily be transferred to juices causing deterioration through spoilage activity and 28% had damaged membranes. Cells not exposed to MEO remained largely polarized with normal efflux pump activity and intact membranes. A decrease of approximately 0.5 log CFU/mL was observed over 251.3 increasing exposure time reducing the population of a cells with depleted membrane efflux activity or membrane damage did not change the population.

Significance: These results show that MEO exerts inhibitory effects through a multi-target mechanism in Salmonella Enteritidis PT4 cells, but continuous exposure appears to create a sub-population of cells able to repair the injuries.

P2-19 Antimicrobial Resistance of Salmonella Recovered from Environmental Samples on Three North Carolina Tomato Farms

Robin Grant Moore, Diane Ducharme, Otis Simmons, Kellie P. Burris, Lee-Ann Jaykay, Jie Zheng, Eric Brown¹ and Rebecca L. Belf³


Introduction: Tomatoes have been repeatedly implicated in foodborne illnesses. The test strains (10⁴ CFU/mL) were exposed to 2.5 µL/mL of MEO in brain heart infusion (BHI) broth or to BHI broth on 12 consecutive days. At the end of the exposure, cells were centrifuged (450g, 10 min, 4°C), washed twice and resuspended in PBS and immediately labeled with the fluorochromes: propidium iodide for membrane integrity, ethidium homodimer for intercellular activity, and 5- and 5.5-didodecylaminotetrazolium chloride for respiratory activity. Flow cytometry data for acquisition was set on 10,000 FSC and CDG were gated for analysis. In each acquisition, 10,000 events were analyzed. Density plots were generated with flowJo software and dot plot analysis of FL1 vs. FL2 was used to establish fluorescence of each population. Cytograms were analyzed using BD Accuri C6 software.

Results: After exposure to MEO, approximately 93% of lactobacilli remained intact, while 5% demonstrated efflux changes and 2% exhibited decreased respiratory activity. In contrast, the percentage of Salmonella Enteritidis PT4 cells demonstrating efflux changes increased to 47%, while 7% showed decreased respiratory activity. Cells not exposed to MEO remained largely polarized, presented intact membranes and normal efflux and respiratory activity.

Significance: Results suggest that MEO exerts activity in distinct cell membrane targets, however, the extent of the effects varies with the intrinsic resistance of sporogenic bacteria.

P2-20 Assessment of Antibiotic Usage and Oxytetracycline Residues in Eggs from Commercial Poultry Farms in Nigeria

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Introduction: The misuse of veterinary drugs is one of the causes of drug residues in animal products.

Methods: A cross-sectional study was carried out to investigate the antibiotic usage patterns in commercial poultry farms and the presence of antibiotic residues in animal products.

Results: Twenty-six (15.4%) of the antimicrobial resistant isolates were found at Farm Site B. A single MDR sample (cefoxitin, amoxicillin/clavulanic, and tetracycline) was found to be resistant against chloramphenicol. Twenty-eight isolates (16.6%) demonstrated resistance to one antibiotic (streptomycin or ampicillin): twenty-six samples (20%) were obtained from random samples of egg drops in the markets and forty-eight (40%) were obtained from marketed eggs. Pooled egg samples (10 from each farm) were analyzed with high-performance liquid chromatography (HPLC) for oxytetracycline residue levels.

Significance: These results show a high level of antibiotic activity in the targeted farms. The respondents (61.5%) frequently administered drugs without a veterinary prescription and 73.3% admitted to self-medication. The results indicate that the use of oxytetracycline by farmers is a public health concern.

P2-21 Effect of Nutrient Enrichment on Antibacterial-resistance-Dependent Antibiotics of Native Bacteria in Orange Juice

Tianyi Zhang, Adnan Alnajjar and Craig E. Miller


Introduction: Administration – Center for Food Safety and Applied Nutrition, College Park, MD, U.S. Food and Drug Administration, Center for Veterinary Medicine, Ibadan, Nigeria

Methods: Field plots were created in an area with little to no impact by antimicrobials or fecal material derived from humans, livestock, or companion animals. Three three-plant replicates were obtained along measurements and dot plot analysis of FL1 vs. FL2 was used to establish fluorescence of each population. Cytograms were analyzed using BD Accuri C6 Software.

Results: After exposure to MEO, approximately 93% of lactobacilli remained intact, while 5% demonstrated efflux changes and 2% exhibited decreased respiratory activity. In contrast, the percentage of Salmonella Enteritidis PT4 cells demonstrating efflux changes increased to 47%, while 7% showed decreased respiratory activity. Cells not exposed to MEO remained largely polarized, presented intact membranes and normal efflux and respiratory activity.

Significance: Results suggest that MEO exerts activity in distinct cell membrane targets, however, the extent of the effects varies with the intrinsic resistance of sporogenic bacteria.

P2-22 Influence of the Presence of a Natural Antimicrobial on Ready-to-Eat, Clean Label, Smoked Pork Sausage during Extended Storage at 4°C and 10°C

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Introduction: Administration – Center for Food Safety and Applied Nutrition, College Park, MD, U.S. Food and Drug Administration, Center for Veterinary Medicine, Ibadan, Nigeria

Methods: The addition of nutrients in the form of sterile bacteriological media led to changes in concentrations from < 1-log CFU/g of soil of tetracycline-resistant (TET) Enterococcus spp. (TET) Enterococcus spp. and 5- and 4-log CFU/g soil, respectively. These results indicate that the esthesiobacterial bacteria are resilient to antimicrobial treatments due to their typical low background flora levels. When nutrient addition was discontinued TET and ERY-resistant populations rapidly decreased to undetectable levels. In both pCR and metagenomic analyses showed that nutrient addition obtained the levels of several gene targets (pCR targets included: blaox, bte, pht, tetr, tetA, tetB, and tetC) of multiple antibiotic resistance classes (families). When using the antibiotic treatment, 88% of Enterococcus spp. treated with erythromycin were killed, while the same bacteria treated with tetracycline only killed 40%. The Bray-Curtis dissimilarity matrix shows clustering of enriched samples with samples from cattle feedlot pen surface material. Significant differences were observed in bacterial communities in the environment without accounting for the ecological dynamics such as outgrowth of native bacterial populations through nutrient supply in the form of fecal deposition.

Significance: This project demonstrates that AMR can flourish in environments in the absence of human-antimicrobial selective pressure.
In the processing environment.

Application of bacteriophages in food processing facilities is still relatively new, thus not much is yet known about possible occurrence of resistant organisms. Therefore, there is a need to more fully investigate the potential risk posed by resistant organisms to food safety. Small-scale studies have been conducted to evaluate the efficacy of bacteriophage application in different processing environments, however there is still a paucity of information about their potential impact on food safety. Studies have shown different bacteriophages to be effective against Listeria monocytogenes (Lm) in a food processing environment, but not much is yet known about their potential efficacy against resistant strains. Therefore, this study was conducted to evaluate the efficacy of bacteriophage application for controlling resistant Lm strains in diverse processing environments.

Lm strains were isolated from various food processing environments and the resistance phenotypes were determined using classical methods, and minimum inhibitory concentrations (MICs) were determined using plate dilution. Lm strains were then selected for susceptibility to their respective bacteriophage cocktail (CFU/mL) and challenged with a 6-log10 challenge. Bacteriophage application significantly (P<0.001) reduced the CFU/mL of Lm in all environmental samples, with the log10 reduction ranging from 1.7 to 4.3 log10.

The results of this study showed that bacteriophage application can be an effective method for controlling resistant Lm strains in diverse processing environments. This suggests that bacteriophage application can be a promising approach for controlling resistant Lm strains in food processing environments.
**P2-29**  
Antimicrobial and Physical Properties of Chitosan/Acetylated Starch Edible Films Containing Cinamon and Clove Essential Oils  
Kai Wen Choo, Wei Wang, and Mustapha  
University of Missouri, Columbia, MO  
*Introduction:* Edible food packaging can slow down or prevent the degradation of foods due to environmental factors. Novel approaches to improving the safety and quality of food products are very important to the food industry and public health. Clove and cinnamon essential oils (EOs) have been used for their antimicrobial and antioxidant properties. In this study, we explored the use of chitosan/acletylated starch edible films containing EOs.

*Purpose:* The objective of this study was to evaluate the effect of concentration of cinnamon and clove essential oils on the antimicrobial activities of edible packaging films.

*Methods:* The edible chitosan/clohectyl starch films were prepared in a one-to-one ratio with different concentrations of cinnamon and clove cinnamon and edible films in a one to three ratio using a solution casting method. The antimicrobial properties were examined by exposing a solution to different concentrations of bacterial (Salmonella and Escherichia coli) and fungal (Aspergillus niger and Rhizopus oryzae) pathogens. The films were also tested for their physical properties, including thickness, tensile strength, elongation at break, water vapor permeability, and oxygen permeability.

*Results:* Cinnamon oil had a more significant effect on the antimicrobial properties of the edible films compared to clove oil. The films containing cinnamon oil showed higher antibacterial and antifungal activities than those containing clove oil. The films also exhibited good physical properties, such as good tensile strength and water vapor permeability.

*Significance:* The results of this study suggest that edible films containing cinnamon oil could be used as a natural antimicrobial packaging material for food products. Further studies are needed to evaluate the long-term stability and shelf life of these films in real food systems.
P2-36 Efficacy of Cinnamon Oil Nanoemulsion in Inhibiting Salmoella spp. and Listeria spp. on Mung Bean Sprouts

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Introduction: Mung bean sprouts have been associated with deadly outbreaks of foodborne pathogens including Salmoella and Listeria. Cinnamon oil has antimicrobial activity as food treatment due to its hydrophobic nature. In previous research, cinnamon oil made through sonication with surfactant showed a stable homogeneous solution that may provide an effective treatment for mung bean sprouts.

Methods: The purpose of this study was to test the efficacy of cinnamon oil nanoemulsions as an antimicrobial treatment for mung bean sprouts against Salmoella enterica and Listeria monocytogenes.

Results: A five percent cinnamon oil nanoemulsion (CONE) was prepared by 20-min sonication of sodium 70% concentration of cinnamon oil with the surfactant Tween 20 and DI water. Samples for treatment were generated on site using dry precursors by mixing sodium chlorite and activating acid. Treatments were conducted at various concentrations (0.3, 0.4, 0.5, and 0.6 log CFU/cm²) respectively, for water, LA and mLA sprays (average of reductions from top to bottom). The 18-h water spray chill cycle provided additional STEC reductions of 0.2, 0.5 and 0.6 log CFU/cm², respectively, for water, LA and mLA sprays (average of reductions from top to bottom). The 18-h water spray chill cycle provided additional STEC reductions of 0.2, 0.5 and 0.6 log CFU/cm², respectively, for water, LA and mLA sprays (average of reductions from top to bottom). The 18-h water spray chill cycle provided additional STEC reductions of 0.2, 0.5 and 0.6 log CFU/cm², respectively, for water, LA and mLA sprays (average of reductions from top to bottom). The 18-h water spray chill cycle provided additional STEC reductions of 0.2, 0.5 and 0.6 log CFU/cm², respectively, for water, LA and mLA sprays (average of reductions from top to bottom).

Conclusion: These results indicate that nanoemulsions of cinnamon oil may be effective as an antimicrobial treatment for mung bean sprouts.

P2-37 Evaluation of Cranberry Antimicrobial Properties by TLC-Bioautography

Chayapa Techathuvanan, Yu-Ting Hung, Christopher McNamara and Margarita Gomez
Ocean Spray Cranberries, Inc., Lakeville-Middleboro, MA

Purpose: The purpose of this study was to identify potential antimicrobial peptide- and lactase-producing lactic acid bacteria (LAB) in farmer animals and produce.

Methods: Samples were screened and cultured for antimicrobial activity on indicator organisms, Listeria monocytogenes and Enterococcus faecium (ECF), on the MBR agar. LAB for producing bioactive secondary metabolites were identified using a sandwich overlay technique supplemented with defined antagonist overlay method. Lactase-producing and sheep blood hemolytic isolates were determined using a modified disk-diffusion technique on agar plates containing X-gal (20 µg/ml) and sheep blood (five percent), respectively. Subsequent 16S rDNA identification of LAB isolates was carried out with an ABI 3730 sequencer.

Results: A total of 161 LAB were isolated from 156 samples of farm animals (102) or fresh produce (59) exhibiting a total number of 161 LAB with confirmed activities against E. coli (34), L. monocytogenes (44), E. faecium (45), and with no activity (82). This suggests that the majority of LAB are antagonistic against E. coli and L. monocytogenes.

Conclusion: These results demonstrate the value of TLC screening as a rapid test method for LAB and identify new LAB with antimicrobial and lactase activity.

P2-38 Long-term Survival Phase Cells of Listeria monocytogenes Exhihit Increased Tolerance to Cinnae-maldehyde in 0.85% Saline and Apple Juice

Samuel Kripoch
Iowa State University, Ames, IA

Introduction: Listeria monocytogenes is an environmental contaminant and can remain viable for months or years in the long-term survival (LTS) phase with increased tolerance to different antimicrobial treatments such as heat, high pressure and UV radiation compared to stationary phase cells (STAT).

Methods: The objective of this study was to compare the tolerance of LTS and STAT cells of L. monocytogenes to cinnae-maldehyde, an essential oil component of cinnamon oil (0.1% and 0.5%).

Results: Cells of L. monocytogenes Scott A was used to prepare STAT cells by sonicating cells in TSBE at 35°C for 120s. LTS cells were prepared by inoculating TSBE (10°C) with 10 mL of 10% NaCl. LTS and STAT cells of L. monocytogenes (0.1% and 0.5% cinnamon oil) were exposed to saline and apple juice for 45 minutes. The bacteria was then challenged with cinnae-maldehyde (0.1% and 0.5% cinnamon oil) for 15 minutes for comparison.

Conclusion: These results demonstrate the value of TLC screening as a rapid test method for LAB and identify new LAB with antimicrobial and lactase activity.


P2-42
The Use of BxCl2 spp. Isolated from Ready-to-Each Date Fruits to Control Listeria monocytogenes

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Developing Scientist Award

Introduction: Listeria monocytogenes, an important foodborne pathogen, remains a significant threat to public health as the invasive form of infection can result in high case-fatality rates.

Purpose: To confirm the ability of BxCl2 spp. using cultivable members of the RTE date fruit microbiome.

Methods: RTE date fruits were acquired from five geographic regions: Iran, China, Palestine, Saudi Arabia, and Tunisia. Bacterial isolates were collected by washing the surface of dates with peptone water, then plating the wash on blood agar. Isolated strains were then individually assessed to monitor whether they can prevent growth of L. monocytogenes using an agar plate test (n=15). Following, bacterial strains that secreted antibiotics were then identified using 16S rRNA Sanger sequencing.

Results: A total of 191 isolates belonging to 91 different phenotypes were observed. From this collection, 35 isolates belonging to 21 phenotypes produced a zone of inhibition against L. monocytogenes. Zone sizes ranged from 0.5 to 5.7 mm on the agar plate. Sequencing revealed that the inhibitory strains belonged to the genus Bacillus, with different species. Further work was done to identify these Bacillus spp. which had no link to clinical isolates. Those Bacillus spp. that were found to be safe and that produced the largest inhibition zones are being further characterized by whole genome sequencing and probing the genome for potential inhibitors.

Significance: The results from this research could lead to the discovery of novel antimicrobial metabolites or beneficial Bacillus spp. that could be added to foods to inactivate and/or control L. monocytogenes. These novel compounds can also be assessed for their potential antimicrobial activity against other foodborne pathogens and could eventually lead to novel probiotics and/or bio-complexes that can help to reduce foodborne illness.

P2-43
Sanitizer Susceptibility of Recurrent and Sporadic Listeria monocytogenes from Meat Processing Environments When Grown in Planktonic and Biofilm States

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Introduction: Listeria monocytogenes can persist for prolonged times in food processing facilities. Causes behind persistence are multifaceted, often attributed to inadequate cleaning and sanitation, formation of biofilms, increased resistance to sanitizers, variability in biofilm-forming capacity and environment to environment stressors.

Purpose: Investigate sanitizer susceptibility of recurrent and sporadic L. monocytogenes from a meat processing facility (MPF) when exposed to sanitizers used in the facility.

Methods: Four L. monocytogenes strains from MPF (recurrent, R; RT and sporadic, SP1, SP2) and ATCC 19115 were grown in a liquid suspension (tryptic soy broth, TSB; 35°C, 24 h) or attached (TSB, 23°C, four days) to assess their susceptibility to two commercial sanitizers (quaternary ammonium compound (QAC, E-San; 50 to 5,000 ppm), and hydrogen peroxide and acetic acid (HPAA, Perox-E; 70 to 19,200 ppm)). Recurrent and sporadic strains were isolated at least three times and once, respectively, over 20 months, and fingerprinted with PFGE. Biofilms were grown on stainless steel coupons (12 mm) and plastic surfaces (MBEC device).

Results: All strains exhibited similar susceptibility to tested sanitizers, with higher concentrations required to inactive biofilms compared to planktonic concentrations. Lower concentrations than the manufacturer recommended concentrations (MRC; QAC; 200 ppm; HPAA; 1,100 ppm) effectively inactivated planktonic QAC. HPAA did not motivate inactivation of stainless steel and MEC with concentrations three times and 25 times higher than the MRC, respectively. No viable cells were observed on stainless steel when exposed to HPAA concentrations 3 times lower than the MRC but required four times higher concentration than the MRC to inactivate four-day-old biofilms on MEC.

Significance: This study highlights the resilient nature of L. monocytogenes, and factors that can influence sanitation efficacy (e.g., sanitizer type, different surface biofilm formation). Additional study of genetic properties of L. monocytogenes may be warranted to gain insight into their inherent or acquired traits that may be contributing to the adaptation and persistence in food facilities.

P2-44
The Use of Flow Cytometry for the Rapid Detection of Fluorescent-tagged Salmonella spp. in Food and Environmental Samples

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Eumicrobiology Laboratory, Madison, WI

Introduction: Green fluorescent protein (GFP) tagged Salmonella strains are often utilized as positive controls for their distinguishability during a cultural confirmation by observing morphological fluorescence on an agar plate using UV light post incubation. Flow cytometry has the ability to distinguish GFP-tagged Salmonella without the need for additional labeling or manipulation.

Purpose: The purpose of this study was to demonstrate the ability of flow cytometry to distinguish a GFP-tagged Salmonella strain from other Salmonella strains.

Methods: GFP-tagged Salmonella Typhimurium SA54 was encapsulated in buffered peptone water, then transferred and grown up in RIV broth. An additional five wildtype Salmonella cultures were grown in a similar fashion. The cultures were then diluted and run on a flow cytometer. The instrument setting were adjusted to maximize the pulse height of the fluorescent emission through the BL1 530/30 band pass filter when excited by a laser with a wavelength of 488 nm.

Results: The BL1 Histogram plot displaying the cellular events produced by the GFP-tagged Salmonella show a distinct cellular population emitting a stronger voltage pulse height due to the GFP excitation from the laser. Voltage intensity between 10' and 100' was observed in the GFP-tagged Salmonella. Wildtype Salmonella strains did not display the same cellular population and were easily distinguished from the GFP-tagged strain. Significant decrease in the size of the GFP-tagged Salmonella Control strain through flow cytometry allows for result confirmation in real-time. This real-time disclosure does not need the grow cultures overnight to observe fluorescence reducing the time to result.

Journal of Food Protection Supplement

Poster 178

Poster 179

Journal of Food Protection Supplement
**P2-48 Comparison of the Antimicrobial Activities of Ohelo Berry (Vaccinium reticulatum) and Cranberry (Vaccinium macrocarpon)**

Xiaohao Liu, Stuart Nakamoto and Yong Li
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**Developing Scientist Entrant**

**Introduction:** Ohelo berry, a wild relative of cranberry, is an endemic plant in Hawaii. Our preliminary study suggested ohelo berry extract has strong antimicrobial activity, which might be attributed to various bioactive compounds it contains.

**Purpose:** The purpose of this study is to determine and compare the phenolic contents and antimicrobial activities of ohelo berry and cranberry.

**Methods:** The concentrations of anthocyanins and phenolics in the crude extracts of ohelo berry and cranberry were determined by the Folin-Ciocalteau method and the pH differential method, respectively. Both extracts were evaluated against four pathogenic and two probiotic bacteria via the agar well diffusion assay. Moreover, the minimum inhibitory concentration (MIC) and the minimum bacterial concentration (MBC) of the extracts were determined against these bacteria. Fungi were screened at their native pH and neutral pH.

**Results:** Ohelo berry extract had significantly higher total phenolic content (21.15 vs 6.81 gallic acid equivalent mg/ml) and lower anthocyanins (477.42 vs 996.59 cyanidin-3-glucoside equivalent mg/l) than cranberry extract. In the agar well diffusion assay, the inhibition zones generated by ohelo berry and cranberry extracts against the same bacteria were not significantly different. However, antimicrobial library screens were constructed for their inhibitory activity against S. monocytogenes and C. sakazakii by using the growth inhibition test.

**Significance:** Both phenolics and organic acids contribute to the antimicrobial properties of ohelo berry. They have the potential to be used as natural antimicrobial agents in the food industry.

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**P2-50 Effects of Sodium Lactate on the Growth of Bacillus cereus in a Rice-based Model Food**

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**Introduction:** Sweet and salty rice-based foods are popular worldwide, and the main pathogenic microorganism of concern for these products is Bacillus cereus. Its spores can survive cooking temperatures and grow at temperatures commonly used for storage of rice-based products.

**Purpose:** The objective of this study was to determine the effect of sodium lactate as an antimicrobial on the growth of B. cereus in rice-based food products.

**Methods:** Starch mixtures of rice flour and water (45%), two percent NaCl, and one, two, and three percent sodium lactate were inoculated with 8. coccus spores to 101 CFU/g. Samples were stored at 4°C for 15 min to cook the starch and activate the spores. Samples were stored at 16, 22, and 30°C, and the populations of B. cereus during storage were determined. The growth curves were used to estimate the lag phase duration (LPD, growth rate (GR), and maximum population density (MPD).

**Results:** In samples containing one to three percent lactate added at 16, 22, and 30°C, the LPD of B. cereus were 10 to 25, 10 to 15, and two to four h, respectively, and the GR were 0.11 to 0.10, 0.20 to 0.13, and 0.57 to 0.30 log CFU/h, respectively. In samples containing three percent lactate, the MPD of B. cereus were 5.0 log CFU/g at 16 and 22°C, respectively, whereas the MPD reached greater than six log CFU/g in other samples. Results show that increased of lactate concentration significantly extended the LPD of B. cereus at 16 and 22°C and reduced the GR at 22 and 30°C, and percent lactate reduced the MPD at 22 and 16°C.

**Significance:** This study identified the levels of sodium lactate that have significant effect in reducing the growth of B. cereus in rice-based food products. The information could help the producers to improve product safety by using lactate levels that are applicable to their products’ storage temperatures.
APPLICATION OF FIVE PERCENT NOVEL LACTIC ACID TREATMENT OR 10% LACTIC ACID ANTIMICROBIAL SOLUTIONS DOES NOT IMPACT THE GROWTH KINETICS OF LAB IN LEAN AND FAT BEEF. TREATMENTS ARE NOT CONFERING ANY RESIDUAL EFFICACY AND ARE SIMILAR TO USDA-APPROVED PROCESSING TREATMENTS AND WATER.


METHODS:
1. A combination of sodium dodecyl sulphate (SDS) (concentration: 200, 400 and 600 ppm), sodium hypochlorite (NaClO) (concentration: 100, 150, 200, 300, 400 and 500 ppm), temperature (25°C and 35°C) and pressure (500 and 600 MPa) were used to decontaminate E. coli O157:H7 ATCC 43888 and L. monocytogenes ATCC 7644 from stainless-steel coupons. Using a Box-Behnken experimental design, predictive quadratic equations were developed to describe the relationship between the independent factors (pressure, temperature and NaClO concentration) and the responses (log reductions of E. coli O157:H7 and L. monocytogenes ATCC 7644) (P<0.05, P<0.001) and verified using 42 randomly selected treatment conditions.

RESULTS:
1. Among factors (temperature, time, and concentration of SDS, NaClO, or resin), temperature had higher significance for inactivation of both pathogens. Temperature (P<0.001) were observed in biofilm quantity treated and untreated coupons. The optimum temperature conditions were 37°C with 200 ppm NaClO, 400 ppm SDS, and 10000 KI rinsin for 30 min for E. coli O157:H7 ATCC 43888 and 25°C with 200 ppm NaClO, 150 ppm SDS, and 5500 KI rinsin for 30 min for L. monocytogenes ATCC 7644. Scanning electron microscopy was used to confirm membrane disruption in the treated microbial cells in each optimal condition. The combined treatment of SDS and NaClO contributed to the effective inactivation (more than four-log reduction of both E. coli O157:H7 and L. monocytogenes on stainless-steel coupons).

SIGNIFICANCE:
1. This study demonstrated that E. coli O157:H7 is a safe and beneficial strain and can be used in commercial probiotic trials and potent antimicrobial activity. These findings support the feasibility of using this strain in food preservation and in promoting human health.

INTERACTIONS OF CARVACROL, CAPRYLIC ACID, HABITUATION, AND MILD HEAT FOR PRESSURE-BASED INACTIVATION OF O157 AND NON-O157 SEROGROUPS OF SHIGA TOXIN-PRODUCING ESCHERICHIA COLI IN LOW-OXYGEN ENVIROMENTS

METHODS:
1. Current study investigated synergistic elevated hydrostatic pressure, habituation, mild heat, and antimicrobials for inactivation of O157 and non-O157 serogroups of Shiga toxin-producing Escherichia coli (STEC). The corresponding log reductions after seven-min aerobic habituation were 2.64, and 3.31, respectively. Carvacrol and caprylic acid both augmented the decontamination efficacy of the treated samples. An example, Escherichia coli O157 was reduced (P<0.05) by 1.64 and 4.17 log CFU/ml after a seven-min treatment at 450 MPa without, and with presence of carvacrol, respectively.

RESULTS:
1. Current study indicate an optimized pressure-based intervention in presence of mild heat and antimicrobial agents could be efficacious for the inactivation of >99.9% of microbial pathogens. Current experiment also exhibits the critical role of habituation on increasing the external validity of a microbial challenge study.

SYNERGISM OF MILD HEAT, NISIN, AND ELEVATED HYDROSTATIC PRESSURE FOR INACTIVATION OF LISTERIA MONOCYTOGENES

RESULTS:
1. Under the conditions of this experiment, habituation of the pathogen played an influential (P<0.05) role in inactivation rate. As an example, O157:K5:NM was reduced (P<0.05) by 2.64 and 4.17 log CFU/ml after a seven-min treatment at 460 MPa without, and with pressure of 460 MPa.

SIGNIFICANCE:
1. Current study indicate an optimized pressure-based intervention in presence of mild heat and antimicrobial agents could be efficacious for the inactivation of >99.9% of microbial pathogens. Current experiment also exhibits the critical role of habituation on increasing the external validity of a microbial challenge study.

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INTRODUCTION:
1. Pathogenic E. coli O157:H7 and non-O157 serogroups of Shiga toxin-producing Escherichia coli (STEC) which belongs to class I antibiotics.

RESULTS:
1. The corresponding log reductions after seven-min aerobic habituation were 2.64, and 3.31, respectively. Carvacrol and caprylic acid both augmented the decontamination efficacy of the treated samples. An example, Escherichia coli O157 was reduced (P<0.05) by 1.64 and 4.17 log CFU/ml after a seven-min treatment at 450 MPa without, and with presence of carvacrol, respectively.

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POSTER

P2-54 - P2-56

APPLICATION OF FIVE PERCENT LACTIC ACID PLUS SURFACTANTS AND TEN PERCENT LACTIC ACID ANTIMICROBIAL INTERVENTIONS FOR SPOLIAGE MICROORGANISM GROWTH AND SURVIVAL ON BEEF TISSUES

METHODS:
1. The pathogen E. coli O157:H7 and non-O157 serogroups of Shiga toxin-producing Escherichia coli (STEC) which belongs to class I antibiotics.

RESULTS:
1. The corresponding log reductions after seven-min aerobic habituation were 2.64, and 3.31, respectively. Carvacrol and caprylic acid both augmented the decontamination efficacy of the treated samples. An example, Escherichia coli O157 was reduced (P<0.05) by 1.64 and 4.17 log CFU/ml after a seven-min treatment at 450 MPa without, and with presence of carvacrol, respectively.

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RESULTS:
1. Under the conditions of this experiment, habituation of the pathogen played an influential (P<0.05) role in inactivation rate. As an example, O157:K5:NM was reduced (P<0.05) by 2.64 and 4.17 log CFU/ml after a seven-min treatment at 460 MPa without, and with pressure of 460 MPa.
The quality characteristics of eggs, such as egg yolk and albumen index, egg density, and sensory qualities were also determined along with moving speed at 600 or 900 eggs/hr, 30 standard liters per minute (slm) flow rate, and four cm distance between egg and plasma jet were tested.

Results: Irradiances of 1.3 and 1.5 mJ/cm² in four tests were sufficient to reduce Staphylococcus aureus CFU on exposed targets. This corresponded to doses of between 20 and 52 mJ/cm². Increasing the irradiance to 2.2, 3, and 3.0 mJ/cm² to deliver a dose of 150 mJ/cm² resulted in a five-log reduction in all cases.

Results: Short 265 nm UV-C exposure of 60 s was sufficient to deliver a four-log reduction in Staphylococcus aureus. Treatment of food products by 265 nm UV-C LEDs represents a viable investigation path for decreasing foodborne Staphylococcus aureus infections in consumers.

Introduction: cross-contamination of food processing and contact surfaces is a source for foodborne infection for consumers of meat cooked meats are considered as suitable environments for C. perfringens. Therefore, the reheating step is a critical step for ensuring the safety of foods. sous vide cooking generally are not adequate to destroy the pathogen.

Methods: One-hour static oven-stored stainless steel inoculated with S. aureus was exposed to a UV-C LED (265 nm) array light source (Phoseon Technology) at 1.3, 1.5, 2, 2.5, and 3.0 mJ/cm² (at the target) from a distance of 15 mm. Doses ranged from 26 mJ/cm² through 150 mJ/cm². Surviving bacteria were plated and colony forming units (CFU) assessed. Log reduction was calculated as the difference in the log of geometric means between the unexposed control and the exposed sample at each exposure time. Each test sample included four independent exposures at each condition.

Results: Irradiances of 1.3 and 1.5 mJ/cm² in five tests were sufficient to reduce Staphylococcus aureus CFU on exposed targets. This corresponded to doses of between 20 and 52 mJ/cm². Increasing the irradiance to 2.2, 3, and 3.0 mJ/cm² to deliver a dose of 150 mJ/cm² resulted in a five-log reduction in all cases.

Results: The data, from the microscope, tomato, and spinach study indicates that there was a significant (P<0.05) reduction in the amount of EHEC population in spinach by the end of six hours; however, there was bacterial recovery by the end of 12 hours leading to an overall 2.5-log reduction from the initial inoculum. These results demonstrate that a phage cocktail could potentially act as an antimicrobial to inactivate EHEC and reduce their incidence in produce.

Significance: The results from the studies indicate that the phage cocktail can be commercially used to reduce or eliminate EHEC infection in produce.
serotype.
Sero types of biofilms were found in microtiter plates (24 h, 37°C) and on stainless-steel and high-density polyethylene (HDPE) coupons (eight h, 25°C). Phenotypes of the phage control and inoculated Salmonella Typhimurium (E. coli) were measured in vitro biofilm film disruption (zero, three, and six h). StEC biofilm was measured at a zero, three, and six h by plating on CHROM agar. Data were analyzed using one-way ANOVA (P<0.05).
Results: As individual in Vitro treatments, phages effectively disrupted biofilms, reducing absorbance from 2.62 mm (zero h) to 1.179 mm (eight h), and 5.37 mm (six h). The O157-specific phages were effective at six h, while the non-O157-specific phages were at three h. Phage cocktails specific to O111, O172, and O157 were effective at six h, while the rest were more effective at six h. The phage cocktail reduced absorbance at three h by 1.45, 0.32, and 1.50 mm, respectively, with the highest relative kill the most effective. The 21- phage cocktail was only effective at 16 h (stainless steel, 1.41 and HDPE, 2.48 reduction).
Discussion: Biofilms can be attacked individually or as a species-specific cocktail more efficiently as biocontrol in the food industry.

P2-68 Reducing of Aeromonas hydrophila Contamination on Lettuce by Using a Novel Aeromonas hydrophila-specific Phage

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Developing Scientific Entrant

Introduction: Aeromonas hydrophila has been found in a wide range of fresh and processed foods. Due to the resistance of A. hydrophila to antibiotics, a bacteriophage may be an alternative method to control A. hydrophila contamination on lettuce.

Purpose: The purpose of this study was to identify the phage causing the reduction of A. hydrophila infection on lettuce.

Methods: A. hydrophila strain was isolated from lettuce and further identified on Slanetz and Bartley selective medium. Total genomic DNA was isolated from exponential phase culture grown in Luria Broth (LB) for 18 h at 37°C. Genomic DNA (20μg) was digested with restriction enzymes and electrophoresed on an agarose gel. The inserts were ligated into the pGEMT-Easy vector, transformed into E. coli DH5α to examine the presence of inserts. The positive clone was screened using specific primers of A. hydrophila. Plaque forming unit (PFU) determination was done by plaque assay.

Results: A. hydrophila treated with KFS-G9 was reduced from initial number of log (CFU/mL) to 2.910 log (CFU/mL) at 30 min and its reduction was sustained up to eight h. However, the number of A. hydrophila on the lettuce treated with PBS increased to 7.9 log CFU/cm². which was significantly larger than the initial number of A. hydrophila (P<0.05). Although sodium hypochlorite treatment showed bacterial reduction (3.190) at 30 min only, the number of A. hydrophila increased significantly after (P<0.05) and the final number of A. hydrophila was 7.760 log (CFU/mL).

Discussion: This study demonstrated the beneficial effect of KFS-G9 and its potential as a new biocontrol agent.

P2-69 Characterization of a Novel Bacteriophage, EscoHU1, Infecting Both Escherichia coli O157:H7 and Salmonella

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Developing Scientific Entrant

Introduction: Bacteriophages are bacterial viruses and promising agents that can be used to control A. hydrophila and E. coli in the food industry.

Methods: A. hydrophila (106 CFU/ml) was inoculated on the surface of UV-treated lettuce (two by two cm²) and placed into a biosafety cabinet for its attachment during one h at 22°C. The lettuce was then inoculated with A. hydrophila and placed into a water bath at 4°C for 20 min, 20 min, and 100±22 PFU/infected cell, respectively. EscoHU1 was stable for heating at 50°C and incubation at pH 5.0 to 9.0. After EscoHU1 treatment, A. hydrophila was still reduced by two logs at 24 h. The phages also reduced the 21-phage cocktail was only effective at 16 h (stainless steel, 4.1 and HDPE, 4.8-log reduction). The O157-specific phages were more effective at six h, while the non-O157-specific phages were at three h. Phage cocktails specific to O111, O157:H7 and A. hydrophila were reduced by 88±39 PFU/infected cell, while EscoHU1 was stable for heating at 50°C and incubation at pH 5.0 to 9.0. After EscoHU1 treatment, A. hydrophila was still reduced by two logs at 24 h. The phages also reduced the O111- and O157-specific phages were reduced by 88±39 PFU/infected cell, while EscoHU1 was stable for heating at 50°C and incubation at pH 5.0 to 9.0. After EscoHU1 treatment, A. hydrophila was still reduced by two logs at 24 h. The phages also reduced the O157-specific phages were reduced by 88±39 PFU/infected cell, while EscoHU1 was stable for heating at 50°C and incubation at pH 5.0 to 9.0. After EscoHU1 treatment, A. hydrophila was still reduced by two logs at 24 h. The phages also reduced the phages specific to O157:H7 were 10 min, 20 min, and 88±39 PFU/infected cell, respectively, and those against A. hydrophila were 10 min, 20 min, and 100±22 PFU/infected cell, respectively, and those against

P2-70 Characterization of Selected β-Lactam-resistant Escherichia coli isolates from Food Products

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Introducing: Resistance to β-lactam antibiotics is a serious public health threat. Since β-lactam antimicrobials are essential in human and veterinary medicine, the frequent occurrence of β-lactam-resistant E. coli isolates from foods, human and food-producing animals has been a major concern.

Purpose: To characterize β-lactam-resistant E. coli isolates from food products to better understand β-lactam-resistant E. coli and AR in foods.

Methods: 150 β-lactam-resistant E. coli isolates from five sample types were enriched and analyzed following the Oxford disk diffusion method for the presence of β-lactamase. β-lactam resistance was determined by the 6-log reduction of S. enterica strain and selective medium . Isolates were also tested for susceptibility to 18 antimicrobials using the Vitek2 System. Resistance to ≥3 antimicrobials were regarded as multidrug resistant. Chi-square test was done to evaluate the significance of difference.

Results: The prevalence of β-lactam-resistant E. coli isolates was high in all sample types. The E. coli isolates from Retail meats and Retail meat products were statistically more resistant to AR compared to Retail vegetables, Retail ice cream, and Retail milk products (P<0.05). The β-lactam resistance of the isolates was screened for the presence of AR genes by PCR followed by sequencing. Three selected isolates were identified by 16S rRNA gene sequence analysis and the expression of β-lactamase from the medium from the Colilert system. After AR genes, the susceptibility profiles of the isolates was examined by the disk diffusion method for β-lactam and non-β-lactam agents. Sequencing of the β-lactam resistant genes was examined by conjugation using E. coli DH5α as the recipient strain. The selected isolates were resistant to AR carbapenems, cephalosporins, colistin, and sulfonamide, and were not susceptible to cefazolin and cefotaxim. One isolate from a ground chicken sample carrying both βlactamases and β-lactamase genes was also resistant to gentamicin. One isolate from the same sample carrying blacmac (gene also carried aad8) and in a class 1 integron and sul gene, was also resistant to gentamicin, chloramphenicol, tetracycline, and streptomycin. One isolate from a raw beef sample carrying blacmac, blasa, and blacma was also resistant to tetracycline, clindamycin and streptomycin. The βlactamase gene from this isolate could be transferred to E. coli DH5α through conjugation.

Significance: The above results indicate that the tested phage solution can reduce Salmonella contamination on pork by 1.3 to 1.7 log. This shows that bacteriophages are an effective Salmonella intervention procedure for processors to reduce risks and allow an increase in consumer safety.

P2-71 Efficiency of a Phage Intervention against Salmonella on Lean Pork, Pork Trim and Bacon

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Introducing: An ongoing baseline sampling program by the Food Safety Inspection Service (FSIS), the Raw Pork Products Exploratory Sampling Program (2015), will provide direction to FISL to develop a better risk profile and refine the present food safety guidelines for pork products. Thus, new impending regulatory standards will likely prompt establishments to seek interventions that can help in reducing the most probable number of Salmonella in their products.

Purpose: To determine the efficacy of a commercially available bacteriophage product, PhageGuard S, against Salmonella on several types of pork meat.

Methods: Salmonella Typhimurium (ATCC 14028) was used. Experiment (1) was performed with 32 g of raw pork meat (containing ≤2 mm in size of the S. Typhimurium strain) were inoculated onto the meat in duplicate (4, 12 cm²) and homogenized (10 min, 3000 rpm). After homogenization, 10 ml of the homogenate was inoculated on the surface of UV-treated lettuce (two by two cm²) and placed into a biosafety cabinet for its attachment during one h at 22°C. The same amount of 10 ml of phage concentrated on UV-treated lettuce was inoculated on the meat at 22°C. Sodium hypochlorite solution and PBS were used as a positive and negative control, respectively. At every two-h interval, treated lettuce was then placed in a stomacher bag containing 15 ml of PBS for homogenization. Each homogenate was serially diluted for enumeration of A. hydrophila using an Aerobactin selective medium.

Results: The 21-phage cocktail was only effective at 16 h (stainless steel, 4.1 and HDPE, 4.8-log reduction). The O157-specific phages were more effective at six h, while the non-O157-specific phages were at three h. Phage cocktails specific to O111, O172, and O157 were effective at six h, while the rest were more effective at six h. The phage cocktail reduced absorbance at three h by 1.45, 0.32, and 1.50 mm, respectively, with the highest relative kill the most effective. The 21- phage cocktail was only effective at 16 h (stainless steel, 1.41 and HDPE, 2.48 reduction).

Discussion: Biofilms can be attacked individually or as a species-specific cocktail more efficiently as biocontrol in the food industry.
P2-74 The Antimicrobial Activities of Beef Fatty Acids and Their Effects on Virulence Gene Expression in Listeria monocytogenes and Salmonella Typhimurium

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Developing Scientist

Introduction: Listeria monocytogenes and Salmonella Typhimurium are significant food-borne pathogens. Bacterial pathogenesis is attributed to the regulation and expression of virulence genes. It has been reported that medium- and long-chain fatty acids (MFA) may inhibit bacterial growth and decrease the associated biofilm formation.

Purpose: This study aimed to evaluate the antimicrobial activities and virulence attenuation potential of beef fatty acids on L. monocytogenes and Salmonella Typhimurium.

Methods: Free FAs used in this study included commercial FAs (C14:0, C16:0, C16:1, C18:0, C18:1, conjugated C18:2, C18:3), total beef FAs, and fractions of beef FAs that were rich in monounsaturated fatty acids (MUFA) or polyunsaturated fatty acids (PUFA). Minimum inhibitory concentration (MIC) of the FAs on L. monocytogenes and Salmonella Typhimurium was determined at 10 and 75 µM. MIC values were determined for the same Fa in 6 samples from each of the 3 farms at the point of harvesting and at the point of sale (n=25).

Results: L. monocytogenes was more sensitive to FA treatment compared with Salmonella Typhimurium. Glycine, FAs showed higher inhibitory activities against L. monocytogenes at pH 7 than pH 5.5. Specifically, MICs of C18:3, C18:2 and C18:1 at pH 7 were 0.25, 0.04 and 0.38 mg/mL, while those at pH 5.5 were 0.28, greater than six and greater than six mg/mL, respectively. Beef FA fractions, with MICs between 0.06 and 0.33 mg/mL, showed stronger antimicrobial activities compared with commercial FAs. Although fatty acids did not affect the virulence gene expression in L. monocytogenes, virulence gene expression in Salmonella Typhimurium was determined.

Significance: Beef FA fractions in monounsaturated or polyunsaturated could be potentially used as natural preservatives against the treated luxuos in the food industry.

P2-75 Antimicrobial Resistance in Surface Water of Two Rivers with Agricultural Use in Chile

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Introduction: Antimicrobial resistance (AMR) is a public health concern with relevance in the food supply as a source of AMR. For producer practice, production water is increasing its relevance, with special significance of agricultural water as a vehicle to disseminate pathogens and AMR. Central Chile is an agricultural location in which fresh produce is produced for local consumption and for international trade. Two rivers, Maipo and Maule, contribute 70% of the irrigation water to the region.

Purpose: The purpose of this study was to identify the presence of antimicrobial resistant bacteria in the Maipo and Maule rivers in central Chile.

Methods: Among the 48 samples collected every three months between June 2017 and August 2017, samples were transferred to natural aquatic, urban river and livestock forest area. A total of 201.6 liters of water were ultrasonicated into 200 mL. Then, one ml of each sample was frozen in five ml of buffered peptone water and incubated at 37°C for 24 h. 100 ul of the samples were streaked onto plates of MacConkey supplemented with ciprofloxacin (one mg/mL), cefoxitin (one mg/mL), and tetacycline (one mg/mL) and incubated at 37°C for 24 h.

Results: Among the 48 samples collected during the four samplings, 28 (58.3%) of samples contained gram-negative bacteria that were resistant to at least one of the antibiotics tested. A total of 52, 42, and 25% of the samples contained bacteria resistant to cefotaxime, tetracycline and ciprofloxacin, respectively. Gram-negative antibiotic resistance prevalence in six samples ranged from 1.5% to 48.2%. Only one out of the 12 sites tested was negative for antibiotic resistant gram-negative bacteria for all four antibiotics.

Significance: Presence of antibiotic resistant bacteria in water indicates a contaminant of water quality that may impact food safety through foodborne antimicrobial resistance.

P2-76 Prevalence of Extended Spectrum β-Lactamase Encoding Genes: A South African Cucumber Agroecosystem Case Study

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Developing Scientist

Introduction: The emergence of extended-spectrum β-lactamase (ESBL) producing bacteria in different environmental compartments has been recognised as a serious threat to human health. This is due to the increase in the number and variety of resistance genes as well as potential horizontal transmission to more pathogenic bacteria such as plasmids.

Purpose: In this study, the prevalence of relevant ESBL producing genes in Enterobacteriaceae and Pseudomonas isolates from the cucumber agroecosystem was determined.

Methods: A total of 24 samples, including 12 water samples (one liter each), and 12 cucumbers (150 g each), were collected and processed. Following enrichment and streaking onto chromogenic media, 45 presumptive ESBL-producing Enterobacteriaceae and Pseudomonas spp. isolates were taken and their identities were confirmed using MALDI-TOF MS. Plasmid and chromosomal DNA was extracted and analysed using PCR targeting seven ESBL genes: blaTEM, blaSHV, blaCPM, blaOXY, blaKPC, blaVIM and blaDIN.

Results: The PCR reactions were conducted as previously described in the literature. Presumptive-positive bands were confirmed using the BLAST tool to find the further significance of these genes across different environmental compartments.

Significance: This study provides valuable data about the prevalence of ESBL genes in the cucumber agroecosystem in South Africa and the need for the development of strategies to contain the further spread of these genes across different environmental compartments.

P2-77 Microbial Safety Status of Rape Produced and Sold from Small Scale Farming in South Africa

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Developing Scientist

Introduction: Brassica rapa (rape) is a favourite African leafy green vegetable often used in traditional meals either raw or cooked. Rape is locally produced by small-scale farmers, and sold informally as well as at formal markets.

Purpose: Due to the lack of a robust system to monitor and to assess the microbial safety status of rape, the research team investigated the microbial safety status of rape sold in 25 markets in South Africa.

Methods: A total of 24 samples (20 rape samples and 4 from Christine farms) were collected in six informal markets where 60% of rape was sold.

Results: A total of 22 out of 24 samples of rape were assessed. Out of the 150 samples examined, 42 (28%) were positive for ESBL-producing bacteria. The 42 isolates were analysed for different antimicrobial resistance (AMR) genes.

Significance: Results revealed that 26/42 (61.9%) of the rape samples were resistant to two or more antibiotics.

P2-78 Beef Contamination with Salmonella spp. and Their Resistance to Antibiotics is a Concern and a Threat to Public Health

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Introduction: Beef contamination with Salmonella spp. and their resistance to antibiotics is a concern and a threat to public health.

Methods: A total of 48 samples were collected from Farm F. The occurrence of ESBL genes was determined using the ESBL-producing Enterobacteriaceae were assessed.

Results: A total of 24 samples, including 12 water samples (one liter × 12), and 12 cucumbers (150 g × 12), were collected and processed. Following enrichment and streaking onto chromogenic media, 45 presumptive ESBL-producing Enterobacteriaceae and the prevalence of ESBL-producing Enterobacteriaceae were enumerated and the toxicity of cefotaxime.

Significance: The occurrence of ESBL-producing Salmonella spp. and their resistance to antibiotics indicates contamination, indicating the necessity to raise awareness of food safety and educate farmers and retailers on good hygiene practices.
Methodology: A total of 385 Salmonella isolates from foods and humans were screened for azithromycin resistance genes by PCR. Antibiotic susceptibility to 18 antimicrobials of the azithromycin-resistant isolates were determined by agar dilution method. PFGE and MLST were used to determine the genetic relatedness and clonal expansion of these isolates. The transferability of antibiotic resistance genes in resistance was tested by conjugation experiments and transformation experiment with E.coli C600 as recipient.

Results: The gene mph was identified in 15 Salmonella isolates from foods and humans. These isolates exhibited concurrently high-level resistance to azithromycin, tetracycline and ciprofloxacin which were determined as the same ST (ST34) and exhibited high similar PFGE patterns. Fifteen transformants were obtained from transformation, and exhibited resistance to azithromycin, tetracycline and ciprofloxacin. These transformants were determined to belong to ST34 with the same PFGE pattern as the parental isolates. In addition, the isolates were also screened for other resistance genes, such as tetA, tetB, tetC, tetD, tetE, tetF, qnr, oqxA, oqxB, oqxC, oqxD, oqxE, oqxF, qepA, claB, and telA, but no other resistance genes were detected.

Discussion: This study revealed that the emergence of resistance to azithromycin to ciprofloxacin and tetracycline in Salmonella isolates was probably due to the spread of the mph gene. The presence of the mph gene in Salmonella isolate indicated the presence of resistance genes, which could contribute to the emergence of resistance to azithromycin in Salmonella.

Significance: This study contributes to the understanding of the distribution and transmission of antibiotic-resistant genes in Salmonella isolates from foods and humans in China, and this was likely due to spread of the mph gene from bacteria to bacteria and the spread of resistance genes to other bacteria.

P2-21 The Cantaloupe Farm Environment Has a Diverse Genetic Pool of Antibiotic-Resistance and Virulence Genes

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Introduction: The aim of this study was to assess the prevalence of azithromycin resistance genes among Salmonella isolates from foods and humans in China, and this was likely due to spread of the mph gene. The presence of the mph gene in Salmonella isolate indicated the presence of resistance genes, which could contribute to the emergence of resistance to azithromycin in Salmonella.

Purpose: The aim of this study was to determine the prevalence and transmission of azithromycin resistance genes among Salmonella isolates from retail foods and farms in China. These genes were sequenced from 75 samples at 20,000 reads per sample and the sequences were analyzed with QIME. Results: Bacteria taxonomy was elucidated using the SILVA 128 16S database.

Methodology: This study aimed to determine the prevalence and transmission of azithromycin resistance genes among Salmonella isolates from foods and humans in China. The presence of the mph gene indicated the presence of resistance genes, which could contribute to the emergence of resistance to azithromycin in Salmonella.

Discussion: This study revealed that the emergence of resistance to azithromycin to ciprofloxacin and tetracycline in Salmonella isolates was probably due to the spread of the mph gene. The presence of the mph gene in Salmonella isolate indicated the presence of resistance genes, which could contribute to the emergence of resistance to azithromycin in Salmonella.

Significance: This study demonstrates the importance of monitoring the emergence of resistance genes in Salmonella isolates from foods and humans in China. This is likely due to the spread of the mph gene from bacteria to bacteria and the spread of resistance genes to other bacteria.
P2-87
Assessment of Veterinary Drugs Present in Pork Kidney Purchased from Four Retail Stores
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Introduction: Kidney is low-cost, in slaughter facilities and an excellent tissue for assessing drug exposures. Rapid on-site infection swab (KIS) tests can screen antibiotic residues efficiently. ELISA can complement KIS screening for non-antibiotic residues or antibiotics that are less sensitive using KIS.
Methods: To determine the extent of commonly used veterinary drug residues in pork kidney obtained from midwestern United States retail markets as well as their correlation with other residues.
Results: A total of 1404 pork kidneys, purchased from four retail stores (a local grocery chain, a butcher shop, two ethnic shops), were screened for veterinary antibiotic residues. Sixty-five samples were obtained from each of the retail locations. The veterinary antibiotic residues identified were amoxicillin (28.25%), trimethoprim-sulfonamide (21.95%), tetracycline (17.77%), enrofloxacin (16.67%), sulfadimethoxine (10.71%), and streptomycin (6.33%).
Significance: Our results indicate that multiple veterinary antibiotic residues are frequently detected in pork kidneys in retail markets. This study provides valuable data for monitoring veterinary drug use and residues in pork kidneys.

P2-88
WITHDRAWN

P2-89
The Presence of Cyclospora cayetanensis, Toxoplasma gondii, and Giardia intestinalis in Potential Sources of Agricultural Water in the United States
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Developing Scientific Evidence: In the United States, livestock and poultry operations face considerable challenges due to climate change. Concerns surrounding freshwater availability and safety have been on the rise, as more outbreaks of foodborne illness have been linked to agricultural water. Protozoan parasites can be isolated from lakes, reservoirs, irrigation water, wastewater, and have a history of association with drinking and recreational water, further substantiating the need to include them in the food safety and quality considerations and evaluation of non-traditional water sources.
Methods: To examine the prevalence of protozoan parasites in potential alternative sources of agricultural water.
Results: Water samples (n=36) were collected (June to October 2017) from surface water (tidal brackish, pond), and recycled water (vegetable processing). Samples were collected in the mid-Atlantic region, and 10 to 20 liters were filtered using an Enrichlock CV Capsule at a filtration rate of two liters. Filters were subjected to the ELISA 16:2013 modified (2013). The DNAmp Water Extraction Kit was used and qPCR was performed with the Quantitative Probe ABI. The relative quantification of the most abundant parasite was performed. Daily data analysis was using q-CR test.
Results: For Cryptosporidium parvum, 65.38% (n=26) of water samples tested positive. By water type, 50.00% (n=10) of recycled water (RW); and 75.00% (n=15) of surface water (SW) tested positive. For Cyclospora cayetanensis, 53.84% (n=20) of the water samples tested positive. By water type, 40.00% (n=10) of RW; and 62.50% (n=20) of SW tested positive. For Toxoplasma gondii, 62.50% (n=26) of water samples tested positive. By water type, 20.00% (n=10) of RW; and 50.00% (n=10) of SW tested positive. Giardia intestinalis, was not detected. There were no statistically significant differences amongst water types for presence of protozoa (Chi²=0.75, p>0.05), for which is more likely to be detected in RW (p=0.043).
Significance: Understanding the prevalence of protozoan pathogens in alternative agricultural waters will enable the establishment of interventions and subsequent safe use of these waters in irrigation.

P2-90
Diverse Shiga Toxin-producing Escherichia coli-specific Bacteriophages Exist in Goat Feces and the Surrounding Environments on an Organic Produce-growing Farm in Northern California, USA
Yen Te Liao, Marion Lennon, Alexandra Salvador, Valerie Lavrenkov, Angelina Hsu and Vivian Chi-Hua Wu
1Western Regional Research Center, Agricultural Research Service, USDA, Albany, CA; 2California State University - East Bay, Hayward, CA
Introduction: Shiga toxin-producing Escherichia coli (STEC) contamination on produce is primarily associated with ruminant feces, which can result in severe foodborne illness. Although numerous STEC-specific bacteriophages (phages) have been isolated from cattle feces, the presence of STEC-specific phages in the top seven STEC (O157:H7) strains tested was not temporally assessed.
Methods: The objective of this research was to investigate the prevalence and diversity of the top seven STEC and STEC-specific phages in ruminant feces and its surrounding environments. Samples were collected from a goat farm in Northern California.
Results: One sample each from three sites (goat feces, cattle feces, and soil) was collected monthly for six months (soil was collected for five months; n=17). Individually, samples were tested for STEC using culture and PCR-based methods and STEC-specific phages using enrichment with a cocktail of the top seven STEC strains followed by purification via plaque assay. The isolated phages were subjected to host range tests and morphological observation by transmission electron microscopy.
Results: Ten samples (five goat, three soil, and one cattle) contained various STEC-specific phages belonging to three families (Myoviridae, Siphoviridae, and Podoviridae). The phages isolated from eight samples (five goat, two soil, and one cattle) showed lytic activity against STEC O114, several of which exhibited a wide range of host specificity. Three phages could not be isolated without STEC of the top seven samples. Two STEC O114 and O75 antigen-negative strain were isolated from one soil and one cattle sample, respectively.
Significance: This study indicates that STEC-specific phages were consistently isolated from goat feces. The prevalence of phages with wide and complex host ranges is against the top seven STECs, resulting on zero isolation of the bacterial pathogens, could be indicative of environmental biocontrol of phages in this niche.

P2-91
Crassphage as a Source Tracking Tool to Investigate Human Stool Contamination
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Introduction: Crassphages are novel bacteriophages that have potential to serve as biocontrol agents in humans. Using Nextseq sequencing, the presence and concentration of phages can be determined in human stool samples. The phages isolated from eight samples (five goat, two soil, and one cattle) showed lytic activity against STEC O114, several of which exhibited a wide range of host specificity.
Methods: Three phages could not be isolated without STEC of the top seven samples. Two STEC O114 and O75 antigen-negative strain were isolated from one soil and one cattle sample, respectively.
Significance: This study indicates that STEC-specific phages can be isolated from other patients. Our data support previous work that crAssphages are uniquely present in human but not animal stool samples making them potentially useful as an indicator of human fecal contamination. Furthermore, we show that crAssphages can be employed to better understand sources of contamination in multiple environments.

P2-92
Virus Recovery Affected by Contact Surface Physicochemistry of Polymer and Glass
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Introduction: Enteric viruses have been recognized as a major causative agent for foodborne illnesses worldwide. Viruses are transmissible via their attachment to a range of contact surfaces on food packaging and storage containers. Enteric viruses can be isolated from food, however, their transmissibility and persistence remains to be determined.
Methods: Abiotic surfaces of polypropylene (PP), polyvinyl chloride (PVC), polyethylene (low and high densities, LDPE and HDPE), and glass (borosilicate and soda lime) were characterized by atomic force microscopy (AFM), profilometry, templometry, and infrared spectroscopy. Observing the MS2 inactivation order from four smooth surfaces was PP > HDPE ≥ LDPE ≥ soda lime glass, with strong hydrophobic PP and PE. AFM revealed that MS2 was more readily attached to silica-rich rough surfaces as compared to smooth, thus decreasing virus recovery. Significantly (P<0.007) more viruses were recovered from smooth compared to rough surfaces. All nine surface recoveries were distributed into six statistical groups, with the highest and lowest being smooth PP (76±12%) and hole-bearing borosilicate glass (52±6%), respectively. The recoveries of PP, PE and PE (including smooth and rough surfaces) were classified into five of the six groups.
Results: The correlation between surface cleanliness could be compromised by scratches or holes occurring during usage or manufacture. Our results illustrated that not all plastic surfaces release attached viruses with equal efficiency; the same was observed for two glass surfaces investigated.

P2-93
The Prevalence of Bacteriophages Lytic against Shiga Toxin-producing Escherichia coli (STEC) and Its Correlation with STEC Bacterial Hosts
Yen Te Liao, Marion Lennon, Alexandra Salvador, Valerie Lavrenkov, Angelina Hsu, and Vivian Chi-Hua Wu
Wuhan National Research Center, Agricultural Research Service, USDA, Albany, CA
Introduction: Lytic bacteriophages are increasingly considered as alternative biocontrol agents of bacterial pathogens due to their killing effect. The most frequently isolated STEC, such as Shiga toxin-producing Escherichia coli (STEC), have emerged as important foodborne pathogens in the United States. STEC-associated infections are often more severe than other foodborne disease outbreaks associated with enterohemorrhagic Escherichia coli (E. coli O157:H7), and Salmonella enterica serotype Typhimurium (S. Typhimurium), which are major foodborne pathogens in the United States.
Methods: In August and September 2018, a total of 370 samples, including water, soil, sediment and animal feces, were collected monthly from an organic farm in Northern California containing two separate farming areas: one with and one without animal activity. Cocktails of three non-pathogenic E. coli (2) and 14 STEC strains (top six non-O157 and O157 serogroups) were used for phage isolation and host range tests. Culture methods and PCR were used to characterize the phages and determine their lytic activity.
Results: The results showed that 31 (8.4%) of the samples were positive for lytic phage activity against STEC strains. Most bacteriophage-positive samples (n=26) were collected from the areas with animal activity. Spring season, particularly April, had relatively high prevalence of bacteriophages, which was likely due to high rainfall precipitation. Additionally, the three most frequently isolated bacteriophages were lytic against STEC O114, O121, and O129. No

P2-87 – P2-90
Journal of Food Protection Supplement
P2-91 – P2-93
Journal of Food Protection Supplement

193
Phages harbored active genes in this study. One STEC O113 was isolated; however, no phages specific to the serogroup were isolated from the particular samples.

Significance: The findings indicate that the prevalence of STEC-specific phages highly correlated with snap crawls collected from the animal-involved areas. Furthermore, the presence of these bacteriophages is important for the negative correlation with the presence of their STEC hosts.

**P2-94 Chemical Inactivation of Encephalitozoon intestinalis and Salmonella Enteritidis**

**Poster**

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**Developing Scientist Entrant**

Introduction: Encephalitozoon intestinalis is a foodborne parasite that causes gastrointestinal illnesses. Currently, there is limited research conducted on the reduction of microbial pathogens via biocides focusing on microsporidia.

Purpose: Chemical inactivation of E. intestinalis is explored with Salmonella Enteritidis as a bacterial control.

Methods: Six disinfectants and sanitizers (peracetic acid, a peracetic/peroxidisocyanic acids, a fatty acid blend, a quaternary ammonium compound, and a fatty acid blend) were tested in suspension at 2000 ppm. Each disinfectant and viability was determined using the TV culture in rabbit kidney cells (RK-13, ATCC CCL-37), Peptone (0.1%) was used as the control for Salmonella and cells were recovered in trypsine soy agar (TSA) containing ampicillin. Log reduction (CFU/ml) of viable spores and bacterial cells were calculated and AMNOVA was used to analyze data from the assay.

Results: E. intestinalis was inactivated by five of the six biocides with no significant differences (p<0.05) between exposure times. Spores treated with the quaternary ammonium compound were not inactivated. Biocides were confirmed with tissue culture. Salmonella Enteritidis had a log reduction range of 5.91 to 7.11 CFU/ml at 23°C and 6.10 to 7.75 CFU/ml at 50°C for all biocides at all exposure times. There was a significant difference (p<0.001) for temperature only for peracetic acid with higher log reductions observed at 50°C (5.9; 5.9, 6.13 CFU/ml for five, 15, and 30 minutes, respectively) than 23°C (7.45, 7.55, 7.51 CFU/ml for five, 15, and 30 minutes, respectively).

Conclusion: Complete chemical inactivation of microsporidia was demonstrated and can be used for food safety interventions in the food industry.

**P2-95 Optimization and Evaluation of a Viralbed Method for Viral Detection in Environmental Source Waters: A Conserved Study**

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**Developing Scientist Entrant**

Introduction: Virus adsorption and elution (VIRADEL) methods originated in the 1970s. The variability of environmental water samples provides challenges for the development of a standard method for viral detection.

Purpose: To optimize the recovery and detection of viruses from environmental source waters used for crop irrigation by modifying and evaluating a VIRADEL method.

Methods: Surface (50 cm²) and reclaimed water (RW) samples were filtered in 25 to 40 L volumes and eluted using 300 ml sodium polystyrene sulfonate. The eluate was concentrated using four 100 kDa filters prior to nucleic acid extraction. Tulane virus (TV), a surrogate for human norovirus, was used as a process control. Initial water samples (n=3), eluates (n=3), and concentrates (n=4) were inoculated to determine efficiency of filtration, concentration, and extraction, respectively. A n=6 aliquot of TV (7.8 log genomic copies/ml) was used to inoculate each initial water and eluate sample, while 0.10 ml was used in each concentration. Detection was performed using RT-qPCR and virus quantified using a standard curve generated from TV RNA. Statistical analysis was performed by one-way ANOVA.

Results: Eluates were successfully concentrated in all inoculated samples (n=10). There was no significant difference in TV genomic copies detected from RW or RW samples inoculated prior to filtration (P=0.65) or eluates (P=0.19), the average detection being 5.62±0.25 and 7.26±0.18 log copies/ml; however, the viral load was significantly lower (P<0.001) in RW than in TW when inoculated into the concentrate (P=0.003). Detected copies from RW samples averaged 7.63±0.18 log copies/ml while RW samples averaged 7.05±0.08 log copies/ml. From these data it was determined that 1.19±0.44 log copies/ml lost was due to the initial filtration step.

Significance: These data show the impact that physicochemical variations of environmental water has on detection of viruses, contributing to the development of a consistent methodology for detection.

**P2-96 Enteric Virus Detection in Leafy Greens**

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**Developing Scientist Entrant**

Purpose: The objective of this study was to validate the use of FDA BAM chapter 268 with the addition of the romaine lettuce and spinach matrices for enteric virus detection including norovirus (NoV) and hepatitis A virus (HAV).

Methods: Following FDA BAM chapter 268, 50 of inoculated romaine lettuce and spinach samples were concentrated using ultrafiltration and RNA extracted from concentrates. The resulting RNA samples were tested for both viruses by one-step RT-PCR using primers specific to the NoV L gene and for Hepatitis A virus by RT-qPCR using primers specific to the HAV pre-S2 gene. Retrotranscriptase PCR was performed to convert viral RNA into cDNA, and samples were co-linear amplified before denaturing. Each PCR product was then used for RT-PCR using primers specific to the NoV L gene and the HAV pre-S2 gene. Primers specific to the L gene were used for a second round of amplification. The resulting PCR products were separated by agarose gel electrophoresis and visualized by staining with ethidium bromide.

Results: The continued occurrence of viral foodborne outbreaks emphasizes the importance of standardized methods. Currently, FDA’s Bacteriological Analytical Manual (BAM) chapter 268 contains a method validated for one matric green onion. Creating a leafy green category, exhibited in ISO 16140-2, can broaden the method’s application.

Purpose: The objective of this study was to validate the use of FDA BAM chapter 268 with the addition of the romaine lettuce and spinach matrices for enteric virus detection including norovirus (NoV) and hepatitis A virus (HAV).

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Results: The continued occurrence of viral foodborne outbreaks emphasizes the importance of standardized methods. Currently, FDA’s Bacteriological Analytical Manual (BAM) chapter 268 contains a method validated for one matric green onion. Creating a leafy green category, exhibited in ISO 16140-2, can broaden the method’s application.
P2-100 Optimization of Traditional and Eco-Friendly Sanitizers against Listeria spp. at Various Temperatures and Organic Loadings

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Introduction: The frequency and variety of recalls linked to Listeria monocytogenes highlight ongoing challenges in control of foodborne illness. A Centers for Disease Control and Prevention (CDC) biofilm reactor was used to grow four-day old multi-strain L. monocytogenes (8 L. innocua, 8 L. seeligeri, and two each of L. monocytogenes, L. innocua, L. seeligeri, and L. innocua). Eight-mm diameter stainless steel coupons were loaded with lettuce, cabbage, and beet nitrates by soaking 24 h, rinsing, and air-drying five min. Manufacturer recommendations for one off (EF) and one chlorine-based sanitizer were tested for their efficacy to inactivate five L. monocytogenes (five to six log CFU) in 100% to the adhered coupons for five min, 24 h and 48 h.

Methods: The EFs of EF sanitizers (AB, CAB) were significantly impacted (P<0.05, Student’s t-test) by treatment temperature, with two-fold higher MBCs at 4°C vs. 30°C for 22 isolates tested. Differences in MBCs were also observed between 15, 20, 25, 20 and 30°C for 22 isolates (16 L. monocytogenes and two each of L. innocua, L. seeligeri, L. wickerhamii). Eight mm diameter stainless steel coupons were loaded with lettuce, cabbage, and beet nitrates by soaking 24 h, rinsing, and air-drying five min. Manufacturer recommendations for one off (EF) and one chlorine-based sanitizer were tested for their efficacy to inactivate five L. monocytogenes (five to six log CFU) in 100% to the adhered coupons for five min, 24 h and 48 h.

Results: The EFs of EF sanitizers (AB, CAB) were significantly impacted (P<0.05, Student’s t-test) by treatment temperature, with two-fold higher MBCs at 4°C vs. 30°C for 22 isolates tested. Differences in MBCs were also observed between 15, 20, 25, 20 and 30°C for 22 isolates (16 L. monocytogenes and two each of L. innocua, L. seeligeri, L. wickerhamii). Eight mm diameter stainless steel coupons were loaded with lettuce, cabbage, and beet nitrates by soaking 24 h, rinsing, and air-drying five min. Manufacturer recommendations for one off (EF) and one chlorine-based sanitizer were tested for their efficacy to inactivate five L. monocytogenes (five to six log CFU) in 100% to the adhered coupons for five min, 24 h and 48 h.

Significance: This study confirmed EF and CB sanitizers are effective against Listeria spp. when following manufacturer recommendations. However, their efficacy may be reduced under conditions relevant to FPs (e.g., temperature, presence of organic material, extended times between sanitization treatments). These findings suggest that sanitation schedules that should be considered when determining suitable sanitizers and sanitation schedules in FPs.

P2-104 Comparison of Chemical Methods for Removal of Listeria innocua Biofilm Attached to a Stainless Steel Surface

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Introduction: Antimicrobial treatment of listeria innocua biofilms does not insure removal of inactive biofilms which may serve as a platform for new biofilm growth, making it difficult to not only sanitize food contact surfaces but also to eradicate the biofilm EPS components and attached dead cells.

Methods: The present study was performed to evaluate different commonly used cleaner/sanitizers for the removal of biofilm components from stainless steel surfaces.

Results: Eight-mm diameter stainless steel coupons were immersed 3.5 cm deep into an L. innocua inoculum at 21°C for 24 h. Coupons were rinsed and subjected to one of eight treatment solutions: no treatment, ≤0.7 pH 7.2, ≤0.6% each alcohol atarray, in water, ≤0 ≤1.5 NaCl at 0°C bleach, ≤0 ≤1.5 NaCl at 0°C bleach, ≤0 ≤1.5 bleach, ≤0 ≤0.05 EDTA at 0°C, ≤0 ≤0.05 EDTA at 0°C, ≤0 ≤0.05 EDTA at 0°C, ≤0 ≤0.05 EDTA at 0°C. The coupons were rinsed and dried and stained using crystal violet or erythrosine B solutions. Stained coupons were photographed and color differences between background and dried area evaluated using an L*a*b* color space. The experiments were repeated three times on three separate days (n=9).

Results indicate the enzyme mixture removed significantly more of the biomass than the remaining treatments (P<0.05). Neither the pH 13 treatment nor commercial solution 2 were significantly more effective in their ability to remove biomass than DI water alone (P>0.05). Treatments with PAA and hydroperoxide provided significantly better bacterial reduction than the untreated control, and commercial solution 1 was minimally, though significantly, more effective at biomass removal (P<0.05) compared to untreated.

Conclusion: This study demonstrates that standard treatments to remove biofilms in a processing environment may be ineffective. Results suggest that the choice of process of enzyme treatment followed by sanitation will greatly improve the ability to both remove and disinfect biofilms from stainless steel surfaces.

P2-105 Effect of Dry Sanitization on Biofilm of Salmonella Strains Isolated from the Peanut Supply Chain

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Introduction: Salmonella is an important foodborne pathogen linked to different foods. In the manufacturing environment of low moisture products it is recommended to employ dry sanitization to avoid Salmenella growth, since the increase in humidity may result in cell multiplication and biofilm formation.

Methods: This study investigated the biofilm formation and the effectiveness of dry sanitization methods on biofilm components from stainless steel coupons. Different conditions were evaluated for optimal growth: temperature (30 and 37°C) and nutrients (tryptic soy broth + 0.6% yeast extract and brain heart infusion broth; BHIB) were added. On stainless steel, biofilm formation was impacted by the time between inoculation and sanitizer application. All LB and diluent I, 50% and 75% of the biofilm were removed by disinfection with 5% geometric mean differences.

Results: These findings highlight variables that should be considered when determining suitable sanitizers and sanitation schedules in FPs.

P2-106 Synergistic Effect of Sodium Hypochlorite and UV Light on the Survival of Listeria monocytogenes Biofilms

Ellen Mendez, Brian Tande and Valentina Tinetti
University of North Dakota, Grand Forks, ND, USA – Food Science Institute, Manhattas, KS

Developmental Scientist Entrant

Introduction: In the food industry, inadequately cleaned equipment represents a potential source for Listeria monocytogenes contamination. This pathogen has shown niche adaptation to different food processing environments and its ability to form biofilms is a hurdle for food safety. Even if sanitation practices can minimize the risk of pathogen survival, existing evidence suggests that food-handling foci in plants are still significant risk areas. The combination of sanitizers with UV light might represent an effective way to control pathogen growth.

Methods: The objective of this research was to study the effect of sodium hypochlorite solution (SHS) and UV light alone or in combination on L. monocytogenes survival on stainless steel (SS) coupons.

Significance: This study is setting stage for future research by determining the optimal concentrations of SHS and UV light alone and in combination with other sanitizers on SS coupons.
heart infusions. 100 (pH for five and 10 min) and UV light (254 nm for 15 and 30 min) were the treatment conditions applied. A control treatment (no sanitizer and UV light) was also evaluated. Experiments were performed in triplicates.

Results: L. monocytogenes biofilm cells on SS coupons ranged from 10^2 to 10^4 CFU/cm² for control samples. A significant log reduction of three log CFU/cm² was observed when samples were treated with SHS for 10 min as compared to controls (P<0.05). After UV exposure for 15 and 30 min, a 1.5 and 1.8-log CFU/cm² reduction was reported. A greater log reduction was observed when the treatment of 10 min SHS and 15 min UV light were combined: four log CFU/cm² (P<0.05).

Significance: This study demonstrates the enhanced effects of SHS and UV light in combination on the survival of L. monocytogenes biofilms on SS surfaces.

P-107 Bacteriophages as Biosanitizers: Using Lytic Phage to Control and Eradicata Listeria monocytogenes Biofilm

Stefan Cucic, Janet Lin, Cezar Khurtiga' and Manya Anany

Introduction: Listeria monocytogenes is a foodborne pathogenic bacterium that causes invasive illness in people with compromised immunity. It can persist at refrigeration temperature, over a wide range of pH, and at high osmolarity. The propensity of L. monocytogenes to produce biofilms and survive in mixed-species biofilms contributes to its persistence in food processing environments. Lytic phages represent a strategy to control sessile L. monocytogenes biofilms.

Purpose: The purpose of this study is to investigate the biosanitation potential of lytic phages against sessile L. monocytogenes biofilms.

Methods: Twenty different biofilms were formed using 10 different strains of L. monocytogenes ATCC serotype 1/2a and ATCC serotype 1/2b on polystyrene using a 96-well plate static culture vial biofilm assay. The strains were inoculated at similar levels in TSB supplemented with one percent glucose and two percent NaCl and incubated for 48 hours at 37°C. For isolation of biofilm-degrading phages, 38 and 33 samples were collected from dairy and poultry processing facilities, respectively, and L. monocytogenes ATCC 19111 (serotype 1/2a) was used as the enrichment strain. Clear plaques with halos were selected for phage purification and characterization. Moreover, the ability of the commercially available Listex P100 phage to degrade a 24-h L. monocytogenes biofilm was investigated at 25°C in static culture.

Results: L. monocytogenes ATCC 19111 (serotype 1/2a) produced ten times more adherent biomass than the other strains tested. Our results showed that concentration of P100 of 10^6 and 10^7 PFU/ml eradicated biomass produced by adherent L. monocytogenes to levels comparable to negative controls. Fourteen phage producing plaques with haloes were obtained from effluent samples collected from dairy and poultry processing facilities.

Significance: These results suggest that phages may be effective to control mono-species biofilms of L. monocytogenes and provide a baseline for comparison with other phage isolates elucidated in this study.

P-108 Effect of Water Wash Matrix on the Correlation between Free Chlorine Oxidation Potential and Potential Deterrent during Fresh Produce Oxidation and Washing Operations

Sam Van Haute, Yaguang Luo2, Bin Zhou2, Patrick Milner2

Introduction: During washing of fresh produce, free chlorine (FC) is added to wash water to avoid pathogen cross-contamination. FC is consumed by organisms released into the water from the produce. FC, FCO (peroxide) during processing is used to indirectly assess the FC residual status during wash processes. ORP is appealing to the industry because it is an easy-to-use technology. Despite this, the industry’s interest in the effect of oxygen transport and matter on the ORP is virtually unknown.

Purpose: Assess how water matrix composition (pH, temperature, acidulant, producer) influences the relationship between FC and ORP during fresh produce washing.

Methods: Fresh-cut wash water (500ml per trial) was generated by washing romaine lettuce (n=6) and romaine residue (n=6) in 3 conditions: (1) UV light, (2) alkaline washed, and (3) control (no washing or UV). The FC concentration and ORP were determined by Foss Vitek-MS System.

Results: The ORP was correlated with the logarithm of FC under all conditions. A decrease in pH or temperature led to a large and small increase in ORP, respectively. Using tap water instead of distilled water to wash the produce significantly changed the ORP. For all types of tested produce, increasing the acidulant during washing also influenced ORP.

Significance: This study demonstrates the enhanced effects of SHS and UV light in combination on the survival of L. monocytogenes biofilms on SS surfaces.

P-109 Multi-Lab Validation for FDA Identification of Salmonella, E. coli and Listeria monocytogenes using the Vitek-MS System

Michael Brown, Lisa Newberry, Thomas Hammack, Kristopher Staraya, Christopher Peters, Amir Alavi, Shannon Ruelle, Gary Hartman, Henry Lau, Elizabeth Reed, Jennifer Hat, Asheeq Ahmed, Stephanie Horton, Tamayo Barnes, Nancy Miranda, Pongsanam Lakanalatha, Michele Pleh, Dana Waggoner, Megan Davis, Rick Bolway and Jason Hert

Purpose: The FDA OIA ORS Pacific Northwest Laboratory (PNL) in coordination with the Center for Food Safety and Applied Nutrition (CFSAN) organized a multi-laboratory study to further validate the technology for these common foodborne pathogens beyond a single laboratory. If successful, this study would support the use of this instrument for bacterial identification of the target organisms.

Methods: Methodology was: A randomized, blinded bacterial panel consisting of 64 traceable, previously characterized environmental and reference sustrates composed of salmonellae and E. coli (16), Escherichia coli (n=16), Listeria monocytogenes (n=16) in triplicates. Each sample was prepared and cultured using Vitek-MS System. A significant log reduction of three log CFU/cm² was observed when samples were treated with SHS for 10 min as compared to controls (P<0.05). After UV exposure for 15 and 30 min, a 1.5 and 1.8-log CFU/cm² reduction was reported. A greater log reduction was observed when the treatment of 10 min SHS and 15 min UV light were combined: four log CFU/cm² (P<0.05).

Significance: This study demonstrates the enhanced effects of SHS and UV light in combination on the survival of L. monocytogenes biofilms on SS surfaces.
Poster

Environments

enzymatic cleaning, and routine enzymatic cleaning. During the period of trials, all factors remained constant and only the cleaning solutions were replaced.

P. fluorescens counts on vegetables with a lesser detrimental effect on their sensory quality.

Results:

Methods:

H. sabdariffa, H. sabdariffa, and water treated cabbages were the more preferred (P<0.05) than chlorine treated ones. Lettuce treated with citric acid and water were more preferred (P<0.05) while salt-treated lettuce was least preferred.

Significance:

A major plant pathogen (Pseudomonas syringae pv. DC3000), a prokaryotic bacterium (Listeria/Escherichia rhommos G) or bacterial strains previously used as biocides are plant disease control in the rice plant.

Enzymatic cleaning and attachment to vegetable seeds.

Results:

Methods:

A plant selected pathogen (Pseudomonas syringae pv. DC3000, a prokaryotic bacterium (Listeria/Escherichia rhommos G) or bacterial strains previously used as biocides are plant disease control in the rice plant. The inhibitory effect of metal-free superabsorbents of competitive bacterial spore cultures on the growth of Salmonella was also evaluated.

Results showed that the mean population of Salmonella in co-culture with E. rhommos G at 37°C was 5.6 log lower than the population in the control and any of the other treatments at the central 25°C. The addition of surface water to both tests had significant (P<0.05) reductions in Salmonella populations. Salmonella cells became undetectable (<1 CFU/ml) at the 12 h sampling point and, in a total of 5-log reduction was achieved after the 24 h incubation period. Although not as effective as L. rhommos G in inhibiting the growth of Salmonella, the biocidal agents were more effective in competing with the pathogens for attachment to vegetable seeds.

When no bacterial competitive strains were present, the mean attachment rates of Salmonella to the four types of vegetable seeds were 10.5%. In the co-cultures with E. coli, the attachment rates were considerably reduced to 1.3 to 1.6 log CFU/coupon.

Significance:

Results of the study will help to strategize interventions for the production of vegetable seeds with microbial qualities.

P2-114 Analyzing Aggregate Environmental Monitoring Data for Listeria spp. in Frozen Food Manufacturing Environments

Methods:

Introduction:

Food processors face serious challenges due to the ubiquity and prevalence of L. monocytogenes in the food processing industry. Developing effective control strategies for L. monocytogenes is a critical challenge in the food processing industry.

Pseudomonas pv. Tomato

ATCC 6051) were mixed individually to co-culture with Salmonella A506, ATCC 6051 in microbiological media or attachment to the seeds of alfalfa, fenugreek, lettuce and tomato. The inhibitory effect of bacteria on the germination of radish seeds was significant.

At the end of shelf life) by classical microbiology and 16S rDNA metagenetic. Statistical analysis (non-parametric tests and principal component analysis) was carried out on with R with different packages.

In the sanitation treatments. A total of 244 samples were analyzed, including surface samples pipes and finished products (jasmine at day zero and at the end of shelf life) by classical microbiology and 16S rDNA metagenetic. Statistical analysis (non-parametric tests and principal component analysis) was carried out on with R with different packages.

Methods:

Introduction:

Historical environmental monitoring data from the frozen food manufacturing industry was compiled and analyzed to evaluate adequacy of environmental monitoring programs. A method was developed to collect anonymous data for analysis to build a strong aggregate data set from multiple facilities. Information included in the data set includes type of food product, location, sampling frequency, test methods and environmental monitoring procedures.

Methods:

A method was developed to collect anonymous data for analysis to build a strong aggregate data set from multiple facilities. Information included in the data set includes type of food product, location, sampling frequency, test methods and environmental monitoring procedures.

Results:

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Results:

Methods:

The initial numbers of Listeria spp. attached to various abiotic surfaces were 5.7 to 6.3 log CFU/coupon. The numbers of L. innocua spores in incubated at 85% RH and 60°C for up to 120 min, regardless of RH.

This study was carried out to develop a seed decontamination method to inactivate E. coli O157:H7 present in radish seeds using a combined treatment of gaseous chlorine dioxide (CD) and mild wet heat.

Methods:

The objective of the present study was to evaluate the influence of bacterial competitors on Salmonella enterica growth in microbiological media and attachment to vegetable seeds.

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This study was carried out to develop a seed decontamination method to inactivate E. coli O157:H7 present in radish seeds using a combined treatment of gaseous chlorine dioxide (CD) and mild wet heat.
Post-119: Escherichia coli O157:H7 Inactivation in Phosphate Buffer by X-Ray with Various Levels of Accelerating Voltage

Yue-ju Wu and Sam Chang

Journal of Food Protection Supplement

Introduction: The purpose of this study was to compare the antimicrobial activities of various organic acids against Escherichia coli O157:H7 and Listeria monocytogenes attached to stainless steel.

Methods: Seven strains of L. monocytogenes were attached to stainless steel coupons. The inhibitory effect of organic acids on the number of bacterial cells was determined using an agar diffusion assay. The lag time for bacterial growth was determined using an agar diffusion assay. The results were analyzed using one-way ANOVA. The significance of differences between treatments was determined using a Tukey’s post-hoc test. A p-value of <0.05 was considered statistically significant.

Results: The results showed that organic acids had a significant inhibitory effect on bacterial growth. The inhibitory effect of organic acids on bacterial growth was concentration-dependent. The inhibitory effect of organic acids on bacterial growth was also affected by the presence of other factors such as pH and temperature.

Discussion: The results of this study indicate that organic acids have potential as preservatives in food systems. However, further studies are needed to determine the optimal conditions for their use.


Developing Scientist Entrant

Post-120: Comparison of Antimicrobial Activities of Organic Acid Vapors Against Escherichia coli O157:H7 and Listeria monocytogenes Attached on Stainless Steel

Ye-jung Shin, Woorim Yoon and Hee-joon Ryu

Department of Biotechnology, College of Life Sciences and Biotechnology, Konkuk University, Seoul, South Korea

Developing Scientist Entrant

Methods: Three strains of E. coli O157:H7 and L. monocytogenes were attached to stainless steel coupons. The inhibitory effect of organic acid vapors on the number of bacterial cells was determined using an agar diffusion assay. The lag time for bacterial growth was determined using an agar diffusion assay. The results were analyzed using one-way ANOVA. The significance of differences between treatments was determined using a Tukey’s post-hoc test. A p-value of <0.05 was considered statistically significant.

Results: The results showed that organic acid vapors had a significant inhibitory effect on bacterial growth. The inhibitory effect of organic acid vapors on bacterial growth was concentration-dependent. The inhibitory effect of organic acid vapors on bacterial growth was also affected by the presence of other factors such as pH and temperature.

Discussion: The results of this study indicate that organic acid vapors have potential as preservatives in food systems. However, further studies are needed to determine the optimal conditions for their use.


Developing Scientist Entrant

Post-121: Reduction of Escherichia coli O157:H7 and Salmonella Typhimurium on Formica Coupons by Switchgrass Extractives, a Value-Added Product

Yung-jun Huang, Yung-hsiang Tsai, Yi-Chen Lee* and Yi-Yin Chen*

National Penghu University of Science and Technology, Penghu, Taiwan, National Kaohsiung University of Science and Technology, Kaohsiung City, Taiwan

Introduction: Scombroid poisoning is a fish-borne intoxication caused by the consumption of seafood containing relatively high histamine content. Various methodologies for inhibiting scombroid intoxication could be applied to reduce the risk of histamine formation by blue LEDs (470 nm) in the inactivation of histamine-producing bacteria. The purpose of this study was to evaluate the antibacterial effect of blue LEDs on the inhibition of histamine formation by blue LEDs (470 nm) in the inactivation of histamine-producing bacteria.

Methods: The blue LED light source providing a photosynthetic photon flux density of 200 µmol/m²/s was used for illumination. Bacterial cultures suspended in phosphate-buffered saline (PBS) were illuminated with blue LEDs at 10, 20, and 30°C for 12 h. The bacterial populations significantly increased at 30°C and on histamine formation.

Results: The results of this study demonstrate the effectiveness of blue LEDs (470 nm) in inhibiting histamine formation and thus demonstrate their suitability as a novel method for reducing the risk of histamine poisoning.

Significance: This data suggests that the combined treatment presents a more efficient means of deamination and maintaining the quality of fresh fruits.


Developing Scientist Entrant

Post-122: Reduction of Escherichia coli O157:H7 and Salmonella Typhimurium in Phosphate Buffer by X-Ray with Various Levels of Accelerating Voltage

Journal of Food Protection Supplement

Methods: The pure culture (phosphate-buffer saline, pH 7.4) was prepared with a three-strain mixture of E. coli O157:H7 (4646, EDL333 and ATCC 48958). Salmonella strains grown in tryptic soy broth with 0.0% yeast extract and inoculated at 37°C for 24 h prior to use and mixed with an equal volume to give approximately 10⁵ CFU ml⁻¹. Pure culture was treated with zero, 250, 500, 750, 1000 or 1250 Gy X-ray at 50, 200 or 350 kV of accelerating voltages with or without one mm aluminum filter. Results: The result of plate count demonstrated a lower susceptibility to X-ray when no filter was applied on the machine (P<0.05). Treatments with 1250 Gy X-ray achieved the reductions of 5.5 log CFU ml⁻¹ at 350 kV or 2.5 and 2.2 log CFU ml⁻¹ at 200 and 50 kV, respectively. With the use of the aluminum filter, E. coli O157:H7 was inactivated to below the detection limit (less than two log CFU ml⁻¹) when X-ray dose was equal or higher than 750 Gy. Treatments with 500 Gy X-ray achieved reductions of 6.6 log CFU ml⁻¹ at 50 kV or 5.9, and 4.8 log CFU ml⁻¹ at 200 and 350 kV, respectively.

Methods: The significance of E. coli O157:H7 detection after varying accelerating voltages and the inclusion of a filter.

Developing Scientist Entrant

Poster
**P2-125 Efficacy of Novel Photo-chlorine Dioxide against Clostridium difficile Endospores**

Muthu Dharmasiri, David Buckle, Hongye Wang and Xiuiping Jiang

**Clowson University, Clowson**

**Introduction:** Clostridium difficile infection are number one cause of healthcare-associated infections in many developed countries. C. difficile endospores pose a significant health risk to the public's health based on their environmental resilience.

**Purpose:** The objective of this study was to evaluate the efficacy of a novel photo-chlorine dioxide disinfectant against C. difficile endospores.

**Methods:** Chlorine dioxide was generated by mixing one percent sodium chlorite with 10 ppm eosin Y. Photo-chlorine dioxide efficacy was assessed against C. difficile endospores in suspension, and on stainless steel laboratory and greenhouse conditions by following EPA SOP No. MB-31-03 protocols. Surviving endospores were enumerated by plate count assay on brain heart infusion agar supplemented with yeast extract, L-cysteine, and sodium taurocholate (BHA-EA-CYS-T) with and without organic soil. All experiments were repeated three times.

**Results:** In the suspension, C. difficile endospores were reduced by 2.25 and 3.65 log CFU within two h with and without soil, respectively. The log reduction of endospores was less than two log with soil. However, under the greenhouse conditions, the reduction of endospores was ca. three log CFU for both with and without soil conditions after 24 h with four doses of toxicity treatment.

**Conclusion:** This study confirms the efficacy of a novel chlorine dioxide generation system against any pathogen. Our results suggest this new system is capable of reducing C. difficile endospores. However, endospores are more resistant to disinfection especially under the soil conditions.

**P2-126 Ultraviolet Light with Grape Seed Extract and Curcumin Inactivates Aichi Virus on Formica Surfaces**

Jackson Craig, Janei Hetu and Doris D’Souza

University of Toronto, Toronto,

**Undergraduate Student Award Recipient**

**Introduction:** Aichi virus (AV) causes human gastroenteritis globally due to the consumption of contaminated oysters. AV of the Picornaviridae family (same family as hepatitis A virus) is a 30 nm, small, non-enveloped, single-stranded, positive RNA virus. Novel methods to control the spread of AV are needed as there is no known vaccine available. The ability to evaluate and control the spread of AV is important for public health. The aim of this study was to evaluate the impact of AV spread on Formica species, host cells in six-well plates.

**Purpose:** The purpose of this research was to determine the ability of PDI-mediated by grapeseed extract and curcumin to inactivate AV on a model food contact surface.

**Methods:** AV (P2-125, P2-126, P2-127, P2-128, P2-129, P2-130) was aseptically dried on sterile Formica coupons (a model food contact surface) in a biosafety cabinet and treated with either water (control) or treated with 0.05 mg/ml GSE or 10 mg/ml curcumin (UV light) at 240-245 nm, UV light with 0.05 mg/ml curcumin or UV light with 10 mg/ml GSE, for up to 30 min at room temperature. Viruses from three replicate treatments were recovered and plaque assayed in duplicate using confluent HeLa host cells in six-well plates.

**Results:** AV showed reduction of 0.66 and 1.26 log PFU/ml with 0.05 mg/ml curcumin and 10 mg/ml GSE, respectively, while UV light alone caused a 2.7 log PFU reduction after 30 min. Both GSE with UV light and curcumin with UV light after 30 min showed increased reduction of 2.88 log and 3.41 log PFU/ml respectively, compared to controls treated with water or 0.05 mg/ml curcumin alone.

**Significance:** Thus, UV light together with natural plant extracts show promise to decrease AV transmission from food contact surfaces.

**P2-127 Effect of High Intensity Light Pulses on the Reduction of Microbial Load in Chia (Salvia hispanica L.) Seeds**

Raúl Avila-Sosa, José Salas Méndez-Aguilar, Patima Reyes-Jurado, Aurelio Lopez-Malo2, Enrique Palou3, Carlos Enrique Ochoa-Velasco2 and Adolfo Ringo-Nazario2

1Benemérita Universidad Autónoma de Puebla, Puebla, 2Universidad de las Américas Puebla, Universidad De las Américas Puebla, Cholula, Mexico

**Introduction:** The harvest of chia seeds (Salvia hispanica L.) provides an excellent environment for the growth of microorganisms; the moisture resulting from the irrigation process, the aerobic conditions, pH and temperatures all contribute to the rapid expansion of microbial populations, including pathogens. Populations are as high as 10⁹ CFU/g have been reported in chia seeds obtained from retail stores.

**Purpose:** The aim of this study was to evaluate High-Intensity Light Pulses (HILP) in microbial load reduction in chia seeds.

**Methods:** High-Intensity Light Pulses (HILP) were performed on chia seeds in a dark room at a distance of 10 cm, using a Philips model TLX 10W/02L/01 lamp and at a power intensity of 1W/cm². Twenty samples of 50 g each had HILP applied at 25 µJ/mm².

**Results:** The mean release of microorganisms from chia seeds was reduced by 2.25 and 3.65 log CFU within two h with and without soil. Under the greenhouse conditions, the reduction of endospores was ca. three log CFU for both with and without soil conditions after 24 h with four doses of toxicity treatment.

**Conclusion:** These findings support HILP as a suitable method for reduction of microbial load in chia seeds.

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**P2-128 Evaluation of Environmental Monitoring Tools for the Release of Microorganisms**

Sarah Jones and Kristin Gibson

University of Arkansas, Fayetteville, AR

**Introduction:** Environmental monitoring of foodborne pathogens (EM) is used to determine harborage sites of microorganisms, assess the effectiveness of surface sanitation practices, and prevent transmission of microorganisms. Surprisingly, most EM tools are not well characterized in their ability to recover and release microorganisms from foodcontact surfaces.

**Purpose:** To characterize EM tools for the release of microorganisms.

**Methods:** Five-hundred microliter of a bacterial cocktail (Listeria monocytogenes, Salmonella Typhimurium) was directly inoculated onto polystyrene flat and cellulose swabs hydrated with PBS. Incubum levels ranged from 10⁵ to 10⁶ CFU per swab. Bacteria were recovered by machine stomaching or manually eluting swabs. Elution experiments were performed using Tula virus, a human coronavirus surrogate, and results were obtained via plaque via study. To study operator efficacy, 10⁵ CFU of microorganisms were inoculated onto both swabs and food matrices for evaluation. Samples were analyzed in duplicate with each system.

**Results:** Microbial release was more consistent using manual elution methods when compared to machine processing. The manual and machine elution methods resulted in mean log losses of 0.5 and 0.71, respectively. Data indicated significantly (P<0.05) greater release of bacteria compared to Tula virus with nearly 20% of the mean release of microorganisms from swabs decreased the inoculum level decreased. For instance, 92% of all microorganisms were released when swabs were inoculated with 10⁵ CFU or PFU/50 µL compared to 76.3% of microorganisms at 10⁴ CFU or PFU/50 µL. Operator variability data indicate there is no significant (P>0.05) difference among operators during manual swab processing.

**Significance:** Findings from this study are important for future development of microbial release, swab material, and elution process type. This variation emphasizes the need for industry standardization and further EM tool evaluation.
P2-132 Changes in AMP, ADP, and ATP Concentrations over Extended Growth Curves for Bacterial Species Significant to Food Hygiene

Nicholas W Smith, Jeffrey Simidan1 and Scott A Rankin1
1University of Wisconsin-Madison, Department of Food Science, Madison, WI, University of Wisconsin-Madison, Department of Animal Science, Meat Science and Muscle Biology Lab, Madison, WI

Introduction: Adenosine triphosphate (ATP) is routinely used as a target for rapid assessments of surface hygiene where prokaryotic and eukaryotic cell contaminants are a concern. There is a strong hypothesis and growing evidence that concentrations of ATP will be depleted as cell contaminants metabolize and dephosphorylate the ATP (ADP and monophosphate (AMP) isomers), thus potentially rendering rapid ATP-based assays less sensitive to contaminants. There are few published reports describing the actual concentrations of adenylate isomers for microbial contaminants.

Purpose: Determine concentrations of ATP, ADP, and AMP (sum of the concentration of all isomers) over extended growth curves for bacterial contaminants of significance to food hygiene.

Methods: Using a luminometer/fuclorin-fuclerise based technique, concentrations of adenylate isomers were calculated against curves of authentic adenylate standards. Specific strains of bacteria included Cronobacter sakazakii, E. coli O157:H7, Listeria monocytogenes, and Bacillus subtilis grown in recommended media. Replicate (n=5) studies were conducted tracking adenylate concentrations from inoculation to up to several weeks of incubation; in complement, growth was estimated using OD600 values. ANOVA was conducted as a means of comparing shifts in adenylate profiles.

Results: In general, initial increases in ATP content followed the growth curve of the organism. ATP was initially the predominante adenylate following OD adjustments but was soon replaced by AMP as the primary adenylate with intermediate levels of ADP. Each bacterial strain displayed unique profiles but isomer generation and degradation.

Significance: Assays for assessing hygiene solely based on the presence of ATP as a means of assessing prokaryotic contaminants may lose sensitivity as the metabolic state of the bacterium changes. Furthermore, the differences between bacteria used in this study suggest that changes in assay sensitivity would also result from the changes in the genera or species of bacterial contaminant.

P2-133 Sanitation Monitoring of Stainless Steel Surfaces Using the Total Adenylates Hygiene Monitoring (TAP) Kit

Natsumi Tanaka, Wataru Saito and Mikiyo Bakke
Kikkoman Biochemifa Company, Noda, Japan

Introduction: ATP rapid hygiene monitoring tests are useful for the implementation of HACCP and HARPC programs. Recently, the ATP-ADP+AMP test was shown to be a powerful tool to reveal improper cleaning and the presence of contamination that conventional ATP tests missed due to the degradation of ATP to ADP and ADP to AMP. The performance of the A3 test was verified using simulated sanitation monitoring and comparison with other conventional ATP methods.

Methods: The A3 test kit (Kikkoman Biochemifa) and three commercially available ATP tests were evaluated. The dry swabs of the A3 test were moistened using nuclease-free water. Foods (ham, raw chicken, yogurt, beer and orange juice) were homogenized and diluted with water. ATP numbers were determined using the kit. A3 test results were compared to independently spiked or naturally contaminated samples, enriched per ISO method and confirmed with the BAX system.

Results: The detection sensitivities of the A3 test were superior. The A3 test showed 157,389 relative light units (RLU) for ham (100-fold dilution); on the other hand, the other commercially-available ATP tests showed 2 to 15,476 RLU (A3) and 20 to 173 RLU (ATP), yogurt (100-fold dilution); 18,371 RLU (A3) and 911 to 3,104 RLU (ATP), beer (tenfold dilution); 10,777 RLU (A3) and zero to 200 RLU (ATP), orange juice (100-fold dilution); 4,568 RLU (A3) and 685 to 1,995 RLU (ATP).

Significance: Food residues on surfaces are the source of nutrients for microorganisms or they can also interfere with the antimicrobial activity of disinfectants. Moreover, they can present a risk of contamination. The results showed that the ATP test may indicate false-negative for food residues and the A3 test is a more accurate tool to verify the levels of hygiene and sanitation.

P2-134 Evaluation of Two Real-time BAX PCR Assays for the Detection of Genus Listeria species and Listeria monocytogenes

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Introduction: Listeria monocytogenes, a widespread, opportunistic pathogen, is the most important contributor to the foodborne illness outbreaks. The occurrence of L. monocytogenes in food/environment samples is of increasing importance highlighted by rising numbers and severity of food safety recalls/outbreaks in recent years. This study aimed to evaluate two real-time PCR methods for their ability to detect L. monocytogenes and exclude other closely related genera or species of bacterial contaminant.

Methods: Fifty target and 30 non-target strains of genus Listeria and L. monocytogenes were tested during inclusivity/exclusivity studies. The inclusivity portion tested 10 cells/225 ml of 24 LE Complete; the exclusivity portion tested pure cultures at ≥10^8 CFU/ml. Lysis of artifically spiked and naturally contaminated food/environmental samples from six sample categories (m44 liedum spp.; m46; 42 L. monocytogenes) in 24 LE Complete Media were prepared. Test kit results were compared to independently spiked or naturally contaminated samples, enriched per ISO guidelines.

Results: The assays were 100% inclusive for all species tested; exclusivity was 100% against closely/distantly related genera. Sensitivity studies showed equal or better performance, compared to GO method, in all six sample categories. L. monocytogenes were all lower than the final AC of 2.5 in all categories.

Significance: The realtime BAX assays exhibit robustness and faster/more accurate sample preparation and cycling times, than endpoint PCR detection. These new assays allow for rapid time-to-results for the testing of food/environmental samples, while pre-serving the ease, accuracy and dependability of the BAX System.

P2-135 The Effect of Food Safety on Customer Satisfaction: Exploring Customer-generated Reviews through Business Intelligence

Jack Hodges, Minh-Long Lee, Agnes DeFranco and Sujata A. Sinrat
University of Houston, Houston, TX

Introduction: Due to the low percentage of restaurant customers who report foodborne illness to public health channels, customer-generated content is becoming increasingly important when monitoring the spread of foodborne illnesses in foodservice establishments. Help.com is the predominant online review marketplace for foodservice businesses, and it has been suggested that information in these reviews could be critical data for scientists.

Purpose: The objectives were to i) identify customers’ voices relating to foodborne illness and restaurant cleanliness issues, ii) quantitatively measure issues related to foodborne illness and restaurant cleanliness, iii) examine the relationship between foodborne illness and restaurant cleanliness issues and restaurant reputation/customer satisfaction; iv) identify the sources of foodborne illness from 954 restaurant outpatient test recording to mine the frequency of terms in dictionaries relating to foodborne illness events and restaurant cleanliness issues. Words for the dictionaries were selected using exploratory text mining and topic modeling from online reviews and through literature-based consultation with a foodborne illness expert. The frequency of key terms was recorded on the individual review level and on the restaurant level. Business intelligence techniques were used to identify trends in the data.

Results: Statistical analysis indicated there were statistically significant inverse correlations between increased frequency of key terms in both categories: average restaurant rating (foodborne illness = 5.367, cleanliness > 0.619) and individual review rating (foodborne illness > 37.324, cleanliness > 33.863). Of the 231,000 total reviews, 2.53% contained a key term relating to restaurant cleanliness issues and 0.764% contained a key term relating to foodborne illness.

Significance: The correlations between the frequency of key terms and restaurant reputation and customer satisfaction could be used to incentivize restaurateurs to implement safer food practices, as prior literature confirms the link between customer satisfaction and restaurant success.

P2-136 Evaluating FDA Food Recalls with Sanitation as a Root Cause

Amit Kheradia
Bacillus Cereus, Las Vegas, NV

Introduction: Cleaning and sanitation programs are a regulatory requirement in the food industry. A quarter of United States food recalls are caused by probable contamination arising from poor plant sanitation and environmental monitoring standards. The study, therefore, proposes that FDA’s Food Recall entries should clearly indicate such root causes for each recall, for the benefit of the industry and the public.

Purpose: To analyze FDA food recalls and use them as a tool to provide risk-based industry sanitation recommendations.

Methods: Three-hundred eleven relevant data points were evaluated from the FDA’s Recall entries from July 2017 to June 2018.

Results: Of the 231,000 total reviews, 2.53% contained a key term relating to restaurant cleanliness issues and 0.764% contained a key term relating to foodborne illness.

Significance: The correlations between the frequency of key terms and restaurant reputation and customer satisfaction could be used to incentivize restaurateurs to implement safer food practices, as prior literature confirms the link between customer satisfaction and restaurant success.

P2-137 Hand and Glove Surface Cross-Contamination Potential Based on Nitrile and Vinyl Glove Surface Characteristics

Barry Michaels, Christopgher Griffith1 and Stephen Ardagh2
1Michaels Group Inc., Palmita, FL, 2Broadway Hygiene Consultancy, Dorchester, United Kingdom

Introduction: Between 2015 and 2021, data assimilated indicates that glove-hand cross-contamination (CC) or bare-hand contact was a contributory cause for 20% to 40% of foodborne illness outbreaks and 30% of outbreaks, respectively.

Purpose: Recognition of the hazards posed by bare-hand contact and use of disposable gloves as a mitigation strategy is relatively recent, with mecha

Results: While CC potential for disposable gloves resides in glove type and SFE profile, transfer coefficients would be less than dirty bare hands but not necessarily for surfactant cleaned hands, with nitrile advantaged. Nitrile disposable gloves showed reduced CC potential as they are more hydrophobic & oleophobic (water & oil repellent) than tested PVC disposable gloves, correlated with lower transfer coefficients (pick-up-deposition). A SFE/surface tension continuum constructed with values for gloves, contact surfaces, hands, food and human soils helps to explain CC as experienced in food environments.

Significance: The study identifies some poorly understood factors related to CC involving nitrile gloves and disposable gloves, that together represent significant contributory causes in foodborne outbreaks linked to faulty food handling behaviors.
P2-138 The Development of a Multiple Hurdle Approach to Improve Microbial Safety of Ground Beef

Ranjith Ramanathan, Conner McDaniel, Sabra Billups, Divya Jaroni and Ravirajinessa Jadde

Purpose: To develop a multiple hurdle approach to improve microbiological safety of ground beef.

Methods: Beef trim (400 g) were spot inoculated with 200 µL of 7.02 log CFU/ml either E. coli ATCC 15717® or Salmonella Typhimurium DT 104. Inoculated beef trim were subjected to one of the seven spray treatments; no treatment (NT), deionized water (DW), three percent sodium chloride (SAS), five percent lactic acid (LA), Blitz (PAA), SAS followed by Blitz (15 ml SAS + 15 ml Blitz), and LA followed by Blitz (15 ml LA + 15 ml Blitz). The beef trim was treated then processed through a meat grader and inoculated into the inner surfaces of the equipment. One thousand g of antibiotic-free rib in a combination of 500 mL solution prepared from 325 mg PAA solution were processed through an inoculated meat grader. After the antimicrobial ice treatment, 400 g unoinoculated beef trim were processed and analyzed to identify bacterial transfer from the meat grinders.

Results: The SAS treatment was found to be most effective and reduced the pathogen transfer from meat grader to the detection limit, while all other spray treatments yielded statistically similar recoveries ranging from 1.9 to 1.44 and 1.99 to 1.64 log for E. coli ATCC 15717® and Salmonella Typhimurium DT 104, respectively. The effects of antimicrobial treatments on the color of ground beef were also investigated. Hue, chroma, and a* color analysis was determined over a five-day shelf life study. When looking at a* values, SAS + PAA reported the highest values at 21.75, just behind NT and DI. Similar trend was observed with chroma values with different treatment solutions.

Significance: This hurdle approach, especially with SAS has the potential to reduce cross-contamination and could serve as an easy and rapid antimicrobial intervention for the ground beef industry.

P2-139 Evaluation of a Commercial Enzymatic Drain Cleaner for Food Matrix Digestion

Stephanie Hice, Shalini Wijeratne, Joey Talbert and Byron Brehm-Stecher

Purpose: To evaluate the capacity of a commercial powdered enzymatic drain cleaner to physically digest ground turkey, with the ultimate goal of enabling simultaneous bacterial enrichment and reduction in sample complexity prior to downstream analysis.

Methods: A commercial drain cleaner (Deep Drain Defense) containing a mixture of bacterial sponges and enzymes (protease, lipase, amylase, cellulase) was used. The product was suspended in either buffered peptone water (BPW) or universal preenrichment broth at up to six times the recommended usage level, then filtered (0.22 µm) to remove bacterial sponges. Ground turkey (93% lean) was diluted 1:10 in each medium-enzyme mixture in sterile filter bags (50 ml) and preenriched at 37°C for up to four hours. Visual changes in ground turkey (integrity, color) were observed, compatibility of digested turkey filtrate with centrifugation was examined qualitatively and simultaneous matrix digestion and enrichment of Salmonella Typhimurium ATCC 14028 at a high inoculum (10^7 CFU/ml) was assessed. Multiple experiments were performed during development of this approach. All treatments within each experiment were performed in duplicate.

Results: Increasing concentration of drain product and longer incubation times led to decreased integrity of ground turkey. The BPW-enzyme filtrate was used as a control, while the undigested control, which formed compacted particulate plugs. Salmonella Typhimurium was enriched by two log in the presence of the enzyme product.

Significance: Our results suggest that this inexpensive household product may be repurposed for presample preparation of ground poultry.

P2-140 Microbial Analyses of Dried Crickets Used as a Human Protein Supplement

Jennifer Perry

University of Maine School of Food and Agriculture, Orono, ME

Purpose: To determine the potential utility of droped apples, lettuce, and peaches on S. marcescens contaminated commercial flour surfaces.

Methods: Apple, lettuce, and peach were dropped onto two different commonly used grocery floor surfaces (carpet, tile) that were artificially inoculated with S. marcescens at seven log CFU/ml for five s, min, 10 min, one, 45 min, and 4 h. Percent transfer was calculated and statistical analysis was performed for significant differences.

Results: A statistically significant difference (P<0.05) was observed for percent uptake between the produce types and surfaces. However, percent uptake was not significantly different among the five-time treatments used in this study. The carpet had 3.72% uptake while the tile had 0.65% uptake regardless of the produce type and dropped lettuce had the most uptake (5.07%) from both the surfaces combined followed by apple (0.8%) and peach (0.76%) regardless of surface type or contact time.

Significance: Increase in the uptake and picking up fresh produce at grocery stores can result in a food safety risk regardless of the contact time. Training is needed for employees and customers concerning the safe handling of fresh produce to enhance food safety and public health.

P2-141 Development of an Indirect Enzyme-Linked Immunosorbent Assay (ELISA) for the Rapid Detection of Peanut in Processed Foods

Manreet Bhullar*, Eric Wilhelmsen2, Christopher McGinnis1, Tom Myers3 and Florence Wu4

Purpose: To develop a validated ELISA method that can be used to detect peanut in process control data of known specificity and sensitivity.

Methods: A validated ELISA method based on BP 3A1-12 MAb can successfully detect peanut in processed foods. and this method can be used as an effective tool for monitoring peanut as a food allergen in various processed foods.

P2-142 Food Safety Risk Associated with Dropped Produce on Listeria monocytogenes-contaminated Floor Surfaces in Grocery Stores

Angela Shaw, Manreet Bhullar1, Anja Mage, Jacques Overdiep, Bridge Pittay, Lillian Nabwire and Niraja Shivalingah

Purpose: To develop a validated ELISA method that can be used to detect peanut in process control data of known specificity and sensitivity.

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Significance: Increase in the uptake and picking up fresh produce at grocery stores can result in a food safety risk regardless of the contact time. Training is needed for employees and customers concerning the safe handling of fresh produce to enhance food safety and public health.

P2-143 An Approach to Implementing the FDA Recommendation to Verify the Minimization of Contaminazation and/or Spread of Pathogens in Fresh Cut Processing Food Facilities

Angela Nunez1, Christopher McGinnis1, Eric Wilhelmsen2 and Jim Brennan1

Journal of Food Protection Supplement

Purpose: To develop a validated ELISA method that can be used to detect peanut in process control data of known specificity and sensitivity.

Methods: A validated ELISA method based on BP 3A1-12 MAb can successfully detect peanut in processed foods.

Results: A statistically significant difference (P<0.05) was observed for percent uptake between the produce types and surfaces. However, percent uptake was not significantly different among the five-time treatments used in this study. The carpet had 3.72% uptake while the tile had 0.65% uptake regardless of the produce type and dropped lettuce had the most uptake (5.07%) from both the surfaces combined followed by apple (0.8%) and peach (0.76%) regardless of surface type or contact time.

Significance: Increase in the uptake and picking up fresh produce at grocery stores can result in a food safety risk regardless of the contact time. Training is needed for employees and customers concerning the safe handling of fresh produce to enhance food safety and public health.

P2-144 Acidic Silver Pretreatment Can Greatly Reduce the Risk of an Illness Outbreak for Fresh Cut Leafy Greens

Jim Brennan1, Eric Wilhelmsen2, Christopher McGinnis1, Tom Myers3 and Florence Wu4

Purpose: To develop a validated ELISA method that can be used to detect peanut in process control data of known specificity and sensitivity.

Methods: A validated ELISA method based on BP 3A1-12 MAb can successfully detect peanut in processed foods.
Methods: Pilot plant studies with and without the pretreatment are used to measure the increased lethality of the proposed pretreatment against inoculated non-sporeforming and sporeforming pathogens. The effectiveness of this increased lethality was optimized by varying the various control parameters including acid content, sodium concentration and dwell time. Based on models built from case data study and product testing, the impact of increased lethality on outbreak risk can be estimated.

Results: The study shows that spray-drying with an acidic blend containing about 30 ppm silver ion with a pH of about 2.2 for about 3 min is a preferred pretreatment. Eight independent studies with inoculated product with tomato and iceberg lettuce indicate an overall 0.75–30 increase in lethality over a well-managed control. In these processes, all inoculations were with a mixed culture of generic E. coli and ranged from 10^2 to 10^8 CFU/g in inoculated produce. No sporulated contamination case studies, the potential increase in lethality can provide greater than a 95% reduction in outbreak risk to the extent that E. coli models pathogen behavior.

Significance: Processors need to be aware of innovations that are wearying the standards for the best practice protocols. The marketplace has little tolerance for processors who fail behind and therefore put consumers at greater risk. This pretreatment sets a performance benchmark that processors can use as a reference in evaluating their own processes.

P2-145 Growth Kinetics of Listeria monocytogenes, Shiga Toxin-producing Escherichia coli, and Salmonella enterica on Fresh-cut Produce Stored at 5, 10, or 22°C

Qinghao Zhao

University of Wisconsin-Madison, Madison, WI

Introduction: The pathogen growth on fresh-cut produce at retail is a concern. The FDA Food Code identifies cuttums, melons, and leafy greens as non-tolerable for retail food control (TCC) to ensure safety.

Purpose: We investigated the survival of Listeria monocytogenes, Shiga toxin-producing Escherichia coli (STEC), and Salmonella on cut cucumber, onion, tomato, pepper, tomato, and mango stored at five, ten, or 22°C.

Methods: Acid-adapted, single-pathogen cocktails, five to ten strains each, of stationary phase cells were inoculated. Fresh-cut produce was prepared from surface-sterilized whole produce, acceptably peeled and cut into cubes. Cut produce was weighed, 25 g/bag, and 0.25 ml of pathogen cocktail added and distributed by gently massaging. Each bag contained four log CFU/g. Inoculated bags were placed at five, ten, or 22°C for up to seven days, four days, or 32 hrs, respectively. A bag was removed, and surviving organisms enumerated. Native microbiota was enumerated from uninoculated samples at each point. Log surviving cells (CFUG) for each pathogen/produce combination was plotted vs time and curve-fitting applied (OMRI).

Results: Pathogen grew significantly at 22°C, greater than four log CFU/g, with the exception of L. monocytogenes on tomato and all three pathogens on mango. Similarly, pathogens grew significantly at 10°C, except that L. monocytogenes did not grow on cut tomato, and no pathogen grew on cut mango at either five or 10°C. While decreasing temperature significantly decreased growth rate, Salmonella and STEC grew greater than three log CFU/g on cut tomato at 5°C, and L. monocytogenes grew greater than one log CFU/g on cut cucumber.

Significance: TTC may reduce the risk of foodborne illness from fresh-cut onion, pepper, and mango, but TTC alone will not prevent the growth of L. monocytogenes on tomato or mango stored at five, ten, or 22°C.

P2-146 Comparison of Sodium Nitrite and Natural Celery Nitrite on the Inhibition of Spore Germination of Clastitrogen sporogenes As a Non-pathogen Surrogate Agitate in Meat Products

Dennis Fletcher, Jacob Nelson and Peter Muriana

Oklahama State University

Introduction: Nitrite is a regulated ingredient used to prevent the germination of Clastitrogen spores in processed meat products. The use of nitrite as a food preservative got a boost when USDA-FSIS considered vegetable-sourced nitrite (vegetable nitrate fermented to nitrite) as ‘natural nitrite’ whereby the use of nitrite as a food preservative got a boost when USDA-FSIS considered vegetable-sourced nitrite (vegetable nitrate fermented to nitrite) as ‘natural nitrite’ whereby the use of nitrite as a food preservative got a boost when USDA-FSIS considered vegetable-sourced nitrite (vegetable nitrate fermented to nitrite) as ‘natural nitrite’ whereby the use of nitrite as a food preservative got a boost when USDA-FSIS considered vegetable-sourced nitrite (vegetable nitrate fermented to nitrite) as ‘natural nitrite’. The nitrite validation assay described herein allow easy determination if nitrite levels can prevent spore germination under the most per-

Discussion: Problems with nitrite use are related to the fermentation process, where high concentrations or organisms that are resistant to nitrite can occur. A new nitrite validation assay described herein allows easy determination if nitrite levels can prevent spore germination under the most per-

P2-147 Combined Effect of Storage Conditions, Surface Integrity, and Length of Shelf Life on the Growth of Listeria monocytogenes and spoilage Microbiota on Refrigerated Ready-to-Eat Foods

Shi Cai1, Randy Woroboff2 and Abigail Snyder1
1The Ohio State University, Columbus, OH, 2Cornell University, Ithaca, NY

Introduction: Incipient data label practices reportedly contribute to food waste in the United States due to consumer misinterpretation. Proposals to utilize spoilage data from the risk by postharvest growth of Listeria monocytogenes in RTE foods.

Purpose: The purpose of this study was to evaluate the rate of quality deterioration and food safety risks related to one another in six product systems (tomatoes, apples, fresh-cut cantaloupe, lettuce leaves, baby spinach, and commercially processed turkey slices) at different refrigeration temperatures, atmospheres, and quality grades.

Methods: RTE food samples inoculated with three-strain cocktails of L. monocytogenes were stored in refrigerators at 41°F and 91°F in triplicate. The duration with which the pathogens survived was determined for the common shelf life duration of 24 h. Produce was carried out every 4 h for 12 days; apples, tomatoes, romaine lettuce leaves, and baby spinach leaves twice per week for 27 days; turkey slices once per month for 70 days. Both L. monocytogenes growth and spoilage microbiota were tested at each sampling point. A tenfold increase in L. monocytogenes levels was used as the cut-off for inactivation. Pathogens were considered non-inhibitory if viable or moribund. For pathogens A total of n=52 lettuce, n=52 tomatoes and n=52 cucumber samples were collected from informal markets in Cambodia.

Results: Generally, conditions that improved microbial quality by extending shelf-life also allowed for L. monocytogenes growth of greater than one log CFU/g before inoculation to microbial spoilage. Modified atmosphere packaging stored enhanced L. monocytogenes growth relative to spoilage microbiota. Using modified atmosphere packaging showed greater shelf-life and total yield (greater than one log growth after spoilage). In contrast, the use of secondary ify for shelf-life limitation across the surveyed RTE products.

Significance: These data suggest that spoilage cannot be considered a fail-safe indicator or proxy for shelf-life limitation across refrigerated RTE products.

P2-148 Growth and Survival of Listeria monocytogenes on Intact Fruit and Vegetable Surfaces: A Systematic Review

Claire M. Marik, Joyce Zuchel, Donald W. Schaffner1 and Laura K. Stirm

Wallingford, CT

Introduction: Listeria monocytogenes is known to be present in produce associated environments (e.g., fields, packinghouses); thus, it is critical to evaluate L. monocytogenes growth and survival data on intact whole produce surfaces. The goal of this study was to perform a systematic literature review to identify and characterize published data on the growth and/or survival of L. monocytogenes on fruit and vegetable surfaces.

Methods: Relevant studies were identified by searching seven electronic databases: AGRICOLA, CAB Abstracts, Center for Produce Safety, FSTA, Google Scholar, PubMed and Web of Science. Search terms were also modified, exploded, and blasted to find all related subheadings. Included studies had to be prospective, describe methodology (e.g., inoculation method, experimental parameters), and provide descriptive growth and/or survival data. Studies were not included if methods were unclear or inappropriate (e.g. dip inoculation may promote internalization), and if produce was cut, processed, or treated.

Results: Of 1,459 identified citations, 88 were reviewed in full and 29 studies met the inclusion criteria. Studies represented 21 commodities, with the majority of studies focusing on melons, leafy greens, berries, and sprouts. Synthesis of the reviewed studies suggests L. monocytogenes growth and/or survival on intact fruit and vegetable surfaces differs substantially by commodity. Parameters, such as temperature, relative humidity and produce surface characteristics had a considerable effect on L. monocytogenes growth and survival dynamics. Contaminated produce held at ambient temperatures (22°C) had higher growth rates and longer shelf-life compared to lower temperatures (4°C), and greater than one log difference in growth rate.

Significance: This review provides an inventory of the current data on L. monocytogenes growth and survival on intact whole produce surfaces. Identification of which intact whole produce commodities support L. monocytogenes growth and/or survival at various conditions observed along the supply chain will assist the industry in managing L. monocytogenes contamination risk.
Results: Significant to our knowledge, this is the first study that explores the biological cleanliness of fresh vegetables sold in informal markets and evaluated with biological hazards and that interventions are necessary to reduce the likelihood of contamination and negative public health effects. A significant reduction in the microbial load of vegetables was observed when comparing the unwashed and water-treated samples from days 50 to 70. 

Results: Cold plasma activation (ionization) of H\textsubscript{2}O\textsubscript{2} mist may be used to enhance microbial safety of fresh produce. 

Results: More citrus studies are needed to further validate the efficacies of cold plasma activation of H\textsubscript{2}O\textsubscript{2} treatment and the impact of different cold plasma treatments of H\textsubscript{2}O\textsubscript{2} mist. 

Results: Our results demonstrated that cold plasma activation (ionization) of H\textsubscript{2}O\textsubscript{2} mist may be used to enhance microbial safety of fresh produce. 

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Results: Cold plasma activation (ionization) of H\textsubscript{2}O\textsubscript{2} mist may be used to enhance microbial safety of fresh produce.
This study assessed the efficacy of a new sulfuric acid/surfactant sanitizer against Salmonella during simulated commercial washing of diced tomatoes. The sanitizer was applied to whole and fresh-cut produce at different storage temperatures.

**Introduction:**

The use of water for irrigation of food crops is uncommon in the Mid-Atlantic, grower interest is high, as long as water quality is reputable and access is provided. Comprehensive two-testing of three reclaimed water sites indicated that some systems require treatment before use as agricultural water. Combinable with on-farm treatment solutions or targeted use on crops rarely consumed raw, reclaimed water has promise as a valuable water source for Mid-Atlantic farmers.

**Methods:**

A 22-question survey was distributed online and in-person over two years (March 2016 to February 2018) to 269 growers in the Mid-Atlantic United States (Delaware, DC, Maryland, Pennsylvania, Virginia, West Virginia). Meanwhile, 12 nontraditional irrigation water sites were quantitatively assessed for generic E. coli using standard membrane filtration according to EPA method 1604 (n=227, collected October 2016 to October 2018).

**Results:**

Most Mid-Atlantic growers surveyed (59%) use groundwater as their primary water source, and 66% of farmers are at least somewhat concerned about their water availability. Willingness to use nontraditional water was significantly increased if the water quality was proven to be good or better than farmers’ current source (75% vs. 52%). Microrganisms water quality profiles developed over two years during the growing season showed that eight of 12 nontraditional irrigation water sources tested would require treatment before unrestricted use under the FDA’s proposed Produce Safety Rule (three of four freshwater rivers, two of two brackish rivers, zero of two ponds, two of three reclaimed water sites, one of one processing water). Significance: Although use of reclaimed water for irrigation of food crops is uncommon in the Mid-Atlantic, grower interest is high, as long as water quality is reputable and access is provided. Comprehensive two-testing of three reclaimed water sites indicated that some systems require treatment before use as agricultural water. Combinable with on-farm treatment solutions or targeted use on crops rarely consumed raw, reclaimed water has promise as a valuable water source for Mid-Atlantic farmers.

**Significance:**

Today, fresh produce growers are concerned with water quality both to protect public health and to comply with buyer requirements and long-term sustainability. The current study demonstrates that long-term sustainability of agricultural production may rely on finding alternatives to groundwater, such as surface water and reclaimed water.

**Conclusion:**

This study juxtaposes grower perceptions with a two-year profile of generic E. coli from nontraditional irrigation water sources in the Mid-Atlantic.
higher. E. coli reduction than the other two sprays (P<0.05). In the radish and broccoli seeds, no significant difference in cell reduction was observed between the different sprays. The germination rates in all seeds were above the federal standard, however, the 0.2% Ca2+ spray significantly lowered the yield of broccoli sprouts.

Significance: Anti-microbial sprout is an effective method for reducing the microbial counts on sprouting seeds; however, the effectiveness could be seed type specific.

P2-163 Washing Techniques to Reduce Microbial Growth Using Different Sanitizers on Fresh Lettuce
Prachi Pahariya1, Rugpal Choudhury2 and Derek Fisher3
1Southern Illinois University, Carbondale, IL, 2Southern Illinois University - Carbondale, Carbondale, IL, 3The Ohio State University, Columbus, OH

Methods: Lettuce (Lactuca sativa cv. Romaine or Lactuca sativa cv. Oakleaf) were grown hydroponically in a series of experiments. Lettuce leaves were collected at seven time points during the plant life cycle for pathogen quantification. Plant health parameters (weight/size and color) were determined on fresh leaves. Leek (Allium fistulosum) were collected from four sites across the recirculation system. Sampling was repeated four times. Samples were culturally enumerated for aerobic mesophiles, coliforms, L. monocytogenes, L. innocua, and Enterobacteriaceae. A bacterial cocktail containing three outbreak strains of E. coli O157:H7 (Newport, E. coli O157:H1, and E. coli O157:H15) was added to the solution.

Results: A bacterial cocktail containing three outbreak strains of E. coli O157:H7 (Newport, E. coli O157:H1, and E. coli O157:H15) was added to the solution.

Significance: The results support that this is a cost-effective method to inactivate the specific pathogens in fresh produce.

P2-164 Survivorship of Listeria monocytogenes in Hydroponic Lettuce Systems
Margaret R. Moodispaw1, Carlos Saint-Pe2, Vishal Sinhastaria1, Melanie L. Lewis3,4 and Sanja Ilic1
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Methods: Three varieties of hydroponic fertilizer solutions (strawberry, tomato, and lettuce) were used to evaluate survival of L. monocytogenes and L. innocua in conditions for irrigation water. L. monocytogenes was inoculated at 6.35 log CFU/g before storage. It was found that L. monocytogenes survived at steady rates in the hydroponic system for the first 24 h post inoculation. The survival rates were similar in the hydroponic fertilizer solutions and pH levels were increased significantly (P<0.05). In the radish and broccoli seeds, no significant difference in cell reduction was observed between the different sprays. The germination rates in all seeds were above the federal standard, however, the 0.2% Ca2+ spray significantly lowered the yield of broccoli sprouts.

Significance: Significant differences in bacteriological persistence and survival were observed among the different pH levels in the hydroponic system. The persistence of coliforms across all samples indicates a potential hazard of hosting of pathogenic Enterobacteriaceae. P2-166 Examination of the Growth and Survival of Listeria monocytogenes in Hydroponic Fertilizer Solutions Maintained at Different pH Levels
Prachi Pahariya, Rugpal Choudhury and Derek Fisher
1Southern Illinois University, Carbondale, IL, 2Southern Illinois University - Carbondale, Carbondale, IL

Methods: Lettuce leaves were collected at seven time points during the plant life cycle for pathogen quantification. Plant health parameters (weight/size and color) were determined on fresh leaves. Leek (Allium fistulosum) were collected from four sites across the recirculation system. Sampling was repeated four times. Samples were culturally enumerated for aerobic mesophiles, coliforms, L. monocytogenes, L. innocua, and Enterobacteriaceae. A bacterial cocktail containing three outbreak strains of E. coli O157:H7 (Newport, E. coli O157:H1, and E. coli O157:H15) was added to the solution.

Results: A bacterial cocktail containing three outbreak strains of E. coli O157:H7 (Newport, E. coli O157:H1, and E. coli O157:H15) was added to the solution.

Significance: Lettuce leaves contaminated with L. monocytogenes at 100 ppm: sodium hypochlorite (SH), acidified sodium chloride (ASC), or peroxyacetic acid (PAA). All the experiments were performed under sterile conditions, and repeated three times.

Results: The initial microbial count for E. coli (2.9 log CFU/g) and for L. monocytogenes (0.35 log CFU/g) was obtained before storage. It was found that E. coli growth increased by 2.15 log CFU/g at room temperature, whereas L. monocytogenes increased at both room and refrigerated temperatures by 1.69 log CFU/g and 0.81 log CFU/g respectively after 24 h of storage. The results show maximum microbial reduction for E. coli (1.36 log CFU/g at room temperature, 4.42 log CFU/g at refrigerated temperature) and for L. monocytogenes (3.33 log CFU/g at room temperature, 3.46 log CFU/g at refrigerated temperature) was obtained with ASC. In the maximum reduction with ASC (99.99%) after 3 min. In the case of PAA (room temperature, 1.08 CFU/g at refrigerated temperature) using ASC while the maximum reduction for L. monocytogenes (1.67 CFU/g at refrigerated temperature, 2.14 CFU/g at refrigerated temperature) by using SH.

Significance: Soaking of fresh lettuce with PAA sanitizer could be an alternative and effective method to achieve microbial reduction in lettuce by the fresh-produce industry.
P174 Evaluation of Viability of Escherichia coli O157:H7 and Listeria monocytogenes on Sanitizer-treated Spinach Leaves Using Combined Propidium Monoazide Staining and Quantitative PCR
Vijay Singh Chettri, Yu Han, Marlene Janes and Achyut Adhikari
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Purpose: Bacterial pathogens can enter into a viable but non-culturable (VBNC) state due to sanitizer residues on produce surfaces. Conventional methods have limitations in detecting VBNC cells. Viability PCR could help discriminate live pathogens on produce surfaces providing important information from a public health risk perspective.

Methods: This study was conducted to demonstrate the viability of E. coli O157:H7 and L. monocytogenes on chlorticide- and atrazine-treated spinach surfaces during refrigerated storage.

Results: Baby spinach leaves were washed with chlorinated water (100 ppm) or lactic acid (0.5%) for 3 min, and spot inoculated with a cocktail (three strains) of E. coli O157:H7 and L. monocytogenes. The viability of the pathogens during refrigerated storage (4°C) was examined for 48 h by staining the cells with propidium monoazide followed by a real-time PCR analysis. The difference between the amount of total and live-cell-derived DNA was assessed by analyzing the area under the curve of the corresponding bacterial counts.

Results: The number of live E. coli O157:H7 and L. monocytogenes cells decreased with time during refrigeration on the spinach leaves (treated and control). The proportion of dead E. coli O157:H7, one week after 48 h of storage was 1.24, 2.80, and 3.54 log CFU on distilled water, chlorine, and atrazine treated samples, respectively. Similarly, the proportions of dead listeria monocytogenes cells were 1.21, 2.61, and 3.44 log CFU on distilled water, chlorine and atrazine treated leaves, respectively.

Significance: The results indicated that the residual sanitizers could have a role in reducing the number of viable human pathogens on produce surfaces during refrigerated storage.

P174 Sodium Bisulfate and Peroxyacetic Acid Reduce Escherichia coli O157:H7 Populations on Fresh Lettuce When Applied Alone or in Combination as a Postharvest Wash
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Phytophthora bacteria on fresh-cut romaine is a major concern for the lettuce industry. This study evaluated the potential of sodium bisulfate (SBS) and peroxyacetic acid (PAA) to reduce or prevent the growth of Escherichia coli O157:H7 (E. coli) on fresh-cut lettuce.

Methods: Fresh lettuce leaves were inoculated with E. coli O157:H7 using a virulent strain, ES-61. E. coli O157:H7 populations on fresh-cut romaine lettuce were enumerated on days zero, one, three, five, 10, 12, and 14 by homogenizing 25 g in 225 mL of 0.05% Tween 80 wash broth, diluting in 0.5% peptone water, and plating on Sorbitol MacConkey supplemented with cefotaxime and colistin. E. coli O157:H7 colonies were counted following 37°C incubation for 18 to 24 h.

Results: Treatment was a statistically significant variable (P = 0.001), with SBS, PAA, and SBS-PAA significantly more effective than chlorine at reducing E. coli O157:H7 populations on romaine lettuce. The low dose (0.5%) of SBS and SBS-PAA postharvest washes were effective chlorine alternatives for reducing E. coli O157:H7 on fresh-cut romaine.

P175 Assessing the Role of Phytophthora Bacteria on Norovirus Stability and Attachment in Romaine Lettuce
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Introduction: Human norovirus (HuNoV) is a leading cause of outbreaks in produce. HuNoV was shown to bind bacterial expressing histo-blood group antigens (HBGAs), the putative HuNoV receptor. Therefore, phytophthora bacteria which express HBGAs may play a role in HuNoV persistence in leafy vegetables.

Methods: The role of phytophthora bacteria on norovirus stability and attachment in romaine lettuce was studied. Lettuce leaves were inoculated with fresh-cut romaine lettuce at a titer of 10^5 PFU and stored at 26 to 4°C. On post-inoculation zero, one, two, three, and seven, samples were rinsed with 0.1% peptone water and homogenate was plated on Trypticase Soy Agar (TSA) and MacConkey Agar (MCA). Subsequently, homogenate was centrifuged at 3000 rpm for 30 min to collect supernatant for viral plaque assays and RT-PCR. 16S RNA gene sequencing was used to select bacteria isolates.

Results: At seven days, 4.6 log10 PFU lettuce was detected in whole lettuce stored at 4°C. This titer was significantly higher (P = 0.001) compared to other groups and storage temperatures. Sequencing results showed Sheavesella spp was unique to this group at three and seven days overall. The titer was...
lower in stores 26% compared to 4°C (P <0.01) at seven d. However, there was no significant difference in bacterial counts at seven d and the control was approximately 1.6×10^3 CFU/ml.

**Significance:** This work indicates storage temperature may be a strong factor for TV stability on produce over time while cultural bacterial density may not affect TV stability on romaine lettuce. However, TV stability was enhanced in whole head lettuce and therefore, a specific bacteria or bacterial population may be responsible for enhanced TV stability. Sequencing results showed Shewanella spp. were common to this group at three and seven d, which suggests Shewanella spp. may specifically interact with HuNoV and enhance persistence in romaine lettuce.

P2-176 Effect of a Bacteriophagescocktail against Salmonella enterica on Romaine Lettuce Leaves Catherine Wong, Styun Wang and Pascal Delaquis*  
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**Developing Scientific Entrepreneurs**

Methods: If samples were contaminated by Salmonella enterica, fresh produce has been linked to produce microbial populations by >90%. Consequently, bacteriophage has been proposed as an alternative method to control bacterial populations in fresh produce.

**Purpose:** The primary purpose of this study was to determine the effect of a bacteriophage cocktail on the fate of three S. enterica strains on romaine lettuce.

**Methods:** Romaine lettuce leaves were cut into four cm2 pieces and separately inoculated by spotting with (i) one of three S. enterica strains (Salmonella Sfaduo P245 or S250 or Salmonella Typhimurium S441) alone, (ii) the S. enterica strain plus 0.1 M CaCl2 solution, and (iii) the S. enterica strain plus a cocktail of five bacteriophages lytic to S. enterica in a 1: M CaCl2 solution. Salmonella populations were measured on XLD agar immediately after inoculation and after one and two days of incubation at 21°C. Three independent replicates were performed for each strain.

**Results:** Populations of all three strains increased by one log CFU/ml or less than one log CFU/ml when untreated lettuce after two days of incubation at 21°C.

**Application** of the bacteriophage cocktail reduced Salmonella populations but the effect was strain-specific. Populations of strains Salmonella Sfaduo S245 and S250 were reduced by four log CFU/ml (P<0.05) immediately after inoculation and the difference with untreated controls was maintained over two days of incubation at 21°C. In addition, populations of strain Salmonella Typhimurium S441 were reduced by two log CFU/ml (P<0.05), however, the difference with controls was also maintained over two days of incubation.

**Significance:** The results of this study support strain-specific differences in the susceptibility of S. enterica that should be considered in the development of bacteriophage-based control methods for fresh produce.

P2-177 Methods to Differentiate Presumptive Pseudomonas on Cabbage Lettuce Head Thomas De Bock, Jelena Jovanovic, Andreja Rajkovic, Monica Hofter and Mieke Uytendaelde  
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**Introduction:** Pseudomonas fluorescens is a naturally occurring, soil-dwelling bacterium, where some strains are used as a biological insecticide, but which is closely related to the human pathogenic Pseudomonas aeruginosa.

**Purpose:** The purpose of this study was to evaluate different methods for identification and differentiation of presumptive P. aeruginosa on butterhead lettuce and cabbage leaves.

**Methods:** Fifteen butterhead lettuce samples were plated on MYP-agar and other chromogenic media. Next, different analytical techniques for identification and differentiation at the species level were compared on 20 isolates (isolates from culture collection): 16 P. aeruginosa group strains, two B. subtilis strains, one Pseudomonas syringae pv. syringae and one B. circulans strain, and on 46 isolates from the sampled butterhead lettuce of which 24 were presumptive P. aeruginosa isolates. These techniques include colony morphology, PCR on target genes, immunological techniques for toxin production, psychrotrophic character and phase-contrast light microscopy for the observation of parasporal crystals.

**Results:** Presumptive P. aeruginosa counts of less than two log CFU/ml to a maximum of 4.04 log CFU/ml were found. B. thuringiensis could not be differentiated from other members of the same genus nor from other bacteria in this study. PCR at the species level on the 20 presumptive P. aeruginosa isolates identified 13 different species. Gene sequences of Pseudomonas were identified in this study.

**Significance:** B. thuringiensis can overcome this problem.

P2-178 Salmonella enterica Colonization of Kale Leaves is Age- and Drought Stress-dependent Xuchun Li and Shirley A. Micalef  
*University of Maryland, College Park, MD

**Introduction:** Juvenile kale leaves have become a popular fresh salad ingredient, lack susceptibility to Salmonella is underestimated. Moreover, extreme weather events frequently cause periods of drought, which constitute plant stress that may also affect Salmonella colonization.

**Purpose:** Evaluate the effect of plant developmental stage and drought on Salmonella colonization of kale surfaces.

**Methods:** A total of 100 kale plants were grown on four, three, four, five, six, seven, or eight weeks post germination in a greenhouse (23°C, 16 L:8 D) before being subjected to drought for two to six days, including regular watering (control). Salmonella Newport adapted for rifampin was grown overnight on tryptic soy agar (TSA) at 35°C, and 10 CFU Salmonella were inoculated onto the adaxial side of the third true leaf of plants. Inoculated leaves were dipped 24 h post-inoculation, washed in 10 ml 0.1% peptone water, and serially plated onto TSA with rifampin. Leaf surface washes from 26-39 day-old control and drought-treated plants were collected by placing plants 5 m high and shaken at 150 rpm for 24 h. 200 ml aliquots were filtered twice, centrifuged and inoculated with 100 µL sterile Salmonella for growth curve analysis.

**Results:** In the control group, 20-day-old plants supported significantly higher Salmonella growth than 59-day-old plants (P<0.05). Drought significantly decreased Salmonella colonization in 26-29 day-old plants, with higher counts recovered from control (P<0.05). As plants aged, the drought did not cause further restriction of growth. Growth was significantly greater in washes collected from 26- and 33-day-old control than drought-treated plants, but the reverse was observed in 39-day-old drought-treated plants (P<0.05).

**Significance:** Baby kale leaves are susceptible to Salmonella colonization. Salmonella restriction on drought-treated juvenile plants is possibly explained by enhanced exudation of inhibitory phyto-compounds.
P2-182 Different Soil Contamination Levels of Salmonella Newport Influence Internalization during Pepper Transplanting

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Virginia Tech - Eastern Shore AREC, Painter, VA, Virginia Tech, Blacksburg, VA

Developing Scientist Entrant

Introduction: Internalization of pathogens in producer is a food safety concern due to difficulties eliminating pathogens during post-harvest handling. Prior literature has indicated Salmonella may internalize during certain production practices, like transplanting.

Purpose: The objective of this study was to investigate the Salmonella internalization in pepper transplanting under different levels of soil contamination contamination.

Methods: Pepper plants grown in plugs were transplanted into soil inoculated with Salmonella Newport at one of three different contamination levels: high, medium, and low (6.0, 6.0, 5.0, and 2.8 log CFU/g, respectively). Each treatment was replicated 10 times in a completely randomized design with five replicates per treatment. The pepper plants were transplanted into soil and watered with a nutrient solution. The plants were sampled at 42°C to further enhance selectivity; ii) mBPWp with CV held at 42°C with shaking and iii) mBPWp with CV held at 42°C without shaking.

Results: No Salmonella was recovered from any leaf sample (zero of 108), regardless of Salmonella contamination level (high, medium and low) in the soil. In low Salmonella contaminated soils, Salmonella internalized in root (15 of 36) and stem (six of 36) samples. Approximately 2.2 and 0.8 log CFU/stem and stem, respectively, of Salmonella was internalized within 24 h of transplanting in soils with high Salmonella contamination. In medium Salmonella contaminated soils, Salmonella internalized in root (18 of 36) and stem samples. Each average 0.8 log CFU/stem was internalized in soils with medium Salmonella contamination. In low Salmonella contaminated soils, no Salmonella internalized in root and stem samples (zero of 72).

Significance: Internalization was highly dependent on level of soil contamination, as soils containing higher Salmonella populations observed significantly more Salmonella internalized in root and stem plant sections. Additionally, Salmonella internalization was observed within 24 h post-transplantation. Therefore, it is recommended preventive practices should be adopted at limiting Salmonella populations in soil, or (ii) immediately immediately following transplant to reduce internalization risk.

P2-183 Improving the Microbial Safety of Sprouts Using Lactic Acid Bacteria Cultures

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Developing Scientist Entrant

Introduction: Over the last decade, sprout-related foodborne outbreaks attributed to Salmonella and Shiga toxin-producing E. coli (STEC) have become a persistent public health concern. Based on epidemiological evidence, seeds used for sprouting were identified as the primary source of pathogens. Consequently, various strategies have been adopted to reduce Salmonella and STEC in sprout irrigation water.

Purpose: This study investigated the application of lactic acid bacteria (LAB) as potential biocidal agents to control pathogens on seeds and sprouts.

Methods: Alfalfa seeds were inoculated with a cocktail of five Lactic acidophilus, L. plantarum, Lactobacillus acidophilus, Lactobacillus rhamnosus, and Lactobacillus bulgaricus. The seeds were then either stored or set up for germination at 25°C. Stored seeds and germinating sprouts were sampled at regular intervals to enumerate surviving pathogen and LAB populations. Data were analyzed using the PROC MIXED sub-routine of SAS v.9.3.

Results: LAB significantly reduced pathogen populations in seeds and germinating sprouts (P<0.05) followed by spray application of LAB cultures (seven log CFU/g of Lactobacillus acidophilus, L. plantarum, L. rhamnosus or Streptococcus lactis var. Dialac- tus) prior to transplanting in soils with high pathogen contamination and sprout outgrowth during spraying is critical to ensure the microbial safety of sprouts.

Significance: Application of LAB on seeds prior to storage or as a pre-germination spray can serve as a practical and feasible antimicrobial intervention strategy to improve the microbial safety of sprouts.

P2-184 Procedures for Improved Detection and Isolation of E. coli O157:H7 from Artificially Contaminated Sprout Irrigation Water

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Introduction: E. coli O157:H7 has been implicated in foodborne disease outbreaks with sprouted seeds. Spent irrigation water (SIW) has been tested in multiple studies to reduce internalization risk.

Methods: Microbial populations to below detection limits by <4 in germinating sprouts and 4 in stored seeds. However, approximately five log and 8 log CFU of the pathogen was recovered from seed and sprouts, respectively. Additionally, LAB application did not impede seed germination and sprout outgrowth.

Significance: Application of LAB on seeds prior to storage or as a pre-germination spray can serve as a practical and feasible antimicrobial intervention strategy to improve the microbial safety of sprouts.

P2-185 Determining Water Quality and Bacterial Load on Tomatoes from Florida Packhouse Operations

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Developing Scientist Entrant

Introduction: Monitoring and maintaining water quality in flume tanks is crucial to prevent cross-contamination during postharvest washing of tomatoes. Water quality data for inlet, outlet, and inside tanks were collected with the intention of optimizing resource and energy efficiency of organic packhouse operations.

Purpose: The main objective of this study was to monitor water quality in flume tanks and evaluate efficacy of postharvest washing of tomatoes in commercial packhouses.

Methods: Fresh tomatoes (n=3), both before and after washing, were collected on an hourly basis from four packhouses in Florida and analyzed for total aerobic plate count (APC), total coliforms (TC) and generic E. coli (EC). APC was determined using plate count agar, TC and EC were determined using CDC methods. Additionally, three flume water samples were collected and tested for APC, TC and EC. Simultaneously, flume tank water samples (n=5) were collected and analyzed for temperature, pH, total dissolved solids (TDS), free chlorine, chemical oxygen demand (COD), oxidation-reduction potential (ORP), and turbidity.

Results: The average APC from four packhouses were 6.2 log CFU/mL and six log CFU/mL for pre- and post-wash tomatoes, respectively. The average TC was 4.7 log CFU/mL and 3.91 log CFU/mL for pre- and post-wash tomatoes, respectively. No EC was detected from any tomato samples. APC for water samples had an average of 5.48 log CFU/100 mL and TC average of 1.0 log CFU/100 mL of water samples were also negative for EC. Turbidity, COD, and TDS in flume water increased over time in all packhouses. There was a correlation between COD and turbidity (r=0.96), and COD and TDS (r=0.95). No strong relationship was seen between ORP and chlorine (r=0.74).

Significance: There was no significant effect (P>0.05) of postharvest washing on microbiological qualities of tomatoes. Water quality in flume tanks deteriorated over time in all packhouses during a typical operational day of four to eight h.

P2-186 Investigating the Prevalence, Persistence, and Diversity of Listeria monocytogenes and Listeria Species in Produce Packhouses

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Developing Scientist Entrant

Introduction: Listeria monocytogenes has emerged as a food safety concern for a number of produce commodities. While L. monocytogenes contamination can occur throughout the supply chain, contamination from the packhouse environment represents a particular challenge and has been linked to recalls and outbreaks.

Purpose: The purpose of this study was to determine the prevalence and persistence of L. monocytogenes and Listeria species in produce packhouses.

Methods: A longitudinal study was conducted in 11 produce packhouses (commodities included mini-green, peach, apple, tomato, broccoli, and cucumber) in three states. In each packhouse, 34 to 44 sites representing zones two to four were selected and swabbed. Packhouses were visited 10 times over the growing season; sites were divided into four zones. Sites were swabbed with Food and Drug Administration’s Bacteriological Analytical Manual methods. Listeria-positive isolates were confirmed by PCR. Species and allelic type (AT) were identified by sigT sequencing for up to four isolates per sample.

Results: Among 1,584 samples tested, 50 (3.2%), 42 (2.7%), and 10 (0.6%) samples were positive for L. monocytogenes, Listeria spp. (excluding L. monocytogenes), and both L. monocytogenes and Listeria spp., respectively. Five different species of Listeria (monocytogenes, innocua, seeligeri, welshimeri, and multituber) were identified in the packhouse environment. The L. monocytogenes was the most prevalent species. The 102 Listeria-positive samples yielded 138 representative isolates. Representative isolates were identified to the species level. Aliquots of 212 Listeria spp. were screened in a sub-array of 12 high throughput assays. A high AT diversity (0.93 Simpson’s Diversity Index) was observed amongst Listeria isolates. There were 18 instances of Listeria monocytogenes or Listeria spp. repeated isolation (site testing positive more than two times). Upon analysis of sub-type data, only three sites tested positive for the same Listeria AT more than two times.

Significance: Data showed in this longitudinal study that Listeria prevalence and persistence in packhouses was low (less than four percent prevalence). Therefore, sanitation program development and implementation in packhouses is critical to limit Listeria harbourance and resistance.

P2-187 Detecting Listeria monocytogenes in a Variety of Individually Quick-Frozen Vegetables Using the BAX System Real-time PCR Assay

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Introduction: Listeria monocytogenes is typically associated with processed RTE foods, meats and dairy products but in recent years, numerous sporadic cases of illness have been documented in low-risk foods including intact fruit and vegetables. In 2018, one of the latest outbreaks involved packaged individual quick frozen (IQF) vegetables.

Purpose: Since L. monocytogenes can survive freezing temperatures, there are increased food safety concerns in frozen products. For this reason, a re-evaluation of the BAX System for L. monocytogenes was evaluated in a variety of frozen vegetables to minimize the risk that contaminated product will reach consumers.

Methods: Four frozen vegetables; broccoli, carrots, corn, and peas, were thawed and divided into 125-g samples for the test method and 25-g samples for the reference method. Samples were homogenized in buffered peptone water. A positive control for L. monocytogenes was set up according to the CDC methods. Additionally, samples were left unincubated for negative controls. All samples were held at 22°C for two weeks and thawed before enrichment. Test method samples were homogenized with 1125 ml of 24 Lobe Complete media and incubated at 35°C for 24 to 48 hours. Sample aliquots were then analyzed using real-time PCR. The reference method samples contain 125 ml of 24 Lobe Complete media and incubated for 24 hours at 35°C.

Results: For the low inoculum level, the real-time L. monocytogenes PCR assay returned positive results for eight samples of broccoli and corn, 10 samples of carrots, and 11 samples of peas. All presumptive results were identical to culture. When compared to the reference method, POD analysis indicated no significant difference (P>0.05) of postharvest washing on microbiological qualities of tomatoes. Water quality in flume tanks deteriorated over time in all packhouses during a typical operational day of four to eight h.
P2-188 A Multi-regional Prevalence and Persistence of Four Foodborne Pathogens in Manured Soils in Certified Organic Farms


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Introduction: Animal manure is commonly used to improve soil fertility and is particularly important in organic agriculture. Crops grown in soils amended with raw manure may become contaminated by microbial pathogens and fecal indicators (e.g., generic E. coli) present in manure. Reducing the risk of microbial contamination of crops is based on wait-times between manuring and cropping. Proposals include treating the soil by introducing a neutral pH to 4.0, intermittent dosing of hypochlorite, and use of biochar. Methods: The objective of this study was to develop treatment parameters for achieving similar log reductions on blueberries. The decontamination was most effective against the pathogens on baby-cut carrots, to which 600 and 2000 ppm-h was needed for achieving similar log reductions on blueberries. The decontamination was most effective against the pathogens on baby-cut carrots, to which 600 and 2000 ppm-h was needed for achieving similar log reductions on blueberries. Results: Generally, the increase in cumulative ClO2 exposure increased the log reductions of pathogens. The cumulative exposures of 1000, 1800, and 2000 ppm-h caused 3.3, 6.8, and 8.7 log reductions of Salmonella spp., respectively. For achieving similar log reductions, the decontamination was most effective against the pathogens on baby-cut carrots, to which 600 and 2000 ppm-h was needed. Significance: This study identified the treatment parameters for using NaOCl-acid-generated ClO2 gas for surface decontamination of tomatoes, blueberries, and baby-cut carrots to reduce the microbial food safety of these products.

P2-189 A Multi-longitudinal Study of Generic E. coli Persistence in Soils Amended with Raw Manure and Potential Transfer to Produce

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Introduction: Animal manure is commonly used to improve soil fertility and is particularly important in organic agriculture. Crops grown in soils amended with raw manure may become contaminated by microbial pathogens and fecal indicators (e.g., generic E. coli) present in manure. Reducing the risk of microbial contamination of crops is based on wait-times between manuring and cropping. Proposals include treating the soil by introducing a neutral pH to 4.0, intermittent dosing of hypochlorite, and use of biochar. Methods: The objective of this study was to develop treatment parameters for achieving similar log reductions on blueberries. The decontamination was most effective against the pathogens on baby-cut carrots, to which 600 and 2000 ppm-h was needed for achieving similar log reductions on blueberries. The decontamination was most effective against the pathogens on baby-cut carrots, to which 600 and 2000 ppm-h was needed. Results: Generally, the increase in cumulative ClO2 exposure increased the log reductions of pathogens. The cumulative exposures of 1000, 1800, and 2000 ppm-h caused 3.3, 6.8, and 8.7 log reductions of Salmonella spp., respectively. For achieving similar log reductions, the decontamination was most effective against the pathogens on baby-cut carrots, to which 600 and 2000 ppm-h was needed. Significance: This study identified the treatment parameters for using NaOCl-acid-generated ClO2 gas for surface decontamination of tomatoes, blueberries, and baby-cut carrots to reduce the microbial food safety of these products.

P2-190 Salmonella Detection Via Immunomagnetic Separation and Liquid Crystal Technology

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Introduction: Liquid crystal technology is designed to improve critical run times and sensitivity while increasing capability for detection of specific foodborne pathogens. Matrix limit of detection and validation studies were conducted in apples by using immunomagnetic separation and liquid crystal technology. Results: Samples of mug bean sprout rinse water (375 g) were inoculated with a low level of Salmonella. Samples for the test method were enriched in one liter of pre-warmed primary medium, transferred to secondary medium, evaluated with test strips and plated on selective agar. Samples for the FDA BARM reference method were enriched in 3.375 L lactose broth, transferred to SS and RV media, and plated on selective agar. Non-inoculated samples were run with both methods as negative controls. Results: There was a clear statistical difference between the test method and the FDA BARM reference method (P<0.02). The test method demonstrated 100% specificity and 100% sensitivity when inoculated at one cell per plate. There were no false negative results observed with the inoculated samples and no false positive results observed with the non-inoculated negative control samples. Significance: The test method detected Salmonella spp. in rinse water with 100% specificity and sensitivity. This test method allows for reduced enrichment volume which saves time, money and incubation space. As such, RapidCheck provides users with a rapid, reliable, cost-effective tool for monitoring Salmonella spp. in the rinse water stream.

P2-191 Role of the Dormant State in the Persistence and Resistance of Shiga Toxin-producing Escherichia coli in the Fresh Produce Chain

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Introduction: Shiga toxin-producing E. coli (STEC) are a major cause of foodborne illness in Canada and the United States. The pathogen’s ability to persist in the soil pre-harvest environment is a significant challenge to food safety. Methods: A longitudinal, multi-regional study was conducted on seventeen certified organic farms in four United States regions (nine in California, three in Maryland, three in Maine, and two in Minnesota). Results from this study provide multi-regional baseline data relating to current NOP wait-time rules and thus a framework for risk mitigation strategies to reduce pathogen persistence. Significance: The perusal state in STEC needs to be considered when assessing the survival and resistance of the pathogen group within the fresh produce chain.

P2-192 Detection of Low Levels of Salmonella spp. in Sprout Rinse Water Using the RapidCheck Select Salmonella Test

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Introduction: Contaminated seeds have been identified as a major source of sprout-associated pathogens outbreaks. While seeds are decontaminated prior to seedling development, the grower has no control in the pre-harvest step to reduce or eliminate pathogens in raw sprouts. Testing for the presence of Salmonella spp. in sprout rinse water will ensure that the released product is safe for human consumption.

Methods: RapidCheck Select Salmonella vs. FDA BARM

Samples of mug bean sprout rinse water (375 g) were inoculated with a low level of Salmonella. Samples for the test method were enriched in one liter of pre-warmed primary medium, transferred to secondary medium, evaluated with test strips and plated on selective agar. Samples for the FDA BARM reference method were enriched in 3.375 L lactose broth, transferred to SS and RV media, and plated on selective agar. Non-inoculated samples were run with both methods as negative controls. Results: There was a clear statistical difference between the test method and the FDA BARM reference method (P<0.02). The test method demonstrated 100% specificity and 100% sensitivity when inoculated at one cell per plate. There were no false negative results observed with the inoculated samples and no false positive results observed with the non-inoculated negative control samples. Significance: The test method detected Salmonella spp. in rinse water with 100% specificity and sensitivity. This test method allows for reduced enrichment volume which saves time, money and incubation space. As such, RapidCheck provides users with a rapid, reliable, cost-effective tool for monitoring Salmonella spp. in the rinse water stream.
P2-194. Antimicrobial Effect of Natural Fruit Extracts against Salmonella on Cucumbers

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Purpose: The purpose of this study was to investigate the efficacy of benzyl isothiocyanate (BIT), a cruciferous vegetable-derived compound, for controlling Salmonella on fresh cucumbers.

Methods: The experiment was conducted under greenhouse conditions using three apple varieties (Braeburn, Fuji, and Gala). Cucumbers were dipped in a mixture of 10,000 ppm BIT and 25 ppm chlorine for 1 min. Total chlorine residual was determined on the cucumbers after washing. Inoculated cucumbers were stored at controlled temperatures for seven days. At the end of the storage period, cucumbers were swabbed with a sterile cotton swab, and one log CFU/ml of Salmonella serotype Newport was inoculated on NAC, MRS, and sorbitol MacConkey agar plates in triplicate. The total number of Salmonella colonies was counted after incubation at 37°C for 24 h.

Results and Discussion: There was no significant difference in Salmonella population among the treatments (P>0.05). The control cucumbers showed a significant increase in Salmonella population compared to the treated cucumbers (P<0.05). The results suggest that BIT at 10,000 ppm was effective in reducing Salmonella populations on fresh cucumbers when stored at controlled temperatures.

Significance: This study is the first to investigate the efficacy of BIT for controlling Salmonella on fresh cucumbers. The results indicate that BIT has potential as a wash treatment for controlling Salmonella on fresh produce.

P2-195. Carvacrol Nanoemulsion Controls Escherichia coli O157:H7 on Fresh Produce

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Purpose: The purpose of this study was to investigate the efficacy of nano-emulsified carvacrol, a natural phytochemical, as a wash treatment for reducing EHEC on fresh produce.

Methods: Cucumbers (n=128) purchased from the local grocery store were sorted for identical sizes and then dip-inoculated with a three-strain cocktail of EHEC. Each cucumber was marked with two one by five 5 cm areas and spot-inoculated with 20 μl of log CFU/ml of EHEC cocktail (Salmonella Newport, P. plantarum, and L. monocytogenes) on days 1, 2, 5, and 7. After inoculation and air-drying, each marked area was swabbed with a sterile cotton swab, and the log CFU/ml of EHEC on the cucumbers was determined on days 0 and 7.

Results: There was no significant difference in Salmonella populations among the treatments (P>0.05). The control cucumbers showed a significant increase in Salmonella population compared to the treated cucumbers (P<0.05). The results suggest that carvacrol nanoemulsion is effective in reducing EHEC on fresh cucumbers when stored at controlled temperatures.

Significance: This study is the first to investigate the efficacy of carvacrol nanoemulsion for controlling EHEC on fresh produce. The results indicate that carvacrol nanoemulsion has potential as a wash treatment for controlling EHEC on fresh produce.
Inoculated tomatoes and debris were washed with uninoculated tomatoes in various concentrations of free chlorine for one minute in a simulated wash water. Pre-wash commingling of non-inoculated tomatoes, inoculated tomatoes, and debris resulted in a high level of contamination which significantly affected firmness, titratable acidity, pH, and soluble solid of berries. Less than 50% fruit loss was observed in ENC treatment after 28 d compared with those in the untreated check. The highest reduction in fruit loss was observed in the ENC treatment after 28 d compared with those in the untreated check. The highest reduction in fruit loss was observed in the ENC treatment after 28 d compared with those in the untreated check.

**Results:**

The effect of ClO₂ on the killing of selected pathogenic bacterial and fungal pathogens and decaying fruits in postharvest storage was investigated. The study included the use of different concentrations of ClO₂ (0.63 to 4.40 ppm-h/g) in fresh fruits such as raspberries, blueberries, and blackberries. The results showed that ClO₂ at concentrations of 0.63 ppm-h/g significantly reduced the incidence of decay and weight loss in all the fruits tested. The efficacy of ClO₂ in controlling postharvest diseases was also investigated in blueberries and blackberries stored at 3°C for 28 d. ClO₂ at concentrations of 0.63 ppm-h/g was highly effective in reducing decay incidence and weight loss.

**Significance:**

The results of this study indicate that ClO₂ is a promising antimicrobial agent for controlling postharvest diseases in blueberries and blackberries. The use of ClO₂ in postharvest management can significantly reduce the incidence of decay and weight loss in these fruits, thereby increasing their shelf life and marketability.

**Poster 204 Evaluation of Viral Infection during the Freezing Process of Berries**

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**Introduction:**

Viruses are known to be present in fruits and vegetables, and their presence can affect the quality and safety of these products. The purpose of this study was to investigate the survival of viruses in frozen raspberries, blackberries, and blueberries.

**Methods:**

Raspberries, blackberries, and blueberries were frozen at 0°C for 28 d, and the viral titers were measured using a plaque assay. The samples were also analyzed for the presence of RNA viruses using RT-qPCR.

**Results:**

The viral titers in raspberries, blackberries, and blueberries were significantly lower after 28 d of freezing compared with the initial titers. The viral titers were also lower in raspberries, blackberries, and blueberries stored at 0°C for 28 d compared with those stored at −20°C. The presence of RNA viruses in the frozen berries was also confirmed using RT-qPCR.

**Significance:**

The results of this study indicate that the freezing process can significantly reduce the viral content of Berries. This finding has implications for the food industry, as it can help in reducing the incidence of viral contamination in fresh produce.
P-207: Effective Pack Practices: Use of Antifungal Packaging Films with Cyanin Analog Neomolecules to Control Postharvest Diseases in Strawberries

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Developing Scientist Entrant

Introduction: Every year 40% of the food items intended for human consumption never reach the table. Produce can be lost due to inadequate storage conditions, disease, or theft. Fruit and vegetable losses cost billions every year; 12.9% of the food lost was due to disease. In the United States and berries tend to be the most susceptible to disease. Producers need cost-effective technologies to help decrease loss and preserve quality and safety until reaching consuming/sell dates.

Purpose: The objective of this study was to evaluate the effectiveness of previously formulated active packaging systems to extend shelf-life and enhance strawberry quality.

Methods: Pullulan films incorporated with cyanin analog neomolecules were selected for this field study. Fresh strawberries were harvested from a local farm and placed into two groups (control and treatment). For each group, 10 strawberries were placed into a mold for digital readout basket and a pullulan film [even by 8 cm] was added at the bottom of each container for the treated group. Samples were stored for up to 10 days at 3°C and 12°C. Every two days microbial, visual, and physiological quality parameters were evaluated. The experiment was run in triplicate.

Results: For treated strawberries stored at 3°C, a reduction of two log CFU/g in yeast and mold population was observed over the 10-day period (P<0.05), as compared to the control. No significant difference (P>0.05) in yeast and mold was seen between controls and treated strawberries stored at 12°C. No significant differences (P>0.05) were reported for either group storage in temperature acid plates. A significant improvement in visual quality was noted for treated berries at both storage temperatures as compared to the control (P<0.05).

Significance: This study demonstrates the effectiveness of pullulan films incorporated with cyanin analog neomolecules to reduce fungal decay in strawberries and enhance desired quality during storage.

P-208: The Influence of Water Antimicrobials and Low Temperature Storage on Inhibiting E. coli O157:H7 and O26:H11 on Strawberries

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Introduction: Foodborne illness caused by O157 and non-O157 Shiga-toxin producing E. coli (STEC) on fresh produce remains a concern worldwide. Fruits contaminated at the point of production, transportation and retail serve as a vehicle for STEC transmission. In this study, commercial and home washing and low-temperature storage practices of strawberries were evaluated for survival of O157:H7 and O26:H11 STEC.

Purpose: Evaluate practices that facilitate STEC inhibition to improve microbial safety of fresh strawberries.

Methods: Strawberries were spot inoculated to achieve six log CFU of O157:H7 and 5.5 log CFU of O26:H11. The inoculated strawberries were washed with 30 s using tap water, electrolyzed water (50 ppm free chlorine) or 50 ppm free chlorine at 20°C. After washing, the strawberries were separately stored at 4°C or 2°C-20°C for 30 d. Samples were processed and plate counting was conducted to determine the population of O157:H7/O26:H11.

Results: There was no significant difference in O157:H7 population reduction between treatments at 4°C for 2 d, or -20°C/-80°C for 30 d. Samples were processed and plate counting was conducted to determine the population of O157:H7/O26:H11 on day 28 of storage.

Significance: This finding indicates that STEC could be inactivated in strawberries and reduce the risk of STEC transmission from strawberry to human consumers.

P-210: Efficacy of Gaseous Chlorine Dioxide in Reducing Salmonella, E. coli O157:H7, and Listeria monocytogenes on Fresh Produce

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Introduction: Use of aqueous sanitizers affects the quality and shelf life of berries. Gaseous chlorine dioxide, a strong oxidizing agent at low concentrations, may be utilized as an economical and effective sanitizing agent when delivered as dry material sachets.

Purpose: The objective of this study was to evaluate the effectiveness of gaseous chlorine dioxide (CD) application in reducing the levels of Salmonella, E. coli O157:H7, and Listeria monocytogenes on strawberries and blueberries.

Methods: Sixty strawberries and blueberries from the same field were spot inoculated with a 0.1 ml inoculum of Salmonella enterica, E. coli O157:H7, and Listeria monocytogenes and dried for 120 minutes inside a biosafety cabinet. The inoculated samples were placed in sealed five-liter chambers containing gaseous ClO2 solutions (63 mg/l for strawberry chambers and 30 mg/l for blueberry chambers) for one, two, and three h with control treatments sampled with no gaseous ClO2 treatment. The concentration of pathogens was determined using a C1-C PORTABLES 8 reader at 20 minutes intervals.

Results: Gaseous chlorine dioxide (CD) treatments for three h in strawberries reduced E. coli O157:H7, Listeria monocytogenes, and Salmonella levels by 2.0±0.3, 3.5±1.8, and 2.0±0.2 log CFU/g, respectively. Listeria monocytogenes levels after three-h treatment were reduced by ≥1.5 log CFU/g compared to one and two h treatments. Three h of gaseous CD treatment on blueberries reduced E. coli O157:H7, Listeria monocytogenes, and Salmonella spp. levels by 2.3, 2.1±0.2, and 2.2±0.3 log CFU/g, respectively. Reductions were similar for E. coli O157:H7 and Salmonella spp. in both strawberry and blueberry samples for all treatment times. Control treatments reported insignificant reductions for all pathogens, averaging 0.20 log CFU/g of pathogen reduction.

Significance: The findings of this study indicate that gaseous ClO2, delivered in gaseous sanitizers at low concentrations (less than five ppm) can significantly reduce pathogens on strawberries and blueberries during post-harvest sanitization.

P-211: Inactivation of Murine Norovirus and Hepatitis A Virus on Strawberry, Blueberry, and Blackberry by High Pressure Processing

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Introduction: Multiple outbreaks associated with foodborne viruses have occurred in recent years due to the consumption of contaminated berries. Recent progress (HPH) has been recognized as a reliable nonthermal processing technology for the food industry capable of inactivating a wide variety of foodborne pathogens including viruses while retaining the organoleptic quality.

Purpose: The objective of this study was to evaluate the effectiveness of inactivating murine norovirus (MNV-1) and hepatitis A virus (HAV) artificially inoculated on strawberries, blueberries and raspberries.

Methods: Fresh berries (25 g) were spot-inoculated with MNV-1 or HAV and 10 to 105 PFU/sample and packaged in high barrier bags filled with 30 ml sterile water. Berries were HPP treated at 200-600 MPa for three minutes with an initial process temperature of 4°C. After treatment, viruses were extracted from the samples and quantified by viral plaque assay.

Results: The initial inoculum level was approximately four and three log PFU/sample of MNV-1 and HAV, respectively, on berries. In the package removal step, 80% of MNV-1/HAV and 90% of MNV-1 were recovered from the external surface of the wrapped berries. HPP treatment at 350 MPa archived >2.4 log reduction of HAV in raspberries immersed in water during HPP. At 400 MPa, both MNV-1 and HAV was reduced to below detection level in strawberries, blueberries and raspberries.

Significance: HPP can be a preventive control for berries while retaining sensing characteristics. However, process optimization will be needed for large scale production due to the need for berries to be immersed in water.

P-212: Behavior of Two Serotypes of Listeria monocytogenes from Outbreaks and Recalls on the Surface of Stone Fruits and Berries during Storage

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Introduction: In 2014, a multidrug outbreak of foodborne illnesses associated with stone fruits contaminated with Listeria monocytogenes was reported. Due to this outbreak and research suggesting that the surfaces of stone fruits should not support the growth of L. monocytogenes, the behavior of L. monocytogenes strains isolated from stone fruits was investigated.

Purpose: Investigate the strain genotypic and fruit cultivar on the survival of L. monocytogenes in stone fruit.

Methods: Two L. monocytogenes strains (4b and 1/2b), isolated from recalled stone fruits from the 2014 investigation were used. Yellow and white peaches were spot-inoculated with 2.5 x 104 CFU/ml L. monocytogenes by submersion into water containing L. monocytogenes, at one of two inoculation levels, for five minutes. Fruits were air-dried for 30 min at room temperature and stored at 4°C for 26 days. L. monocytogenes levels were enumerated by a rinsing and direct plating method developed in our FDA laboratories. Counts were obtained by plating onto LA/BA (BrainHeart) and RAPIDZ/mini (BIO-RAD) agar. Twelve treatments (four replicates x three stone fruit + water control) were performed.

Results: The L. monocytogenes populations sharply declined in the first three and then declined more slowly until day 21. The maximum decline was 1.77 log CFU/fruit on yellow peaches inoculated with the serotype 4b strain at ~5000 CFU/fruit. On Day 21, the highest level of L. monocytogenes (2.20 log CFU/fruit) was observed on yellow peaches that had been inoculated with a high level of the serotype 4b strain. The L. monocytogenes remained viable until the end of storage (day 26), but the levels were not significantly different from those on day 21. The highest level of L. monocytogenes on day 26 was observed on white peaches with a low inoculum level and the high inoculum level had a negative correlation with X-ray storage (r = 0.757, p < 0.05). Although X-ray treatment results indicated a significant increase in pathogen levels by (P<0.05), no conclusive trend was observed.

Significance: Low-energy X-ray can be utilized to deliver a phytosanitary dose of 0.4 kGy for fresh blueberries while reducing risk of foodborne illness and maintaining quality.

P-207 – P-209

Poster

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Characterization of Tree Fruit Bacterial Communities during Harvest

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Introduction:
Under FUMA the tree fruit industry is required to validate onsite sanitizing water systems, but food safety issues limit the use of active foodborne pathogens for testing. Therefore, we hypothesize that the native microbiome present on tree fruits can be utilized as surrogates to validate sanitation water systems.

Purpose:
Current tree fruit microbiota data is limited, particularly throughout the growing season, thus we are establishing the microbiome of three tree fruit types, apples (granny, peaches, and stone), and navel oranges (citrus) with the goal of establishing an onsite validation bacterial index.

Methods:
A larger area (0.92 m²) was sampled for the presence of Enterobacteriaceae on all fruits, and these taxa represent potential surrogates for onsite validating tree fruit sanitizing systems.

Results:
The 2015 listeriosis caramel apple outbreak, linked to apples contaminated during packing, has increased the focus on the prevalence of pathogens within tree fruit packinghouses and the need for reliable rapid testing methodologies to quickly respond with recleaning during a sanitation event.

Discussion:
Limited information on the survival of Salmonella in peach wash water contacts due to field packing operations exists.

Introduction:
Five food contact surfaces (cotton, nitrile, rubber gloves, cotton rags, and stainless steel) coupons were autoclaved (clean) or rubbed with a can- taloupe leaf for 20 s (fouled), and inoculated with a five-strain cocktail of Salmonella Newport or L. monocytogenes (approximately seven log CFU/mL) for 30 min. Inoculated discs were plated using a bioblot for 30 min to allow the bacteria to attach on the rinse surface. The samples were vortexed for 15 s to recover loosely attached bacterial cells and sonicated for two minutes to recover strongly attached bacterial cells. Samples were serially diluted and plated on xylose lysine desoxycholate (XLD) agar to enumerate Salmonella and on modified Oxford FMD (MD) agar to enumerate L. monocytogenes. Attachment strength was calculated using the formula: Attachment strength (strongly attached bacterial + strongly attached bacterial + loosely attached bacterial + non-attached bacterial) / 100. The highest average attachment strength for Salmonella O157:H7 on apples.

Significance:
The evaluated rinsing wash water met the regulatory standard. The prevalence of Salmonella and E. coli on cantaloupe surfaces and enterococci on gloves and rice water indicates a potential risk of contamination on peaches.

References:

Survival of Salmonella spp. on Cantaloupe Field Pack Food Contact Surfaces

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Introduction:
Limited information on the survival of Salmonella on food contact surfaces during field packing operations exists.

Purpose:
The survival of Salmonella on cantaloupe field food contacts was studied, using five different conditions of food contact surfaces, was studied.

Methods:
Five food contact surfaces were sampled (cotton, nitrile, rubber gloves, cotton rags, and stainless steel) coupons were autoclaved (clean) or rubbed with a cantaloupe leaf for 20 s (fouled), and inoculated with a few-strain cocktail of Salmonella (or log CFU/mL). The wet inoculum was spot inoculated (100 µl) onto Salmonella selective agar plates, incubated at 35°C for 24 h. DNA was extracted from each salmonella isolate using the FastDNA Spin Kit for Soil (MP Biomedicals). The samples are enumerated on xylose lysine desoxycholate (XLD) agar, or non-selective plates to confirm isolates. Samples were serially diluted and plated on XLD agar to enumerate Salmonella and on Selective Oxidase Agar (XO) to enumerate L. monocytogenes.

Results:
Under all conditions, Salmonella populations peaked following a wet inoculum significantly higher (P<0.05) than those following a dry inoculation over 8 h (cotton glove, 3.50 and 0.75 log reduction for wet and dry, respectively). The expected Fox and cotton were killed off, wet (0.85-log reduction) and dry (1.32-log reduction). Salmonella population decline on clean surfaces, regardless of instument type or material, were generally less than 4 log CFU/mL. For example, reductions on dry rubber glove (1.25 and 0.33 log), and nitrite glove (3.02 and 1.0 log) for clean and fouled, respectively.

Significance:
The expected Fox and cotton were killed off, wet (0.85-log reduction) and dry (1.32-log reduction). Salmonella population decline on clean surfaces, regardless of instument type or material, were generally less than 4 log CFU/mL. For example, reductions on dry rubber glove (1.25 and 0.33 log), and nitrite glove (3.02 and 1.0 log) for clean and fouled, respectively.

Introduction:
Salmonella may attach to the surface of papaya through soil, animal feces, and post-harvest handling. Chlorine dioxide (ClO2) is a strong oxidizer and produces few carcinogenic byproducts. Washing papaya with ClO2 may reduce the risk of Salmonella contamination.
**P2-220 Prevalence, Virulence and Antimicrobial Resistance of Salmonella Isolated from Mango "Ataulfo"**

**Angelita Godínez-Oviedo and Montserrat Hernández-Bercial**

**Universidad Autónoma de Querétaro, Querétaro, Mexico**

**Introduction:** Salmonella is one of the most foodborne pathogens in the world, causing a number of outbreaks related to fresh produce. In 2012 a food-borne outbreak linked to mangos imported from Mexico occurred in Mexico. In Mexico, information regarding the presence of the pathogen in mangos at the point of sale and of the contamination that has not been reported.

**Purpose:** The main goal of this study was to detect, quantify and characterize Salmonella strains isolated from Ataulfo mangos purchased in Queretaro, Mexico.

**Methods:** During July and August 2018, 60 samples of Ataulfo mangos were collected from retail shops, supermarkets and markets located in Queretaro City. The detection of Salmonella was conducted using the 3M Detection Molecular System and positive samples were confirmed by traditional method established in the Bacteriological Analytical Manual, and quantified by the most probable number technique. In the isolated Salmonella strains the presence of thirteen virulence genes (hly, ags, eae, spa, spa, spa, spa, qacE, bcrABC, bcr, bcr, bcr, bcr, bcr, bcr) were identified by PCR amplification of target genes.

**Significance:** The obtained data in this study is important to establish the risk to the Mexican population associated with the consumption of fresh mangos. Decontamination methods should be evaluated to control the presence of Salmonella in mango in Mexico.

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**P2-221 Internalization of Salmonella spp. in Mangos (Mangifera indica) Variety Tommy Atkins**

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**Introduction:** In recent years, significant cases of salmonellosis caused by contaminated fruits were reported. Cross-contamination of fruit surfaces and post-harvest internalization of pathogens may result in increased risks to consumer health. However, little is known about the internalization of Salmonella in mangos.

**Purpose:** This study investigated the internalization capacity of Salmonella spp. in mango variety Tommy Atkins, under different experimental conditions.

**Methods:** Mature mangos were pre-evaluated for absence of Salmonella spp. on the surface using the swab technique. Salmonella negative fruits were challenged with Salmonella typhimurium ATCC 14028 and Salmonella enteritidis ATCC 10076 and their internalization was evaluated after spot contamination of the peel with the respective Salmonella strain and incubation at 8°C. The growth of Salmonella strains at refrigerator temperature (2°C) could also be used to determine the efficiency of a control Salmonella strain and Salmonella enteritidis was used as a control system.

**Results:** Salmonella ATCC 14028 grew by 2.13 log CFU at 21°C within 14 days, while it remained stable at 4°C. HCl (2.0% buffer) and citric acid produced 780 ppm and 270 ppm of ClO₂ during two separate time intervals. Analysis indicated such growth niches were potentially attributed to an uncleanable location and a damaged floor, with a great growth/survival potential if Salmonella contaminates mango. P2.223.

**Significance:** There is a great survival potential if Salmonella contaminates mango. P2.223 offers an effective and environmentally friendly alternative to chlorine bleach for controlling Salmonella on mango.

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**P2-222 Stress and Sanitizer Resistance Characterization of Persistent and Transient Listeria monocytogenes Genes Isolates from a Cold-Smoked Salmon Processing Facility**

**Joshua Gurtler, Renato Orsi1, Bala Jaganath1,2, Martin Wiedmann1,2 and Carmelina Viterbo1,2**

**Cornell University, Ithaca, NY, 1Novel Research, Luzern, Switzerland**

**Introduction:** Listeria monocytogenes is an pathogen in food, as well as in the food processing environment, that leads to foodborne illness and is a significant public health concern worldwide.

**Purpose:** The current study investigated the use of blue light to inactivate three strains of E. colii (MM4243, MM4164, and MM4245) known to grow on plant surfaces.

**Methods:** E. coli cultures grown overnight in tryptic soy broth (TSB) with nalidixic acid were resuspended in PBS. The inoculum was placed in four identical glass petri dishes each containing a min-stir bar. Each petri dish was placed on a stir plate under a lamp with 400 nm, 460 nm, 460 nm + 600 nm, or held in the dark. The plates were held in the dark. The plates were placed from each dish and plated on TSA and MacConkey agar at intervals for up to 24 hours.

**Results:** In this study, over the course of six trials, 400 and 460 nm blue light were shown to be most effective in killing E. coli in PBS at close range. Of three strains tested on TSA using 460 nm light, M4243 (three trials) was reduced by four log after seven hours, M4244 (two trials) was undetectable after eight hours, and M4245 (one trial) was undetected within six hours. The reductions were considerably less for 460 nm light treatment but still significant compared to the dark control. During each trial the inoculum was sampled at six to 10 time points.

**Significance:** Blue light in the range of 400 to 470 nm has been demonstrated to have antimicrobial effects and is also important for vegetative plant growth and enhanced production of various phytoantinbacterials in specific plant species during cultivation.

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**P2-223 Effects of Sweetgrass Fast Fry Biosol Biochar Generation Temperature on Survival of E. coli 0157:H7 in Soil**

**Joshua Gurtler, Akwari Boating1 and Charles Muller2**

**1U.S. Department of Agriculture–ARS, Beltsville, MD, 2University of Maryland, Department of Nutrition and Food Science and Center for Food Security and Safety, College Park, MD**

**Introduction:** Blue light in the range of 400 to 470 nm has been demonstrated to have antimicrobial effects and is also important for vegetative plant growth and enhanced production of various phytoantinbacterials in specific plant species during cultivation.

**Purpose:** This study investigated the use of blue light to inactivate three strains of E. coli (MM4243, MM4164, and MM4245) known to grow on plant surfaces.

**Methods:** E. coli cultures grown overnight in tryptic soy broth (TSB) with nalidixic acid were resuspended in PBS. The inoculum was placed in four identical glass petri dishes each containing a min-stir bar. Each petri dish was placed on a stir plate under a lamp with 400 nm, 460 nm, 460 nm + 600 nm, or held in the dark. The plates were placed from each dish and plated on TSA and MacConkey agar at intervals for up to 24 hours.

**Results:** In this study, over the course of six trials, 400 and 460 nm blue light were shown to be most effective in killing E. coli in PBS at close range. Of three strains tested on TSA using 460 nm light, M4243 (three trials) was reduced by four log after seven hours, M4244 (two trials) was undetectable after eight hours, and M4245 (one trial) was undetected within six hours. The reductions were considerably less for 460 nm light treatment but still significant compared to the dark control. During each trial the inoculum was sampled at six to 10 time points.

**Significance:** Blue light in the range of 400 to 470 nm has been demonstrated to have antimicrobial effects and is also important for vegetative plant growth and enhanced production of various phytoantinbacterials in specific plant species during cultivation.
**P2-226 Effects of Manuring on Survival of *E. coli* in Certified Organic Field Soils and Transfer to Fresh Produce in the Delmarva Region**

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*1University of Maryland Eastern Shore, Princess Anne, MD, 2University of California, Davis, CA, 3Western Center for Food Safety, University of California-Davis, Davis, CA, 4US Department of Agriculture–ARS-NEA-ARC, Beltsville, MD

**Purpose:** Key data are needed to ensure safe production of fresh produce in certified organically-managed crop systems using untreated animal manure. Current National Organic Program (NOP) standards stipulating a 90 to 120 day waiting period between manuring and crop harvest remain a concern.

**Introduction:** To evaluate *Escherichia coli* survival in soil and transfer to fresh produce harvested 90 to 120 after application of raw manure to certified NOP field.

**Methods:** From 2017 to 2018, certified NOP sandy-loam field plots were amended with dairy manure(DM), horse manure (HM), poultry litter (PL), or left unmanured (UR), before spraying-inoculation with a mixture of rifampicin-resistant *E. coli* O157:H7=stx1 and *Salmonella* Typhimurium. Composite core samples were enumerated (MPN at d 0, 30, 60, and 120 post-inoculation), with harvested produce enumerated on d 90 and 120. Recovery of rifampicin and aconitase resistant *E. coli* O157:H7 populations declined from 100% positive for all treatments by d 90 to 120 (≤4 log). However, there was 100% persistence on bulbs for all 2017 treatments. In 2018 PL and DM amendments were 50% positive (≤16) for transfer of *E. coli* O157:H7 to 120 bulbs. For tomato soils, *E. coli* O157:H7 populations were 100% positive (≤32) at d 77, 81 (SAS application – cover crop), respectively. Army warm-fruits reduced in 100% contamination of all tomato fruits (=32). In 2018, *E. coli* O157:H7 declined to 10-20% by d 90 with no transfer to tomato fruits. *Salmonella* spp. populations varied substantially between 2017 to 2018 in all treatments (2-32). In 2017, 30-grams samples per plot (≤32) in 2017, 2018 and 2019.

**Significance:** These first *E. coli* survival data for certified-NOP coastal Delmarva soils and produce will aid risk-assessed management of manure application-to harvest wait-times for fresh produce safety. This study paves the way for future evaluations of microbiological and physiochemical factors contrib- ing to safety of fresh produce.

**P2-227 Survival of Desiccation-resistant *Salmonella* on Apple Slices after Dehydration and Following Anti-microbial Immersion Treatments**

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**Introduction:** Salmonella is capable of surviving dehydration processes of various foods including dried fruit. Dried fruit, including apple slices, have been the subject of recalls due to possible contamination with Salmonella. Our study is the first to document the survival and population densities of *Salmonella* on apple slices under various dehydration and anti-microbial immersion treatment conditions.

**Methods:** Six varieties of apples (Envy, Gala, Red Delicious, Fuji, Pink Lady, Granny Smith) were cored and sliced into 0.4 cm rings, inoculated with a five-strain combination of *Salmonella* and *Listeria monocytogenes* at 1 log CFU/mL and both directly plated on TSA Agar and incubated at 35°C, or further enriched and plated on TSA Agar. Immunomagnetic separation (IMS) of the non-selective enrichment and both direct and IMS enriched samples was performed using the magnetic separation system. Plate counts were enumerated on TSA Agar.

**Results:** The survival of *Salmonella* populations varied according to apple variety and IMS enrichment. Inoculation densities (log CFU/g) were Envy (1.69), Gala (2.09), Red Delicious (2.77), Fuji (2.93), Pink Lady (3.15), and Granny Smith (3.77). Greater numbers of *Salmonella* (≤0.05) were inoculated on Granny Smith, Pink Lady and Fuji apples than on Gala and Envy apples. Survival of *Salmonella* on Gala apple slices (log CFU) following anti-microbial treatments and dehydration were untreated control (5.58, Ps 0.05), 580.90, AA 0.33, LA 0.28, GA 0.23, FA 0.17, and SAS 0.05. Lower survival rates were achieved by pretreating apple slices with either FA or SAS before dehydration. All other anti-microbial treatments may provide these results for increasing the inactivation of *Salmonella* during the dehydration process of apple slices applicable for the food industry.

**Significance:** This information would allow manufacturers to understand the survival and population densities of *Salmonella* on apple slices under various dehydration and anti-microbial immersion treatment conditions. Further studies are needed to evaluate the effect of dehydration conditions on the survival and population densities of *Salmonella* on apple slices.
Journal of Food Protection Supplement

P2-232 A Longitudinal Study Using 16s rRNA Gene Sequence Analysis of Soil Amended or Unamended with Heat-treated Poultry Pellets Contaminated with Salmonella Newport

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Introduction: Salmonella Newport is a foodborne pathogen frequently associated with outbreaks in produce. Several factors, such as the use of manure, temperature, and soil moisture, have been shown to influence its survival in soil. 

Purpose: The purpose of this study was to investigate soil microbiomes by 16s rRNA amplicon sequencing, over a two-month period, in heat-treated poultry pellets with and without Salmonella Newport contamination.

Methods: Soil was inoculated at ~7.5 log CFU per gram dry weight into a total of six plants, containing heat-TPP-amended soil and three without. Six similar plants were also set up without Salmonella Newport inoculation. Soil samples were collected in duplicate from each plant on days 1, 14, and 21., and duplicate samples from bulk and rhizosphere soil were also collected at days 1, 21, and 28. 16s rRNA gene sequencing was performed on extracted metagenomic DNA.

Results: Amended and unamended soils harbored very diverse microbiomes, with >1,500 taxa identified over all samples. The addition of HTPP reduced, or added certain taxa; namely, Paenibacillus, P. entomophila and Bacillus (such as B. amyloliquefaciens, B. subtilis, and B. licheniformis); and Rhodoplanes/Devosia (such as R. stabekisii, D. tarandus, and members of the Acidobacteria) and the Rhodopila, while others increased in proportional abundance, such as Novosphingobium, Devosia, Rhodobacter, and Devosia to rich soil. 

Significance: The soil inoculation resulted in an increase in abundance of some of the microbiome without an overall impact on the diversity and provided survival of Salmonella Newport.

References:

P2-233 Factors Affecting Salmonella Newport Survival in Soil and Subsequent Transfer to Spinach Plants

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Introduction: Microbial and Food Safety Laboratory, Beltsville, MD

Purpose: The purpose of this experiment was to assess the impact of HTPP as an amendment on the survival of Salmonella Newport surviving on spinach leaves after simulated transfer.

Methods: Soil was sieved, and autoclaved twice for one hour. Soil moisture was adjusted to a 10% volumetric water content. Triplicate samples were inoculated with ~7.5 log CFU/ml of Salmonella Newport and maintained in a biosafety cabinet. Soil samples were collected on days 0, 14, and 21. Heat-treated poultry pellets (HTPP) were applied to plots (200 g applied per plot) and evaluated for their influence on the flora of the soil.

Results: Survival of Salmonella Newport was quantified by culture method and PMA-qPCR over 91 days in three experimental replicates. Higher populations of Salmonella Newport from HTPP amended soil transferred to and survived on spinach leaves than that from unamended soils (P < 0.05).

Significance: HTPP amended soils provided a favorable environment for Salmonella Newport survival in soil and transfer to spinach plants when introduced in irrigation during spinach cultivation.

References:

P2-234 Serotypes and Antimicrobial Resistance of Salmonella Recovered from Chicken Liver in Florida Operations

Alan Gutierrez, Jaycansan Der and Keith Schneider
University of Florida, Gainesville, FL

Introduction: The development of antimicrobial resistance (AMR) in foodborne pathogens is a major public health concern, and identifying the antimicrobial resistance (AMR) patterns of isolates to various antimicrobial agents is critical to controlling the spread of AMR and limiting its impact on human health.

Methods: Salmonella isolates (n=47) recovered from chicken liver were tested in the USDA National Veterinary Services Laboratories for serotyping, antimicrobial susceptibility testing, and two different qPCR methods. Salmonella isolates were serogrouped using a microblot serotyping method. Antimicrobial susceptibility testing was performed using a broth microdilution method. Two different qPCR methods were used to detect salmonella with a panel of 14 antimicrobial agents: amoxicillin/clavulanic acid, ampicillin, azithromycin, cefoxitin, ceftriaxone, chloramphenicol, ciprofloxacin, gentamicin, levofloxacin, nalidixic acid, norfloxacin, ampicillin, cephalaxin, and tetracycline, and tetracycline (such as B. amyloliquefaciens, B. subtilis, and B. licheniformis); and Rhodoplanes/Devosia (such as R. stabekisii, D. tarandus, and members of the Acidobacteria) and the Rhodopila, while others increased in proportional abundance, such as Novosphingobium, Devosia, Rhodobacter, and Devosia

Significance: The soil inoculation resulted in an increase in abundance of some of the microbiome without an overall impact on the diversity and provided survival of Salmonella Newport.

References:

P2-235 Survival of Escherichia coli O157 from Bovine Manure in Autochlorated and Un autoclaved Florida Sandy Soil

Christina S. Thysen-Jones, Thinyoung Lee, Jaycansan Der, Kwangcheol Casey Jeong and Keith Schneider
University of Florida, Gainesville, FL

Purpose: Evaluate the potential effect of two different chlorination methods (15 ppm and 50 ppm NaOCl) on the survival of Escherichia coli O157 in bovine manure amended with different soil types.

Methods: Four different amendments were applied to twenty plots (3m x 3m each). Composite soil samples were collected weekly over a 120-day period and evaluated for Escherichia coli O157 survival. 

Results: The average linear death rate (log CFU/g day-1) for all amending factors were applied to twenty plots (3m x 3m each) and evaluated for Escherichia coli O157 survival on uninoculated (0, 0.16, and 0.19) and autoclaved (0.01, 0.09, and 0.11) soils. Microbial populations in soils shifted over time with a higher abundance of Acidobacteria and Alphaproteobacteria observed in uninoculated, and a predominance of Firmicutes and Proteobacteria in autoclaved soils.

Significance: This study provides insights into the microbial survival in sandy soil based on natural microbial populations.

References:

P2-236 Bacterial Survival as a Factor of Variation in Extrinsic and Intrinsic Soil Parameters with Soil Amendments of Animal Origin

Pushpinder Kaur Litt, Alyssa Kelly1, Quinn Riley2, Alexis Omar3, Gordon Johnson4, Manan Sharma2 and Kali Kriel5
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Purpose: To assess the impact of different soil amendments on the survival of bacteria on soil parameters may clarify their role as potential indicators of pathogen survival in manure-amended soils.

Methods: Using one-way ANOVA (P<0.05).

Results: Average values for soil moisture (15.4±0.4%), temperature (27.3±0.7°C) and solids content (54.0±0.4%) did not significantly vary among amendment types (P>0.05). Significant differences in soil moisture content between treatments and controls were observed. Other significant differences in survival were observed between the strains. Cell densities determined by PMA-qPCR and culture methods were similar (P>0.05), as was survival on spinach leaves with/sinificant pathogen populations.

Significance: Identifying and measuring soil parameters may clarify their role as potential indicators of pathogen survival in manure-amended soils. Results suggest soluble carbon could be a critical factor to understanding potential for contamination.

References:

P2-237 Application of Rhizobacteria as a Biocontrol by Tackling Pathogen-Pathogen Interactions

Pushpinder Kaur Litt, Alexis Omar, Manan Sharma2 and Kali Kriel5
1University of Delaware, Newark, DE, U.S. Department of Agriculture–ARS, Environmental Microbial and Food Safety Laboratory, Beltsville, MD

Purpose: To assess the impact of different soil amendments on the survival of bacteria on soil parameters may clarify their role as potential indicators of pathogen survival in manure-amended soils.

Methods: Soil and bacterial inoculum were mixed and applied to plots of soil, and evaluated for their influence on the flora of the soil.

Results: Survival of Salmonella Newport was quantified by culture method and PMA-qPCR over 91 days in three experimental replicates. Higher populations of Salmonella Newport from HTPP amended soil transferred to and survived on spinach leaves than that from unamended soils (P<0.05).

Significance: HTPP amended soils provided a favorable environment for Salmonella Newport survival in soil and transfer to spinach plants when introduced in irrigation during spinach cultivation.

References:

P2-238 Developing Scientis...
PIE-238: Estimating Salmonella and Campylobacter risk in Animal Densities and Their Potential Impact to Significant Contamination Events
Tahl Levit, Jennifer Allen, and Jorge J. Couto
Oregon State University, Corvallis, OR

Introduction: Significant foodborne outbreaks have been linked to the presence of wildlife and/or domesticated species in and around agricultural environments. Despite these linkages, there is little information about the likelihood of pathogens to be associated with certain species or about the load that may be present in fecal or excretal samples.

Purpose: To estimate the quantity of Salmonella and/or Campylobacter in animal feces to identify species and/or individual animals that could contribute to the contamination of agricultural products.

Methods: Dropped feces were identified and collected as part of wildlife research at the intersection of free-range cattle, mule deer, and rocky mountain elk in southwestern Oregon (n=27 per species). Total DNA was extracted using a modification of the Qiagen DNeasy Blood and Tissue kit. DNA ex- tractions were performed using Qiagen DNeasy kit. PCR primers used for detection of genes; bla ששתז (in 29R; 65% homology to the GenBank sequence JF950950.1) and invA_176F 5’-CAACGTTTCCTGCGGTACTGT; in 30X coverage, followed by raw read assembly using the A5 pipeline, and gene annotation using PATRIC.

Results: Campylobacter and Salmonella were detected in elk at 25% and 6%, respectively. The top three incidences for coagulase positive staphylococci were 44%, 32%, and 24%, and those for Campylobacter were 92%, 58%, and 27%, respectively. The top three incidences for coagulase positive staphylococci were 44%, 32%, and 24%, and those for Campylobacter were 92%, 58%, and 27%, respectively. The top three incidences for coagulase positive staphylococci were 44%, 32%, and 24%, and those for Campylobacter were 92%, 58%, and 27%, respectively. The top three incidences for coagulase positive staphylococci were 44%, 32%, and 24%, and those for Campylobacter were 92%, 58%, and 27%, respectively.

Significance: These results confirm that cattle, deer, and elk can be significant carriers of Campylobacter and Salmonella, however, species and individual animals differ in their potential for contaminating agricultural products. Further work is necessary to understand how these and other species contribute to Campylobacter and Salmonella contamination in agricultural environments.

PIE-239: Isolation and Characterization of Extended Spectrum β-Lactamase (ESBL) Producing Non-Shiga-toxicogen Escherichia coli (nSTEC) from Healthy Food Animals and Their Environment
Shivasharanappa Nayakvadi, Dhananjay Desai1, Shivaramu Kettela2, Paula J. Fedorka-Cray3, Chethan Kumar HB4 and Eakin B Chakravorty
1Washington State University, Kangas, WA, 2Central State Agricultural Research Institute, Goa, India, 3Department of Population Health and Pathobiology, CMU, NC State University, Raleigh, NC, 4North Carolina State University, Raleigh, NC

Introduction: The occurrence and dissemination of extended spectrum β-lactamase (ESBL) producing E. coli among food animals is a global public health concern.

Purpose: The purpose of this study was to determine the prevalence of multidrug-resistant ESBL E. coli from apparently healthy food animals and their associated environment in west-coast India.

Methods: A total of 235 fecal samples were collected from healthy young calves (n=23), goats (n=155), pigs (n=20), buffaloes (n=7), ducks (n=30) and their associated environment (n=18) in west-coast region of India. Isolation and antimicrobial susceptibility testing of ESBL E. coli was carried out according to standard protocols using four µg/ml ceftazidime in MacConkey for the screen. Multiplex-PCR assay was optimized to detect ESBL, nSTEC and virulence (sfa, vps, genes.

Results: Overall, the prevalence of ESBL E. coli was 110 (47.4%) of 235 among fecal animals of cows. The ESBL E. coli in young calves was 87 (76.26%) of 23, goats 115 (75.52%) of 155, pigs 18 (90%) of 20, buffaloes 4 (57.14%) of 7, ducks 27 (54%) of 30 and their associated environment was 12 (72.22%) of 18. Antimicrobial susceptibility testing of all isolates (n=110) showed a higher frequency of resistance to penicillin (110 of 110) following by cephalaxin (99), trimethoprim (97), ciprofloxacin (96) and ampicillin (95). The most common resistance pattern was Ampicillin-cefoxitin-ceftazidime. Multiplex-PCR assay of 110 isolated detected ESBL resistance determinants in 67 (60.91%), sfa in 88 (7.27%), vps in 90 (10.09%). All isolates were negative for presence of virulence gene stx. However, stl was found in three (2.72%) and eae in five (4.54%) of the samples.

Significance: The study highlights that apparently healthy food animals are potential reservoirs of non-virulent multidrug-resistant E. coli which harbors ESBL genes which may pose a risk to humans via food chain.

PIE-240: Incidence of Coagulase-positive Staphylococcus and Staphylococcus aureus on Farms from Cattle Sources
Luiya Zhao and Jinu Chen
Department of Food Science and Technology, The University of Georgia, Griffin, GA

DEVELOPING SCIENTIST ENTRANT

Introduction: Human staphylococcal food poisoning is sometimes caused by consuming foods originating from animals that are reservoirs of coagulase-positive staphylococci and Staphylococcus aureus, such as cattle. Feces can be transmitted to milk via the teat canal or from cattle to humans.

Purpose: The purpose of this study was to identify the prevalence of coagulase-positive-staphylococci and S. aureus in feces from cattle farms.

Methods: Feces (50 from each farm) were captured using fly traps at nine cattle farms interspersed through Georgia (United States). Coagulase-positive Staphylococcus and S. aureus were isolated from the internal surface and external tissue of the flies following a method outlined in the FDA Bacteriological Analytical Manual with slight modification. Presumptive coagulase positive staphylococci and S. aureus colonies were subsequently confirmed.

Results: Coagulase-positive staphylococci were isolated from seven (70%) of the farms where 52 (20%) of 265 captured flies were found to be positive for coagulase-positive staphylococci. S. aureus was isolated from seven (70%) of the farms where 50 (20%) of 265 captured flies were found to be positive for coagulase-positive staphylococci.

The incidence of coagulase-positive staphylococci and S. aureus-positive flies varied from farm to farm ranging from ten to 44% and zero to six percent, respectively. The top three incidences for coagulase positive staphylococci were 44%, 32%, and 24%, and for S. aureus were six, four, and two percent, respectively.

Significance: These data suggest that flies are active carriers of coagulase-positive staphylococci and S. aureus on cattle farms.
P2-244 Antimicrobial Susceptibility Monitoring of Bacterial Pathogens Isolated from Korean Black Goat
Woo Kyung Jung1, Sook Shin2, Chan Lan Kim3, Kun Taek Park4 and Yong Ho Park5

1Seoul National University College of Medicine, Seoul, South Korea
2Animal and Veterinary Science, College of Agriculture, National University of Singapore, Singapore
3College of Animal Sciences, Pukyong National University, Busan, South Korea

Introduction: Mastitis is an inflammatory disease of the mammary glands which may cause partial or full damage to the udder. Subclinical mastitis is one of the most important diseases and considered a constant risk for whole herd and their environment. Staphylococcus spp. and other coagulase-negative staphylococci are the main causative agents of mastitis.

Purpose: The present study was conducted to isolate and identify bacteria that cause mastitis and diarrhea in Korean black goats and evaluate the anti-microbial susceptibility.

Method: A total of samples were, from 20 Korean black goats (Capra hircus corenae) which were identified subclinical cases in Namwon, Korea, collected in 2018. Subclinical mastitis was confirmed by a somatic cell count (SCC) > 300,000 cells/ml (Bentley Instruments, Chaska, USA). Feaces from 32 goats which showed diarrhea were also sampled. Identification of the isolates was achieved using matrix assisted laser desorption ionization-time of flight (MALDI-TOF) spectrometry (Bruker Daltonics, Bremen, Germany). Antibacterial resistance was evaluated for Staphylococcus spp. and Enterococcus spp. with 14 antibiotics by the agar dilution method described by Clinical and Laboratory Standards Institute standards.

Results: A total of 118 isolates were recovered (43 from milk and 21 from feces from 20 goats with subclinical mastitis and 54 from 32 goat feces with diarrhea). In milk samples, 11 (26%) Staphylococcus aureus, 28 (65%) and one E. coli were found while three E. coli, nine coagulase-negative staphylococci and one Strepococcus pneumoniae were isolated from feces samples. One Staphylococcus aureus, eight coagulase-negative staphylococci from milk and seven coagulase-negative staphylococci from feces showed metillin resistance. For Staphylococcus spp., resistance to ampicillin and penicillin was seen in 11% of isolates and amoxicillin/clavulenate, tetracycline and cephalaxin were 21% resistant. Resistance was absent or very few for erythromycin, marbofloxacin, gentamicin, cindamycin and chloramphenicol.

Significance: Even as the size of Korean black goat mastitis is growing, this type of study is rare; it showed antimicrobial resistance and the need for surveillance.

P2-245 Sporeformer Presence in a Milk Fractionation Process
Kristi Gowans, Reece Larsen, Tina Lin, Jeremy Arbon, Greyden Clark, Frost Steele and Bradley Taylor Brigham Young University, Provo, UT

Undergraduate Student Award Entrant

Introduction: Because various thermophilic endospores are capable of surviving heat treatment steps common to milk processing, the implementation of spore-reduction technologies is difficult without a proper understanding of the heat classifications, characteristics, and identities of common sporeformers in dairy processing and powder production.

Purpose: To enumerate thermophilic and mesophilic endospores at multiple points in a commercial powder milk fractionation process using temperature classifications and presence/absence of O2.

Methods: Product samples were collected at 12 different points in the milk fractionation and drying process and thermally shocked (80°C) to select for thermophilic endospores. The samples were then plated and enumerated using a pour plate method on skim milk plate count agar and incubated at mesophilic (31°C) and the thermophilic (55°C) temperatures under aerobic and anaerobic conditions (n=4, total samples=48).

Results: A total of 98 colonies were identified in the process (200 CFU/ml in fluid milk, 200 CFU/ml in fluid skim milk, 20 CFU/ml in powder, 20 CFU/ml in final product), particularly following post- to post-ultrafiltration (250 CFU/ml compared to 1000 CFU/ml) and pre- to post-drying (830 CFU/ml compared to 3070 CFU/ml), possibly due to decreased metabolic activity, increased concentration of the solutes, and outgrowth. Mesophilic aerobic were the most prevalent sporeformers identified throughout the process, at 39.8% (730 CFU/ml) of the total count with mesophilic anaerobes as the next highest proportion at 27.1% (590 CFU/ml).

Significance: We suspect sporeformer survival during HTST pasteurization and further germination and outgrowth during regeneration sections of unit operations during long production runs since both mesophilic and thermophilic sporiferous counts increased throughout the production process. Because thermophilic bacter is an adverse impact on powdered milk quality and limited secondary applications of the product, this bacterial growth may be controlled for in order to limit concentration of endospores and outgrowth during processing, resulting in the production of sporefree milk powders.

P2-246 Determining the Effect of Individual or Combined Protective Cultures on the Growth of Listeria monocytogenes and Shiga Toxin-Producing Escherichia coli in Raw Milk
Sulaiman Alajias1, Catherine Genderson1 and Dennis D’Amico1

1University of Connecticut, Storrs, CT, 2University of Connecticut, Department of Animal Science, Storrs, CT, 3University of Connecticut, Storrs, CT

Introduction: Dairy products with selective toxic-producing Escherichia coli (STEC) and/or Shiga toxin-producing Escherichia coli (STEC) (e.g., both O157:H7 and O157:H7 serotypes, are pathogens of concern in the production of raw milk. Protective bacterial cultures (PCs) with limited acidification and flavor production represent a potential control strategy.

Purpose: This purpose of this research was to determine the antimicrobial efficiency of commercially produced PCs, used individually or in combinations, against L. monocytogenes and STEC when inoculated in raw milk following an inoculation time and temperature profile similar to cheese-making and ripening.

Methods: Six-strain cocktails of L. monocytogenes, O157:H7 or non-O157 STEC were inoculated to - two log CFU/ml in raw milk and stored at 4°C for 24h. Freeze-dried PCs were then added individually or in binary combinations (1:1 ratio) at - seven log CFU/ml. Control samples were inoculated at 37°C for 4h, followed by 20°C for 2h, and then held at 12°C for seven days. Experiments were conducted in duplicate and were repeated at least three times.

Results: After seven days, L. monocytogenes counts were 5.16, 3.09 and 3.21 log CFU/ml lower than control when co-inoculated with Lactococcus lactis subsp. lactis B5-10, Lactobacillus plantarum Holdac, Listeria or Lactobacillus plantarum LUPAL, respectively. Inoculation with LALCLUST Protect ADF01 at 8x B16 inhibited non-O157 STEC and O157:H7 STEC growth with differences of 3.91 and 1.86 log CFU/ml compared to control, respectively. Moreover, potential synergy against L. monocytogenes was 1.56-1.5 log CFU/ml in B5-10 and Pediococcus acidilactici B-LC-20 whereby counts were below the limit of enumeration (less than one log CFU/ml) by day four.

Significance: The study demonstrates the efficacy of commercial protective cultures for controlling the growth of L. monocytogenes and STEC in raw milk. It also identifies a potential synergistic combination as a potential strategy for enhancing the safety of raw milk products. Determining the effectiveness of these cultures and their combinations in the production of raw milk is critical to the safety of raw milk products.
When used as the main or composite substrate for fermented foods, they give rise to foods containing beneficial probiotics, digestive enzymes and health-promoting bacteria. The mash was then divided into three parts and two parts were heated with hot water. The portions were mixed together at 70°C. Spore formers and moulds were observed in them during the third and fourth week of storage.

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Purpose: The objectives of this study were to characterize the microbial ecosystems of traditional farmstead cheese production and evaluate the role of microbial diversity to product microbiota composition through production and aging.

Methods: A total 108 products (e.g., raw milk, curd, cheese) and environmental (e.g., tools, working surface, air) samples were collected along the cheese-making continuum. DNA was directly extracted and amplified targeting bacterial 16s rRNA and fungal ITS genes. Sequencing was conducted on the Illumina MiSeq. Amplicon data were analyzed using DADA2. SourceTracker was applied to estimate the role of the environment in microbial transfer.

Results: High microbial diversity was found across all sample types. Among environmental samples, the wooden barrel was dominated by Lactobacillus species. The cheese samples were dominated by Bacillus, followed by Lactococcus and Streptococcus. The core of the cheese was dominated by lactic acid bacteria, while the rind was dominated by yeasts and molds.

Significance: This study demonstrates the role of the environment in influencing cheese microbiota composition, highlighting the importance of providing optimal storage and handling conditions to maintain the desired microbial profile.

P2-256 Source Tracking and Succession of Microbial Communities during the Production of a Farmstead Cheese

Lang Sun1 and Dennis D’Amico1
1University of Connecticut, Storrs, CT, 2University of Connecticut, Department of Animal Science, Storrs, CT

Developing Scientist Entretainer

Purpose: Historically, cheese production was conducted on farms using raw milk and wood boards, which led to a high degree of microbial diversity. The microbial composition of cheese varies depending on the type of cheese and cheese-making processes.

Methods: A total of 108 products (e.g., raw milk, curd, cheese) and environmental samples were collected along the cheese-making continuum. DNA was directly extracted and amplified targeting bacterial 16s rRNA and fungal ITS genes. Sequencing was conducted on the Illumina MiSeq. Amplicon data were analyzed using DADA2. SourceTracker was applied to estimate the role of the environment in microbial transfer.

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Significance: This study demonstrates the role of the environment in influencing cheese microbiota composition, highlighting the importance of providing optimal storage and handling conditions to maintain the desired microbial profile.
Purpose: To elucidate the diversity of oxalate-in resistant isolates recovered from fresh cheese of artisan production in Costa Rica.

Methods: One hundred sixty cheese samples that were selected in three different days at five locations across four provinces. The Vitek system was used to identify the isolates and obtain antibiotic susceptibility profiles. Furthermore, 10 isolates with phenotypic resistance to oxolin were whole genome sequenced to determine their type sequences, spa types, and SCCmec structures.

Results: A total of 154 isolates were obtained, of which 15 were resistant to oxolin with MICs>4 or ≥48 μg/ml. Positive samples were derived from three of the four provinces visited. Three of the 10 sequenced isolates belonged to the widespread, human-associated, ST810 type and have a type IVa (2B) SCCmec element. The other seven sequenced isolates did not have this type sequence. Three of them were classified as ST325 (59), which is of bovine origin, and the rest were classified as novel sequence types (ST127, ST1127, and ST11028 strains).

Significance: The finding of human and bovine strains indicates bad hygiene and inadequate pasteurization during production. In addition, our results likely reflect zoonotic MRA transmission.

P3-03 Genetic Analysis of Natural Microflora in Stored Joraengyi Rice Cakes and Their Capability for Propionic Acid Production

Heeadae Park1, Jung Kyu Chae1, Ighal Hossain1, Sazzad Hossen Touchchi1, Ha Lim Jeong1 and Sang-Doo Ha2

Advanced Food Safety Research Group, Brain Korea 21 Plus, Chung-Ang University, Ansan, South Korea, 2Chung-Ang University, Ansan, South Korea

Introduction: In the Republic of Korea, rice cakes account for the largest portion in the sales of rice processed foods. Propionic acid (PA) is a preservative that can be produced by natural fermentation. However, a widespread fermentation of microorganisms originating from raw materials during storage, although they are not artificially added.

Methods: The purpose of this study was to verify the natural origin of PA in rice cakes by analyzing the population of natural microflora present in deteriorated joraengyi rice cakes and investigating microbial growth and capability for PA production.

Results: A total of 98 microbial strains were isolated from microflora that grew after the expiration of shelf life of joraengyi rice cakes. The lactobacillus group and yeasts were the dominant groups. 39.3% were found in cheese originating from three of the four provinces visited. 10 sequenced isolates derive from the same province, which had a widespread, human-associated, ST810 type and have a type IVa (2B) SCCmec element. The other seven sequenced isolates did not have this type sequence. Three of them were classified as ST325 (59), which is of bovine origin, and the rest were classified as novel sequence types (ST127, ST1127, and ST11028 strains).

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P3-03 Genetic Analysis of Natural Microflora in Stored Joraengyi Rice Cakes and Their Capability for Propionic Acid Production

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gene expression of moldy soybean protein (Ma et al., 2013). On the other hand, digestion of mycotoxin-contaminated feed by intestinal microorganisms decreases the mycotoxin load in the system, as shown by the results of our study. In our study, moldy soybean protein was treated by simulated digestion (Ma et al., 2013). The results showed that the mycotoxin load in the system was significantly lower in the treated group compared to the raw group. This suggests that the mycotoxin load in the system can be reduced by digestion of moldy feed.

Conclusions:

The results of our study indicate that the mycotoxin load in the system can be reduced by digestion of moldy feed. This suggests that the mycotoxin load in the system can be reduced by digestion of moldy feed.

Acknowledgments:

This study was supported by the grant from the Ministry of Agriculture, Food and Rural Affairs, Republic of Korea (PJ011542).

References:


Poster

P3-13 - Reduction in A. orcinus in Rice and oat Porridge by an Indirect Steaming Process with Baking Soda

Yu-Jung Lee, Keija Gu, Shufang Li* and Doojin Ryu*
*University of Iowa, Moscow, DC

Introduction: Ochratoxin A (OTA) is one of the most important mycotoxins owing to its widespread occurrence and toxicity including nephrotoxicity and proteinuria. OTA contamination in rice and oat products has been detected in processed food products due to its heat stability while significant reduction of the toxin may be achieved under higher temperature and alkaline conditions during food processing.

Purpose: In this study, the effects of retort process on the stability of OTA in spiked (20 ug/kg of dry weight basis) rice and oat porridge (10% solid content) in the presence and absence of baking soda was investigated using a laboratory indirect steaming system.

Methods: The samples were heated in a pot at 85°C central temperature for 10 min to be gelatinized followed by drying in 50°C oven overnight. The entire experiments were conducted in triplicates. Results were analyzed for OTA by high pressure liquid chromatography with ultraviolet detection (HPLC-FLD).

Results: The reduction of OTA in inoculated rice and oat porridge was 59% (8.2 ng/g) and 14% (17.2 ng/g), respectively, while greater reduction of OTA was observed with addition of baking soda. The reduction of OTA in the rice porridge with 0.5% and 1.0% baking soda were 78% (4.4 ng/g) and 64% (6.4 ng/g), respectively. In the oat porridge, reduction of OTA was also evident to result in 58% (8.4 ng/g) and 72% (5.6 ng/g) with 0.5% and 1.0% of added baking soda, respectively.

Significance: These results suggest that OTA in rice and oat may be reduced by significant by indirect steaming process. In addition, adding baking soda may positively impact the reduction of OTA.

P3-14 - Detection of Salmonella Typhimurium in Environmental Sponge Swab Enrichment Cultures Using the bioMérieux VIO3D SLM, EZ-SLM and the FDA BAM Cultural Assay

Ryan Zimmerman, Leahne Halln, Sue Kelly, Laurie Post, Brian Fairn, Charles Baue, Patrick Rule, Stan Bailey and John Mills

1Deibel Laboratories, Inc., Madison, WI, 2Deibel Laboratories, Inc., Bethlehem, PA, 3Deibel Laboratories, Inc., Gainesville, FL, 4Deibel Laboratories, Inc., Lincoln, IL, 5BioMérieux Inc., Newkiroy, MO

Introduction: Environmental testing for Salmonella is a vital to a food safety program. Individual sponge swabs are collected from multiple sites in order to take action when screened positive but requires more time and cost . Creating secondary samples post-enrichment could address both while allowing

Methods: Sponge swabs hydrated with DE neutralizing broth were coinoculated at a fractional level with approximately 1.5 CFU S. Typhimurium and approximately 1.5 CFU E. coli O157:H7 (competitor) (n=20). Sponges (n=80) were inoculated with 15 CFU S. Typhimurium and enrichments from each were mixed with the coinoculated enrichments in a 1:5 ratio. Positive (2≥5 CFUs/vial, n=5) and negative (uninoculated, n=5) sponges were included. Inoculated sponges were incubated 18 h to simulate standard enrichment conditions prior to enrichment and screening using the VIO3D SLM, EZ-SLM or the FDA BAM reference method. All sample enrichments were streaked to selective/differential isolation media and the plates examined for typical colonies.

Results: Individual sponge enrichments demonstrated identical recovery to the five-sponge mixed enrichments tested by VIO3D SLM, EZ-SLM and reference methods. The proportion of positive sponges was 15/20 (POD 0.75) for Lactose broth enrichments and 16/20 (POD 0.80) for BPA enrichments. The difference in probability (POD) between methods was determined. No significant difference was observed between individual sponge enrichments and secondary sponge mixed enrichments when compared to the average of any of the tested methods. All confidence intervals (59%) included 0. All five high inoculated sponges tested positive (POD 1.00) and all 5 negative sponges tested negative (POD 0.00).

Significance: A five-sponge mixed enrichment will provide a comparable recovery to testing individual environmental sponge enrichments.

P3-15 - Test for Detection of Listeria spp. from Environmental Surfaces without Enrichment

Naval Bakri, Quyen-Nhi Le*, Preetha Biswas, Brooke Roman*, Mark Mozola, Robert Donofrio*, Benjamin Bastin, Nicole Kliss* and Pat Walsh-Bird

1Aegon Corporation, Lansing, MI, 2IQ Laboratories, Inc., Cincinnati, OH

Introduction: This method is a novel, enrichment-free test for detection of Listeria spp. in swab samples taken from environmental surfaces. Results are available in less than one hour. In a previous Performance Tested Method study, the test was validated for stainless steel and sealed concrete surfaces (PTM 414501).

Methods: Performance of the test method was compared to the U.S. Food and Drug Administration Bacteriological Analytical Manual Performance Tested Methods kit in a AOAC Performance Tested Methods Test Kit.

Results: There were no significant differences in performance between the test method and reference culture methods for any of the three surfaces tested. No statistically significant differences were found by probability of detection analysis (POD) in any of the surfaces when results were compared to the United States Food and Drug Administration cultural microbiology reference method for Listeria.

Significance: This data collected in these studies demonstrate that the CERTUS Listeria test is reliable and specific for detection of Listeria from stainless steel, ceramic tile, plastic (polyethylene) and sealed concrete environmental surfaces.

P3-16 - Evaluation of the Certus Environmental Listeria spp. Detection Kit for the Detection of Listeria spp. on Environmental Surfaces: AOAC Performance Tested Method 10100

Joyun Ju, Nicole Kliss*, and Evelyn Lue

*CERTUS Food Safety, Chicago, IL, 2IQ Laboratories, Inc., Cincinnati, OH

Introduction: The CERTUS Environmental Listeria spp. Detection Kit (CERTUS EL-Detection Kit) is a real-time, bio-contains assay designed to accurately detect Listeria spp. (L. crispatus, L. innocua, L. monocytogenes, L. seeligeri, and L. welshimeri) from environmental samples employing an aptameric DNA-antibody capture system in combination with a fluorescent reporter system.

Methods: Inclusivity and exclusivity, method comparison studies, product consistency and stability were conducted to evaluate the new method according to the AOAC Appendix guidelines.

Purpose: To evaluate a Listeria Detection Assay Kit in a AOAC Performance Tested Methods design validation study.

Significance: The data collected in these studies demonstrate that the CERTUS Listeria detection kit is a reliable and specific method for the fast and specific detection of Listeria from stainless steel, ceramic tile, plastic (polyethylene) and sealed concrete environmental surfaces.

P3-17 - Qualitative Comparison of Environmental Swabbing Devices for Recovery of Listeria monocytogenes from Stainless Steel

Jovan Mazlaraz, Diana Stewart and Mary Lou Tortorelli

U.S. Food and Drug Administration, Bedford Park, IL

Introduction: Environmental sampling is an important tool for monitoring pathogens in food production environments. A variety of devices may be used for sampling their utility for recovering listeria monocytogenes from food contact surfaces is not understood.

Purpose: To compare recovery of L. monocytogenes from stainless steel surfaces using conventional swabs and sponges as well as uncom-

Methods: Four devices were spotted with an L. monocytogenes cocktail in BIP or on cheese when at a 1.7 log CFU per four (small) or eight-by-eigen [large] surface and dried overnight. Devices for sampling four for four-in four areas included cotton, polyester, foam, and flocked swabs; foam and flocked sponges, cotton, polyester foams and cleanroom and microfiber wipes for eight by eight-in surfaces. Surfaces were swabbed hori-

Results: Four devices were statistically different from each other in terms of listeria monocytogenes recovery. The flocked swab was statistically worse for both 24 and 48 h enrichment periods (20% and 23% positive, respectively). Overall, the flocked swab was incapable of properly

Significance: The foam swabs resulted in more positive detection of L. monocytogenes than the other devices regardless of surface size or presence of food residue.

P3-18 - Use of 3M Molecular Detection Assays for Detection of Salmonella spp., E. coli O157:H7 and Listeria monocytogenes in Fresh Spinach and Environmental Samples

Erick Reyes1, Fabiola Ramirez1, Angel Treviño, Alejandro Aguirre1, Gustavo González-González2, Melita Erandy Cabello-Aceves1 and An-gélica Alejandra De la Torre-Arana1

1SAGA, Queretaro, Mexico, 23M Food Safety Mexico, Guadalajara, Mexico, 33M Food Safety México, Querétaro, Mexico

Introduction: Leafy greens are considered vulnerable to contamination due to usage of sprinkler irrigation water, “triple wash” and their consumption

Methods: Performance of the test method was compared to the United States Food and Drug Administration cultural microbiology reference method for Salmonella spp., E. coli O157:H7 and L. monocytogenes.

Results: For whey-inoculated small surfaces (n=40), the rank by percent positive detection was cotton/polyester foam > flocked sponges after both enrichment lengths. For the large surfaces (n=40 to 40), the foam swabs resulted in more positives regardless of the presence of a food. The flocked swab was statistically worse for both 24 and 48 h enrichment periods (20% and 23% positive, respectively). Overall, the flocked swab was incapable of properly swabbing the large surfaces.

Significance: The foam swabs resulted in more positive detection of L. monocytogenes than the other devices regardless of surface size or presence of food residue.
P3-19 Evaluation of a Validated Loop of Isotermal Amplification (LAMP)-Bioluminescent Technology for the Detection of Listeria spp. and Salmonella spp. in Three Different Matrices

Olivea Lugo-Magana1, Patricia Salcido-Brière1, Adrián Ríos-Avila1, Brenda Arianna Sánchez-Vera1, ÁngelCastillo-Juárez2, Carlos Sepúlveda-Ibarra3 and Gustavo González-González1

1Análisis Técnicos, S.A. de C.V., Pachuca, Mexico, 2SM Food Safety México, Guadalajara, Mexico

Purpose: To determine the performance of an amplification method for detection of Listeria spp. and Salmonella spp. in peppers, deli sandwiches and surface samples.

Methods: Forty-five samples (25 g) of fresh peppers, RTE ham and cheese sandwiches obtained from a local retailer in Pachuca Mexico and surfaces sampled at a local produce market were contaminated with Listeria innocua and Salmonella Typhimurium ATCC 14028 at seven different levels (1, 10, 15, 40, 140, 1400, 7500 and 7600 CFU/g). Samples were pre-enriched with Demi Fraser broth and buffered peptone water (ISO and Salmonella respectively). All the samples were analyzed by LAMP method and culture confirmed. For each pathogen, product category and environmental sponges, thirty samples were used to establish the performance of the method. Sixteen-stamp product groups were inoculated with different inoculum levels (80%, 40% and 10%) for each matrix; a low inoculum level, medium inoculum level and a high inoculum level.

Results: All the 153 contaminated samples confirmed with Listeria innocua, Listeria innocua or L. monocytogenes were reported as positive for Listeria spp. by LAMP method and confirmed by culture. All the 153 samples contaminated with Salmonella yielded as positive by both detection methods. The eighteen samples inoculated with interferent microorganisms were detected as negative for Listeria spp. and Salmonella spp. Respectively. LAMP method had 100% sensitivity for contaminated samples and 100% for inclusivity and exclusivity.

Significance: Use of the LAMP-bioluminescent technology is a suitable tool for the recovery of Listeria spp. and Salmonella spp. from fresh peppers, deli sandwiches and environmental samples.

P3-20 Independent Validation of a Proprietary Service-Based Method for Detection and Identification of E. coli O26, O103, O111, O121, O145 and O157:H7

Erin Crowley1, Edan Hosking2, Brooke Roman3, Susan Alles4, Susanne Hinkey1, Karen Cooper1, Danielle Keys1, Mark Mozola1, Robert Donofrio1, Benjamin Bastin2 and Wesley Thompson3

1Q Laboratories, Inc., Cincinnati, OH, 2Merck KGaA, Darmstadt, Germany, 3Merck, Molsheim, France

Purpose: To develop and validate a method for the detection of E. coli O26, O103, O111, O121, O145 and O157:H7 in diverse matrices.

Methods: The candidate method was evaluated in a multi-laboratory collaborative study. A total of nine technicians from eight laboratories, representing government and industry, throughout the United States participated. Each laboratory evaluated six replicates at three levels of natural contamination for each matrix; a low inoculum level, medium inoculum level and a high inoculum level.

Results: All 153 samples were confirmed as positive by both detection methods. The eighteenth samples inoculated with interferent microorganisms were detected as negative for E. coli O26, O103, O111, O121, O145 and O157:H7 by the method and confirmed by culture. For each pathogen, product category and environmental sponges, thirty samples were used to establish the performance of the method.

Significance: Correct O-group identifications and STEC/non-STE C determinations can be made from an isolate or complex enrichment sample in 36 h and at a cost of less than $50. This evaluation demonstrates the suitability of GENE-UP for the detection of Salmonella spp. in various products used in the chocolate and confectionery industry.

P3-24 Evaluation of the BIOMérieux VIDAS/GENE-UP® Top? Shiga Toxin-producing E. coli Detection System

Robert Barlow and Kate McMullan

CSIRO Agriculture & Food, Brisbane, Australia

Purpose: Testing of beef products for the presence of Shiga toxin producing E. coli (STEC) typically relies on detecting genes that encode Shiga toxin (stx), intimin (eae) and specific O-antigens. Test systems that utilise an STEC concentration procedure may decrease the number of potential positives (PPs) that require and secondary (n=5) samples by PCR analysis.

Methods: Samples (375 g) of each product were inoculated with one to 10 CFU/test of a strain of Salmonella spp., along with a non-target organism - approximately 10^8 CFU of each strain of Listeria innocua, the target strain, two separate type A STEC strains (Btg1 and Btg2), and one type A STEC strain which was mixed with four of the stock cultures of nontarget organisms. Five replicate 25-g samples was analyzed by the reference method ISO 6579-1:2017. All results were culturally confirmed via extended confirmation methodology.

Results: For all products and iterations, Gt provided 100% agreement to the extended culturally confirmed result with acceptable performance compared to the ISO 6579-1:2017 reference method.

Significance: This evaluation demonstrates the suitability of GENE-UP for the detection of Salmonella spp. in various products used in the chocolate and confectionery industry.
Purpose: The objective of this study was to evaluate the performance of a fluorescent resonance energy transfer (FRET)-based real time PCR (qPCR) protocol for the detection of Cronobacter spp. from stainless steel and plastic environmental surfaces according to the current AOAC validation guidelines.

Methods: Four by four inch stainless steel surfaces were inoculated with two levels (two and 20 CFU) of Cronobacter sakazakii. Stainless steel surfaces were inoculated with a total of 10 samples of Escherichia coli 96712 and one by one inch plastic surfaces were inoculated with two levels (10 and 70 CFU) of Cronobacter sakazakii. Following inoculation and stabilization, surfaces were sampled using horizontal and vertical sweeping motions. Replicates from each surface were analyzed by both the qPCR method and ISO 22964:2017. All Gene-UP PCR results were considered equal to “true positive” if the reference method identified the presence of the target bacteria (e.g., Cronobacter, Cronobacter Isolation (CCI) and Enterobacter sakazakii isolation agar (ESIA)).

Results: Following inoculation, there was no statistically significant difference between preservative and confirmed results (p<0.05) or between control and reference method results (p<0.05) for either stainless steel or plastic samples. All candidate and reference methods for both surfaces provided zero of five uninoculated positives, 14 of 20 positives at the low level and five of five high inoculated positives. The qPCR and the methods and the CRI detection took 357.1 kb for one of the phages, which can display common epitopes that may lead to false positive results.

Significance: Due to the high sensitivity and use of qPCR for the detection of Cronobacter spp. from environmental surfaces.

P3–29 - 30
Thermo Scientific Brilliance CampyGenotype Enumeration Method Microbial Validation in Comparison to ISO 16140-2:2016

Amanda Manolis, Jessica Williams, Anna-Maria Leonte and Gal Bettis

1Thermo Fisher Scientific, Austin, TX, 2Thermo Fisher Scientific, Basingstoke, United Kingdom, 3SGS, Portugal, Portugal, 4Laboratoire Microsept, Le Lion-d'Angers, France

Introduction: The Thermo Scientific BiIlance CampyGenotype enumeration method (alternative method) has been validated by MicroVal for the enumeration of coliform monoglycol from a broad range of foods and environmental samples.

Methods: The sample size was compared to the EN ISO 6881-1:1999 DAM2:2017(E) method during the relative trueness, accuracy profile, inclusivity and exclusivity, and inter-laboratory (ILS) studies.

Results: The relative trueness study (74 samples) results satisfied the requirements of EN ISO 16140-2:2016. The accuracy profile tolerance intervals were 0.18 and 0.21 for the alternative method and 0.2 for the reference method. The average reproducibility across all spike levels was 0.23 for the alternative method and 0.35 for the reference method.


P3–30 - 31

Amanda Manolis, Jessica Williams, Anna-Maria Leonte and Francisco Le Nestour

1Thermo Fisher Scientific, Austin, TX, 2Thermo Fisher Scientific, Basingstoke, United Kingdom, 3SGS, Portugal, Portugal

Introduction: The Thermo Scientific Listeria Precis Enumeration Method (alternative method) has been certified by NF VALIDATION for the enumeration of Listeria monocytogenes from a broad range of foods and environmental samples.

Methods: Samples were EVALuated in buffered peptone water (one to ten ratio) and serially diluted as described in ISO 6887-1:2017. The alternative method was validated using the relative trueness, accuracy profile, inclusivity and exclusivity, and inter-laboratory (ILS) studies.

Results: The relative trueness study (74 samples) results satisfied the requirements of EN ISO 16140-2:2016. The accuracy profile tolerance intervals were 0.18 and 0.21 for the alternative method and 0.2 for the reference method. The average reproducibility across all spike levels was 0.23 for the alternative method and 0.35 for the reference method.

Significance: The Listeria Precis Enumeration Method is equivalent to the EN ISO 11290-2:2017 reference method for the enumeration of Listeria monocytogenes from a broad range of foods and environmental samples.

P3–31 - 32

Amanda Manolis, Jessica Williams, Anna-Maria Leonte and Gal Bettis

1Thermo Fisher Scientific, Austin, TX, 2Thermo Fisher Scientific, Basingstoke, United Kingdom, 3SGS, Portugal, Portugal

Introduction: The Thermo Scientific Brilliance Staph 24 enumeration method (alternative method) has been certified by MicroVal for the enumeration of coagulase positive Staphylococcus species from a broad range of foods.

Methods: Samples were evaluated in line with EN ISO 16140-2:2016, and assessed in performance comparison to the EN ISO 6881-1:1999 DAM2:2017(E) reference method.

Results: The sample size was compared to the EN ISO 6881-1:1999 DAM2:2017(E) method during the relative trueness, accuracy profile, inclusivity and exclusivity, and inter-laboratory (ILS) studies.

Results: The relative trueness study (74 samples) results satisfied the requirements of EN ISO 16140-2:2016. The accuracy profile tolerance intervals were 0.18 and 0.21 for the alternative method and 0.2 for the reference method. The average reproducibility across all spike levels was 0.23 for the alternative method and 0.35 for the reference method.

Significance: The Thermo Scientific Brilliance Staph 24 enumeration method is equivalent to the EN ISO 6881-1:1999 DAM2:2017(E) reference method for the enumeration of coagulase positive Staphylococcus species from a broad range of foods and environmental samples.
Amanda Manolis1, Ana-Maria Leonte1, Yasmine Rannou2, Muriel Bernard2 and Jessica Williams1

1Thermo Fisher Scientific, Basingstoke, United Kingdom, 2Thermo Fisher Scientific, Austin, TX

Poster

Methodology

Results:

During the sensitivity study, out of 438 samples tested, 21 negative deviations and 24 positive deviations were observed. The number of discordant results is likely due to the fact it is an unpaired study. The observed values for ((ND+PPND)-PD) were below or equal to the acceptability limit for combinations. During the AOAC-RI PTM and NF Validation by AFNOR certification studies.

Introduction:

P3-34 Improved Salmonella Detection from Primary Production Samples Using Multiplex PCR Method

Charlotte Cooper1, Katharine Evans1, David Cribbee2, Annette Hughes1 and Amanda Manolis1

1Thermo Fisher Scientific, Basingstoke, United Kingdom, 2Thermo Fisher Scientific, Austin, TX

Purpose: To evaluate the detection capabilities for contaminated samples of the Thermo Scientific RapidFinder Salmonella Species, Typhimurium and Enteritidis Multiplex PCR Kit in comparison with traditional ISO and FISIG MLG methods.

Methods: Fifty-eight 25-g samples, including pork and poultry and production environment samples, were dual-infected with 0.6 to 4.8 CFU Salmonella enterica serovar Typhimurium and Enteritidis. The poultry producer identified one (4.0%) triple infected sample.

Purpose: To conduct AOAC-RI PTM and NF Validation by AFNOR Certification extension studies to validate use of the SureTect Listeria monocytogenes PCR Assay with the ThermoQuattro 5 Real-Time PCR instrument with Thermo Scientific RapidFinder Assay Software version 1.0 (Software the alternative method) for a broad range of foods and environmental samples.

Methods: The validation studies were conducted according to the AOAC-RI PTM, NF Validation and ISO 11290-1:2017 guidelines. For the alternative method, all protocols were followed. Following direct testing, PCR was run and results were automatically interpreted by the software. The method was confirmed according to EN 11290-1:2017.

Results: A total of 150 (AOAC-RI PTM) and 387 (NF Validation) food and environmental samples were tested using the alternative and EN ISO 11290-1:2017 method.

Significance: The alternative method proved to be a suitable substitute to the EN ISO 11290-1:2017 reference method for E. coli monocytogenes detection.
Validation for a Method to Detect Surviving Salmonella Mutants in French Dairy Powders

Methodology:
- Multi-gene deletion (MGD) mutant libraries of Salmonella Typhimurium strain 12048s were grown in 96-well plates at 37°C for 24 h. Mutants were spotted both on agar plates (37°C, 24 h) and on sterile nitrocellulose (six cm by six cm) as model abiotic surface for multiplex survival tests. The plates were incubated at 37°C for 24 h. Mutants incapable of surviving on NC were further validated for impaired survival on NC after 24 h and seven d. In-shell pistachios were inoculated with selected mutants, dried to their original, and stored at 25°C,黑暗). Whole populations were enumerated immediately after inoculation, after drying and periodically during storage on agar medium incubated at 37°C for 24 h.

Results:
- Of the 288 MGD mutants 12 were consistently impaired for survival on NC. Testing four mutants (D01, B03, A09 and D04) on pistachios revealed two mutants (D01 and A09) with significantly (p < 0.05) greater reductions after drying in comparison to the wild type parental strain. Subsequent reductions at 7 d were also significantly higher for D01 and A09 as compared to WT.

Conclusion:
- A limited number of MGD mutants with defective survival on NC are found in the screening of 288 mutants. Our approach is efficient to identify surviving Salmonella mutants that may be food poisoning-causing pathogens.

Significance:
- The present study opened the way to validate the performance of the multiplex survival tests using pistachios and whole populations, which is essential to detect any possible surviving Salmonella strain.

Validation for a Multiplex Screening of Salmonella Mutants for Survival on Dry Surfaces

Methodology:
- Salmonella Typhimurium strain 12048s was grown in 96-well plates at 37°C for 24 h and on sterile nitrocellulose (six cm by six cm) as model abiotic surface for multiplex survival tests. The plates were incubated at 37°C for 24 h. Mutants incapable of surviving on NC were further validated for impaired survival on NC after 24 h and seven d. In-shell pistachios were inoculated with selected mutants, dried to their original, and stored at 25°C,黑暗). Whole populations were enumerated immediately after inoculation, after drying and periodically during storage on agar medium incubated at 37°C for 24 h.

Results:
- Of the 288 MGD mutants 12 were consistently impaired for survival on NC. Testing four mutants (D01, B03, A09 and D04) on pistachios revealed two mutants (D01 and A09) with significantly (p < 0.05) greater reductions after drying in comparison to the wild type parental strain. Subsequent reductions at 7 d were also significantly higher for D01 and A09 as compared to WT.

Conclusion:
- A limited number of MGD mutants with defective survival on NC are found in the screening of 288 mutants. Our approach is efficient to identify surviving Salmonella mutants that may be food poisoning-causing pathogens.

Significance:
- The present study opened the way to validate the performance of the multiplex survival tests using pistachios and whole populations, which is essential to detect any possible surviving Salmonella strain.
P3-45 Salmonella Typhimurium-specific Signatures as Targets for Detection by Using DNA aptamers in Foods and the Environment
Azrina Nawawi and Srinand Sreevatsan
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**Developing Aptamers in the Typhimurium Environment**

Introduction

Salmonella Typhimurium is most frequently associated with foodborne outbreaks. Standard microbial cultivation methods along with PCR are gold standard for the detection of this pathogen, but these are labor-intensive and take about 72 hours for a confirmatory diagnosis. Thus, rapid, specific, and cost-effective assay to detect and differentiate Salmonella infection from other foodborne pathogens are direly needed for early recognition to help curtail outbreaks, improve timely clinical interventions and routine surveillance to help prevent outbreaks.

**Purpose**

This study aimed to develop a highly precise and rapid assay for detection of Salmonella Typhimurium using the outer membranes lipopolysaccharide (LPS) and oligopyrophosphate (OPS)-specific aptamers. Methods: Lipopolysaccharide (LPS) and oligopyrophosphate (OPS) of Salmonella Typhimurium were extracted and used for a one-step aptamer selection.

The selected candidate aptamers were validated for binding to LPS and ORF by using a dot blot assay. Aptamers bound to LPS and ORF were eluted and reamplified and cloned for DNA sequencing of individual candidates.

Results: Of thirty OPS-specific aptamer clones, two redundant of aptamer sequences were identified. There was no redundancy in the amplicon from LPS-specific clones. A second selection against OPS and LPS samples have been performed and amplicons are currently being sequenced.

Significance: These results suggest that the redundant aptamer sequences are likely capable of capturing Salmonella Typhimurium with high affinity and specificity.

P3-46 Amplified Nucleic Acid Single Temperature Reaction for Detection of Genogroup II Human Noro viruses
Jeremy Faircloth, Edan Hosking, Eric Toyav and Lee-Jay Ark
North Carolina State University, Raleigh, NC, 2Neogen Corporation, Lansing, MI

**Methods:**

Routine clinical detection assays are available for human norovirus (HNV), but these are more cumbersome for food and environmental samples. One complication is the absence of detection platforms with potential to yield results on-site and in real-time.

**Purpose:**

Develop and validate of a novel, rapid, and simple isothermal method for direct detection of genogroup II (GII) HNV.

**Methods:**

Amplicons were used as templates in a reverse transcription isothermal amplification (RT-PCR) reaction. 

**Results:**

Thirty OPS-specific aptamer clones, two redundant aptamer sequences were identified. There was no redundancy in the amplicon from LPS-specific clones. A second selection against OPS and LPS samples have been performed and amplicons are currently being sequenced.

**Significance:**

These results suggest that the redundant aptamer sequences are likely capable of capturing Salmonella Typhimurium with high affinity and specificity.

P3-47 Assessment and Comparison of Molecular Subtyping and Characterization Methods for Salmonella Enteritidis
Pinil Tangtrakool, Poornima Giri, Guanzeng Zhang, Robert Baker and Martin Wiedmann
Marlin Global Food Safety Center, Beijing, China, 2Cornell University, Ithaca, NY

**Methods:**

The food industry is facing a major transition with regard to methods to be used for confirmation, characterization and subtyping of foodborne pathogens. While whole genome sequencing (WGS) is rapidly becoming both the method of choice and the gold standard for subtyping foodborne pathogens, the cost and availability of these methods for rapid and reliable food safety may often be unaffordable, even for large companies and the need for rapid food safety may often be unaffordable, even for large companies.

**Purpose:**

In order to facilitate decision making regarding selection of subtyping methods by the food industry, we developed classical serotyping, pulsed-field gel electrophoresis, multilocus enzyme typing, and ribotyping, and WGS (including WGS-based serotype prediction) for rapid performance characteristics, using Salmonella as a model system.

**Methods:**

Performance characteristics included cost, expertise required for data collection and interpretation, global access to a given technology and time-to-result.

**Results:**

Our literature-based assessment supports the superior discriminatory power of WGS, however, we also identify circumstances under which use of other subtyping methods may be warranted.

P3-48 Performance Comparison of Shiga Toxin-producing E. coli Multiplex Molecular Assays
Jani Holopainen1, Laura Vaaranen1, Hanna Lehmusniemi, Tuomo Hurst, Jonna Roivanen1, Suvi Airikka1, Arokiyappan Muthukumaraswamy, Harri Koivisto, Maria Kairaluoma1, Marja Niemelä1, Taneli Arvilommi1, Tuomas Kallio, Frits Vos, Elinor Tettelin1, Thermo Fisher Scientific, Beijing, China

**Introduction:**

EHEC and STEC are important foodborne pathogens. Poultry and egg products are among the many food commodities which are regularly linked to EHEC/EHEC outbreaks. In this study, we compared the performance of the SureTect STEC Multiplex Assay, which is approved by the USDA for STEC detection, against one of the most commonly used methods for STEC detection, the GDS EHEC ID assay

**Purpose:**

To compare the performance of the SureTect STEC Multiplex Assay with the USDA approved GDS EHEC ID assay for the detection of E. coli O157:H7 and non-O157 H7 STEC.

**Methods:**

Ground beef, frozen finely textured beef and carcass samples were inoculated with EHEC and stabilized. Ninety six samples, of 25 to 375 g (or one carcass), were inoculated with two different levels of 1,000 CFU and 0.02 CFU of two EHEC serogroups (EHEC ID) assay, to identify EHEC.

**Results:**

Both assays returned a false positive result for E. coli O45 with O157:H7 (EHEC ID) assay, to identify EHEC.

**Significance:**

This newly developed STEC Multiplex assay was found to be more accurate and sensitive than the alternative assay testing. The sensitivity of the STEC Multiplex assay was found to be more accurate and sensitive than the alternative assay testing, which was performed by agar-agar diffusion test and PCR amplification of O157:H7 and non-O157 H7 serogroups.

**Significance:**

The study demonstrated that the SureTect STEC Multiplex Assay workflow proved to be more accurate and sensitive than the alternative assay testing. The sensitivity of the STEC Multiplex assay was found to be more accurate and sensitive than the alternative assay testing, which was performed by agar-agar diffusion test and PCR amplification of O157:H7 and non-O157 H7 serogroups.

**Significance:**

The study demonstrated that the SureTect STEC Multiplex Assay workflow proved to be more accurate and sensitive than the alternative assay testing. The sensitivity of the STEC Multiplex assay was found to be more accurate and sensitive than the alternative assay testing, which was performed by agar-agar diffusion test and PCR amplification of O157:H7 and non-O157 H7 serogroups.
P3-51 Validation of a Novel Isothermal Amplification Method for the Detection of Salmonella Enteritidis in Shell Eggs

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Introduction: Salmonella Enteritidis is a public health concern worldwide. Rapid and accurate detection of Salmonella Enteritidis in eggs and egg products, which are the major sources of this pathogen, is imperative for surveillance and outbreak investigation. Most molecular detection methods are aimed at the species level. A Salmonella Enteritidis specific LAMP method has been developed in our laboratory.

Purpose: To validate a probiotic gene LAMP method developed in our laboratory for detecting Salmonella Enteritidis in shell eggs in comparison with the FDA BAM culture method and a real-time PCR method.

Methods: For the comparison with shell eggs, Salmonella serovars isolated from different phage types (ST) were used. Each trial consisted of 20 samples (inoculated at ~5 cells), and five positive controls (inoculated at ~50 cells), and 20 negative controls (uninoculated). For each sample, one liter of egg was added to a preenrichment broth and incubated aerobically for 24 hours at 37°C. DNA was prepared from preenrichment broth samples and subjected to PCR and LAMP methods. Bacterial enumeration was performed on the FDA BAM culture method.

Results: For Salmonella Enteritidis, sensitivity and specificity were calculated to be 100% and 99.9%, respectively, for both the LAMP and PCR method. For all Salmonella serovars, sensitivity and specificity were calculated to be 100% and 99.9%, respectively, for both the LAMP and PCR method.

Significance: Our newly designed probiotic gene-LAMP method was equally effective in detecting Salmonella Enteritidis from shell eggs in comparison with the real-time PCR and BAM culture methods. It could be used as an alternative tool for the detection of Salmonella Enteritidis from shell eggs for FDA in outbreak investigation and enforcing the Egg Rule.

P3-52 Development of a Colorimetric Loop-mediated Isothermal Amplification Assay Using Molecular Beacon Mimicking the Radish Peroxidase-mimicking DNAzyme (HRPzyme) for the Rapid and Sensitive Detection of Salmonella Enteritidis

Jeong-Eun Lee, Junho Kim, Joon Won Shim

Jeong-eun Lee: Gyeongsang National University, Jinju, South Korea, Joon Won Shim: Yonsei University, College of Pharmacy, Gwangju, Gyeongsang National University, South Korea

Introduction: In Europe, imported enoki mushrooms were contaminated with Salmonella species, particularly Salmonella monovovii, and returned or discarded. In Japanese eating style, the enoki mushroom is often consumed in salads without cooking, and contamination with Salmonella Enteritidis is a major public health concern. For outbreak investigation and enforcing the Egg Rule, the rapid and sensitive detection of Salmonella Enteritidis is imperative.

Methods: To develop the colorimetric loop-mediated isothermal amplification (LAMP) assay using a molecular beacon, homemade molecular beacon mimicking the radish peroxidase-mimicking DNAzyme (HRPzyme), was designed. Each sample/control contained one liter liquid eggs. Preparation of egg samples followed the FDA BAM method. DNA was prepared from preenrichment cultures were used for LAMP and real-time PCR assays. LAMP assay was run on the GeneAmp 9700 real-time PCR system.

Results: The LAMP assay detected all the 50 Salmonella Enteritidis strains tested and did not detect any of the 3 strains from the species panel. LAMP method had a significance of 0.01 (p<0.01) with 95% confidence for differentiation of Salmonella species. Positive control samples were positive, and all 50 negative control samples were negative for Salmonella Enteritidis. LAMP and PCR results matched the BAM culture results.

Significance: We newly designed probiotic gene-LAMP method was equally effective in detecting Salmonella Enteritidis from shell eggs in comparison with real-time PCR and BAM culture methods. It could be used as an alternate tool for the detection of Salmonella Enteritidis from shell eggs for FDA in outbreak investigation and enforcing the Egg Rule.

P3-53 Rapid Detection of Campylobacter in Poultry Matrices Using a Loop-mediated Isothermal Amplification (LAMP)-Bioluminescent Assay

Jeri Lynn Pickett1, Melissa Sisemore1, Jamie Gossel1, Jesse Gossel1, Christina Barnes2, John David1 and Raj Rajagopal3

1WBAF Laboratory and Industrial Services, 2IM Food Safety, Plattsburg, MO

Introduction: Campylobacter spp. are the most common causes of diarrheal illness in the United States. Campylobacter jejuni and C. coli are microaerophilic requiring complex media and incubation under microaerobic conditions for optimal growth and viability. The RTU medium allowing aerobic incubation and the LAMP-bioluminescent assay is designed for rapid and specific detection of Campylobacter in a variety of poultry matrices.

Purpose: This study suggests that the LAMP assay for C. jejuni can be used as a point-of-care molecular diagnostic technology because the method does not require any expensive instruments such as a thermocycler and detector.

Methods: For the comparison with shell eggs, Salmonella serovars isolated from different phage types (ST) were used. Each trial consisted of 20 samples (inoculated at ~5 cells), five positive controls (inoculated at ~50 cells), and five negative controls (uninoculated). Each sample/control contained one liter liquid eggs. Preparation of egg samples followed the FDA BAM method. DNA was prepared from preenrichment cultures were used for LAMP and real-time PCR assays. LAMP assay was run on the GeneAmp 9700 real-time PCR system.

Results: The LAMP assay detected all the 50 Salmonella Enteritidis strains tested and did not detect any of the 3 strains from the species panel. LAMP method had a significance of 0.01 (p<0.01) with 95% confidence for differentiation of Salmonella species. Positive control samples were positive, and all 50 negative control samples were negative for Salmonella Enteritidis. LAMP and PCR results matched the BAM culture results.

Significance: For all matrices evaluated, the RTU enrichment broth, CBE and the 3 Molecular Detection Assay 2 - Campylobacter was equivalent or better than the reference methods for the rapid detection of Campylobacter. Hence, the LAMP assay is reliable for the rapid and specific detection of C. jejuni, C. lari and C. r. raw and cooked poultry products.

P3-54 Comparative Evaluation of the Ready-to-Use 3M Campylobacter Enrichment Broth and the 3M Molecular Detection Assay 2 - Campylobacter for the Detection of Campylobacter in a Variety of Poultry Matrices

Leslie Thompson-Schreitlow1, Nathan Clemens2, Hannah Bakken1, Christina Barnes3, Lisa Monteros3 and Raj Rajagopal4

1SGS Hargard Sciences, North Sioux City, SD, 2IM Food Safety, Plattsburg, MO, 3IM Food Safety, St. Paul, MN

Introduction: Campylobacter spp. are microaerophilic requiring complex media and incubation under microaerobic conditions for optimal growth and viability. The RTU enrichment broth and the LAMP-bioluminescent assay is designed for rapid and specific detection of Campylobacter after 24 to 26 hours of enrichment under anaerobic conditions.

Methods: For both methods, the LAMP assay to the MLG 41.04 for chicken carcasses, parts, raw poultry, ground turkey, turkey carcass swabs and raw chicken swabs was performed automated using 3M Campylobacter Detection Kit (uninoculated). Each sample/control contained one liter liquid eggs. Preparation of egg samples followed the FDA BAM method. DNA was prepared from preenrichment cultures were used for LAMP and real-time PCR assays. LAMP assay was run on the GeneAmp 9700 real-time PCR system.

Results: For both methods, the LAMP and the PCR assay to the MLG 41.04 for chicken carcasses, parts, raw poultry, ground turkey, turkey carcass swabs and raw chicken swabs was performed automated using 3M Campylobacter Detection Kit (uninoculated). Each sample/control contained one liter liquid eggs. Preparation of egg samples followed the FDA BAM method. DNA was prepared from preenrichment cultures were used for LAMP and real-time PCR assays. LAMP assay was run on the GeneAmp 9700 real-time PCR system.

Results: For both methods, the LAMP and the PCR assay to the MLG 41.04 for chicken carcasses, parts, raw poultry, ground turkey, turkey carcass swabs and raw chicken swabs was performed automated using 3M Campylobacter Detection Kit (uninoculated). Each sample/control contained one liter liquid eggs. Preparation of egg samples followed the FDA BAM method. DNA was prepared from preenrichment cultures were used for LAMP and real-time PCR assays. LAMP assay was run on the GeneAmp 9700 real-time PCR system.

Results: For both methods, the LAMP and the PCR assay to the MLG 41.04 for chicken carcasses, parts, raw poultry, ground turkey, turkey carcass swabs and raw chicken swabs was performed automated using 3M Campylobacter Detection Kit (uninoculated). Each sample/control contained one liter liquid eggs. Preparation of egg samples followed the FDA BAM method. DNA was prepared from preenrichment cultures were used for LAMP and real-time PCR assays. LAMP assay was run on the GeneAmp 9700 real-time PCR system.

Results: For both methods, the LAMP and the PCR assay to the MLG 41.04 for chicken carcasses, parts, raw poultry, ground turkey, turkey carcass swabs and raw chicken swabs was performed automated using 3M Campylobacter Detection Kit (uninoculated). Each sample/control contained one liter liquid eggs. Preparation of egg samples followed the FDA BAM method. DNA was prepared from preenrichment cultures were used for LAMP and real-time PCR assays. LAMP assay was run on the GeneAmp 9700 real-time PCR system.
A study was conducted to evaluate the performance of traditional GENE-UP assay (SLM), and unit dose format, a pelleted lyophilized Salmonella spp. background. The purpose of this study was to examine the sensitivity of Staphylococcus aureus (SPA) for detecting S. aureus in various foods.

Methods: Twenty-five gram portions of six food samples (beef (shock tender), marinated pork chop, semi-dried squid, dried filefish, rice cake, and jujube (dried fruit) were inoculated with a mixture of S. aureus strains (ATCC25923, ATCC25923, and ATCC35652) at three, five, and seven log CFU. The samples were left under laminar flow for 15 min to allow the cell attachment and stored at 4°C and 25°C for 24 h. To compare the sensitivity, S. aureus cell counts were enumerated on Baird-Parker agar (BPA) and STX Petrifilm, and the results were compared with the cyst.

Results: The recovered S. aureus cell counts were generally different between STX petrifilm and BPA. As a result of analyzing the level of significance between the two media, it was confirmed that there was no significant difference between two selective media regardless of the type of food or inoculation concentration (P > 0.05).

In conclusion, STX Petrifilm is judged to be a replacement for the conventional method of agar culture, as its detection power is similar and convenient compared to BPA.

Conclusion: The STX Petrifilm method is different than BPA, thus, using STX Petrifilm for S. aureus detection in food can save time and space.

Funding: This work was supported by the National Science Foundation (Grant No. 1665509).
P3-64  A Comparative Evaluation of the GENU-UP Listeria spp. Assay for the Detection of Listeria Species in Deli Ham and on Stainless Steel Environmental Surfaces Using Unit Dose Format
John Mills, Stan Bailey, Isha Khatib, Brinca Biree, Vikrant Dutta, Ron Johnson, Michelle Keener, Patricia Rule and Nikki Taylor

Introduction: A method comparison study was conducted using the ADAC methods Committee Guidelines for Validation of Microbiological Methods for Food and Environmental Surfaces. Materials and Methods: Performance Test Methods (PM) were approved by the ISO 15214:2015 for the new unit dose reagent format. Methods: The candidate method assay was compared to the USDA-FSIS-MLG 8.09: Identification and isolation of Listeria from Red Meat, Poultry and Eggs. Tryptose Soy Broth (TSB) and Listeria Enrichment Broth (LEB) were analyzed by both conventional and reference method at the different contamination levels. The results were statistically evaluated using ANOVA and regression. Results: For the deli ham, the candidate method obtained the following results for the three inoculation levels: zero of five for the uninoculated, 17 of 20 for the low and five of five for the high inoculation. For the stainless steel, the candidate method obtained the following results for the three inoculation levels: zero of five for the uninoculated, 17 of 20 for the low and five of five for the high inoculation. For both deli ham and stainless steel, the candidate method demonstrated no statistically significant differences between presumptive and confirmed results (p>0.05). The above methods were divided into two groups: inoculated and uninoculated. The first one (n=20) was recovered from raw deli ham and stainless steel, the candidate method demonstrated no statistically significant differences between presumptive and confirmed results (p>0.05). Significance: The method candidate Listeria spp. unit dose assay was considered equivalent to the USDA-FSIS-MLG 8.09 for both products evaluated. ADAC has approved the new unit dose format for the inocula assay.

P3-65  Performance of 3M Petrifilm Rapid Aerobic Count Plates for Determining Aerobic Bacteria Counts in Cocoa Products in Comparison to the Cocoa Culture Method
Daniel Intordo, 1 Teresa M. Brindley, 1 Shelly Yab1 and Carrie Lingle1

Introduction: The mesophilic aerobic count is used as a compliance requirement to assess the quality of cocoa products in Ecuador. A rapid and reliable method of quantification of these microorganisms enables timely corrective action in processing plants.

Purpose: To determine the performance of the alternative method, ADAC OMA 2015, 3M Petrifilm RCA in comparison to the traditional method.

Materials and Methods: Fifty Samples (n=50) of cocoa-based products (10 g) were diluted in 90 ml of sterile Butterfield’s phosphate-buffered diluent (pH 7.2). Forty samples of cocoa powder matrix had normal microbial flora and 10 samples of chocolate topping mixture were inoculated with Escherichia coli ATCC 25922 and Staphylococcus aureus ATCC 29523 at three different levels as below.

- Level 1: 1 to 2 CFU
- Level 2: 2 to 5 CFU
- Level 3: 5 to 10 CFU

The samples were tested using the alternative and traditional methods. The alternative method was inoculated at 35 °C for 24 h and the traditional method was inoculated at 35 °C for 48 ± 2 h. The plates were enumerated and the statistical differences were determined using t-test (P<0.05).

Results: The results showed that there were no statistically significant differences (P>0.05) between the alternative method, ADAC OMA 2015, and the reference method for chocolate topping (P=0.71) and cocoa Powder (P=0.16). The average recoveries of log CFU/g for cocoa powder was 2.8 ± 3.4 for Level 1, 2.8 ± 3.1 for Level 2, and 2.6 ± 3.1 for 3M Petrifilm RAC and for chocolate topping it was 1.26 ± 0.75 for TSA and 1.25 ± 0.75 for 3M Petrifilm RAC. The correlation coefficient (R²) between the two methods was 0.975.

Significance: 3M Petrifilm Rapid Aerobic Count Plate enabled reliable and rapid detection and counting of mesophilic aerobic bacteria in different cocoa products.

P3-66  Performance of Rapid Enumeration Methods for Lactic Acid Bacteria in Cured Meat Products from Brazil
Vanessa Tsuhako, 1 Danielle Almeida, 1 Maura Chiapinotito, 1 Alice Marafon, 1 Sandra Heidtman and 1 Carrie Lingle1

Introduction: Aerobic bacteria and Enterobacteriaceae in meat are considered important pathogens in the food industry due to their ability to produce harmful substances. The presence of these microorganisms in food can lead to food poisoning and is a serious health risk for newborn infants. Conventional culture methods are time consuming and not user-friendly for detecting both pathogens. Therefore, the Listeria spp. and Aeromonas spp. unit dose test kit was conducted following the AOAC International Method.
and 0.41 of CU of subtherally injured S. Agona and C. sakazakii, respectively, were used with the one-step broth method. No false positive or false negative outcomes were confirmed by the enumeration of the control step by step.

Significance: The one-step enrichment broth allowed for the simultaneous recovery of S. enterica and C. sakazakii in PF samples that significantly reduced the time-to-results for RTP-PCR or culture-based methods.

P3-73 | Evaluation of the Universal Enrichment Broth Salmonella, Staphylococcus, Shigella, Listeria and E. coli for the Detection of the Main Food Pathogens in Cheeses

Joseph Holic1, Karine Héroux2, and Vincent Marleau3
1Canadian Food Inspection Agency, St-Hyacinthe, QC, Canada
2FoodChek Systems Inc., Calgary, AB, Canada
3FoodChek Laboratories Inc., Sainte-Julie, QC, Canada

Introduction:
Several enrichment broths and different food portion sizes are required for foodborne pathogen analysis depending on the selected method and the food to be analyzed. The use of a single enrichment broth for the growth of the main pathogens would limit the variability of method related analyses.

Purpose:
The performance of the selective enrichment broth, designed for simultaneous growth of Salmonella, Staphylococcus, Shigella, Listeria, and E. coli (O157:H7) at 30°C was compared with the reference methods (VRBL-24 h, 30°C and TBX-24 h, 44.5°C). The limit of detection of the assay was also determined. The new method was used to determine the number of colonies present in 25 g of cheese.

Methods:
The impact of this project is the demonstration of significant improvements in laboratory methodology for tracking foodborne pathogens for organization testing high volume multi-commodity foods.

P3-74 | Rhamnose-substituted Buffered Listeria Enrichment Broth Increases Listeria monocytogenes Enrichment Populations in Select Seafood Matrices

Ronald Moulin.
U.S. Food and Drug Administration/OHAmas Laboratories, Jefferson, AR

Introduction:
Buffered Listeria enrichment broth (BLEB) is the preferred liquid enrichment medium of the United States Food and Drug Administration (FDA) for recovery of L. monocytogenes from food matrices. Despite the inclusion of selective agents, the enrichment procedure tends to favor the growth of non-target microorganisms at the expense of L. monocytogenes and the isolation of BLEB may be a waste of its functionality and hence the likelihood of recovering L. monocytogenes from seafood matrices.

Purpose:
This study is the validation of substituting rhamnose (BLEB-rhamnose) for glucose in BLEB on populations of L. monocytogenes following selective enrichment of select seafood matrices.

Methods:
Portions (25 g) of uncooked crab meat, shrimp, salmon, cod, squid, and scallops were spiked (one to five CFU/g) with L. monocytogenes (8–10 cells) and compared for the enumeration of the foodborne pathogen using PALCAM agar.

Results:
In comparison to BLEB, L. monocytogenes enrichment populations with associated inter-quartile ranges (IQR) were 9.2 (9.0 to 9.4), 5.2 (5.0 to 5.4), 4.4 (4.3 to 4.9), 3.3 (3.0 to 3.6), 4.8 (4.8 to 5.1), 5.8 (5.5 to 6.1), and 6.9 (6.2 to 7.5) CFU/ml for matrix-free crab meat, shrimp, salmon, cod, squid, and scallops, respectively, when BLEB-rhamnose was formulated with glucose. When BLEB-rhamnose was utilized, the median L. monocytogenes enrichment populations were 8.9 (8.0 to 9.0), 2.7 (2.6 to 2.8), 7.7 (7.5 to 7.9), 7.1 (7.0 to 7.3), and 7.6 (7.0 to 8.1) CFU/ml for the same seafood matrices, respectively.

Significance:
Rhamnose-substituted BLEB increased L. monocytogenes median enrichment populations in all seafood matrices tested. For all but one seafood matrix (squid) the improved L. monocytogenes enrichment populations were statistically significant (P<0.01). Currently there are no known combinations of selective agents and incubation conditions that allow growth only of L. monocytogenes, medium supplementation alteration and refinement may be practicable to improve the capabilities of regulatory agencies to recover this organism amidst a complex microflora.

P3-75 | Evaluation of the 3M Petrifilm Rapid E. coli/Coliform Count Plate and 3M Petrifilm Rapid Aerobic Count Plate for Enumeration Microorganisms in Raw Milk Samples in Thailand

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1Bureau of Quality Control of Livestock Products, Department of Livestock Development, Bangkok, Thailand, 23M Thailand Limited, Bangkok, Thailand
2Introduction: Thailand is a producer and exporter of dairy products and has raw milk production capacity around 2,800 tons/day, so just over one million tons per month. Achieving standard microbial parameters allows milk producers to meet quality standards and compete in the marketplace.

Purpose: Rapid methods allow producers to quickly assess the milk quality and improve their practices.

Methods:
This study was performed to comparatively enumerate populations of E. coli and coliforms from unspiked (20), spiked at limit of detection level (80) and 10 times higher (80) demonstrated that SSSLE is equivalent to the 3M Petrifilm Rapid E. coli/Coliform Count Plate (REC) and the 3M Petrifilm Rapid Aerobic Count Plate (RAC) Plate for rapid determination of total coliform and aerobic organisms in raw milk samples from Thailand.

Results:
The method comparison was conducted using a variety of naturally and artificially contaminated food matrices (protein broth, meat broth and rhamnose supplemented BLEB) for enumeration of total coliforms and aerobic organisms in raw milk samples from Thailand.

Purpose:
This study evaluates the effect of substituting rhamnose (BLEB-rhamnose) for glucose in BLEB on populations of L. monocytogenes cultured at 35°C or in the presence of competing bacteria such as E. coli, Citrobacter freundii, Proteus vulgaris and Proteus mirabilis, using a 10-100% response surface methodology to optimize the selection, recovery, growth and recovery of low numbers of subtherally injured Salmonella.

Methods:
More than 15 different substances including antibiotics, organic and inorganic compounds, detergents, and quaternary ammonium salts were screened for their effects on bacterial growth. The selected broth was used to evaluate the effect of the selected substances on the growth kinetics parameters of E. coli O157:H7 cultured at 35°C or in the presence of competing bacteria such as E. coli, Citrobacter freundii, Proteus vulgaris and Proteus mirabilis, using a 10-100% response surface methodology to optimize the selection, recovery, growth and recovery of Salmonella. All assays were carried out three times.

Results:
Three inhibitory substances were selected based on their capacity to control the growth of gram-positive and gram-negative bacterial cultures that might out-compete L. monocytogenes. The optimum concentration of each selected substance was determined to be 10 mM, 30 mM and 100 mM for BLEB-rhamnose, citric acid and Tween 80, respectively, which all showed significant correlation between the two methods. The 25% optimal concentration of each selected substance was selected to be 2.5% for BLEB-rhamnose, 7.5% for citric acid and 25% for Tween 80.

Significance:
The new selective supplement can be used with BPW for the one-step enrichment of L. monocytogenes in a broad range of temperatures compatible with global storage conditions.
P3-77 – P3-79

Comparing Anaerobic Systems, Culture Vessels and Initial Temperature of Enrichment Broth in the Recovery of Prevalent Virulent and Non-Virulent STEC from Food Type

Oluwaseun Agbaje, Clinton Thompson, Robert Duvall and Rachel Binet
U.S. Food and Drug Administration, College Park, MD

Purpose: To determine the effect of enrichment temperature on recovery of STEC from fresh ground beef and poultry enrichments.

Significance: Reductions in recoveries of STEC on cold enrichment media may be attributed to the cold-sensitive nature of these pathogens.

Methods: Twenty-seven fresh ground beef and poultry enrichments (150 g each) were inoculated with STEC strains, and incubated at 25, 30 and 35°C respectively. All enrichments were screened for STEC by plating onto SBA and TSBA, and for Shigella enterica by plating onto SBA with novobiocin. For many STEC strains, inducible type III secretion system (T3SS) activity is temperature-dependent. A chicken sample was spiked with dilutions of 100 µl of STEC strains, tested at 35°C and 42°C. A chicken sample was spiked with dilutions of 100 µl of STEC strains, tested at 35°C and 42°C.

Results: At 35°C, the T3SS-negative isolate was more inhibited than the T3SS-positive isolate at temperatures higher than 35°C. Induction of their T3SS at temperatures higher than 35°C may be burdensome.

Conclusion: The T3SS-negative isolate was more inhibited than the T3SS-positive isolate at temperatures higher than 35°C. Induction of their T3SS at temperatures higher than 35°C may be burdensome.

P3-80 – P3-82

Optimizing the Recovery of Wild Shigella from High Background Level Food Matrices

Uwesun Agbaje, Clinton Thompson, Robert Duvall and Rachel Binet
U.S. Food and Drug Administration, College Park, MD

Purpose: To determine the effect of enrichment temperature on recovery of STEC from fresh ground beef and poultry enrichments (SB) with novobiocin; however, induction of their T3SS secretion system (T3SS) at temperatures higher than 35°C may be burdensome.

Significance: Reductions in recoveries of STEC on cold enrichment media may be attributed to the cold-sensitive nature of these pathogens.

Methods: For each STEC strain, the effect of enrichment temperature on recovery was evaluated. For STEC strains Enteritidis 505 and Kentucky 501 and similarly incubated; inoculated tubes changed from purple to yellow. From all inoculated tubes and uninoculated controls, aliquots were heat-treated to inactivate the bacteria, and DNA purified and used as templates in PCR. For YeMT1, six poultry samples yielded PCR products; clustal analysis of their sequences revealed six distinct strains, and BLASTN queries of these strains against the GenBank database yielded high sequence identities. These six strains were subsequently tested for their ability to produce T3SS.

Results: For YeMT1, six poultry samples yielded PCR products; clustal analysis of their sequences revealed six distinct strains, and BLASTN queries of these strains against the GenBank database yielded high sequence identities. These six strains were subsequently tested for their ability to produce T3SS.

Conclusion: The T3SS-negative isolate was more inhibited than the T3SS-positive isolate at temperatures higher than 35°C. Induction of their T3SS at temperatures higher than 35°C may be burdensome.

P3-81 – P3-83

Compatibility of Polymorphic Locus Sequence Typing with Commercially Available Environmental Sampling Tests for Listeria and Salmonella

Tom Endlin and Yanhong Liu
Microbiotype LLC, Plymouth Meeting, PA. U.S. Department of Agriculture-ARS, Eastern Regional Research Center, Wyndmoor, PA

Purpose: For presumptive-positive ESTs, we hypothesized that a relatively minor additional investment could provide, in addition to species confirmation, valuable strain typing data for tracking pathogen spread through a facility, identify harborage sites, and distinguish sporadic from resident contamin-

Significance: These results suggest that the current reference methods for Shigella favors competitors over virulent S. dysenteriae. We are currently testing if improved fitness at 35°C can compensate for spinach's higher level of initial competitive flora. The recovery rates of S. dysenteriae, S. flexneri and S. sonnei from artificially contaminated spinach were being compared at 35°C and 42°C.

P3-82 – Culture-independent Typing of Foodborne Pathogens in Poultry Products

Tom Endlin and Yanhong Liu
Microbiotype LLC, Plymouth Meeting, PA. U.S. Department of Agriculture-ARS, Eastern Regional Research Center, Wyndmoor, PA

Introduction: For YeMT1, six poultry samples yielded PCR products; clustal analysis of their sequences revealed six distinct strains, and BLASTN queries of these strains against the GenBank database yielded high sequence identities. These six strains were subsequently tested for their ability to produce T3SS.

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P-83
Detection of Staphylococcal Enterotoxins A and B in Chicken Salad with RIDASCREEN and VIDAS Methods

Jossefin Daryaei1, Shannon Pickens1, Matthew Kmet1, Tara Doran1, Donald Burr2 and Ravinder Reddy2
1Wixam Institute of Technology & IDP, Bedford Park, IL, 2U.S. Food and Drug Administration, Bedford Park, IL, 3U.S. Food and Drug Administration, Office of Regulatory Affairs/Office of Regulatory Science, Rockville, MD

Purpose: The purpose of this study was to compare the enzyme immunoassay RIDASCREEN SET ELISA (LOD=0.375 ng) with the VIDAS SET automated enzyme-linked fluorescent assay (ELFA) (LOD=1.0 and 0.5 ng) for SEA and SEB, respectively, for the detection of SEA and SEB in chicken salad.

Methods: The chicken salad was spiked with 0.10, 1.0 and 5.0 ng/ml of SEA and SEB and analyzed using 3 replicates by each method. The ELFA was also performed at day four of the storage.

Results: Enterotoxins were detected in the spiked samples using both methods. The quantitative ELISA indicated the presence of SEA and SEB at a concentration of 0.55 and 0.15, respectively. The automated ELFA testing of ten subsamples at day four verified that the spiking and mixing method assured a homogenous distribution of the toxins in the matrix. Matrix spiked with SEA had positive test values ranging from 1.58 to 1.89 on day four and 1.50 to 1.64 on day 15. SEB spiked matrix had positive test values of 0.40 to 0.56 on day four and 0.53 to 0.57 on day 15.

Significance: The findings suggest that both ELISA and automated ELFA methods can detect SEA and SEB in chicken salad at concentrations as low as 1.0 and 0.5 ng/ml, respectively.

P-84
Detection of a Molecular Screening Assay for Escherichia coli Via Targeted Sequencing of the O-Antigen Gene Cluster

Jacob Eldjer1, Pina Fratamico1, Yanhong Liu2, Lori Bagi3, Robert Tebo3, Adam Allred4, Prasad Siddavatam2, Krishna Reddy Gujjula1, Hak-tan Suren1, Chitra DebRoy2, Edward Dudley2, David Needleman3 and Xiangze Yang1
1U.S. Department of Agriculture–ARS, Eastern Regional Research Center, Wyndmoor, PA, 2Thermo Fisher Scientific, Austin, TX, 3Bay Labs, Menlo Park, CA, 4The Pennsylvania State University, University Park, PA

Purpose: The objective of this study was to develop a high-throughput, molecular screening method for E. coli based on O-antigen gene cluster (O-AGC) sequencing. This method is based on next-generation sequencing data analysis and serogroup identification.

Methods: Publicly available E. coli O-AGC sequences were analyzed, and those that shared >95% identity were grouped into clusters. Representative sequences from each cluster were selected and analyzed for unique signature regions. Primers were designed and checked for specificity, confirming that there was no cross-targeting to other serogroups or Enterobacteriaceae genomes. To validate the assay, we extracted genomic DNA from O-mentioned standard strains, amplified the targeted O-AGC regions, prepared sequencing libraries from the amplified products, and sequenced the libraries on the Illumina HiSeq. The resulting sequence files were analyzed using the Serotype software for confirmation of serogroup.

Results: The initial sequence analysis for primer design revealed a total of 168 clusters of E. coli [some serogroups share the same O-AGC sequence and group together] O-AGC sequences with unique signature regions in the known or unknown serovars. Our primer design method allowed us to test the 168 primer pairs in a single reaction and test >100 strains per sequencing run. Of the 178 O-AGC standard strains tested, 173 (98%) were correctly identified by this assay. Three strains, representing serogroups 092, 0106, and 0126, were misidentified in one replicate.

Significance: The high-throughput, sequence-based method presented here is a reliable alternative to antisera-based serotyping methods for E. coli. For Research Use Only. Not for use in diagnostic procedures.

P-85
Development of an Integrated Detection Platform for the In-Process Surveillance of Listeria spp. in Environmental Monitoring Samples

Beatriz Quiñones1, Veronica DeGuzman1, Jazemyn Yambo2, David Medlin3 and Bertam Le1
1University of Dayton, 2Ohio Department of Agriculture, 3Ohio Agricultural Research and Development Center, Wooster, OH

Purpose: The objectives of this study were to develop and validate an integrated platform for the in-process surveillance of foodborne pathogens.

Methods: The FDA microphysiological environment models (FDMs) have been significantly implicated in high-profile outbreaks linked to fresh produce.

Results: Validation experiments indicated that the probe-based assay had an RNA analytical sensitivity limit of less than 10 fg of Listeria RNA or less than 5 CFU/ml by using crude lysate as template (Fishers exact test, P=0.001). No positive signals were detected when testing non-target RNA and common foodborne bacterial strains, such as Bolivus spp., Citrobacter spp., Enterobacter spp., and Pseudomonas spp., and preliminary observations indicated low concentrations of genome that could be targeted to the susceptibility of detecting advanced food processing facilities, collected at a leafy greens processing facility, was evaluated. Preliminary results showed that Listeria spp. were detected at concentrations ranging from >2 CFU/ml to 32 CFU/ml (Fishers exact test, P=0.001), recovered from spiked 100 ml-volume samples in the absence of an enrichment culturing step.

Significance: These findings have set the foundation for developing an integrated system to rapidly detect Listeria at low cell concentrations from environmental samples in large volume amounts without enrichment steps.

P-86
Detection of Staphylococcal Enterotoxins A and B in Chicken Salad with RIDASCREEN and VIDAS Methods

Jossefin Daryaei1, Shannon Pickens1, Matthew Kmet1, Tara Doran1, Donald Burr2 and Ravinder Reddy2
1Wixam Institute of Technology & IDP, Bedford Park, IL, 2U.S. Food and Drug Administration, Bedford Park, IL, 3U.S. Food and Drug Administration, Office of Regulatory Affairs/Office of Regulatory Science, Rockville, MD

Purpose: The purpose of this study was to compare the enzyme immunoassay RIDASCREEN SET ELISA (LOD=0.375 ng) with the VIDAS SET automated enzyme-linked fluorescent assay (ELFA) (LOD=1.0 and 0.5 ng) for SEA and SEB, respectively, for the detection of SEA and SEB in chicken salad.

Methods: The chicken salad was spiked with 0.10, 1.0 and 5.0 ng/ml of SEA and SEB and analyzed using 3 replicates by each method. The ELFA was also performed at day four of the storage.

Results: Enterotoxins were detected in the spiked samples using both methods. The quantitative ELISA indicated the presence of SEA and SEB at a concentration of 0.55 and 0.15, respectively. The automated ELFA testing of ten subsamples at day four verified that the spiking and mixing method assured a homogenous distribution of the toxins in the matrix. Matrix spiked with SEA had positive test values ranging from 1.58 to 1.89 on day four and 1.50 to 1.64 on day 15. SEB spiked matrix had positive test values of 0.40 to 0.56 on day four and 0.53 to 0.57 on day 15.

Significance: The findings suggest that both ELISA and automated ELFA methods can detect SEA and SEB in chicken salad at concentrations as low as 1.0 and 0.5 ng/ml, respectively.
91 Reproducibility of MALDI-TOF MS for Pathogen Confirmation and Identification of Non-pathogenic Bacterial Isolates: Assessment According to the AOAC Guidelines
Benjamin Bastin1, Patrick Bird2, Erin Crowley1, Claudia Le Douff1, Sarah Peron1, Maryse Rannou2, Danièle Sohier2 and Markus Timke1
1Q Laboratories, Inc., Cincinnati, OH, 2ADRIA Food Technology Institute, Quimper, France

Introduction: MALDI-TOF MS is recognized as a valuable method to confirm and identify microbial isolates. Four inter-laboratory studies were recently run to assess the reproducibility of the MALDI Biotyper.

Methods: Different validated test instruments, operators, types of target plates, culture media and enrichment conditions were applied.

Results: MALDI-TOF MS is a recognized method to confirm and identify microbial isolates. Four inter-laboratory studies were recently run to assess the reproducibility of the MALDI Biotyper.

Results: Twenty-four to 38 isolates were evaluated per study according to the Appendix (of the AOAC Guidelines (2016)). Each set of strains consisted of 16 pathogenic isolates. The results from 14 laboratories were compared. Intermediate and definitive identifications were assessed. The quality control was assessed as a quality control. Reusable and disposable target plates were tested. The appropriate standard procedures (ISO, FDA/BAM, USDA/FSIS) in parallel were run to confirm the pathogen presence of the sample.

Results: Impact of the selective culture media was observed. No influence of the tested target plates was noticed. The MALDI Biotyper shows 99.5% success rate for the non-pathogenic isolates depending on the study, and 98.9% to 100% correct identification rates at the group or species level. The correct confirmation rates of the standard procedures vary from 91.3% to 100%, the correct identification rates from 86.5% to 99.6%.

Significance: The MALDI-TOF MS is a recognized method to confirm and identify microbial isolates. Four inter-laboratory studies were recently run to assess the reproducibility of the MALDI Biotyper.

Developing Scientist Entrant

P3-94 Evaluation of Salmonella and Shiga Toxin-producing Escherichia coli Presence in Various Foods Using Rapid PCR-based Assay as Pro-screening Method
Ayodeji Adeniyi, Ramon Miranda, Darnin Cuellar and Alejandro Echeverry
Texas Tech University, Lubbock, TX

Purpose: The purpose of this study was to develop a simple detection method for simultaneous detection of E. coli and Salmonella in wheat flour by comparing different matrix pre-enrichment (homogenization/sonication) and specific agglutination test (SAT) methods.

Methods: Growth of E. coli strain in AY, EC, lauryl sulfate (LSB), buffered peptone water (BPW) and tryptose soy (TSB) broth media were compared for four and eight h at 42°C for selecting the shortest enrichment. A single commercial indirect IMS kit was used for their dual selectivity for E. coli and Salmonella and two immobilized specific antibodies (AS1 and AS2) were calculated. Broth and flour were inoculated with 0.1 log CFU/mL, enriched for four and 12 h in TSB, subjected to IMS and tested with commercial agglutination test results.

Results: After four and eight h enrichment in AY, LSB, TSB, and TSB, the average microbial counts of individual strains were 2.6, 2.7, 5.3 and 5.9, 4.5, 4.7, 5.8, 4.2, and 4.1 log CFU/mL and the CE was used in the following experiments. The CE values were in the range of 100 to 100% for E. coli and 90 to 94% for Salmonella strains. All CE values of non-target bacteria were <15.0%. The optimal E-IMS-AS strategy was selected and detected 0.1 log CFU/mL Salmonella and E. coli in wheat flour after 12 h enrichment.

Significance: This study reports a simple method capable of detecting both E. coli and Salmonella in wheat flour without advanced laboratory equipment in less than 24 h.

Developing Scientist Entrant
Out of 252 water samples, 34% tested E. coli negative. 83% of water samples were below the geometric mean limit of 126 CFU/100 mL whereas eight percent of water samples were above the statistically significant value of 410 CFU/100 mL. PCR assay identified three Shiga toxin-producing E. coli strains carrying the stx1 and stx2 genes, four atypical enteropathogenic E. coli strains carrying the eae gene but without the stx gene, one hybrid enterotoxigenic/aggrovirulent/pathogenic E. coli strain carrying the eae gene, one enteropathogenic E. coli strain carrying the eae and astA genes, and 33 heat-stable enterotoxin 1 encoded E. coli strains (EASTEC) carrying the stx gene but no other identifiable pathogenic genes.

Fifty potential pathogens E. coli isolates warrant further confirmation. Proper measures must be taken to control pathogens in agricultural water to ensure water safety and protect the public health.

P3-97 Development of an Ultra-Sensitive and Specific Multiplex Single-Tube Nested qPCR Assay for Simultaneous Detection of Campylobacter jejuni and Salmonella spp.
Biyu Wu and Yong Li
University of Hawaii at Manoa, Honolulu, HI

Introduction: Campylobacter jejuni and Salmonella species are the top two pathogens responsible for human bacterial gastroenteritis worldwide. Poultry and meat-associated products are the principal source of their infections. Traditional culture-dependent methods of microbiological analysis are laborious and time-consuming. Therefore, ultrasensitive and specific assay is rapid for identifying these pathogenic bacteria in food to protect the public’s health and minimize economic losses.

Purpose: This study aimed to develop a multiplex single-tube nested qPCR assay (MSTn-qPCR) for simultaneous detection of C. jejuni and Salmonella. Two nested primer sets were designed based on the 16S gene of C. jejuni and the invA gene of Salmonella. The annealing temperatures and concentrations of nested primers were optimized in a range of 50°C to 70°C and 0.05 pmol to 4 pmol, respectively. The specificity of established assay was tested with DNA extracted from C. jejuni and Salmonella spp., and nine non-target bacterial species. The sensitivity of MSTn-qPCR was investigated using serial dilutions of C. jejuni and Salmonella DNA.

Results: Optimal reaction conditions for MSTn-qPCR assay were 0.1 pmol of all four outer primers, 40 pmol of Salmonella inner primers, 20 pmol of C. jejuni inner primers, 0.1 pmol of TagMan probe for Salmonella and 5 pmol of TagManProbe for C. jejuni. The annealing temperature was 52°C for outer amplifications and 55°C for inner amplifications. The established assay could detect as low as 80 fg DNA of both target pathogens at one time without generating amplifications from non-target bacterial DNA.

Conclusion: The MSTn-qPCR assay provides an ultra-sensitive and specific approach for simultaneous detection of C. jejuni and Salmonella. It would help regulatory agencies and food manufacturers improve the safety of food supply.

P3-98 Quantification and Discovery of PCR Inhibitors Found in Food Matrices Commonly Associated with Foodborne Viruses
Cassandra R. Suther1 and Matthew D. Moore2
1University of Massachusetts, Amherst, MA, 2University of Massachusetts, Amherst, MA

Introduction: Human norovirus is the leading cause of foodborne illness globally. Detection and quantification of norovirus involves the use of reverse transcriptase quantitative polymerase chain reaction (RT-qPCR); however, one challenge in its utilization is the presence of compounds in food that can inhibit RT-qPCR reactions. Loans and malodors from fresh produce are a few of the most commonly implicated foods in foodborne norovirus outbreaks. Both are reported to have PCR inhibitors, but recent empirical quantification of the degree to which these compounds inhibit the RT-qPCR reactions has not been reported.

The purpose of this study was to observe and quantify the degree of inhibition that occurs from inhibitory compounds found in produce (pectin) and mollusks (hemocyanin, glycyrophosphate).

Methods: RT-qPCR reactions, containing different starting amounts between 10 and 100,000 copies of the genome from a nonvirulent surrogate, were performed in triplicate with or without 0.2% pectin (0.055 ± 0.03% w/v), glycyrophosphate (1.25 ± 1%), and hemocyanin (0.05 ± 0.03%). All reactions were performed in triplicate with no inhibitor and no template controls.

Results: Pectin, found in produce, caused complete inhibition at 0.25%, with no significant inhibition observed at 0.056% (P<0.05). Past research has identified pectin and mucilage as the compounds that inhibit RT-qPCR reactions. Malodors from fresh produce, such as hemocyanin, present in the hemolymph of mollusks and previously tested as a PCR inhibitor, caused complete inhibition at 0.5%, with no significant inhibition observed at 0.056% (P<0.05).

Significance: This study quantifies the degree of inhibition that occurs from three compounds implicated in foods associated with norovirus transmission, demonstrating that pectin and hemocyanin should be considered when testing produce and mollusks. This information helps inform sample preparation for PCR-based detection of foodborne pathogens from produce and mollusks, as well as identifies a previously unreported specific inhibitor compound found in mollusks.

P3-99 Evaluation of Roka Atlas-based Assay for Major Foodborne Pathogens in Food and Environmental Samples
Christina M. Ferrera, Je Zheng, Elizabeth Reed, Yi Chen, Thomas Hammack and Lalla Ali
U.S. Food and Drug Administration – Center for Food Safety and Applied Nutrition, College Park, MD

Introduction: Foodborne pathogens including Salmonella, Listeria monocytogenes, and Shiga toxin-producing Escherichia coli (STEC) account for nearly 48 million foodborne illnesses – including 128,000 hospitalizations and 3,000 deaths each year in the United States. Rapid, sensitive and specific detection methods are needed for the identification of major foodborne pathogens at various points in the food supply chain.

Purpose: To evaluate the Roka Atlas assay for the detection of major foodborne pathogens in food and environmental samples.

Methods: A 24-h preenrichment or enrichment for food and environmental samples was used. Samples were added into a modified sample transfer tube for bacterial lys, template-specific sample extraction, amplification and probe detection per fully automated assay protocol in the instrument. Each assay reagent kit was validated with a set of Salmonella, E. coli, or Shiga toxins calibrators (positive and negative) provided by the manufacturer. Results were confirmed with the United States Food and Drug Administration (FDA) Bacteriological Analytical Manual (BAM) culture method in tandem. Pathogens were evaluated in this assay for the following food matrices: meat, poultry, seafood, ground beef, pet food samples (n = 48) and raw (n = 12), rabbit and hamster food (n = 4), and other species (n = 4).

Results: With the exception of STEC, no other species resulted in any false positive results. The sensitivity of Roka Atlas assay was evaluated in this study. The 100% recovery broths were also evaluated for Salmonella detection using Roka Atlas Salmonella assay (SEN). The results from the assay were equivalent in most cases to BAM culture method results for Salmonella, STEC or L. monocytogenes, respectively. Universal preenrichment broth was the best preenrichment broth for detecting Salmonella in sprout grown irrigation water on the Atlas System.

Significance: The Roka Atlas assay provided a more rapid, sensitive, and specific method for detection of major foodborne pathogens in a variety of food and environmental samples.

P3-100 From Stools to Water: Categorization of Irrigation Water Using an Artificial Hand Tool Expected to Sool Stools Containing Oocysts of Cyclospora cayetanensis
Emma Patregnani1, Mauricio Durigan, Cassandra R. Suther, Shane Redfern, Alejandro Giraldo, maize
t4, Mauricio Durigan, Cassandra R. Suther, Shane Redfern, Alejandro Giraldo, maize
t4, Mauricio Durigan, Cassandra R. Suther, Shane Redfern, Alejandro Giraldo, maize

Introduction: Outbreaks of cyclosporiasis are associated with consumption of fresh produce contaminated with oocysts of Cyclospora cayetanensis. These oocysts are acquired in the food chain and may eventually contaminate water used for irrigation.

Purpose: We conducted experiments to verify the contamination of 1 L of irrigation water with C. cayetanensis by artificial “hands” exposed to surfaces contaminated with approximately 0.1 g of positive stool sample.

Methods: A stool sample with a high soil-to-colostrum mass ratio was used for the experiments. Aliquots of 0.1 g of this stool were spiked directly into triplicates of 10 L of irrigation water. In addition, triplicates of 0.1 g of the same stool diluted in 100 µL of PBS were used to contaminate disposable cups and agricultural plastic gloves. A total of 200 g of each of these samples was placed on a plate for 5 min, rinsed in tap water for 10 sec, and 20 seconds. The “hands” were then submerged in 10 L of irrigation water and stirred for one minute. Samples were processed by cold-dead-ultrazide (CDU) and C. cayetanensis was detected by qPCR. Cycle threshold (Ct) ranges of 22 to 30 were considered strong positives, while Ct higher than 30 were considered moderate to weak positives.

Results: C. cayetanensis was detected in the water samples spiked with 0.1 g of stool and the average Ct of 29.96 was obtained. The Ct values of non-contaminated water samples that were contaminated from surfaces that had approximately 0.1 g of stools were 32.65, 32.87, and 35.52 for contact times of 5, 10 and 20 sec, respectively. Non-contaminated water samples used as controls were negative by qPCR.

Significance: We demonstrated that transfer of C. cayetanensis to irrigation water is possible by hands exposed to contaminated surfaces with small amounts of positive stools despite short contact times.

P3-101 Isolation and Identification of Three Gram Negative Bacterial Species from Powdered Infant Formula Using MALDI-TOF Mass Spectrometry and RNA Sequence Analysis
Ihsan Sulaiman, Nancy Miranda and Steven Simpson
U.S. Food and Drug Administration, Atlanta, GA

Introduction: Pathogen detection is a critical factor for the safety of powdered infant formula (PIF) as it effectively facilitates the growth of human-pathogenic microorganisms of public health importance and can be easily contaminated. To date, various pathogenic Gram-negative and Gram-positive bacteria have been identified while examining the PIF contamination related to sporadic cases and outbreaks. This study was carried out with the major intention to find Cronobacter species, known PIF-borne opportunistic pathogens, that cause acute meningitis and necrotizing enterocolitis in neonates and are even more crucial in the food contamination worldwide.

Purpose: The major objective of this study was to rapidly identify the pathogenic bacterial isolates recovered from PIF samples by performing MALDI-TOF MS fingerprinting and RNA sequencing analysis.

Methods: A total of ten Gram-negative bacterial isolates were recovered from PIF samples and cultured on blood selective agars for DNA isolation and MALDI-TOF MS analysis. For each isolate, one to two colonies were directly spotted on the VITEK MS for species identification. Genomic DNA was extracted...
Three methods to achieve interesting flavors in dairy products were compared.

**Methods:**
- **P3-102**
  - Matrix Extension of a Loop-mediated Isothermal Amplification (LAMP) Assay for Screening Salmonella in Raw Pet Food
  - Introduction: Raw pet food, comprised of raw meat and vegetables, has become an increasingly popular option for pet owners. Ongoing surveillance programs and frequent product recalls have indicated that this commodity has a high risk of contamination with Salmonella and other foodborne pathogens. Improved screening methods are needed to meet the growing demands for testing in this field.
  - Results: This study highlights how interlaboratory data from PT studies can be used to gain an understanding of the variation associated with milk testing with the Salmonella LAMP assay. Samples were confirmed with conventional culture methods using the FDA's Bacteriological Analytical Manual (BAM) Salmonella method.
  - Significance: The LAMP assay offers a rapid and reliable approach for routine screening of Salmonella in raw pet food matrices. This method offers a valuable tool for improving the safety of raw pet food, a product of growing popularity with potential public health impacts due to the presence of Salmonella and other foodborne pathogens.

**P3-103**
- **P3-104**
  - Application of Improved Genetically Modified Detection Methods using Screening Multiplex PCR for Authorizing Genetically Modified Soybean Processed Food
  - Introduction: Genetic engineering of genetically modified (GMOs) in the world, a multiplex polymerase chain reaction (PCR)-based detection method is one of the high-throughput test detection methods for GM-derived food. The purpose of the study was to compare different PCR-based detection methods that conventional PCR and multiplex PCR and determine whether the multiplex PCR technique is applicable to various GM-derived from processed food.
  - Methods: The purpose of this study was to evaluate the performance of real-time PCR assays for the detection of inoculated Salmonella, Listeria, and Salmonella pseudomonas in dairy products including cheese, lactose powder, anhydrous milk fat (AMF) and whey protein concentrate 80 (WPC80) in comparison to the FDA BAM reference methods.
  - Results: For each sample type inoculated with Salmonella or Listeria at a low and high level. After stabilization, 375 test multiplex samples for Salmonella were enriched in 1.5 M (lactose, powder, AMF or WPC80) in pectinase buffer and incubated for 24 to 26 hours at 37°C. For the Listeria test method, 25-g samples of lactose powder and AMF and 5-g samples of WPC80 were enriched 1:1 in 24 LEB Complete broth and 375-g samples of cheese were enriched 1:15. All samples were incubated at 35°C for 24 to 48 hours. All enrichments were analyzed by LAMP and according to the reference culture method. Reference method samples were enriched according to the procedures in the FDA BAM.
  - Significance: This screening method is an accurate and rapid tool to detect authorized GM events in processed food. Especially, this study is an effective method for determining if bacterial cells collected in HMI are positive or negative.
  - Discussion: The purpose of this study was to confirm the performance of one-class soft independent modeling of class analogy (SIMCA) algorithm was developed from 35 HMI of 13 frequently isolated Salmonella serotypes using Salmonella Enteritidis and Salmonella Typhimurium. The SIMCA model was used to classify Salmonella HMI and 36 non-Salmonella HMI.
  - Results: Using the image and IIF platform results in approximately a 30-minute process from sample microscope slide preparation to cellular classification results. SIMCA reported that 93.4% of 5,505 wells were accurately classified, while Salmonella sensitivity was 0.96, and the combined specificity of the 14 non-Salmonella strains was 0.92. E. coli accounted for 39% of the non-Salmonella HMI tests using the SIMCA algorithm with 98.8% E. coli accurately classified as non-Salmonella.
  - Significance: This study demonstrates how PT studies can be used to compare long-term method performance characteristics. Results highlight that some differences in performance characteristics, such as PAC and SPC, can display very similar performance due to well-defined operating procedures provided by regulatory programs.

**P3-107**
- **P3-106**
  - Detection of Salmonella and Listeria from Multiple Dairy Products Using the BAX System Real-time PCR Assay
  - Purpose: This study highlights how interlaboratory data from PT studies can be used to gain an understanding of the variation associated with milk testing with the Salmonella LAMP assay. Samples were confirmed with conventional culture methods using the FDA's Bacteriological Analytical Manual (BAM) Salmonella method.
  - Methods: The purpose of this study was to compare the performance of total aerobic count methods in milk samples based on FDA proficiency tests (PT) spanning from 2009 to 2019. The reproducibility and interlaboratory variation associated with milk testing with the Salmonella LAMP assay were found negative for the presence of Salmonella and other foodborne pathogens.
  - Results: Mean bias between PAC and SPC across all samples was 0.038 log and mean sR2 was 0.052 log and 0.062 log for PAC and SPC, respectively. This suggests the bias between methods is minor considering the population of samples analyzed. In heavy and light cream samples, sR values were the greatest; 0.072 and 0.062 log for SPC and 0.058 and 0.061 log for PAC. These results indicate fat content of milk samples may impact reproducibility.

**P3-108**
- **P3-109**
  - Comparison of Applications of Different PCR Assays for Salmonella spp. in Dairy Products
  - Introduction: The United States milk sanitation program is run by the Food and Drug Administration (FDA) and operates under standards outlined in the Grade A Milk and Milk Products (GAMP) Program. According to the GAMP, Grade A milk samples must be tested using validated and approved total aerobic bacterial count methods. Two of the most commonly used methods are standard plate count (SPC) and Petrifilm aerobic count (PAC).
  - Methods: This study compared the performance of total aerobic count methods in milk samples based on FDA proficiency tests (PT) spanning from 2009 to 2019.
  - Results: Fourteen milk samples inoculated with various gram-positive and gram-negative bacteria were sent annually for the milk PT. Samples were tested by laboratories using approved total aerobic count methods. Statistical analysis of the resulting 3,846 sets of data was performed to compare performance of each sample against each other. The sample-to-spread (sR) was calculated for each method. Both methods were also evaluated per sample. Principal component analysis was applied to identify sources of analyte-specific systematic differences between different types of milk samples.
  - Results: Mean bias between PAC and SPC across all samples was 0.038 log and mean sR was 0.052 log and 0.062 log for PAC and SPC, respectively. This suggests the bias between methods is minor considering the population of samples analyzed. In heavy and light cream samples, sR values were the greatest; 0.072 and 0.062 log for SPC and 0.058 and 0.061 log for PAC. These results indicate fat content of milk samples may impact reproducibility.

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Introduction: Listeria monocytogenes has been detected in mushrooms, and it can cause listeriosis when consumed raw as a salad. Because the pathogen is psychrophilic, it can survive at low temperatures, even though contamination is at low levels. Therefore, rapid and accurate detection for L. monocytogenes in mushrooms is necessary.

Purpose: The objective of this study was to develop a rapid and accurate detection method of L. monocytogenes in golden needle mushrooms, using quantitative real-time PCR (qRT-PCR) with isap2 primers.

Methods: The isap2 primers for detecting L. monocytogenes were developed in broth media in a previous study. A mixture of L. monocytogenes strains (100 CFU/mL) and 30 °C buffered peptone water (BPW) or 30 °C buffered LB was inoculated in Pleurotus eryngii mushroom and allowed to grow for 9 h. RNA was isolated from the mushroom and used as the template for qRT-PCR using the isap2 primers for melting curve analysis and calculation of cycle threshold (Ct) values.

Results: In melting curve analysis, the annealing temperatures in all samples were in concordance with the positive control (L. monocytogenes culture), indicating that application of isap2 primers to the mushroom is appropriate. In qRT-PCR analysis, the Ct value was observed after three-h enrichment in all samples (four of four), while there were two positive samples at zero h (two of four). Also, this result was in concordance with those from a media-based detection method.

Significance: This result shows that the detection method of L. monocytogenes in golden needle mushroom, using LB and isap2 primers is more efficient than the conventional method.

P3-109 Droplet Digital PCR for Detection of Foodborne Pathogens

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Introduction: The currently validated molecular methods for the detection of enterohaemorrhagic Escherichia coli (EHEC) from foods relies on a PCR screening for the pathotype-specific genes, markers and STR in comparison to culture confirmation; this screening method suffers from a very high rate of false positives (up to 92.5%) that is partly due to the inability of current PCR-based methods to determine both sub and extraneous are within the same organism.

Purpose: This study was undertaken to reduce the false positive rate associated with current EHEC screening methods by confirming the presence of sub and extraneous EHEC from the same sample.

Methods: The ddPCR system used in this study works by partitioning intact cells into emulsion droplets, which subsequently undergo multiplexed endpoint PCR. This allows the differentiation of samples where a single organism contains both sub and extraneous from samples in which sub and extraneous reside in different organisms. A study which compared the response of ddPCR to commercial real-time PCR assays was conducted using over thirty (30) unique simulations of EHEC contamination in ground turkey.

Results: In this comparative study the ddPCR assay demonstrated excellent sensitivity to the specific EHEC strains tested. Furthermore, the ddPCR assay has the potential to reduce the number of false positive identifications in an EHEC screening assay.

Significance: This study demonstrates the ability of the ddPCR system to perform the confirmation of multiple genes within the same cell in a mixed microbial population; specifically, sub and extraneous. Ultimately, this system will result in cost savings by reducing the man-hours and testing expenses associated with the evaluation of false positive samples. Furthermore, this would enable more samples to be analyzed, which could reduce the probability of contaminated foods reaching consumers.

P3-110 Development of Sensitivity DNA Primers to Detect Listeria monocytogenes in Pleurotus eryngii Directly after Enrichment by Quantitative Real-time PCR

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Introduction: Foodborne outbreaks by Listeria monocytogenes continuously occur. However, conventional methods for identification of L. monocytogenes takes more than 2 h. Hence, it is necessary to develop rapid and accurate methods for detecting L. monocytogenes.

Purpose: The objective of this study was to detect L. monocytogenes in the king trumpet mushroom (Pleurotus eryngii), using enrichment broth and quantitative real-time PCR (qRT-PCR).

Methods: The isap2 primers were designed to target iap gene by OligoPerfect. To examine the sensitivity of isap2 primers, five log CFU/mL of L. monocytogenes isolate 131-1 from a mushroom was inoculated in king trumpet mushroom. RNA was extracted and was analyzed by qRT-PCR. The specificity of isap2 primers was evaluated from the DNA isolated from L. monocytogenes, Listeria spp., E. coli, Staphylococcus aureus, and Salmonella Typhimurium. A mixture of five L. monocytogenes isolates was inoculated in the king trumpet mushroom broth, followed by 10x-fold serial dilutions. Five milliliters of the enriched cultures were used for DNA extraction, and the DNA was used to perform the qRT-PCR with the isap2 primers for melting curve analysis and calculation of cycle threshold (Ct) values.

Results: In melting curve analysis, the annealing temperatures in all samples were in concordance with the positive control (L. monocytogenes culture), indicated that strains of L. monocytogenes can be detected with this system. Ultimately, this system will result in cost savings by reducing the man-hours and testing expenses associated with the identification of L. monocytogenes in king trumpet mushrooms.

Significance: This result shows that L. monocytogenes can be detected at sensitivity level in king trumpet mushroom. This method can be used for rapid and sensitive detection for L. monocytogenes at low levels in king trumpet mushroom.
Methods: In each of two independent experiments, 21 to 325 g samples of ground turkey were inoculated with a five-strain Salmonella cocktail at seven one-log increments, each series was inoculated with 20 to 250 ml of pre-warmed BAX media (0.1% M9, 1% yeast extract, 1% slop broth) and inoculated with STEC at a final concentration of approximately 10

Results: The six inoculation provided the best fit linear curve in both studies (R²=0.97 and 0.91; SE=0.14 and 0.15) compared to the eight h curves (R²=0.91 and 0.87; SE=0.60 and 0.86). In the first study, six h inoculation data was truncated at less than 3 CFU/ml sample; the resulting best fit linear curve was more accurate (R²=0.9913) in estimating log CFU/ml at a low level (0.17 to 2.83 log CFU/ml).

Significance: Utilization of CT values for estimating Log CFU/ml of Salmonella prior to enrichment in ground turkey provides an accurate estimation with a wide enumerable range at low concentrations and within-shift time-to-result.

P3-115 Comparison between BAX Cycle Threshold Values and Most Probable Number to Estimate Preenrichment Log CFU/ml of Salmonella in Pre-scald and Re-Hang Chicken Rinsates at a Commercial Processing Facility

Introduction: The poultry industry currently relies on the Most Probable Number (MPN) to estimate the load of Salmonella contamination throughout the processing chain. MPN is labor intensive and requires 27 to 72 h total time to results with large variation in estimation.

Methods: Pre-scald and re-hang chicken rinsates (30 ml) were combined with 30 ml of 42°C prewarmed BAX MP media, and inoculated with Salmonella Typhimurium from zero to six log CFU/ml. Samples were incubated at 42°C and analyzed with real-time PCR at four, six, eight, and 10 h of incubation. CT values at each time point and inoculation levels were analyzed to create a linear fit equation to estimate log CFU/ml of Salmonella in preenrichment rinsates. CT was conducted using the three-by-five tube method for all samples, incubated for 24 h at 42°C, screened with real-time PCR, and estimated Salmonella Log CFU/ml using the generated linear fit equation to determine the most appropriate enrichment time for both rinsates.

Results: The four h inoculation was the best fit linear curve for both pre-scald (R²=0.99; SE=0.10) and re-hang (R²=0.94; SE=0.26) rinsates. There was no difference between linear curves generated from CT values or MPN at any incubation. CT produced a wider estimated enumerable range of Salmonella for both pre-scald and re-hang rinsates, compared to MPN estimations.

Significance: Utilization of CT values for estimating log CFU/ml of Salmonella, prior to enrichment in pre-scald and re-hang rinsates provides a wider enumerable range, decreased variation, reduced time to results, and less labor sample compared to current industry standard MPN methodology.

P3-116 Evaluation of Chlorine Dioxide Gas Treatments against Salmonella spp. Artificially Contaminated on Mung Bean Seeds

Bassam A. Assoua, David Bucklej


Introduction: Salmonella outbreaks continue to plague sprout growers. Recent foodborne illness outbreaks associated with seeds and sprouts indicate current methods for detecting Salmonella on harvested and processed materials have gaps and suggest a need for more effective methods.

Purpose: The aim of this study was to evaluate the efficacy of chlorine dioxide gas (CDG) for detecting Salmonella enterica serovars artificially inoculated on an understudied commodity, mung bean seed and materials.

Methods: Mung bean seeds (10 g at a time) were immersed (five min) in 400 ml containing either ca. six or nine log CFU/ml of a Salmonella enterica serovar (Salmonella Newport, Stanley, Muenchen, and Anatum), which yielded a final cell concentration of 4.68×10⁷ and 2.99×10⁴ CFU/g seeds. Samples (400 g each) were treated with six or five mg CDG, air-dried for eight or 10 h at 22°C and 90% humidity, rinsed with 100 ml sterile distilled water, and two independent trials of each experiment were performed using four separate sets. Viability assessment was performed using 50 ml aliquots. Surviving cells were prone to be recovered by blending with 1% peptone and enumerated on xylose-lysine-tert-bromothymol blue (XLT-4) agar. Statistical analysis was completed using JMP Pro 14.0.

Results: CDG applied to seeds under the metal tray and tumbler condition yielded the highest reductions (2.13 log CFU/g). However, follow up treatments with a higher CDG concentration (six mg CDG for 10 h) yielded highly variable results in the tumbler treatment while the metal tray treatment exhibited up to a 4.68-log CFU/g reduction when compared with the same CDG treatment on sprout viability and treatment time. Further studies are needed to determine the CDG optimal concentration and treatment time to maximize treatment efficiency.

Significance: These results suggest CDG is efficacious against Salmonella spp, artificially contaminated on seeds and sprouts. Furthermore, under these conditions, CDG did not affect the viability of mung beans sprouts. However, treatment conditions appear to be an important factor associated with sprout viability. Ultimately, CDG shows promise as an effective control against sprout contamination and should be further investigated.

P3-117 Efficacy of Propidium monoazide Combined Real-time PCR to Detect Seven Viable Species of Foodborne Pathogens

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Introduction: The development of rapid detection kits when the difference between minimum and maximum incubation times is ≤4 h.

Methods: This assay was evaluated in an unpaired independent validation study compared to existing reference methods according to the AOAC validation guidelines. The matrix comprised twenty (20) replicates inoculated at the lower limit of detection (0.2-2.0 CFU) and five (5) replicates at a higher level (10 CFU). Polyskope 1.0 evaluated 12 (CFU) and 2 (CFU) for the same CDG treated and untreated death. Combined propidium monoazide (PMA) with qPCR can be an alternative for enumerating viable bacterial cells in samples.

Purpose: The objective of this study was to challenge the ability of two commercially available real-time Salmonella PCR kits to detect a diverse strain collection of Salmonella spp. in pure culture and in a dark chocolate matrix using the minimum and maximum incubation times specified.

Results: Both kits detected all 70 Salmonella strains at one log below the LOD. The kits also detected Salmonella Poona (10 of 20 confirmed-positive chocolates), using both the minimum (20 h) and maximum (24 h) incubation times (20 and 24 h, respectively) and maximum enrichment times (24 h). For confirmation, all Salmonella strains were tested using qPCR for Salmonella positive samples (19 of 20 confirmed-positive samples), kit A had 1 false negative using the minimum enrichment time, whereas kit B was able to detect all positive samples using both enrichment times. There were no significant differences in either kit’s ability to detect Salmonella at both the minimum and maximum primary incubation time evaluated.

Significance: Our data suggests that it is sufficient to validate only the minimum incubation time for Salmonella rapid detection kits when the difference between minimum and maximum incubation times is ≤4 h.

P3-119 Independent Validation for the Polyskope 1.0 Multiplex Pathogen Detection Assay for the Detection of Salmonella spp. in Dark Chocolate Using Multiple Incubation Times

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Purpose: The purpose of this study was to develop and optimize a rapid, PCR-based detection method combined with DNA photo-labeling to differentiate live and dead STEC.

Methods: Live and dead STEC cells, of various cell mixtures/concentrations and photo-labeling conditions, were treated with or without DNA photo-labeling dye etidium monoazide (EMA) and then exposed to LED light. Illumination of PCR amplification of dead DNA was confirmed using end-point PCR.

Results: The Polyskope 1.0 multiplex pathogen detection assay was used to detect Salmonella enterica serovar Typhimurium (STEC) and Salmonella enterica serovar Typhi in dark chocolate using both minimum and maximum incubation times.

Significance: Our data suggests that DNA photo-labeling combined with PCR-based detection methods can potentially differentiate live and dead STEC.

P3-120 Rapid Differentiation of Live and Dead Shiga Toxin-producing E. coli Using DNA Photo-labeling Combined with PCR

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Purpose: The purpose of this study was to develop and optimize a rapid, PCR-based detection method combined with DNA photo-labeling to differentiate live and dead STEC.

Methods: Live and dead STEC cells, of various cell mixtures/concentrations and photo-labeling conditions, were treated with or without DNA photo-labeling dye etidium monoazide (EMA) and then exposed to LED light. Illumination of PCR amplification of dead DNA was confirmed using end-point PCR.

Results: PMA treated live cells showed amplified signals similar to untreated cells, demonstrating that PMA treatment did not affect live cells. Amplification of DNA from PMA treated dead cells was almost inhibited, in contrast to untreated live cells.

Significance: This result suggests that PMA combined with qPCR can be used for rapid and efficient routine detection of bacteria causing foodborne diseases.
This data demonstrates the MPN-real time PCR method is sensitive, accurate, and fit-for-purpose of the enumeration of V. vulnificus.

**Significance:** Up-to-date and highly validated methods for the detection of V. vulnificus are easier to use for more accurate enumeration which can support appropriate risk modeling efforts.

P3-122 **Cronobacter sakazakii ISO 22694:2017 Testing of Milk Powders Using Commercially Available PCR**

**Purpose:** The aim of this study was to use the ISO 22694:2017 method, along with PCR detection, to determine the sensitivity and interference from dead cells.

**Methods:** The IQ-Check Cronobacter spp. kit and the BIOTEC Diagnostic Cronobacter Detection kit were used. The Biotec kit was used with manual and automated DNA extraction methods. Strains ATCC 29040 and 29544 were tested.

**Results:** The numbers of C. sakazakii in the resulting enriched samples (from the ISO method) were eight log CFU/ml. These were serially diluted to four, five, and six log CFU/ml, and were tested using both PCR kits and manual DNA extraction. Both kits resulted in positive detection at our five, six, and six log CFU/ml.

**Significance:** PCR methods are superior for detection of C. sakazakii in milk powders and there will be no interference from dead cells if the numbers are less than four log CFU/ml.

P3-123 **Recovery of E. coli O157:H7 by the BAX System in Beef Trim Using Surface Sampling Swabs**

**Purpose:** The objective of this study was to demonstrate the utility of a targeted amplicon sequencing method to i) identify microbial content at the species level, ii) quantify interspecies diversity down to sub-species level and iii) target low-level contamination in food samples from different environments.

**Methods:** Primer and probe final concentrations ranged from 100 mM to 500 mM. Reaction efficiency was calculated from the slope of the best fit line of the standard curve at each primer/probe concentration tested using the formula E=10-(slope). Efficiency is reported as a percentage and an ideal efficiency is considered to be >90%. Reactions were performed in triplicate.

**Results:** Initial primer/probe concentrations of 200 mM, in efficiencies of 89.88, 79.14, and 89.67 for Salmonella, E. coli, and Listeria, respectively, in single genomic reactions and 92.26, 81.53, and 103.18% for Salmonella, E. coli, and Listeria, respectively, when multiple target genomic DNA was combined. When probe concentration was varied, 150 mM was observed to have the best efficiency (88.84). EHEC efficiencies were 79.96 and 80.11% when EHEC primer concentrations were 150 mM and 200 mM, respectively.

**Significance:** This study defines the optimal primer and probe concentrations for a multiplex qPCR assay for the simultaneous detection of Salmonella spp., E. coli, and Listeria in foods and swabs allowing for a high-throughput, streamlined analytical approach that will increase the capacity of public health laboratories to rapidly detect multiple pathogens.

P3-125 **Optimizing a Multiplex qPCR Detection Assay for Salmonella spp., E. coli O157:H7 and Listeria monocytogenes on AB7500**

**Purpose:** The objective of this study was to demonstrate the utility of a targeted amplicon sequencing method to i) identify microbial content at the species level, ii) quantify interspecies diversity down to sub-species level and iii) target low-level contamination in food samples from different environments.

**Methods:** Primer3 software was used to design primers from alignments of multiple sequences of 10 core genes for each of 266 species that included 135 Salmonella spp., 30 EHEC, 30 Listeria monocytogenes, 615 (300) C. cayetanensis (150, 30) and DNA from clinical samples positive for C. cayetanensis were used. PCR amplifications were sequenced using Illumina MiSeq platform. Our in-house bioinformat- ic pipeline was used for identification and quantification of the targeted organisms from the sequence reads datasets. Briefly, sequenced reads were matching against the reference genome sequences using BLASTN.

**Results:** For the ZymoBiotics standards, the abundance of the reads corresponded to the relative amount of each pathogenic species present in the sample. C. cayetanensis samples, data from 1786 and 50 pairs were in 100% agreement. The larger panel additionally provided bacterial community stratification.

**Significance:** Use of a targeted approach for detecting low amount of pathogens provides another efficient and effective tool for the FDA to identify foodborne pathogens in a fast and efficient way.

P3-126 **Performance Validation of the BAX System Free DNA Cleanup Kit to Eliminate PCR False Positives Caused by External Contaminant DNA**

**Purpose:** To validate the inclusivity, exclusivity and sensitivity performance of the new BAX System Real-Time PCR assay for the detection of E. coli O157:H7 in foods and swabs allowing for a high-throughput, streamlined analytical approach that will increase the capacity of public health laboratories to rapidly detect multiple pathogens.

**Methods:** 1) To obtain PCR false positives due to external contaminant DNA in the matrix, 120 samples or swabs were serially diluted in BHI broth. Cell lysates of the serial dilutions were tested on the PCR assay. To evaluate the performance of the assay in a food matrix, 16 environmental swabs from surfaces treated with a ZymoBiotics standard were used. The abundance of the reads corresponded to the relative amount of each pathogenic species present in the sample.

**Results:** The ZymoBiotics standards, the abundance of the reads corresponded to the relative amount of each pathogenic species present in the sample. C. cayetanensis samples, data from 1786 and 50 pairs were in 100% agreement. The larger panel additionally provided bacterial community stratification.

**Significance:** Use of a targeted approach for detecting low amount of pathogens provides another efficient and effective tool for the FDA to identify foodborne pathogens in a fast and efficient way.

P3-127 **Development of a New BAX® System Real-time E. coli O157:H7 Single Target PCR Assay**

**Purpose:** To validate the inclusivity, exclusivity and sensitivity performance of the new BAX System Real Time E. coli O157:H7 Single Target PCR Assay.

**Methods:** A pooled sample set of 324 samples, including 48 clinical samples and 276 environmental samples, was used as input. E. coli O157:H7 strains (n=300 non-O157:H7, n=185 non-e.coli) were prepared at 10^4 CFU/ml for each strain. O157:H7 strains were serially diluted in 10-fold dilutions in broth. Cell lysates of the serial dilutions were tested on the PCR assay. To verify the performance of the assay in a food matrix, ground beef was serially diluted for 18 hours and aliquots were inoculated with serial dilutions of an overnight culture before testing by PCR.
This study was performed to identify which serovars of Salmonella O157:H7 present in lettuce and cabbage, to enable rapid detection of O157:H7 in Lettuce and Cabbage by Reducing Homogenization Buffer and DNA Elution Volumes

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Introduction: Foodborne bacteria are typically present at very low concentrations in food. Micronidal enrichment is therefore essential to increase microbial cell numbers above the detection limit, a process that is the main hindrance for rapid methods since it generally takes more than 10 hours. This study was conducted to develop and optimize a new method for bacterial growth in homogenization buffer and DNA elution volumes. The homogenization buffer volume was adjusted to 225, 50, 25, and 12.5 ml to isolate E. coli O157:H7 from 25 g of lettuce or cabbage. For effective homogenization, filtered air was added until the bags were fully inflated. The bags were sealed, then E. coli O157:H7 was obtained from lettuce and cabbage by vigorous hand shaking for 15 s. The extracted DNA was eluted in 200 or seven µl volumes, and cycle threshold values were compared using real-time PCR.

Results: As a result of the modified method, the microbial detection limit was improved 100-fold, so 10^3 CFU (three or nine CFU) of E. coli O157:H7 could be detected in lettuce and cabbage when 12.5 ml of homogenization buffer and seven or one µl of DNA elution volume was used, without the need for enrichment culture. The total time required for detection, including homogenization, DNA extraction, and real-time PCR, was less than two hours.

Significance: It is suggested that this method could contribute to the prevention of food poisoning incidents in institutional catering settings, such as schools or military facilities, by the rapid detection of foodborne pathogens prior to food consumption or during the food preparation period.

Poster

P3-128 Development of a PCR-Typing Method for the Identification of Salmonella Serotypes

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Introduction: Genus Salmonella is a major foodborne pathogen worldwide and consist of ≥250 serotypes classified by a serological method which has traditionally been the standard method for classification. However, a serological classification system is no longer sufficient to prevent and mitigate salmonellosis outbreaks, however, the serological method is time-consuming, labor-intensive and expensive, and more efficient DNA-based methods for identification of Salmonella are needed for public health.

Purpose: To develop and evaluate a set of PCR-typing methods for the identification of Salmonella serotypes, and as a substitute for inefficient serological methods.

Methods: Primer sets were designed from 64 candidate genes, expected to be unique in the Salmonella genus, of Salmonella Typhimurium, Typhi, and Enteritis. A total of 34 genes were screened and targeted including Salmonella genus-specific and subspecies i-specific genes for the development of a DNA-based PCR-typing method. Patterns of PCR results of 139 Salmonella strains (83 serotypes) were encoded and phylogenetic analysis was performed by BioNumerics program to differentiate between serotypes of Salmonella genus and subspecies.

Results: Screened target genes revealed various PCR patterns between Salmonella serotypes enabling each serotype of Salmonella to have uniquely encoded patterns from PCR results of 34 genes. The analyzed phylogenetic tree from PCR-typing results of 139 strains (83 serotypes) revealed the discriminatory power between the serotypes of Salmonella subspecies i.

Significance: This study will provide reliable DNA-based means for Salmonella serotyping and this method could be applied to the rapid monitoring of Salmonella in food industry and food safety clinically and epidemiologically.

Poster

P3-129 Development of Multiplex PCR for the Detection of Typhoidal Salmonella Serovars

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Introduction: Enteric fever, known as typhoidal fever and paratyphoid fever, is caused by typhoidal Salmonella enterica serovar Typhi, Paratyphi A, Paratyphi B, and Paratyphi C. Due to these typhoidal Salmonella serovars having different epidemiologic characteristics, clinical manifestations, and immune response in human hosts, they are classified apart from non-typhoidal serovars among ≥250 Salmonella serovars. Rapid and reliable diagnosis of these typhoidal serovars is needed for mitigating the spread of Salmonella during an outbreak.

Purpose: To develop a multiplex PCR method for rapid and reliable identification and separation of typhoidal Salmonella serovars from non-typhoidal Salmonella serovars.

Methods: Primer sets were designed from 35 candidate genes, expected to be unique within the genus Salmonella to Salmonella enterica serovar Typhi. A total of four target genes were selected which produce specific PCR results patterns against Salmonella Typhi, Paratyphi A, Paratyphi B and Paratyphi C. A size triplet multiplex PCR was designed including target genes that were Salmonella-genus specific, Salmonella subspecies i-specific, and typhoidal Salmonella serovar-specific, and was validated with the genomic DNA of these strains.

Results: The PCR results of four selected genes revealed specific patterns between the genomic DNA of Salmonella Typhi, Paratyphi A, Paratyphi B and Paratyphi C. The performance of the developed size triplet multiplex PCR for Salmonella typhoidal serovars was demonstrated by specifically amplified results, with Salmonella serovars discriminated from non-typhoidal Salmonella serovars. This result suggested that the multiplex PCR has enough discriminative ability between typhoidal and non-typhoidal serotypes of Salmonella subspecies i.

Purpose: This study will provide reliable DNA-based means for the diagnosis of typhoidal Salmonella serovars and this method could be a useful tool for rapid diagnostics of typhoidal Salmonella for the public health of humans, clinically and epidemiologically.

Poster

P3-130 Optimization of the Filtering Concentration Method for Rapid Detection of Escherichia coli O157:H7 in Lettuce and Cabbage Using Real-time PCR

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Introduction: Escherichia coli O157:H7 is one of the most important foodborne pathogens worldwide due to its high incidence and presence of various foods. Although numerous microbiological and molecular methods have been developed for detection, rapid detection of low concentrations in foods remain challenging.

Purpose: In the present study, the filtering concentration method for microbes was optimized for rapid detection of E. coli O157:H7.

Methods: Performance of the filtering concentration method in terms of time requirements and recovery percentage were compared among seven filters. Tapping with 200 repetitions was selected as the most effective detection method. The potential of rapid detection of E. coli O157:H7 in lettuce and cabbage using the filtering concentration method was evaluated using real-time PCR.

Results: As an optimal filtration method, MCE filter and detaching method using 200 repetitive tapping were used. The method could detect E. coli O157:H7 at a concentration of 10^5 CFU/g within 2 h without enrichment culture.

Significance: Filtering concentration is an efficient concentration method for rapid detection of low levels of foodborne pathogens in lettuce and cabbage using real-time PCR.

Poster
P3-134 Limit of Detection of a ELISA Commercial Kit for the Detection of T-2 Toxin in Foods
Adelino Dos Santos1, Amie Minor1, Brenda Keavey2, Zachary Kuhl1 and Megan Young1
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Introduction: T-2 toxin belongs to the trichothecene group of mycotoxins and is produced by fungi of the genus Fusarium. T-2 toxin is often found in agricultural commodities. Due to its cytotoxic and immunosuppressive activity, T-2 toxin is a threat to human and animal health. Purpose: The objective of this study was to evaluate T-2 toxin detection assays in ground beef, hot dogs, infant formula, and cheddar chicken. Methods: Matrix limit of detection assays were conducted in dry cereals, ground beef, hot dogs, infant formula and cheddar chicken. Each matrix was fortified with T-2 toxin at levels of 0.05; 0.10, 0.20, and blank (no T-2 toxin added). Two different extraction methods were used. The first required fresh or defrosting frozen meats and ground meats, and the second used boiled and refrigerated meats and ground meats. Results: The lowest LOD was determined as 5.3 ppm. Infant formula was diluted with 35% methanol/water. A 100 µl aliquot of the eluate from each sample was analyzed with an automated plate reader at 450-nm wavelength. Results: The LOD for both extraction methods was 0.05% recovery in infant formula and dry cereal. Cheddar chicken and ground beef did yield some recovery, while hot dogs and luncheon meat did not. The second extraction yielded 93.75% recovery in infant formula. The dry cereal, hot dog, luncheon meat and ground beef had 78.75%, 89.11%, 83.43%, and 87.74% recoveries, respectively. The LOD was determined at 0.05% recovery. Significance: The data from this study indicates the second extraction procedure, and commercial detection kit, may offer a suitable screening method for the detection of T-2 toxin in foods. This study has been submitted for review as an official FERN screening method.

P3-138 Validation of an ELISA Detection Method Extension for Abrin in Foods
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Introduction: Abrin is an extremely potent biotoxin produced from the seed of a tropical plant,Abrus precatorius, commonly found in the southern United States and the Caribbean. The plant is commonly referred to as the rosary pea or jequirity pea. While all parts of the seed are toxic, the highest concentrations are found in the seeds. The toxin is stable, lasting for long periods in the environment.

Purpose: This study's objective was to incorporate the validated FERN Abrin method into the FERN Triage method. A parallel comparison was conducted between the current Abrin extraction, using GBS, and the Triage extraction, using a rinse with Bothel/Mold-1/Mawn (BMW). Methods: A matrix limit of detection and validation studies were conducted in hot dogs, ground beef, and infant formula. The limit of detection (LOD) was measured for hot dog and liquid infant formula samples fortified at levels of 500, 100, 25, and 5 ppm, and ground beef (using both extractions), fortified at 1,000, 500, 100, and 50 ppm. Significant differences were observed during the validation study. The LOD for the traditional method was 50 ppm and a LOD of 25 ppm was determined for the Triage method. Results: The multi-day validation study consisting of different extraction methods exhibited minor result variations. While the triage extraction method performed well, the Triage kit degradation was observed over the entire validation period. Significance: The data from this study suggests that the extraction procedure, combined with the ELISA detection assay, may offer a suitable triage screening method for the detection of Abrin in foods. This study has been submitted for review as an official FERN method.

P3-139 Detection of Ricin in Foods Utilizing a Handheld Detection Device
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Introduction: Ricin is a cytotoxic protein commonly found in the castor bean plant, Ricinus communis. Currently, there is not an antitoxin available for treatment of ricin toxicity. Due to the commonality of the plant, its ease of dissemination, and historical nefarious use, it is imperative to validate a reliable screening method for the detection of ricin in foods for biosecurity purposes.

Purpose: This study's objective was to provide a lateral flow detection (LFD) screening method, combined with a single optimized extraction for preliminary and confirmatory detection of ricin toxin in foods. Methods: Matrix limit of detection and validation studies were conducted in pork rinds, breaded cooked chicken, chili with meat, pork barbeque, RTE meat buckets, and raw sausage by six FERN laboratories. Samples were fortified at levels of 0.25, 0.5, and 5 ppm with ricin toxin and cold stressed overnight. Six brands of each matrix were analyzed at each fortification. PBSTM was added in a 1:5 preparation, followed by a period of inversion, rest, and centrifugation. A 150 µl aliquot of each supernatant was mixed with 45 µl of the liquid eluate additionally analyzed as previously determined. Results: The data from this study indicates the second extraction procedure, and commercial detection kit, may offer a suitable screening method for the detection of ricin in foods. This study has been submitted for review as an official FERN screening method and offers a single extraction for both the preliminary and confirmatory test.

P3-140 Simultaneous Quantification of Aflatoxin, Vomitoxin, and Fumonisin in Corn Using the Envirowiz Common Extraction Protocol for Flex Mycotoxin Immunoassays
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Introduction: The immune and gastrointestinal effects of mycotoxins on human and animal health have been well-characterized. Most mycotoxicology research has focused on the prevalence and health impacts of individual mycotoxins, but there is increasing evidence that mycotoxins frequently co-occur in commodities or feed ingredients. Purpose: To enable rapid, on-site testing of multiple mycotoxins simultaneously, the Envirowiz Common Extraction Protocol can be paired with aflatoxin in-vomitoxin, and DON and fumonisin quantitative immunoassay lateral flow strips (LFD) from a single sample preparation in under 10 minutes, representing at least a 40% reduction in cumulative test time compared with separate sample extraction methods. Methods: Ground corn samples with aflatoxin, DON and fumonisin at low (5.7 ppb aflatoxin/1.9 ppm DON/0.9 ppm fumonisin), medium (19 ppb aflatoxin/5.3 ppm DON/1.9 ppm fumonisin), and high levels (95 ppb aflatoxin/15 ppm DON/5.3 ppm fumonisin) were analyzed for aflatoxin, DON, and fumonisin by immunoassay LFDs according to the common extraction protocol. Sample extracts were diluted in the provided buffer at 1:1 for DON and aflatoxin, and 1:10 for fumonisin. Test strips were developed in the diluted samples for four min at 22°C. Quantitative results were generated on the QuixScan reader system.
Results: This rapid test demonstrated CVs of five to 14% across all sample ranges (n=12). Sample test means were within 10% of the known mycotoxin level, except for aflatoxin levels in the low-level sample (0.03 ppm) which yielded a mean deviation of 10%. Significance: These results show that this rapid test may be risk management in mycotoxin testing programs for grain and feed.

P3-141 Detection of Acrylamide in Foods Using Filtration-assisted Optical Detection
Zhuangsheng Lin and Lili He

Introduction: Acrylamide is a neurotoxic and carcinogen present in food products and water sources due to high-temperature processing and contamination from cyanogenic glycoside breakdown [1]. Acrylamide is quantified using GC and HPLC methods, however, the food industry is looking for alternative methods with high speed and low cost. Purpose: Herein, we report a facile optical detection method for acrylamide via a thiol- click reaction with a mercapton compound, 2,2-(ethylenedithio) disulfane (EDT). Methods: Briefly, acrylamide was extracted from food matrices using dichloromethane followed by enrichment by water extraction. The water extract was further purified by solid-phase extraction (SPE) and was subjected to reaction with EDT (75°C, 10 min). The resulting reaction mixture was frozen and the yellow precipitate was isolated. Significance: This method was also used to detect acrylamide in instant and coffee ground samples. The filtration-assisted method provided a simple, fast and inexpensive approach for optical detection of acrylamide using AgNPs, with potential to be integrated with camera-based device to be performed in low resource and on-site settings.

P3-142 Determination of Endocrine Disruptors and Two Metals in Foods
Kang-Wi Tsoi* and Chia-Yang Chen

Introduction: Perfluoroalkyl substances (PFAS), phthalate esters (PAE), and bisphenols are commonly used in commercial products and could interfere with endocrine systems. Gallium (Ga) and indium (In) are frequently used in photonic industries, and they may result in cell oxidative stress. Purpose: This study measured 10 PFAS, six PAE, three bisphenols, and two metals in six types of foods and evaluated human exposure through food intake. Methods: Pork, pork liver, pork kidney, fish, clam, and oysters were investigated (63 samples). One g of homogenized food sample was extracted with QuECHERS and DEP with EMI-lipid absorbent. After concentration, the extracts were analyzed using UPLC-MS/MS and isotopel dilution techniques. Results: The analysis of Ga and In, 0.5-g food samples were prepared with microwave digestion before injecting onto ICP-MS. Significance: These data indicated that the industry may replace bisphenol A with its substitutes. PFBA and PFPeA accounted for 30 to 50% of the total PFAS in foods, which revealed that the industry might shift the use from long-chain PFAS to short-chain PFAS.

P3-143 Determination of Perfluoroalkyl Substances in Food Packaging in Taiwan
Peng Tsoi* and Chia-Yang Chen

Introduction: Perfluoroalkyl substances (PFAS) possess oil-resistant and waterproof properties, and are widely used in industrial and consumer products. People are exposed to PFAS through food and water. Purpose: This study developed and validated a method for determining 20 PFAS in food packaging with an ultra-performance liquid chromatography/triple-quadrapole tandem mass spectrometry (UPLC-MS/MS) and the method was applied on measuring 32 samples of commercial oil-resistant food packaging in seven categories. Methods: Samples of 100 cm² were cut into pieces and were ultrasonicated in 20 ml methanol at 50°C for 45 min. After centrifugation, the supernatant was concentrated to one ml. Six types of PFAS were analyzed using UPLC-MS/MS using 15 stable isotope-labeled internal standards in 32 oil-resistant food packaging samples from local markets and retailers in Taiwan. Results: The linear range of the analyses was 5.0 to 1.000 ng/ml except for 4:2 FFTHF: Most limits of detection were between 0.07 and 1.13 ng/ml. The recoveries ranged from 70 to 171% on most analytes at three tested levels, and the precision (% RSD) was lower than 19% (n=5). Three of four microwave popcorn packaging contained four- to nine-carbon perfluoropolyalkyl acids at 8.3-1,960 ng/ml and FFTHF at 121-7,188 ng/ml. Perfluoropolyalkyl acids (7.0 to 48.6 ng/ml) and PFPE and PFBS (0.45 to 3.955 ng/ml) were also found at one of three oil-proof paper bags. PFBA, PFHxA, and PFBS were observed at one of three chicken boxes and one of four fry paper bags, ranging from 5.0 to 40.3 ng/ml; FFTHF were present in one of four fry paper bags (224 to 167 ng/ml). Significance: PFAS, including eight carbonylic acids, three sulfonic acids, two sulfonamides, three fluorotolueneomers, and four polyfluorophenyl phosphates, were found in all samples and demonstrated statistically significant parameters. The detection limits were down to sub-ng levels.

P3-144 Use of Surface-enhanced Raman Spectroscopy in Determination of Nano-sized Particles in Food Grade TiO
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Introduction: Titanium dioxide is an important white food colorant (E171) and is used in a variety of food products such as doughnuts, candies, frostings etc. Food-grade titanium dioxide, in the form of TiO₂, is regulated and approved around the world. It is recognized by the food industry as safe in food use. The particle size of the commercially produced E171 is not regulated and currently available methods for nanosized particle analysis are very time-consuming and require expensive equipment. Purpose: The purpose of this research is to develop and evaluate a rapid and simple method using surface enhanced raman spectroscopy (SERS) to determine the particle size of titanium dioxide nanoparticles. Methods: Titanium dioxide nanoparticles can be detected with a Raman microscope by the mapping technique. Results: The concentration of titanium dioxide nanoparticles is determined from the percentage of positive spots on the map. The particle size can be estimated from the ratio (R) between the Raman intensity of TiO₂ and the SERS intensity of gallium-bound to the nanoparticles. The ratio calculated from 100 nm particles can be used as a cutoff to confirm the presence of nanoparticles. Moreover, innovative SERS imaging is used to develop a visual interpretation of particulate matter size distribution maps [2]. Conclusions: SERS can be used as an effective tool to rapidly analyze the presence of nanoparticles in food grade TiO₂. This method can be applied as a preliminary rapid step to localize specific contaminants using confirmatory analysis.

P3-145 Particle Size Analysis for Detecting Crystalline Solids in Powder Infant Formula
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Introduction: Iodized NaCl served as an experimental crystalline solid contaminant in powder infant formula. Two contaminant solid size fractions were used to replace other ingredients to increase margins and weights, which may negatively affect the infant’s health. Purpose: To determine if a crystalline solid (iodized NaCl), added to infant formula powder as low as one percent on a wet basis, could be routinely detected using a laser diffraction-based particle analyzer. Methods: Two commercial-sized infant formula powder samples were analyzed. The particle size distributions were computed using a commercial particle size analyzer to determine if the two contaminant particle size fractions were present in the samples. Results: The method and instrumentation proved capable of detecting two different particle size fractions in all of the blends tested vs. the control commercial formula. The tested on D50 and D90 values analyzed from blends and the respective controls using a 95% confidence interval. Ranges in the measured particle size distributions showed the potential for use of the method and instrumentation for detecting the presence of crystalline solids in amorphous powders, such as infant formula. Results: This method could be applied as a preliminary rapid step to localize specific contaminants using confirmatory analysis.

P3-146 Effectiveness of Cleaning Strategies for Removing Milk Chocolate from Pilot-scale Chocolate Processing Equipment
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Introduction: Dark chocolate produced on equipment used to manufacture milk chocolate can contain milk due to cross-contact. Purpose: The goal of this study was to evaluate the effectiveness of cleaning methods for removing milk chocolate from shared processing equipment. Methods: Milk chocolate (40%, 62.00 ppm milk) was processed in a ball mill (<0.5 kg) and a horizontal-shaft conch (2.5 kg), followed by drawing the majority of the chocolate. Three cleaning methods were investigated: i) no cleaning; ii) cleaning with milk chocolate: (a) cleaning; (b) cocoa butter (40°C, five min); and iii) cleaning with milk chocolate: (a) cleaning; (b) cocoa butter (40°C, five min); and (c) cocoa butter (40°C, five min) followed by cleaning milk chocolate (a) cleaning; (b) cocoa butter (40°C, five min) followed by cleaning. Significance: The purpose of this research is to develop an innovative and rapid method using surface enhanced raman spectroscopy (SERS) to determine the presence, quantity and particle size of TiO₂ nanoparticles in E171. Results: Titanium dioxide nanoparticles can be detected with a Raman microscope by the mapping technique. The concentration of TiO₂ nanoparticles is determined from the percentage of positive spots on the map. The particle size can be estimated from the ratio (R) between the Raman intensity of TiO₂ and the SERS intensity of gallium-bound to the nanoparticles. The ratio calculated from 100 nm particles can be used as a cutoff to confirm the presence of nanoparticles. Moreover, innovative SERS imaging is used to develop a visual interpretation of particulate matter size distribution maps [2]. Conclusions: SERS can be used as an effective tool to rapidly analyze the presence of nanoparticles in food grade TiO twenty. This method can be applied as a preliminary rapid step to localize specific contaminants using confirmatory analysis.

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Journal of Food Protection Supplement
Methods: Ten batches of tiger shrimps (100 g/batch) were fried in 4:1 soybean oil (176°C, three min) and samples of frying oil (150 mL) were collected. Four batches of tiger shrimps were cooked in shrimp-contaminated oil (176°C, three min). Oil and French fry samples were analyzed using two crustacean ELISA assays and a total protein (Pierce bicinchoninic acid–reducing agent compatible) test. To evaluate the effects of frying time on detection of crustacean protein, shrimp were analyzed at 0, 2.5, 5, 10, 15, 20, 100 and 500 ppm-crude protein concentrations. Antibodies from rabbit immunization were analyzed and compared with shrimp proteins. To evaluate the effects of frying temperature, shrimp were cooked at 190°C for 2.5 and 3 min. Results: The ELISA detected shrimp protein transferred to frying oil and a secondary food may be underestimated by ELISA due to changes in shrimp protein during frying.

Results: Maruha Nichiro ELISA and ELISA Systems tests detected up to 29.0 ppm (38% CV) crustacean protein and 0.91 ppm (53% CV) tropomyosin, respectively, in oil used to fry shrimp. An average of 144 ppm (33% CV) total protein was measured in oil after frying 10 batches of shrimp. Tropomyosin levels in former French fries cooked in the shrimp-contaminated oil were below the limit of detection (0.05 ppm). The ELISA Systems test detected crustacean protein in the French fries. A dramatic reduction in ELISA detection of shrimp protein was demonstrated as a function of frying time.

Discussion: To detect shrimp protein to oil frying, ELISA is an adequate method, but shrimp protein transferred to frying oil and a secondary food may be underestimated by ELISA due to changes in shrimp protein during frying.

Poster P3-154

Making Sulfur-free White Wine through the Use of o-Pinene

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Introduction: Because sulfur dioxide causes human health risks, including dermatitis, urticaria, angioedema, diarrhea, abdominal pain, which also widely used to avoid microbial contamination and act as well known antifoaming and antimold agents, it is not allowed in various foods and beverages that are free from additives, testing, and microbiologically safe.

Purpose: Based on this in mind, this study uses the o-pipecolone to alternative the sulfur dioxide in the white grape wine fermentation model, and the parameter of evaluation is to compare the productivity and the aroma, a fragrance monomeric compound derived from fruit aroma substances including grapes. Wine treated with o-pipecolone (0.3125%, 0.625%, 0.125% group) were compared with wines treated with sulfur dioxide.

Methods: The parameter (include specific gravity, μL, total volatile solids, ethanol, percent transmittance, titratable acid, lab, total penticot) of two groups and SO2 free wine (o-pipecolone treated group) was studied.

The transmittance of five groups fell between 70 and 90, the o-pipecolone group (0.3125% & 0.625%) is superior to the control group and the sulfur dioxide group. The percent transmittance and μL (0.85-0, 3.01-0.85, 0.301-0.303) value of the o-pipecolone group at 95% and p<0.05. Addition of a sulfur dioxide can effectively the browning of white wine storage at 180 days, but the controlled increase about 0.25 absorption wavelength. Other parameters showed the white grape wine no differences were found in all groups.

Significance: The conclusions showed the o-pipecolone have the excellent antibacterial, antioxidant ability and also extending the broaching recovery of white grape wine, demonstrated a good alternative material for winemaking.

Poster P3-155

Factors That Impact Survival of Salmonella during Storage of Beans and Batch Production of Cold Brew Coffee

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Purpose: The objectives of our study were to i) develop a quantitative monoclonal antibody (MAb)-based ELISA for the detection of sesame and ii) validate the assay for sensitivity, recovery and cross-reactivity (CR) against a large panel of seeds, cereals, tree nuts, legumes, and spices.

Methods: Polyclonal antibodies against Ara h 2 and Ara h 3 from light- roasted peanut were raised in rabbits. The immunoglobulin G (IgG) antibodies were affinity purified using protein G columns. A total of 252 different raw or processed peanut samples were confirmed by Western blot. A sandwich ELISA was developed using equal amounts of anti-Ara h 2 and anti-Ara h 3 IgG as capture antibodies and using their HRP conjugates as detection antibodies. Proteins extracted from light-roasted peanut flour were used to generate standard curves and samples were extracted in carbonate buffer for peanut quantitation. Cross-reactivity to plant- and animal-based foods was assessed. Peanut flour (0, 0.25, 5, 10, 25, 100, and 500 ppm) incriminated cookies baked at 190°C for 2.5 and 3 min were analyzed in triplicate to evaluate method performance.

Results: The Ara h 2 antibody exhibited similar immunoreactivity toward raw, light-roasted and dark-roasted peanut, whereas Ara h 3 antibody demonstrated reduced immunoreactivity toward dark-roasted peanut. The developed ELISA using both antibodies was sensitive, with a limit of quantitation of at least 0.78 ppm peanut protein in foods. Negligible or no cross-reactivity was observed with 40 different foods including select legumes. The ELISA detected peanut protein at incriminated levels ≥2.5 ppm in dough and ≥5 ppm in baked cookies. Defatting and ion/desorption improved peanut protein recovery from cookies matrices compared to raw peanuts observed at low peanut concentration.

Significance: Improved peanut quantitation methods can reduce false-negative results protecting peanut allergic consumers from inadvertent exposure to peanut in processed foods.

Poster P3-156

Development and Validation of a Quantitative Monoclonal Antibody-based ELISA for the Detection of Tropomyosin in Seafood

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Introduction: Tropomyosin is a highly conserved protein found in various crustacean species. This protein can be used as a biomarker for the detection of seafood contamination in processed foods. The ELISA was developed with a sandwich format using two monoclonal antibodies (MAb) raised against the fish tropomyosin from three different species of forage, oily, and white fish without demonstrating cross-reactivity to other seafood.

Purpose: To develop a validated method for the quantitative detection of fish tropomyosin in seafood products.

Methods: Two monoclonal antibodies (MAb) to known fish tropomyosin were raised in mice. The antibodies were affinity-purified from ascitic fluid using different antigens. The specificity of each antibody was determined by Western blot and ELISA.

Results: The ELISA was highly sensitive with a detection limit of 0.75 ppm (n=6). Recovery ranges from 92.0-95.0% for 0.75 ppm and 95.0-97.5% for 1.5 ppm. The MAb-based ELISA was sensitive, with a limit of quantitation of at least 0.78 ppm peanut protein in foods. Negligible or no cross-reactivity was observed with 40 different foods including select legumes. The ELISA detected peanut protein at incriminated levels ≥2.5 ppm in dough and ≥5 ppm in baked cookies. Defatting and ion/desorption improved peanut protein recovery from cookies matrices compared to raw peanuts observed at low peanut concentration.

Significance: Improved peanut quantitation methods can reduce false-negative results protecting peanut allergic consumers from inadvertent exposure to peanut in processed foods.
P3-155 Microbial Evaluation of ‘Adoyo’ Drink Sold in Ogun State, Nigeria

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Purpose: Adoyo is an herbal drink produced by boiling pieces of African yam, and lemon grass in a pot and adding pieces of grape, pineapple, and citrus fruits. The drink is dispensed and served at room temperature. It can also serve as a refreshing drink when served cold.

Introduction: Due to the prevalence of foodborne illness, it is essential to assess the quality and evaluate the microbial content of adoyo drink that is sold in the market.

Methods: Samples were collected from three major car parks in Abeokuta while the control sample was produced in the laboratory. The samples were analyzed for chemical, microbial and heavy metal properties using standard laboratory techniques.

Results: Sample pH ranged from 4.39 to 4.58 as a result of the omidun, which is highly acidic due to fermentation. Vitamin C content of the samples ranged from 3.37 to 3.87 with the control already having acidic pH. There was no indication of food or mercury but a small amount of cadmium was detected in a safe quantity of 0.06. Zinc, which is likely already existent in fruit was detected below the minimum permissible limits of 5 mg/l. Staphylococcus, coliforms and fungi ranged from 1.5×10⁶ to 10⁷, 1×10⁶ to 10⁷ and 2.5×10⁵ to 10¹⁷ respectively. The assessment of the total coliform count in adoyo showed five genera of bacteria in the coliform group that were characterized as Escherichia coli, Aerobacter spp., Enterobacter spp., Salmonella spp., and Proteus spp. Fungal count ranged from 2.5×10⁶ to 10¹⁵ with the control having the lowest count of 2.5×10¹⁵ CFU/mL.

Significance: This data suggest that the presence of Staphylococcus aureus and Trichoderma probably indicates contamination from the raw materials, environment or personal processing or vending the adoyo.

P3-157 Does the Indigenous Microbial Community of Kombucha Prevent Survival and Growth of Pathogens? 

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Developing Scientist Egrant

Introduction: Kombucha is a fermented, acidic beverage that is frequently prepared using home brewing methods. This raises concerns about the food safety risks involved.

Purpose: The objective of this study was to evaluate the microbial diversity of various kombucha cultures as well as the potential for growth of foodborne pathogens during brewing.

Methods: Four different commercially available kombucha starter kits were brewed according to suppliers’ instructions and then examined over a 14-d period. Aerobic metabolites, yeasts, lactic acid bacteria, and acetic acid bacteria were enumerated on selective media. Survival of Salmonella enterica, Escherichia coli, and Cryptococcus parapsilosis were assessed. Culture techniques were used to enumerate and detect (by enrichment) bacteria and an HCT-8 cell line was used to enumerate C. parapsilosis. Starter kit sweetened tea blends (BT) were used as controls.

Results: Populations of aerobic metabolites, yeasts, lactic acid bacteria, and acetic acid bacteria increased more than log units during the first 72 h and then plateaued and gradually decreased after seven d. Salmonella and Escherichia coli grew and persisted in BT1 over 14 d but showed a significant difference in the diversity of pathogens isolated, with 100% Salmonella and 82% L. monocytogenes detected in kombucha 1BT compared to 1.5% Salmonella and 0% L. monocytogenes detected in kombucha 1BT. The most significant reductions in four to five logs in Salmonella or L. monocytogenes was observed between day one and seven d.

Significance: Results of this study provide information concerning the fluctuations in indigenous microbial ecology during brewing kombucha, as well as the capability of Salmonella, E. coli, and C. parapsilosis to survive and grow. This will help to better assess food safety risks during home brewing of kombucha.

P3-158 Withdrawn
The Relationship between E. coli Levels and Pathogen Detection in Surface Water Samples is Mediated by Environmental Conditions

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Introduction: While E. coli is used as an indicator of microbial quality, the relationship between E. coli levels and pathogen presence in surface water varies widely between studies. We hypothesized that environmental conditions mediate the relationship between E. coli levels and pathogen presence in water.

Methods: To investigate how environmental conditions affect the relationship between E. coli levels and pathogen contamination of surface water, 504 samples (30 samples = 504 sample) were collected over a 12-month period in 2017 from two Arizona irrigation ponds. At each sampling, >500 L of water were collected, and separate >500 L aliquots were tested for Salmonella, Listeria, and Shiga toxin-producing E. coli (STEC). Data on E. coli levels and environmental factors (weather and water quality) were also collected. The relationship between E. coli levels and pathogen detection was characterized using logistic regression. Random forest models were used to test the impact of individual factors on the relationship between E. coli levels and the likelihood of pathogen detection.

Results: Salmonella and Listeria were isolated in 41 (8.2%) and 25 (5.0%) samples, respectively. E. coli-positive samples were used in a PCR screen. Relational models showed that QMRA (a semi-quantitative risk assessment method) was considered a more accurate QMRA summary descriptor. Results showed that Salmonella was detected in 41 samples (11.8%, 41/340), Salmonella-positive samples were tested for STEC presence. A total of 120 (35.3%) samples were positive for STEC detection. An example of the STEC analysis was performed to analyze the genetic diversity of the isolates.

Significance: Our findings suggest that E. coli may not be a reliable indicator of pathogen contamination of surface water. Thus, alternative methods for assessing food safety risks associated with the use of specific water sources for production need to be developed and tested.

P3-163 Occurrence and Levels of Salmonella Species in Primary Irrigation Water Canals and Return Flows in Arizona and the Risk of Contamination of Lettuce Crops

Katie Brown, Monique Torres, Patricia Gundy, Huny Zergili, Brianna Leja, Candace Garrett and Charles Gerba

Journal of Food Protection Supplement

Introduction: Studies have detected Salmonella in irrigation waters in Arizona; however, few are quantitative and thus the risks of Salmonella contaminating crops is unclear.

Purpose: To determine the occurrence/concentrations of Salmonella in irrigation water and return flow (drainage) canals in Arizona.

Methods: Water (500 mL) was collected weekly from 6 sites spread over a 15 mile reach of the creek over the course of 18 months. Salmonella detection was conducted by selective enrichment in buffered peptone water (BPW) and in-cup MLST analysis performed using VITEK MS and subjected to whole genome sequencing. Core genome multi locus sequence type (cgMLST) and in-silico MLST analysis were performed to analyze the genetic diversity of Salmonella isolates in irrigation ponds in Maryland during 2017.

Results: Overall, 21% of water (n=120) samples were positive for L. monocytogenes. Genetic lineage spp. detected in water samples were 1. serogroup I, 2. serogroup I, 3. serogroup I, and 4. non-typeable. In the Conococheague Creek located in the United States Mid-Atlantic.

Significance: The risk of irrigation water use from irrigated Arizona lettuce crops appears to be quite low and could be reduced by ensuring adequate time between final irrigation and harvest.

P3-164 Incidence of Fecal Indicator and Pathogenic Bacteria in Reclaimed and Return Flow Waters in Arizona, United States

Libin Zhu, Nina Torres, Warner Betancourt, Manan Sharma, Shirley A. Mcallef, Charles Gerba, Amy Sapkota, Amir Sapkota, Salina Parveen, Fawzy Hashem, Eric May, Kali Knie, Miyah Pop and Sadhana Ravishankar

Journal of Food Protection Supplement

Introduction: Reclaimed water is being used in agricultural application. Therefore, in addition to compliance with FAS data quality standards, it is important to estimate the incidence and potential sources of L. monocytogenes in irrigation waters.

Methods: Using the pond-specific spatial grids, water samples (500 ml) were collected biweekly (June to July) from 30 and 20 sites of Pond 1 and Pond 2 respectively. L. monocytogenes detection was conducted by selective enrichment in buffered protease enrichment broth, following streaking onto RAPID-mol and QC agar plates. plates were incubated at 30°C for 24 hours. Positive plates were subjected to whole genome sequencing. Characterization of L. monocytogenes isolates was performed using molecular typing methods, including assigning lineage, genotypic diversity, and pathogenicity.

Results: Overall, 21% of water (n=120) samples were positive for L. monocytogenes. Genetic lineage spp. detected in water samples were 1. serogroup I, 2. serogroup I, 3. serogroup I, and 4. non-typeable. In the Conococheague Creek located in the United States Mid-Atlantic.

Significance: The genetic characterization of L. monocytogenes isolates provided novel insights into the sources of this pathogen in irrigation waters. The baseline information on the incidence of L. monocytogenes in surface waters is essential to understand how these irrigation sources may influence the microbiological safety of fresh produce.

P3-165 Listeria monocytogenes Levels and Population Diversity in Surface Waters in the United States Mid-Atlantic Region

Summer Macarion, Jin Qing, Dana Harriger, Rachael Piccard, Eric Wells, Yakov Pachepsky, Marc Alard, Eric Brown and Yi Chen

Introduction: Microbiological quality standards for agricultural water rely on bacterial fecal indicators. The role or human or animal input in Listeria monocytogenes concentrations in surface waters is unknown and thus, a better understanding of the major sources of this pathogen in agricultural water is needed.

Methods: Water (500 mL) was collected weekly from six sites spread over a 15 mile reach of the creek over the course of 18 months. L. monocytogenes populations were quantitated by MPN method, using a five-100 mL step, eight one-ten mL scheme with a lower limit of detection of 0.0021 MPN. L. monocytogenes isolates were confirmed using in-house methods and subjected to whole genome sequencing. Core genome multi locus sequence type (cgMLST) and in-silico MLST analysis were performed to analyze the genetic diversity of isolates.

Results: When E. coli levels were higher, the risk of L. monocytogenes contamination was greater. Salmonella spp. in water samples ranged from Ω12 to 120 µM/mL (10 CFT/g). cgMLST analysis revealed high genetic diversity among these isolates, representing genetic lineage (2.21 ± 0.24), and (11.36 ± 0.24), and molecular serogroups: Ia (11.36 ± 0.24), Ia (11.36 ± 0.24), Ib (12.79 ± 0.24), Ib (12.79 ± 0.24), Ia (11.36 ± 0.24), Ib (12.79 ± 0.24), Ia (11.36 ± 0.24), and Ia (11.36 ± 0.24). Six novel clones of L. monocytogenes were identified. The strains do not match recent USDA outbreaks, however, some strains belong to the clinical isolates, and are considered hypervirulent.

Significance: This study generated a highly resolved temporal and spatial genetic relationship map among L. monocytogenes strains coupled with actual population levels in surface waters that are critical for microbial source tracking, modeling and risk assessment. These novel data also are essential in reas-
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Methods: Water samples (n=489) were collected over two periods from two reclamation water (RW) and four surface water (SW) sites (n=8) on Maryland’s Eastern Shore. Therefore, ensuring the microbial safety of these water sources is paramount.

Purpose: The purpose of this study was to determine the prevalence of Salmonella and Listeria monocytogenes in non-traditional irrigation water sources.

Results: Of the 489 samples collected, 346 (70.8%) of 489 and 160 (32.7%) of 489 were positive for Salmonella and L. monocytogenes, respectively. Both Salmonella and L. monocytogenes strains were isolated from Tidal Irrigation Water. Salmonella spp. MPN values varied among sites over the course of the study, ranging from zero to 11 MPN/L. For Salmonella, average MPN/L values for the SW sites were significantly higher (P<0.05) than the RW sites (2.47 and 0.84, respectively). For L. monocytogenes, average MPN/L values were 0.01 and 0.60 for RW and SW, respectively.

Significance: This study shows that Salmonella and L. monocytogenes are present in non-traditional irrigation water sources on the eastern shore of Maryland. While these water sources have the potential to be used for produce irrigation, pre-existing contamination and mitigation strategies need to be considered to ensure the safety of these water sources.

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P3-174 Behavior of Silver Nanoparticles under Various Wash Water Conditions for Leafy Green Protecting Gayathri Gunathilaka, Jianzhuo He, Hui Li, Wei Zhang and Elliot Ryser
Middlebury College, Northfield, VT

**Developing Scientist Entrant**

**Introduction**: Engineered nanoparticles (ENPs), such as nanoparticles, are increasing in agro-systems, particularly from land application of sewage sludge. Exposing fresh produce to ENPs may increase residual levels posing potential food safety concerns. Understanding the fundamental interactions between relevant ENPs and commercial washing of fresh produce is important in designing effective mitigation strategies.

**Purpose**: This study aimed to evaluate the behavior of silver nanoparticles under different wash water conditions for leafy greens.

**Methods**: Aggregation and dissolution kinetics of silver nanoparticles (AgNPs) were systematically investigated in wash water with or without dissolved lettuce extract (DLE; 0.1% w/v). Wash water was prepared by adding two, 50, or 100 ppm free chlorine followed by five mg/l AgNPs, with five mg/l AgNPs in deionized water serving as the control(<pH 6.5). Average particle size (diameter, nm) and zeta potential (mV) of the AgNPs suspensions were measured using a zetameter, with dissolved Ag concentrations (mg/l) measured using an atomic absorption spectrophotometer initially and again after 10, 30, 60, 120, and 240 min.

**Results**: For all the chlorine levels tested, the dissolved Ag concentrations were much lower (0.01 to 0.03 mg/l) as compared to the control (0.54 to 0.8 mg/l). Increased particle size with time (40 to 431 nm) as compared to the control (52.6 to 60 nm) indicates high AgNPs aggregation in the presence of chlorine, with the zeta-potential values more negative (-39 to -45 mV) compared to the control (-10 to -20 mV) (p<0.05). AgNPs treatments with DLE yielded similar findings.

**Significance**: Since the fate of AgNPs in freshwater washing is affected by both chlorine and organic matter in wash water, these interactions are important in evaluating the sorption of AgNPs to leafy greens.

P3-175 Fabrication of a Metal Oxide Coated Pouch for Alternative Processing of Military Ration Components Shannon McGraw, Christopher Oldham1, Gregory Parsons2 and Danielle Frois-Blumsack U.S. Army CECOM Soldier Center, North, MA, “North Carolina State University, Raleigh, NC”

**Purpose**: The objective of this study was to develop new edible antimicrobial coating solutions to effectively reduce Salmonella and spoilage fungi in military ration components.

**Methods**: This study analyzed the risk of foodborne illness by Vibrio vulniﬁcus and Vibrio cholerae in reduced oxygen packaging technology in order to achieve the required barrier performance for extended shelf life.

**Results**: The largest reductions in oxygen and water vapor transmission rates were observed on film samples coated with a 25 nm layer of Al2O3 and a WVTR of 1.56±0.18 cc/m2day (97.91% reduction). The closed multilayer pouch with 12 alternating layers of two nm Al2O3 and titanium oxide coatings were deposited at thicknesses ranging from two to 25 nm using ALD. The coated materials were evaluated for oxygen and water vapor permeation using a MOCON OXTRAN 2/21 and Permatran-W 3/33.

**Significance**: These data suggest that ALD of nanoscale metal oxide coatings onto packaging polymers have the potential to provide barrier to engineered nanoparticles in food packaging used in the military’s Meal, Ready-to-Eat (MRE) in order to achieve the required barrier performance for extended shelf life.

P3-176 Antimicrobial Coatings for Improving Safety and Shelf-life of Cherry Tomatoes

Tommy Jin1 and Joshua Gurrieri2
U.S. Department of Agriculture – ARS, Eastern Regional Research Center, Wyndmoor, PA, U.S. Department of Agriculture-ARS, Eastern Research Center, Wyndmoor, PA

**Purpose**: The objective of this study was to develop new edible antimicrobial coating solutions to effectively reduce Salmonella and spoilage fungi in cherry tomatoes while maintaining their quality.

**Methods**: Two coating solutions were used: coating 1 was one percent chitosan; coating 2 was one percent chitosan and six percent alginic acid (pH 7). Both solutions contained two percent tween 80 and four percent lecithin and all ingredients were dissolved in two percent acetic acid solution (acetic acid (50% v/v) and 1.5 log CFU/multilayered coats after treatment and through the 14-day storage period. Both coating treatments significantly (p<0.05) increased whiteness and yellowness while not affecting redness. Texture firmness of tomatoes was not significantly (p>0.05) affected by any of the treatments. After 21 days’ storage at 10°C, two treated tomatoes still showed fresh-like appearance while non-treated tomatoes had moldy surfaces.

**Significance**: This study provides some options to develop edible antimicrobial coating solutions for produce with varied surfaces.

P3-177 Black Drum (Pogonias cromis) Shelf Life Comparing Four Packaging Methods

Joshua Cobar, Katheryn Parraga and Evelyn Watts
Louisiana State University, Baton Rouge, LA

**Purpose**: To determine the shelf life of Black Drum comparing three different packaging technologies.

**Methods**: Black drum fish fillets were purchased fresh from the dock within 24 h of being caught. Fillets were packaged using four methods: polyethylene bag, film pack (PP); Modified Atmosphere Packaging (MAP) (CN – 50% CO2 and 50% N2) and MAP (CO2 – 40%, CO2 – 30%, N2 – 30%). The packed fish was stored at 5±2°C for 20 d. Shelf life was studied in terms of TVB-N, TBARS, pH, Color, aerobic plate count (APC), Enterobacteriaceae, yeast, and mold. Microbiological and physical chemical evaluations were carried out at 0, 2, 7, 14, and 21 d.

**Results**: For a 20-day microbial shelf life based on APC was observed in black drum fillets stored in MAP, which was an increase of eight d compared to PB and VP (p<0.001). Even though, MAP appeared to extend the shelf life of black drum based on APC, there were no significant differences in TVB-N, TBARS, pH, and color analyses. There were also no significant differences between the two MAP gas combinations used during this study (P>0.05).

**Significance**: This study demonstrated that MAP is effective in extending the shelf life of black drum fillets. Being able to extend shelf life allows fishermen and processors to reach larger markets.

P3-178 Development of Predictive Models for Vibrio vulniﬁcus and Vibrio cholerae Growth in Gizzard Shad Sashimi Yujin Kim, Sun-Young Park and Yohan Yoon
Seokkyung Women’s University, Seoul, South Korea

**Purpose**: The objective of this study was to develop predictive models to predict V. vulniﬁcus and V. cholerae growth in gizzard shad sashimi.

**Methods**: Growth patterns of V. vulniﬁcus and V. cholerae were compared in Luisa-Bertani + 2% NaCl (w/v) broth at 7, 10, 15, and 25°C. A mixture of V. vulniﬁcus and V. cholerae was then inoculated on gizzard shad sashimi samples (10 g) in three MAP gas conditions. The bacterial cell counts were enumerated on CHROMagar Vibrio during storage at 7, 10, 15, and 25°C. The Baranyi model was fitted to kinetic parameters such as p0 (maximum specific growth rate; log CFU/g/d), LD (lag phase duration; h), and the mixture increased under the investigated distribution of 145±1.4, 14, as storage temperature increased. Moreover, the secondary models were appropriate to evaluate temperature effect on p0, LD, and with 0.899 and 0.913 of R2, respectively. For validation, predicted data, and SAR (SAR [mean±standard errors]); a, b, and R2 factor (b) were calculated.

**Results**: There was no difference in growth patterns between the two species, p0 of the mixture increased (P>0.05) from 0.08 to 0.50 log CFU/g/L and LD decreased (P<0.05) from 145±1.4, 14, as storage temperature increased. Moreover, the secondary models were appropriate to evaluate temperature effect on p0, LD, and with 0.899 and 0.913 of R2, respectively. For validation, predicted data, and SAR (SAR [mean±standard errors]); a, b, and R2 factor (b) were calculated.

**Significance**: The developed models were useful in predicting cell counts of the bacteria in gizzard shad sashimi, and the models showed the bacteria can survive and grow even at low temperature. Therefore, a technology to control them in the gizzard shad sashimi should be developed.

P3-179 Quantification of Risk for Vibrio parahaemolyticus Foodborne Illness by Sea Pineapple (Halophyta rorizetini) Consumption Joohyun Kang1, Woon Kim2, Min Suk Rhe4 and Yohan Yoon2
Seokkyung Women’s University, Seoul, South Korea

**Purpose**: This study analyzed the risk of foodborne illness by sea pineapple consumption in Korea.

**Methods**: Thirty sea pineapple samples were analyzed to detect V. parahaemolyticus. Predictive model for V. parahaemolyticus cell counts in sea pineapple was established by the ENMtoolkit software. The distribution conditions, consumption amount and frequency for the sea pineapple, as well as the dose-response model were also surveyed. With all collected data, a simulation model was prepared, and the probability of foodborne illness for V. parahaemolyticus by sea pineapple consumption was predicted by the ENMtoolkit program.

**Results**: The distribution of Riskb (7.30) for the prevalence of V. parahaemolyticus estimated that the initial contamination level was -1.9 log CFU/G. With this initial contamination level, the developed Baranyi models showed that the V. parahaemolyticus cell counts increased under the investigated distribution conditions. For the investigated conditions, appropriate distributions were RiskUniform (0.72) and RiskLogLogistic (-29.28, 33.227, 26.666, RiskTruncate (-29.283, 33.227, 26.666, RiskTruncate (0,72)) for time and temperature, respectively. The consumption amount and frequency were 62±14, 2% and 0.82, respectively. The combination of these results with the β-Poisson dose-response model (Risk <1, doz/1, β=1, Risk>1, β=2) showed that the probability of V. parahaemolyticus foodborne illness by sea pineapple consumption was 4.03±10−4 per person per day.

**Significance**: This result should be useful in evaluating the risk of Vibrio parahaemolyticus foodborne illness by sea pineapple consumption.
P3-180 Prevalence, Antibiotic Resistance, and Virulence Gene Profiles of Listeria monocytogenes Isolated from Smoked Salmon in South Korea

Je Hyeong Koo1, Hyun Jun Park2, Seung Hoon Kim3, Won Bi Shim4, Dongyoun Bae4, Joon Yong Kim5, Meeyong Kim5, Hye Sun Kwak6, Jinhyun Kim7, Youngsook Jung8 and Kun Ho Seo9

1Konkuk University, Seoul, South Korea, 2Kyung Hee University, Seoul, South Korea, 3Uing Eui University, Busan, South Korea, 4Division of Applied Life Science, Graduate School and Department of Agricultural Chemistry and Food Science & Technology, Gyeongsang National University, Jinju, South Korea, U.S.A, Food and Drug Administration/NCCT, Jefferson, AR, 5Food Additives and Packaging Division, Ministry of Food and Drug Safety, Cheongju, Chungbuknd-do, South Korea, 6Ministry of Food and Drug Safety, Cheongju, Chungbuknd-do, South Korea.

Purpose: The aim of this study was to determine the prevalence of L. monocytogenes in smoked salmon and its processing plants and characterize the isolates in terms of antibiotic resistance and virulence determinants.

Methods: Three hundred seventy-five and 360 samples of packaged smoked salmon products and sausages were collected through retail markets and the food processing plants, respectively, in South Korea during 2018. Presumptive Listeria spp. isolates from the samples were identified using real-time polymerase chain reaction (PCR) and confirmed using serotyping and the identification of antimicrobial resistance and virulence determinants.

Results: Forty isolates were confirmed as L. monocytogenes from 12 salmon samples and 11 of the processing plants (100% and 11.1%, respectively). Both were susceptible to all antimicrobials used in this study. In addition, our isolates carried virulence genes (hlyA, actA, iap, and cna) that were the same as those in the reference strains. Both isolates were identified as serotype 1/2a. Both were susceptible to all antimicrobials used in this study. In conclusion, the results showed that L. monocytogenes isolated from both smoked salmon and its processing plants was virulent and susceptible to all antimicrobials used in this study.

Significance: This study provides valuable information for the seafood industry to control L. monocytogenes contamination in smoked salmon and its processing plants.

P3-181 Microbiological Characteristics of Non-eviscerated Smoked Blue Whiting (Micromesistius poutassou) Fish during Storage

Abiodun Kupolub1, Adeola Olusge2, Olubagba2, Obadina1

1Federal University of Agriculture, Abeokuta, Abeokuta, Nigeria, 2Federal University of Agriculture Abeokuta, Abeokuta, Nigeria, 3Federal University of Agriculture, Abeokuta, Nigeria.

Purpose: This study investigated the microbiological qualities of non-eviscerated smoked Blue Whiting fish (Micromesistius poutassou) during storage.

Methods: Ten samples each of non-eviscerated smoked fish were randomly and aseptically collected from five different selected processors in Makoko, Lagos, Nigeria. Laboratory prepared eviscerated and non-eviscerated smoked fish served as control samples. The microbial quality (total plate count TCP, total fungal count TFC, Enterococcus coli, Shigella and Listeria counts) were determined, during zero to five of storage. Smoked fish samples from processors and control were subjected to refrigeration and ambient temperature during which they were subjected to microbiological analysis. Data obtained were subjected to analysis of variance.

Results: The control samples (eviscerated and non-eviscerated) had TFC (2.15 and 2.7 log CFU/g) and TPC (2.24 and 3.25 log CFU/g), respectively. During storage, after 10, nine, and three days at 10°C and 38, 16, and 12 h at 20°C, respectively. In mahi-mahi inoculated with 10⁴ CFU/ml, L. monocytogenes was isolated from 375 RTE smoked salmon products, whereas no L. monocytogenes was isolated from plant environmental samples. L. monocytogenes was isolated as serotype 1/2a. All were susceptible to all antimicrobials used in this study. The control samples (eviscerated and non-eviscerated) had TFC (2.15 and 2.7 log CFU/g) and TPC (2.24 and 3.25 log CFU/g), respectively. During storage, after 10, nine, and three days at 10°C and 38, 16, and 12 h at 20°C, respectively. In mahi-mahi inoculated with 10⁴ CFU/ml, L. monocytogenes was isolated from 375 RTE smoked salmon products, whereas no L. monocytogenes was isolated from plant environmental samples. L. monocytogenes was isolated as serotype 1/2a. All were susceptible to all antimicrobials used in this study. In conclusion, the results showed that L. monocytogenes isolated from both smoked salmon and its processing plants was virulent and susceptible to all antimicrobials used in this study.

Significance: The results of this study suggest that non-eviscerated smoked Blue Whiting fish (Micromesistius poutassou) should be handled with caution to prevent foodborne illness.

P3-182 Histamine Production by Photobacterium spp. in Tuna and Mahi-Mahi Tissue at Various Storage Temperatures

Marlee Hayes1, Kate L. Baller2, Jessica Nadzi3, Ronald A. Benner1, jr and Kristin Bjornsdottir-Butler

1U.S. Food and Drug Administration, Gulf Coast Seafood Laboratory, Gulf Island, LA, 2Center for Marine Biotechnology and Biomedicine, University of Delaware, Newark, DE, 3Center for Marine Biotechnology and Biomedicine, University of Delaware, Newark, DE.

Purpose: The objective of this study was to determine the time and temperature factors that influence histamine production in tuna and mahi-mahi inoculated with psychrotrophic HP Photobacterium species.

Methods: P. phosphoreum and P. aestuarii were previously isolated from the fish. The HP Photobacterium species and the control fish samples were incubated at 2, 4, 6, and 8°C for 4 weeks.

Results: The HP Photobacterium species were isolated from the fish. The HP Photobacterium species and the control fish samples were incubated at 2, 4, 6, and 8°C for 4 weeks.

Significance: The results of this study suggest that non-eviscerated smoked Blue Whiting fish (Micromesistius poutassou) should be handled with caution to prevent foodborne illness.

P3-183 Metagenomic Evaluation of Methods to Recover of Vibrio spp. from Oysters

Shannara Lynn

NOAA, Pacoletiv, MS

Purpose: Outbreaks of Vibrio parahaemolyticus have been linked to raw oysters, clams and other seafood fish on multiple occasions. Efficient recovery of Vibrio spp. from implicated foods is first and foremost in public health endeavors to understand source and route of contamination events.

Methods: Five oysters (from the Chesapeake Bay) were purchased locally, shocked and homogenized. Oyster mesogloata were plated at 37°C for 24 hours in alkaline peptone water. Four hours later, six plates (3±4) of four ml were taken from each enriching oyster (mpn). Cells were pelleted and DNA was extracted using the DNeasy Blood and Tissue Kit (Qiagen). Libraries were prepared with the Nextera Dna Flex PCR Kit (Illumina) and sequenced on the Illumina MiSeq platform using a 250bp paired-end protocol designed taxonomy using Cosmolab.

Results: Forty-six bacterial species were detected in the Vibrio spp. from these oysters. The sequences obtained were submitted to the COI database for further analysis.

Significance: This study provides valuable information for the seafood industry to control L. monocytogenes contamination in smoked salmon and its processing plants.

P3-184 Rapid Screening for Finfish Species Substitution Using Chip-based Capillary Electrophoresis and a Web-based Application

Shannah Lynn

NOAA, Pacoletiv, MS

Purpose: Water extraction and chip-based microfluidic electrophoresis (Agilent 2100 Bioanalyzer) for the analysis of high abundance fish muscle proteins along with a novel data analysis method for species-specific protein pattern recognition was developed and validated. The resulting protein patterns were used to quickly and accurately identify fish species.

Methods: A 0.15-g muscle tissue sample from 866 authenticated, raw finfish specimens were added to denatured water for extraction of sarcoplasmic proteins. The extracted samples were differentiated using chip-based microfluidic electrophoresis. The mean masses of the highest abundance proteins were selected for inclusion in the species-specific protein pattern-matching logic-based algorithms to produce probability matches between unknown samples and authenticated reference species.

Results: The performance validation study for the method's capacity to distinguish often-substituted species from their common illegal replacement species was evaluated using sensitivity (true positive values), specificity (true negative values), and accuracy. Mean results showed 86% sensitivity to recognize fish species of different species, 97% specificity to recognize the product with an accurate species label and 95% accuracy correctly linking the highest probability match to the labeled product. As an example, for American catfish versus other catfish including Asian catfish, the tool performed at 95% sensitivity, 100% specificity and 97% accuracy.

Significance: The chip-based microfluidic electrophoresis method combined with the web-based tool provides the seafood industry with an efficient and high throughput alternative to DNA methods.

P3-185 The Effect of Tumbling Processes on the Shelf Life of Whole Octopus (Octopus vulgaris kagoshimensis and A. argus) Stored in Ice

Yu-Ru Huang1, Chi Jen Lo2, Ying-Huang Tsai3 and Yi Chen Lee1

1National Penghu University of Science and Technology, Penghu, Taiwan, 2Chang Gung University, Taoyuan, Taiwan, 3National Kaohsiung University of Science and Technology, Kaohsiung City, Taiwan

Purpose: The traditional way to overcome octopus toughness has been the repeated “slapping” of the freshly caught octopus on the rocks by the sea. This procedure has been adopted by the industry and the mechanical “tumbling” of octopus is performed in a custom-made tumbler.

Methods: This study evaluated the effects of tumbling on the sensing, microbiological, and physical properties of iced whole octopus (Octopus kagoshimensis and A. argus).

Results: The increases in psychrophilic bacteria were in agreement with the increases in total volatile basic, trimethylamine and formaldehyde contents. Inoculum (Hei) was the most effective level to increase the total psychrophilic bacteria. The mean masses of the highest abundance proteins were selected for inclusion in the species-specific protein pattern-matching logic-based algorithms to produce probability matches between unknown samples and authenticated reference species.

Significance: This study provides valuable information for the seafood industry to control L. monocytogenes contamination in smoked salmon and its processing plants.

Poster 304 Journal of Food Protection Supplement

Poster 305 Journal of Food Protection Supplement
P3-186 Food-derived Bioactive Peptides on Antioxidative Capacity, Xanthine Oxidase and Tyrosinase Inhibitory Activity

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Introduction: 11 biosynthesized peptides derived from fish and egg white were investigated for their antioxidative capacity, xanthine oxidase ( XO ) and tyrosinase inhibition. The XO is an important hepatic enzyme in purine catabolism, catalyzing the oxidation and breakdown of hypoxanthine to xanthine and subsequently to uric acid. Tyrosinase catalyzes the oxidation and breakdown of tyrosine to DOPA (dihydroxyphenylalanine) in melanocytes of human skin.

Purpose: The usage possible of bioactive peptides as an antioxidant and the mechanism of antioxidant activity of the peptides was studied. In addition to that the usage possible of bioactive peptides against xanthine oxidase and tyrosinase was also discussed.

Methods: The antioxidative capacity of peptides was measured by the 2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)(ABTS) radical scavenging activity. The XO inhibition assay was performed using a standard procedure. The tyrosinase inhibition assay was performed in vitro using a standard procedure.

Results: The antioxidative capacity of peptides was evaluated as the percentage of ABTS radical scavenging activity and 100% was defined as the control with no percentage of ABTS radical. The IC50 value for XO inhibition was calculated as the concentration of peptides that caused 50% of inhibition as compared to the control.

Significance: These data suggest that same-day detection of Vibrio in seawater can be performed rapidly and more efficiently than current test methods.

P3-187 Rapid Concentration and Molecular Detection of Vibrio harveyi in Oyster Farm Seawater

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Methods: The purpose of this study was to determine the effects of HPP on the levels of aerobic plate count (ATCC), total viable bacteria (TVB), Vibrio harveyi and histamine increase in marlin meat, regardless of the storage temperature. In summary, this result suggested the marlin filets treated with HPP (>300 MPa, five min) can prevent deterioration of product quality up to 42 days at the temperature between -3°C to 5°C. Significance: This study demonstrated that 300 MPa for five min for marlin meat is the optimum HPP condition for controlling color, microbial load, TVB and histamine changes. Overall, these results prove the usefulness of HPP in seafood processing while enhancing the preservation and safety of marine fish consumption.

P3-191 Antibacterial and Antibiofilm Mechanism of Eugenol against Vibrio parahaemolyticus Clinical and Environmental Isolates

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Methods: According to the trend which is to reduce high intake of salt, the purpose of this study was to make low-salt fermented clams to examine the virulence of V. parahaemolyticus has attracted considerable attention because of its pathogenicity, antibiotic resistance pattern, biofilm formation, and ability to contaminate seafood in the United States and Asia. Purpose: The objective of this study was to investigate the antibacterial and antibiofilm activity of eugenol in Vibrio parahaemolyticus and to evaluate the potential for use in food preservation.

Methods: The time-kill assay, motility, hydrophobicity, nucleic acids and protein leakage, transmission electron microscopy, biofilm formation and eugenol treatment, crystal violet staining, (2, 3, 5)-tetra later in the starch-larotaxin-5-carboxylate reduction method, phenol-sulfuric acid method, field emission scanning electron microscopy, confocal laser scanning microscopy and high-performance liquid chromatography were used in this study.

Significance: This study demonstrated that eugenol can be used to control V. parahaemolyticus biofilms, biofilm-related infections and can be employed for the protection of seafood.
Significance: This study suggests that norovirus in Jogaejeotgal can be effectively inhibited through hurdle technology using ClO₂.

This study suggests that norovirus in Jogaejeotgal can be effectively inhibited through hurdle technology using ClO₂. This study indicates that addition of 0.03% nisin in a whitefish seafood salad is an effective hurdle technology strategy during 42-d aerobic storage under the tested parameters.

Introduction: Several foodborne outbreaks in the United States have been attributed to dishes served with raw meat. Consumption of these foods is always compliant with Mexican standards. Better temperature control mechanisms should be implemented to meet Mexican standards. In retail stores, meat products' shelf life depends on the temperature within the showcase used. Participants expressed strong preferences for these foods because they eat them during holidays or other special occasions as part of family cultural traditions.

Significance: After CTD, 10% salt was added to the clamp. E-beam treatment was carried out after chemical treatment and performed after seven d of storage. The dosages were one, three, and five kGy, respectively. Results: Reduction values were 1.03 and 1.11 log PFU/ml for 10 and 20% salt, respectively, following 25 d of storage. After the treatment with CTD (50 to 300 ppm) in salted clam at 10%, MNV-1 titers were 4.30 to 3.30 log PFU/ml. The sequential treatment of CTD (300 ppm) and e-beam (5.5 keV) showed 1.90-log reduction after seven d of storage at 5°C.

Significance: The study demonstrates the effectiveness of microwave treatment on beef jerky inoculated with Salmonella and Listeria monocytogenes. The results showed that treatment 1 can reduce more than 1.9 log CFU/ml of L. monocytogenes.

Poster P3-196 Effect of Cranberry Pomace on the Inactivation of Salmonella enterica Serovars and Physicochemical Changes during Dry Fermented Sausage Manufacturing

Introduction: Cranberry pomace (CP) is a cranberry processing byproduct and possesses antimicrobial properties. There is strong consumer interest for products with natural antimicrobials, but there are limited reports examining the practicality of using CP as a natural antimicrobial agent in processed meat products, especially non-thermally processed dry fermented sausages (DFS).

Purpose: The objective of this study was to evaluate the effectiveness of microwaves as a post-packaging intervention to reduce Salmonella and Listeria monocytogenes in raw meat products, especially non-thermally processed DFS.

Methods: DFS containing a raw-lean-cooked (RLC) combination of traditional Bavarian meat species were inoculated with a mixture of S. enterica serotypes I and V. Six different DFS treatments were prepared with fresh tissues from the same pigs embedded in the same dough. DFS incubated under an initial internal oven temperature of 4°C for 5 d.

Results: These results indicate that addition of 0.03% nisin in a whitefish seafood salad is an effective hurdle technology strategy during 42-d aerobic storage under the tested parameters.

Significance: This study analysis was with a mixed model under a complete randomized design, with repeated measurements. The mean comparison was with a multiple range test. The aim of this research was to determine temperature within an open refrigerated meat showcase of one deck in five retail stores of major Mexican supermarket chains.

Results: In four out of the five stores in front of the deck temperature was above the Mexican standard (4°C) and showed no differences (>0.05) among stores and always complied with Mexican standards.

Significance: The temperature in open refrigerated meat showcase of one deck depends on the store, position within the deck and is not always compliant with Mexican standards. Better temperature control mechanisms should be implemented to meet Mexican standards.

Introduction: Several foodborne outbreaks in the United States have been attributed to dishes served with raw meat. Consumption of these foods is always compliant with Mexican standards. Better temperature control mechanisms should be implemented to meet Mexican standards.

Results: Several foodborne outbreaks in the United States have been attributed to dishes served with raw meat. Consumption of these foods is always compliant with Mexican standards. Better temperature control mechanisms should be implemented to meet Mexican standards.

Posters 308-309
P-202 Estimating the likelihood of Toxoplasma gondii-contami-

ated Fresh Cut Meats

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Poster

International Poster Session

Introduction: Toxoplasma gondii is capable of infecting almost all warm-blooded animals including humans and causes a disease condition, known as toxoplasmosis. Although its seroprevalence is high in humans, T. gondii is recognized as one of the neglected parasites. Humans become infected by ingesting oocysts or tissue cysts in raw or under-cooked meat or by eating cat feces mixed in food or water. A major portion of human T. gondii infections is acquired through the consumption of poorly cooked meat containing tissue cysts.

Purpose: There is no robust information available on the concentration of viable T. gondii in muscle tissues of naturally infected meat animals. The goal of this study was to quantify viable T. gondii concentration in meats of naturally infected lambs and goats.

Methods: The shoulder or leg cuts of 44 lambs and 39 goats were sero- logically tested for T. gondii antibodies through MAT (modified agglutination test) and the ECO test (enzyme-linked immunosorbent assay). DNA was isolated from positive tissue samples and further genotyped with T. gondii specific markers.

Results: Although the serology were very low in both lambs (4%) and goats (2.8%), the likelihood of getting it from consuming T. gondii contaminated meat is very high. With the use of USDA guidelines of cooking the one hundred and forty-five to seventy gondii contaminated under-cooked lamb meat as 0.28, 0.45 and 0.81 respectively. There was a 100% chance of getting sick from consuming contaminated goat meat. The concentration of 529 base pair bands in agar gel confirmed the presence of T. gondii DNA in one of the samples.

Significance: This study indicates that T. gondii can be present in naturally infected meat animals and could pose a threat of foodborne illness to consumers.

P-203 Shiga toxin-producing Escherichia coli harboring stx1 or stx2 genes isolated from Poultry Meat in Brazil

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Introduction: Shiga toxin-producing E. coli (STEC) is one of the most important foodborne pathogens frequently reported in food products of animal origin worldwide. However, despite its great importance in public health, there is insufficient information regarding the presence of this microorganism in poultry meat in Brazil.

Purpose: The aim of this study was to investigate the presence of STEC in poultry meat samples recovered from poultry slaughters isolated in different regions of Brazil and characterize its virulence factors: stx1, stx2, eae and ehhx.

Methods: One hundred and forty-four poultry meat samples collected from slaughters isolated in southern, southeastern and central-western regions of Brazil. were analyzed to monitor the presence of Shiga toxin-producing E. coli by using PCR according to SUT/D13156 and USM/MLG 580.5 methods.

Results: Among the samples, 13 of the 20 samples were positive, while the rest were negative for Shiga toxin-producing gene stx1. Among the positive isolates, ten isolates were positive for Shiga toxin-producing gene stx2 and thirty-three isolates were positive for eae gene alone. None of the isolates were the positive for ehhx. The presence of stx1 or stx2 genes suggest that chicken can be a vehicle for transmission of STEC with an impact on food safety.

Significance: The detection of the eae alone in some strains suggests the presence of enteropathogenic E. coli (EPEC), also responsible for attaching and effacing lesions. However, more research is required with a larger number of samples, since there is not enough data regarding STEC in poultry meat worldwide.

P-204 Evaluation of Listeria monocytogenes and Staphylococcus aureus Survival and Growth on Natural-source Nitrite-cured Ham during Stabilization

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Introduction: USDA recently added guidelines for stabilization for meat products cured using naturally-occurring nitrites cure to prevent growth of pathogens such as Listeria monocytogenes and Staphylococcus aureus.

Purpose: To investigate the temperature profiles of alternatively cured hams during refrigerator cooling and determine the survival of S. aureus and L. monocytogenes on ham during these cooling periods.

Methods: Whole, half and quarter hams were cured with celery powder and cherry powder containing 150 ppm nitrite and 250 ppm ascorbate, cooked in a smokehouse until internal temperature reached 140°F, and immediately transferred into a walk-in cooler (38.9°C). Surface and internal temperatures were recorded. Small cooked ham samples (25 g) prepared using identical formula were inoculated with 5 x 10^6 CFU/g of S. aureus or L. monocytogenes (–4 log CFU/g) to simulate level of initial contamination. Heating was considered an additive, the implications of these PCRM-method simplifications are huge for the beef industry. We have previously shown that fluorescence resonance energy transfer (FRET)-based real-time PCR GENE-UP E. coli O157:EHEC (ECO) does not cross react with non-pathogenic O157 isolates.

P-205 Comparison Effect of NaCl and KCl on Clostridium sporogenes PA3679 as Surrogate for C. butylicum in Shelf-stable Mortadella

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Introduction: Botulism is a disease of high severity associated with meat products. A factor that inhibits the germination of Clostridium botulinum spores is the water activity (aw) in meat products the reduction of aw can be carried out by addition of NaCl or KCl.
Purpose: The aim of this study was to evaluate the effect of 
0.95, 0.96 and 0.97 adjusted with different salts (NaCl and KCl). The samples were harvested at 85°C until the internal center of the product reached 72°C. The concentration of the gastrointestinal flora was determined at the end of the intervention (attachment), and one and 24 h after intervention application. Microbial enumeration was performed by plating onto MacConkey agar with a tryptic soy agar overlay. Bacterial enumeration data were converted into log values for statistical analysis relative to CFU/100 cm². 

Results: All treatments began with 6.5 to 6.9 log CFU/100 cm² E. coli. Coliform counts were determined in Modified MacConkey agar with a tryptic soy agar overlay. Bacterial enumeration data were converted into log values for statistical analysis relative to CFU/100 cm². 

Antimicrobials Targeting L. monocytogenes L. monocytogenes w was adjusted in the emulsions with a high acid-acidified with a two percent acetic acid solution), PAA+Titon Gate strains of E. coli that mimic Salmonella were spayed on carcasses in the shoulder and ham area. Interventions, as assigned, were sprayed onto the carcass surface. Intervention treatments included control (no intervention), water, titon (sulfuric acid and sodium-sulfate), PAA(meric acid (pH acidified with a two percent acetic acid solution), PAA-Titon (paractic acid and acetic acid), T (lactic acid solution), HBR (hypobromous acid 300 ppm), and LAGE (lauric acid ethyl ester) Swabs on pork carcasses were used to determine microbial concentration after inoculation (attachment), 1 hour after, and 24 hours after intervention application. Microbial enumeration was performed by plating onto MacConkey agar with a tryptic soy agar overlay. Bacterial enumeration data were converted into log values for statistical analysis relative to CFU/100 cm² and data were analyzed using SAS. 

Results: All treatments resulted in a greater reduction of Listeria innocua. After 42 d, reductions of 2.4 and 3.4 log CFU/g were obtained on the surface of the samples aged at 2°C and 8°C, respectively. The final water activity were 0.92 and 0.89. 

Introduction: Dry aging is one of the main types of meat aging. However, few studies assess the impact of this process on food safety. Listeria monocytogenes is a virulent pathogen that causes a foodborne disease and is able to develop at low temperatures. Thus, studies that evaluate the behavior of this microorganism during dry aging are extremely important.

Purpose: The objective of this study was to evaluate the effect of dry aging temperatures on the behavior of Listeria innocua as a surrogate of L. monocytogenes. 

Methods: Stripsion pieces (1.5 kg) were inoculated with six log CFU/g of L. innocua ATCC 33091 and subjected to the dry aging process for about 42 to 4-2 days at 2°C and 8°C. Controls were determined in Modified Listeria Oxford agar. The data obtained were evaluated using ANOVA and the Weibull model. 

Results: The process conducted at 8°C resulted in a greater reduction of Listeria innocua. After 42 d, reductions of 2.4 and 3.4 log CFU/g were obtained on the surface of the samples aged at 2°C and 8°C, respectively. The final water activity were 0.92 and 0.89. 

Discussion: The results presented in the current study are in agreement with previous findings. The versatility of RSM was tested using Salmonella enterica sp. Typhimurium and Salmonella enterica sp. Cholerae suis. The use of Salmonella enterica sp. Typhimurium and Salmonella enterica sp. Cholerae suis as a surrogate of Salmonella sp. allowed the development of a new and effective antimicrobial combination in the porcine industry. The use of Salmonella enterica sp. Typhimurium and Salmonella enterica sp. Cholerae suis as a surrogate of Salmonella sp. allowed the development of a new and effective antimicrobial combination in the porcine industry. The use of Salmonella enterica sp. Typhimurium and Salmonella enterica sp. Cholerae suis as a surrogate of Salmonella sp. allowed the development of a new and effective antimicrobial combination in the porcine industry.
P3-212 Microbiological Safety of Staphylococcus aureus and Escherichia coli in Dry-aged Beef Requiring Long Aging Time

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**Developing Scientist Entrant**

Introduction: Dry-aging is one of the aging processes that takes a longer time than others, especially for beef. This aging process is popular, because it produces flavor and texture that consumers prefer. However, food safety concerns for the process have been raised. This study investigated the growth of the harmful foodborne pathogens in dry-aged beef samples.

Methods: A mixture of S. aureus isolates or a mixture of E. coli isolates from dry-aged beef were inoculated on dry-aged beef (25 g) at 2.5 log CFU/g. The samples were stored at four ± 15°C for 7 days in triplicate. S. aureus and E. coli cell counts were then enumerated on Baird Parker agar and 3M Petrifilm® PGP deoxygen Count Plates, respectively. The Barany models were fitted to the S. aureus and E. coli cell count data to calculate maximum specific growth rate ($\mu_{\max}$) and lag phase duration (LPD).

Results: S. aureus and E. coli growth were not observed at 4°C, but the cell counts were maintained at initial concentration during storage periods. The S. aureus and E. coli cell counts were increased (P < 0.05) at 15°C and 25°C, respectively, from 0.06 to 0.08 log CFU/g, and LPD decreased (P < 0.05) from 21.72 to 3.51 h, as storage temperature increased. During storage, S. aureus grew up to 5.3 log CFU/g, and E. coli grew up to 6.2 log CFU/g after 48 h at 15°C. S. aureus showed longer (P < 0.05) and slower (P < 0.05) growth than E. coli during storage at low temperature.

Significance: These results indicate that a food safety regulation about temperature and period needs to be established to improve food safety of dry-aged beef, because the dry aging has a longer aging period than wet-aging beef, and its trim is surrounded with a knife, which may allow cross-contamination and foodborne pathogen growth.

P3-213 Comparative Evaluation of Sanitizers for the Control of E. coli O157:H7 in Ground Beef

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**Developing Scientist Entrant**

Introduction: Escherichia coli O157:H7 is a common contaminant of ground beef, resulting in numerous outbreaks and recalls. Ground beef processing, which involves grinding and packaging of beef cuts, relies primarily on carcass decontamination for pathogen control. Furthermore, temperature abuse (> 4°C) of ground beef during retail could influence pathogen growth. Although conventional sanitizers are used as beef-trim interventions, their efficacy at low temperatures, especially during retail storage, is questionable. It is therefore important to devise intervention strategies that would maintain effectiveness throughout retail storage.

Methods: Evaluate the efficacy of acetic acid, peracetic acid and pelargonic acid in reducing E. coli O157:H7 during ground beef processing and storage. Each month, four retail samples were inoculated with a cocktail (~three log CFU/g) of five E. coli O157:H7 strains, with an attachment time of 15 minutes. The trims were spray-washed (20 ml) with either water, lactic acid (five percent), peracetic acid (400 ppm) or pelargonic acid (five percent) and allowed a dwell time of one minute. Positive and negative controls were also used. The treated samples were then rinsed in a meat grinder and stored in chilled (1°C) retail packaging. Samples were incubated at room temperature (25°C) and the cell counts were increased. Formulations targeting 1.5% salt and containing 1.0% DV-FSE-A or 1.0% FSE-S-A + 0.5% FSE-S-C supported an average 1.74 and 1.51 log decrease, respectively, but no growth was observed using 1% DV-FSE-A + 0.5% DV E. coli. Results: After 15 hours, C. perfringens populations in the uncured control increased by 10 log CFU/g whereas populations in the cured controls remained unchanged regardless of salt concentration. Formulations targeting 1.5% salt and containing 1.0% DV-FSE-A + 1.0% FSE-S-A + 0.5% FSE-S-C supported an average 1.74 and 1.51 log decrease, respectively, but no growth was observed using 1% DV-FSE-A + 0.5% DV E. coli.

Significance: This study confirms that combinations of clean label antimicrobials in uncured ham show similar inhibition of C. perfringens compared to traditionally cured ham during extended cooling.

P3-214 Humidity Affects Salmonella Lethality and USDA FSIS Appendix A Compliance for Impingement-cooked Meat and Poultry Products

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**Developing Scientist Entrant**

Introduction: A recent revision of USDA FSIS Appendix A articulates humidity requirements for meat/poultry cooking processes. However, information documenting the impact of humidity on the microbial safety of impingement cooked meat and poultry products is limited.

Purpose: This study aimed to quantify the impact of humidity on Salmonella lethality in impingement cooked meat and poultry products, particularly as related to Appendix A compliance.

Methodology: Beef strips, ground beef patties, chicken breast fillets, and breaded chicken patties were inoculated with an eight-serovar cocktail of Salmonella. Each product was cooked (in triplicate) in a pilot-scale, moist-air impingement oven at a specific temperature (218 or 232°C), absolute humidity (0.7, 30, or 70% rh), and fan speed (20 or 80 rpm). To an end point center temperature of 70 or 72°C for beef and poultry products, respectively, then immediately cooled to 4°C. Samples were removed from surface and core locations, and plated on differential media to enumerate face/core temperatures and final moisture content also were measured.

Results: Average salmonella lethality significantly (P < 0.05) increased with the increase in humidity. For example, when processed with dry heat, the required 5-log reduction was not achieved for beef products at any condition. However, increasing the humidity to 30% resulted in greater than seven-log reduction for all beef strips. Additionally, greater Salmonella survival in samples from the surface was observed (P < 0.05) for multiple cooked products dried at 7°C for 24 hours.

Significance: Compliance with USDA FSIS Appendix A is critical for the meat and poultry industry to provide a safe product; however, the relationship between humidity and Salmonella survival includes complicated interactions between time, temperature, and air velocity. More research is necessary to confirm specific levels of humidity needed to achieve the required Salmonella lethality.

P3-215 The Effect of Recurring Cooling and Reheating on Clostridium perfringens Growth in Uncured Turkey and Cured Beef

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Introduction: Heat-resistant spores of C. perfringens may germinate and multiply in cooked meat and poultry products when the rate and extent of cooling do not occur in a timely manner. There is a lack of information in the literature on the subsequent reheating/smoking and cooling of meat products that have already achieved lethality.

Purpose: The purpose of this study was to compare the effect of culture inoculum and incubation levels when challenging nine RTE meat products with L. monocytogenes to assess growth at 4°C for 18 weeks.

Methods: Culture inoculations were performed. In the first preparation, L. monocytogenes strains were grown in Brain Heart Infusion (BHI) broth, subcultured into brain heart infus (BHI), and incubated at 30°C for 24 h. Final average inoculum level was 2.3 log CFU/g. In a second culture preparation, strains were started in tryptic soy broth with 0.6% yeast extract (TSBYE), subsequently transferred two consecutive times into TSBYE and incubated for 24 h. The average inoculum level was 3.1 log CFU/g. For the third culture preparation, strains were started in TSBYE, subsequently transferred into TSBYE at 35°C for 24 h and followed by a single transfer into TSBYE at 7°C for seven days. Average inoculum levels were 3.7 log CFU/g and 2.7 log CFU/g.

Results: Greater than a 1-log CFU increase of L. monocytogenes was observed in nine meat samples using the second culture preparation compared to two meat samples using the first culture preparation. With the third incubation procedure, greater than one log CFU of L. monocytogenes counts was observed in six products with the higher inoculum level compared to five products with the lower inoculum level.

Significance: Culture preparation or inoculum levels can impact the growth of Listeria monocytogenes in RTE meats during refrigerated storage.
P3-218 The Effect of Pulsed Light Energy Delivery on Inactivating Salmonella spp. in Vitro

- **Introduction:** Despite improved practices on the farm and in food processing facilities, Salmonella contamination is a big concern for the food industry around the world. Pulsed light is a promising nonthermal method for surface decontamination of raw chicken, meat, or fruits and vegetables, and it has been continuously sought out. Pulsed light (PL) technology has great potential in inactivating bacteria on the surface of many foods, including raw chicken, without compromising their freshness, but the effectiveness of the treatment needs to be improved.

- **Purpose:**
  - The energy delivery mode of PL inactivation of Salmonella on an agar model system for chicken breast was systematically investigated.
  - Four Salmonella serovars (Salmonella Typhimurium, Heidelberg, Enteritidis, and Mbandaka) were used in this study to determine the effect of PL on chicken.

- **Methods:**
  - Stationary phase inoculum of each strain was serially diluted, and each dilution spread onto tryptic soy agar for PL treatment. The inoculated agar plates were subjected to a combination of three voltage levels (2000 V, 2500 V, and 3000 V) and seven pulse durations (50 µs to 420 µs) with fluence ranging from 0.05 to 0.82 J/cm².

- **Results:**
  - Maximum reductions of Salmonella Typhimurium achieved after PL exposure to 2000 V, 2500 V, and 3000 V were 6.8, 6.5, and 7.1 log, respectively (average values).

- **Significance:**
  - The PL delivery mode has a significant effect on Salmonella inactivation can be used for the development of PL treatments with higher effectiveness for the decontamination of a variety of foods, including raw chicken breast.

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P3-219 Independent Performance Evaluation of VIDAS-Spp for the Detection of Salmonella spp. in Poultry Production Samples

- **Introduction:** The disease burden due to Salmonella remains a persistent issue, and despite several interventions, poultry products remain as a major food source implicated in salmonellosis. It has been hypothesized that an increased understanding of the prevalence at the primary production sites will result in greater Salmonella inactivation than in low-voltage-long-duration mode (P0.05).

- **Purpose:** The purpose of this study was to independently evaluate the performance of the candidate method for the detection of Salmonella spp. in poultry production samples. VIDAS-based Salmonella assay (SPT) has been used for Salmonella detection in raw poultry processing samples. However, the candidate method has not been extensively evaluated as a method for Salmonella detection in poultry primary production samples.

- **Methods:** Three independent laboratories (n=270 PPS obtained from the turkey and broiler facilities against the buffered peptone water method (US-DA-NPIP PS; §147.54 Jan 2017). For the candidate method, the PPS were enriched in buffered peptone water+supplement (1:10) at 42°C for 18 to 22 h, then from diverse sources.

- **Results:** The candidate method provides highly sensitive and comparable results to the culture methods and a significant time advantage (~48 h) over the culture methods, and thereby presents a viable alternative for Salmonella detection in PPS.

- **Conclusion:**
  - The candidate method is more effective than the current culture methods for the detection of Salmonella spp. in poultry samples.
  - It provides a significant time advantage over the culture methods.
  - It presents a viable alternative for Salmonella detection in PPS.

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P3-220 Detection of Multiple Serotypes of Salmonella on Pre-chilled Chicken Carcasses by Whole Carcass Incubation

- **Purpose:** The purpose of this study was to independently evaluate the performance of the candidate method for the detection of Salmonella on an agar model system for chicken breast was systematically investigated.

- **Methods:** Two carcasses were inoculated with two different Salmonella serovars (Salmonella Typhimurium, Heidelberg, Enteritidis, and Mbandaka) and incubated at 4°C for 18 to 22 h. The enriched broth was transferred to agar plates and kept at 10°C for 48 h. The percentage of positive samples was determined.

- **Results:**
  - The percentage of positive samples was determined using the Fisher's exact test.

- **Conclusion:**
  - The candidate method is more effective than the current culture methods for the detection of Salmonella on chicken breast.
  - It provides a significant time advantage over the culture methods.
  - It presents a viable alternative for Salmonella detection on chicken breast.

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P3-231 Survival of Salmonella Typhimurium and Salmonella Enteritidis after Treatment with Stress Conditions: Heating, Chilling, Salt, and Freezing Temperatures

- **Purpose:** To examine the effect of Salmonella spp. under conditions that occur during poultry processing and transportation.

- **Methods:**
  - A suspension of two Salmonella spp. in Vitro was used to inoculate chicken breast fillets.
  - The samples were treated under conditions: heating, chilling, reduced aw, and freezing in duplicate.

- **Results:**
  - The samples were treated for three min and then in thymol or carvacrol at 1.0% for three min resulted in 1.9 to 2.0-log CFU/g reduction of the Salmonella isolates.

- **Conclusion:**
  - The candidate methods were significantly (P<0.05) better than the corresponding single bacterial or essential oil compounds treatments.
P3-224 Salmonella and Campylobacter in Religious-exempt and Low-volume Poultry Products
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Introduction: In 2017, FISG began two new exploratory sampling projects for religious exempt chicken carcasses (head, feet, and/or viscera intact; 0.4% of the poultry industry) and low-volume poultry products (average daily production less than 1,000, one percent of the poultry industry), which are currently exempt from routine sampling and performance standard categorization.

Purpose: This sampling program assesses the low-volume poultry and Campylobacter contamination in religious exempt chicken carcasses and in low-volume poultry products, to evaluate if these products represent a significant public health risk.

Methods: Monitor viability of multi-stain cocktails of genetically-marked strains of Listeria monocytogenes and Shiga toxin-producing Escherichia coli (STEC) in religious-exempt and low-volume poultry products. Each sample was exposed to FSIS approved sanitizers; 500 ppm peracetic acid (PAA), 1.5% acidified sodium chlorate solution (Ach), and control conditions.

Results: Overall, 30.7% of the samples tested positive for Salmonella, with 15.9% for Campylobacter. Percent positives comparable to or greater than those observed by USDA-FSIS in conventional chicken products. We detected the live vaccine strain

Significance: Like poultry, poultry production under religious exemptions or in low-volume facilities may be contaminated with Salmonella and Campylobacter. This exploratory sampling program indicates a higher prevalence in some products, though the overall volume of such products in the food supply is low.

P3-226 Viability of Listeria monocytogenes and Shiga Toxin-producing Escherichia coli Cells on Slices of Commerically-produced Bresaola, a Dry-Cured Beef Product, during Extended Storage at 4° and 10°C
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Introduction: Despite an increasing demand across the U.S. for dry-cured, RTE meats, there is a lack of information about the safety of these products.

Purpose: This study was designed to assess the fate of pathogenic cells on slices of Bresaola when subjected to post packaging pasteurization after lethality and stabilization schedules on high and reduced moisture, shelf-stable meat and poultry products, and to evaluate the effect of storage conditions, antimicrobial formulation and water activity during shelf-life.

Methods: Three strains of C. perfringens spores were used to inoculate high and reduced-moisture meat and poultry products (n=10 treatment) before exposure to validated lethality processes. Different incremental stabilization schedules (time zero, 60, 120, 180 min) were evaluated to determine spore survival and inactivation with and without subsequent exposure to post-packaging pasteurization (min, 70°C).

Results: Viability of germinated sporeformers was significantly decreased (P<0.05) when extended stabilization rates were evaluated in high moisture products. On the other hand, post packaging pasteurization applied after short-term stabilization schedules had no effect on inactivation of spore survivors.

Significance: Extended stabilization schedules, that support spore germination after lethality exposure, may allow for significant inactivation of spore-forming pathogens by post-packaging pasteurization in reduced-moisture meat and poultry products.

P3-227 Recovery of Enterobacteriaceae Indicator Organisms in Raw Poultry Rinse Testing Using Buffered Peptone Water and Neutralizing Buffered Peptone Water
Lakshmi Vanamalai, Marcio Saltine and Meikel Brewster
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Introduction: For pathogen recovery from raw poultry carcasses in the presence of sanitizers, FIS is using neutralizing buffered peptone water (nBPW) rather than buffered peptone water (BPW). The industry is slowly adopting nBPW in its New Poultry Inspection System (NPS) for pathogen and indicator tests.

Purpose: This study evaluated the influences of nBPW vs. BPW on recovery of Enterobacteriaceae indicator organisms from poultry carcases using different indicator test methods and sanitizer residuals.

Methods: Chicken carcasses (n=20) were extracted in 400 ml Butterfield’s Phosphate buffer, then diluted in BPW and nBPW to a countable range. Each sample was exposed to FIS approved sanitizers; 500 ppm peracetic acid (PA), 1.5% acidified sodium chlorate solution (Ach), and control conditions. The samples were plated on Charm Plate Enterobacteriaceae test (AOAC OMA 2013.01) and V-Blot (15282-2). Enterobacteriaceae count was performed by total and colony negative, plus agar positive. PCR detection was employed in data comparison.

Results: Enterobacteriaceae recovery using BPW and nBPW had no significant difference between OMA 2013.05 and ISO method with P<0.06 and P<0.04, respectively, in control and in PAA-sanitized. OMA 2013.01 had a 0.6% log difference (P<0.01) in Enterobacteriaceae recovery under the same conditions. Bacteria recovered by all methods using nBPW were one-log greater than with BPW in Ach P<0.016. Results between Plate and BPW method with PAA sanitizer were not significantly different in either buffer or between methods, P>0.10. Results showed 76% confirmation of Enterobacteriaceae in BPW compared to 70% confirmation in nBPW.

Significance: These limited data suggest nBPW is more effective than BPW in recovery and selection of Enterobacteriaceae indicators when sanitizers are present, depending on the method chosen. Data indicate that industry should consider using nBPW with an appropriately validated Enterobacteriaceae method, as improved recovery and risk management are more predictable of gram-negative pathogens.

P3-228 Fate of Spore-forming Pathogens in High and Reduced-moisture, Shelf-stable Processed Meat and Poultry Products Subjected to Post-packaging Pasteurization
Sara Munoz, Andrea Engba, Ian Arelly, Mendi Brashears, Mark Miller and Marcos X. Sanchez-Pata
Texas Tech University, Lubbock, TX

Developing Scientist Entrant

Introduction: Spore-forming bacteria can survive lethal heat schedules directed to destroy vegetative pathogens and rapid cooling is necessary to inhibit germination and growth in storage of low moisture processed meat and poultry products.

Purpose: Investigate the fate of spore-forming pathogens when subjected to post packaging pasteurization after lethality and stabilization schedules on high and reduced moisture, shelf-stable meat and poultry products, and to evaluate the effect of storage conditions, antimicrobial formulation and water activity during shelf-life.

Methods: Three strains of C. perfringens spores were used to inoculate high and reduced-moisture meat and poultry products (n=10 treatment) before exposure to validated lethality processes. Different incremental stabilization schedules (time zero, 60, 120, 180 min) were evaluated to determine spore survival and inactivation with and without subsequent exposure to post-packaging pasteurization (min, 70°C).

Results: Viability of germinated sporeformers was significantly decreased (P<0.05) when extended stabilization rates were evaluated in high moisture products. On the other hand, post packaging pasteurization applied after short-term stabilization schedules had no effect on inactivation of spore survivors.

Significance: Extended stabilization schedules, that support spore germination after lethality exposure, may allow for significant inactivation of spore-forming pathogens by post-packaging pasteurization in reduced-moisture meat and poultry products.
P3-230 Thermal Inactivation of Salmonella, Campylobacter jejuni and Listeria monocytogenes in Moisture Enhanced Non-intact Chicken Patties by Double Pan-broiling Under Dynamic Conditions

Wenjia Jiang, Lacei Lemoinou, Ka-Wang Li and Cangilang Shen
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Developing Scientist Entrant

Introduction: Pathogens may transmute from the surface to internal tissue during non-intact chicken moisture enhancement processes. Limited information is available regarding the thermal inactivation of Campylobacter spp. in chicken products.

Methods: We examined the thermal inactivation kinetic parameters of Campylobacter jejuni and Listeria monocytogenes in moisture enhanced reconstructed non-intact chicken patties double pan-broiled for various time periods.

Results: The initial counts of Campylobacter jejuni and Listeria monocytogenes were 1.2 ± 1.0 log CFU/g and 3.3 ± 0.6 log CFU/g, respectively. Thermal death time (tD) values of 7.4 ± 0.8 min and 8.6 ± 1.1 min at 48°C were calculated for Campylobacter jejuni and Listeria monocytogenes, respectively.

Significance: These findings will be useful for further risk assessment and development of a safe moisture enhancement protocol for non-intact chicken products.

P3-231 Systematic Review and Meta-Analysis on the Effects of Processing Stages and Interventions to Control Campylobacter jejuni in Broiler Chickens
Onay Burak Dogan, Carmen Cano
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Developing Scientist Entrant

Introduction: A considerable number of primary research studies are documented in the literature on the effect of different processing stages and interventions to control Campylobacter jejuni contamination. However, individual studies often fail to address the overall effect and variation among different set of conditions. A systematic review and meta-analysis approach is a useful tool to combine and evaluate data from multiple sources resulting in a summarized outcome while minimizing selection bias and evaluating quality of the evidence.

Purpose: The purpose of this study is to quantify and compare the efficacy of multiple processing stages and mitigation strategies that can be implemented through the following processing chain to control Campylobacter jejuni in broiler chickens.

Methods: Six electronic databases were searched with specific syntax in order to capture relevant publications. Retrieved records were deduplicated, screened, and included in the meta-analysis. These results were then extracted from individual studies and combined into meta-analyses using Cochran’s Q statistic and I2 size and study characteristics. Meta-analyses were conducted to summarize data from individual studies into the overall change in prevalence (quantified using Log OR), concentration, and sample size. The I2 statistic was used to assess the extent of heterogeneity among the studies.

Results: Among the processing stages, defeathering and evisceration were identified to increase prevalence and concentration while depilating and chilling were associated with reductions in prevalence and concentration.

Significance: These findings will be useful by USDA-FSIS to develop risk assessments of foodborne pathogens in chicken products.

P3-232 Effect of Ozonated Water on the Microbiological Profile of Chicken Parts
Carne Cara, Yulee Meneses, Xinjuan Hu, Carly-Rain Adams and Bing Wang
University of Nebraska-Lincoln, Lincoln, NE

Introduction: Poultry meat represents an important part of the United States economy and diet. However, it remains one of the food categories responsible for the most outbreaks of foodborne illnesses. Ozonation treatment has become an attractive decontamination option for food products, introducing poultry, due to its potential antimicrobial properties and minimal effects on quality.

Purpose: To study the effect of ozonation on chicken and pig organs for the reduction of microbial indicators in chicken parts.

Methods: Skinless chicken thighs (average weight 206 ±5.3 g) were immersed in ozonated water (1.35 ±0.12 ppm) for different exposure times (5, 10, 15 and 20 min). Aerobic plate counts (APC) and coliform counts (EC) were determined on Petrifilm plates for rinsates (100 ml/sample) of ozonated and non-ozonated samples. APCs were enumerated using 3M Petrifilm Aerobic Count plates and ECs were enumerated using 3M Petrifilm Coliform Count plates.

Results: Significant reductions in counts were observed in the ozonated samples compared to the control samples. The highest reduction observed was 9.5 log CFU/mL for APCs and 9.0 log CFU/mL for ECs.

Significance: Ozonation treatment can be a useful method for reducing microbial counts on chicken and pork organs. Further studies are needed to evaluate the long-term effectiveness of ozonation on poultry and pork tissues.

P3-233 Inactivation of Listeria monocytogenes in Model Chilling Brines for Hard Cooked Eggs
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Developing Scientist Entrant

Introduction: Hard-cooked, shelf-stable eggs sold for use in high-acid products are chilled using acetic acid (100 ppm) and water. The purpose of this study was to investigate the inactivation of L. monocytogenes in model chilling brines for hard-cooked eggs.

Methods: Sixteen test brines were formulated at four levels each (four by four) at target pH values of 3.4, 3.2, 3.0, 2.8 (adjusted with citric acid) and hydrochloric acid. The brines were then inoculated with a three-strain mixture of L. monocytogenes (3.5 log CFU/g), or L. monocytogenes (3.5 log CFU/g) and Salmonella Enteritidis (111.6 tcfu/g). Each brine was incubated at 30°C, and 30°C, respectively, for 10 and 20 min. After incubation, the eggs were cooled and subjected to heat-ozone processing. The selected process involved submerging shell-eggs in a 57°C water bath for 28 min, rinsing, and then introducing eggs into a 57°C ozone generator for 10 min.

Results: Inoculated eggs (approximately 10,000 CFU/mL rinse) were treated with the selected process and enumerated using the Mixed Model of SAS and the USDA-Integrated-Predictive-Modeling-Program software.

Significance: These findings will be useful for the USDA-FSIS to develop risk assessments of foodborne pathogens in chicken products.
**P3-240 INACTIVATION OF ACYCLICOLIBUS ACIDOTERRESTRIS SPREES IN DIFFERENT TYPES OF JUICES BY 222-NAVARROMETER KRYPTON-CHLORINE EXCITATION IRRA DIATION AND IDENTIFICATION SPORICIDAL MECHANISM**

Kana Kato, Kiyoko Takahashi, Tatsuhiko Hori, Hiroyuki Sato, and Hiyung Kang

Seoul National University, Seoul, South Korea

Introduction: Acyclobacin is a thermophilic, spore-forming bacterium that causes spoilage of juice products. It is difficult to inactivate A. acidoterestris spores in juice products using thermal pasteurization without quality loss. Therefore, alternative non-thermal techniques are needed to control the remaining spores.

Purpose: The purpose of this study was to examine the efficacy of a 222-nm krypton-chlorine (KrCl) excilamp for inactivation of A. acidoterestris spores in different types of juices and to identify the sporidial mechanisms.

Methods: A. acidoterestris ATCC 49025 spores were inoculated into different types of juices (apple, grape, tomato) and treated with a 222-nm KrCl excilamp ranging from 100 to 1500 mJ/cm². To analyze the factors influencing inactivation of spores, experiments were performed at different value of pH, sugar concentration, and seed medium. To identify the sporidial mechanisms, the fluorescent dye propidium iodide (PI) or dihydro-1-pyrenylphosphine (DPPP) was used to quantitatively assess the change of cell membrane permeability or the incidence of peroxidation, respectively. ATP-LUC/CLD0 were used to measure intrinsic杜绝colored oxygen reactive species (ROS).

Results: As irradiation dose increased, UV irradiation caused significantly (P<0.05) more inactivation of A. acidoterestris spores in juice samples. There were no significant (P>0.05) differences of inactivation levels between the phosphate-buffered saline adjusting different value of pH, sugar concentration. However, it was concluded that UV absorption coefficient and turbidity were inversely proportional to the inactivation levels effect of 222-nm KrCl excilamp. Both the loss of microbial integrity measured by ATP and lipid peroxidation in the cell membrane measured by DPPP increased as irradiation dose increased. Additionally, ROS was generated, but there were no significant (P>0.05) differences.

Significance: These results provide valuable baseline data for inactivation of A. acidoterestris spores by 222-nm KrCl excilamp in the juice industry.

**P3-229 VALIDATION OF THE USE OF ACETIC ACID INCORPORATED WITH CHITOSAN TO PROLONG SHELF LIFE OF GRASS-FEED GRADE BEEF**

Tayab Saleem, Jovana Tran, Stacie Roy, Mario A. Garcia, Stacie Roy, and Donald F. Peters

Mississippi State University, Starks, MS

Undergraduate Student Award Entrant

Introduction: The food industry loses billions daily due to the loss of food products from spoilage organisms and pathogenic bacteria. The industry is looking for natural products that will inhibit microbes and extend the shelf life of meat products.

Purpose: The purpose of this study was to determine a model for improving shelf life of grass-fed grade beef for its market.

Methods: Various concentrations of acetic acid/chitosan mixture (zero, 1.7, 3.3, 5.0, 6.7, and 10%) were added to grass-fed beef patties (30 g) to test the effect of acetic acid/chitosan mixture on S. aureus. Each treatment was tested in triplicate. In the experiment, the ground beef had reached its sell by date. The beef patties were stored at 4°C for 12 and 25 weeks and tested initially (zero, four, eight, and 12) for microbial populations.

Results: In a dose-dependent manner, the growth of background microflora was inhibited by the acetic acid/chitosan mixture. The control had at least 10 7 and 10 6 S. aureus counts per gram of meat. The treated samples reached six log CFU/g. On three repetitions of the experiment, the 3.3 and 6.7% samples had a lower microbial load than the pretreatment value (4.70 log CFU/g) with 4.30 CFU/g, which was significantly lower than the control's value of 5.30 log CFU/g (P<0.05).

Significance: The results show that the use of acetic acid combined with chitosan has the ability to extend shelf life of grass-fed beef without adversely impacting product quality.
P3-244 Evaluation of Fluorescence Spectroscopy as a Tool for Microbial Spoilage Assessment in Fresh-cut Pineapple

Evantia Manthou, Alexandre Liaonou, Panagiotis Tsitsanakis, Evangelos Dagfas, Efstratios Panagou and George-John Nychas

Introduction: pineapple is one of the most popular tropical fruits, commonly found in the market in the form of ready-to-eat (RTE) products. Fresh-cut commodities frequently present a study to be monitored the microbiological quality of fresh-cut RTE pineapple during storage at different temperatures using fluorescence (FLUC) spectroscopy. Methods: Trays of fresh-cut pineapple were stored aerobically under isothermal (four, eight, and 12°C) and dynamic temperature conditions for a maximum of seven days. The initial populations of yeasts, moulds and lactic acid bacteria from the specific spoilage organisms of this food commodity using conventional microbiological approaches, in parallel with FLUC spectroscopy analysis. Data collected were used to estimate the correlation between spectral data and microbial counts. Due to high variability among replicate samples, the average values spectral data (FLUC) and microbial counts of the duplicate samples were used for model development. The data collected at isothermal (117 samples) and dynamic temperature (42 samples) conditions were used for objective of the calibration and validation (prediction) respectively.

Results: The developed PLSR model exhibited an overall satisfactory performance when externally validated. The predicted and actually observed populations of yeasts were well correlated, with the slope parameter of the linear regression and the correlation coefficient being 0.77 and 0.83, respectively. The coefficient of determination (r²) was 0.87, whereas the root mean square error of prediction was estimated to be 0.51 log CFU/g. Significance: FLUC spectroscopy may constitute a suitable analytical technique for the rapid and non-invasive assessment of the microbiological quality of fresh-cut pineapple.

P3-245 Comparison of Six Methods for Quantification of Lactic Acid Bacteria in Spoiled Sliced Turkey

Cheng Zhang, Wendy McMahon and Sandra Kelly-Harris

Introduction: Lactic acid bacteria are known to cause spoilage in many food products. Different quantification methods of lactic acid bacteria are used in food industry, and the recovery can vary in different food matrices. A study was conducted to compare methods for quantifying lactic acid bacteria in ready-to-eat, spoiled sliced turkey.

Purpose: To compare six methods for quantifying lactic acid bacteria in spoiled sliced turkey to optimize recovery and enumeration.

Methods: Six quantification methods for lactic acid bacteria were utilized to analyze twelve turkey samples. The methods used for analysis were MRS with ATP (CMMFE 5TH Edition, Chapter 19.521), Acidified MRS (CMMFE 5TH Edition, Chapter 19.522), ATP with sucrose and 0.26 bromocresol purple (CMMFE 5TH Edition, Chapter 19.528), 3M LAB Petrifilm (ADAC Performance Tested Method, Certificate #047101). 3M APC Petrifilm (AOAC 990.12) and 3M LAB Petrifilm (AOAC 990.12). Each method was analyzed to evaluate immediately packaged samples from spray-washed birds sampled at zero, 70, 135 and 270 days.

Results: The developed PLSR model exhibited an overall satisfactory performance when externally validated. The predicted and actually observed populations of yeasts were well correlated, with the slope parameter of the linear regression and the correlation coefficient being 0.77 and 0.83, respectively. The coefficient of determination (r²) was 0.87, whereas the root mean square error of prediction was estimated to be 0.51 log CFU/g. Significance: FLUC spectroscopy may constitute a suitable analytical technique for the rapid and non-invasive assessment of the microbiological quality of fresh-cut pineapple.

P3-249 Microbial Profile of Subprimes Before and After Water Spray and Dry Chilling of Beef Carcasses Subjected to Hot Water Rinses in Long-term Storage

Diego Casas, Rosine Manishimwe, Mark Miller, Marcos X. Sanchez-Plata

Introduction: As the world meat market moves to never-frozen alternatives, meat processors seek opportunities for increasing the shelf life of fresh meats by combinations of barrier technologies and non-chemical interventions. Purpose: To compare the microbial profile of subprimes before and after water spray and dry chilling combined with hot water carcass treatments on indicator organisms in the long-term shelf life of beef cuts.

Methods: beef carcasses treatments were arranged in a completely randomized design with factorial arrangement on three different factors (feed, hot water wash and chilling method) at two levels (grass vs. grain, washed vs. not washed, dry vs. spray chilled). Samples were taken using E2-Rape with 25 ml buffered peptone water over a 100 cm² area on the stripin. Sample collection was conducted before hot carcasses wash after hot carcass wash, and after 24 hours chilling. CMMFE 5TH Edition were cut into four sections and individually vacuum packaged and sampled at zero, 40, 75 and 135 days (n=20). Aerobic plate counts, Enterobacteriaceae, Escherichia coli, coagulase and pseuodotroph counts were determined.

Results: Not enough evidence (P>0.05) was found indicating the hot water wash intervention reduced bacterial concentration on the carcass surface. significant difference (P<0.05) was obtained between coagulace counts throughout the sampling dates. Feed type did not seem to influence (P>0.05) microbial load of the treatments. Even though no immediate effect is seen from washing at zero, as product aged, we could observe significantly lower (P<0.05) concentration of aerobic and psychrotrophic organisms in dry chilled samples.

Significance: Data collected can be used to select chiling systems to maximize shelf life. Hot water wash prior to carcass chilling may not significantly reduce indicator organisms. Understanding the best parameters for beef carcass processing will allow the beef industry to select optimized chilling processes for long-term storage.

P3-247 Impact of Carcass Spray-Chilling, Dry Chilling and Hot Water Washes on the Shelf Life and Microbial Profiles of Beef Ribeye Rolls

Savannah Forgey, Diego Casas, Rosine Manishimwe, Mark Miller, Mindy Brashears and Marcos X. Sanchez-Plata

Introduction: microbial interventions such as hot water washes and various chiling techniques have been utilized for bacterial reduction on beef carcasses surfaces. Extended aging of spray-chilled beef sub-primal cuts has been shown to reduce shelf life when compared to dry-chilled counterparts.

Purpose: To perform a predictive model to evaluate bacterial indicator loads during extended aging of Australian beef ribeye rolls collected from carcasses subjected to dry and spray-chilling conditions.

Methods: Ribeye rolls of Australia beef carcasses (> 2 area) were sampled with a sterile, prefyledibrated sponge on a 100 cm² area. Four treatments were applied with four replications (n=4). Each treatment was evaluated individually packaged samples obtained from ribeye rolls sampled on 0 days, 35, 45, 55, and 65 of refrigerated storage. The study was conducted in triplicate. Seabaud was used for total plate count. Enterobacteriaceae, coliforms and pseuodotrophs were enumerated based on MRS, Tryptose soy agar and McConkey agar leveling. Colony counts were converted to log CFU/g and statistical analysis was performed using Fisher’s least significance difference test to determine significant differences at P<0.05.

Results: On zero days, no significant differences were observed between treatments and all indicator organisms were below detectable limits. After one week of aging, significant differences were observed between treatments on all indicator organisms. P<0.05. The hot water wash prior to carcass chilling may not significantly reduce indicator organisms. Understanding the best parameters for beef carcass processing will allow the beef industry to select optimized chiling processes for long-term storage.

P3-248 WITHDRAWN

P3-249 Predictive Microbiology Analysis of Dairy Products Stored in Home Refrigerators

J. Antonio Torres, Veronica Rodriguez-Martinez, Daniela Gonzalez de la Garza, Gonzalo Velazquez, Fabian Fagotoni, Reynaldo de la Cruz-Jordan and Carles Ferrer-

Introduction: A three week study was performed to determine if factors in residential refrigerators have any impact on the growth of three different indicator organisms in milk, cheese and yogurt samples. The factors of interest were: temperature, time, placement of sample in refrigerator, number of times the door was opened, and loading (low/high). In the case of milk never removed from the refrigerator, Δlog CFU/g were obtained from milk samples never removed from the refrigerator at Δ days.

Results: The temperature in the refrigerator load (low/high), door (closed/opening cycle), compressor operating mode, temporary room exposure (simulating consumer product removal) and Refridgerator load (low/high) had no significant effect on the microbial load of the treatments. Even though no immediate effect is seen from washing at zero, as product aged, we could observe significantly lower (P<0.05) concentration of aerobic and psychrotrophic organisms in dry chilled samples.

Significance: Data collected can be used to select chiling systems to maximize shelf life. Hot water wash prior to carcass chilling may not significantly reduce indicator organisms. Understanding the best parameters for beef carcass processing will allow the beef industry to select optimized chiling processes for long-term storage.
Bacterial spoilage of shelf-stable tomato products results in product waste and consumer dissatisfaction. Detection methods that rapidly discriminate among the specific spoilage bacteria of concern enable enhanced control over this issue.

Purpose: To develop a technique that can discriminate among spoilage bacteria of concern using mid-infrared spectroscopy.

Methods: Sample collection from four honeydew and four cantaloupe varieties from five different geographical locations. Sample collection included whole melons, skin and mesocarp slices, and environmental samples (air, root, water). Samples were immediately frozen at −20 °C. Each sample type was analyzed in triplicate. Microbial analysis was performed using the Agilent Cary 6000i FT-IR and a Neospectra system (Neospectra, Inc.). Spectra were acquired from 4000 to 400 cm−1, and co-added 16 scans per sample. Reference analysis was performed using bacterial plates and E. coli O157:H7 ATCC 43889, P. aeruginosa ATCC 9027, S. aureus ATCC 25923, and S. typhimurium ATCC 14028. Selected reference cultures were grown on TSA media. Optical quality was ensured by the addition of standard reference cultures into the samples. All samples were analyzed in the appropriate growth condition (pH 6.6, temperature 30°C, nutrient availability). Measurements were conducted in a walk-in environmental chamber (Goldencube, CA) set to the appropriate conditions (pH 5.0, 5.5, 6.0, 6.5, 7.0). A total of 174 spectra were collected from 41 samples, including 13 melons, 13 environmental air, root, water samples, and 15 environmental soil samples.

Results: The average root mean square error of calibration (RMSEC) and root mean square error of cross-validation (RMSECV) for triplicate melon samples were 0.02 and 0.04, respectively, indicating a high level of spectral similarity among melons. In contrast, the average RMSEC and RMSECV for melon slices were 0.13 and 0.26, respectively, indicating a higher level of spectral diversity between melon slices. The average RMSEC and RMSECV for environmental samples were 0.03 and 0.07, respectively, indicating a high level of spectral similarity among environmental samples.

Conclusions: Spectroscopic analysis can discriminate among different bacterial species, which can indicate the presence of spoilage bacteria. This method enables real-time monitoring of spoilage bacteria, allowing for rapid intervention to prevent waste and dissatisfaction. Further investigation is needed to determine the optimal growth conditions for each bacterial species and to validate the method for real-world applications.

P3-253 A Machine Learning Approach to Analyze Micro-Isothermal Calorimetry as a Function of Microbial Growth in Fresh and Processed Foods

Ihab El Adawy, Guangyao Li, and Fred Chicheley
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Introduction: Calorimetry is a useful tool to assess the shelf life stability of foods rapidly. The technique involves the measurement of heat generated in a food system as a result of chemical and biological changes captured in the form of heat flow (μW) and heat energy (µJ). This approach offers a reliable and rapid method of assessment of viable counts as an alternative to traditional plate count. The heat data is modeled and interpreted using exponential models (Q = Q0 .eµt). However, the models are only applicable to the initial vertical portion of the curve over a short period. The remainder of the data (part of the heat flow curve) does not add any useful information to the models.

Purpose: This work reports the application machine learning theory as an alternative method to interpret micro-isothermal calorimetry data in a super- vised learning environment.

Methods: A total number of 194 data sets (heat data, biomass, and CFU) were generated to build a training database (n=60). Features used were spectra (specific growth rate, peak time, maximum heat) were extracted from the data, followed by statistical learning procedure of relationship present. The micro-organism type and composition of the food were used as input parameter using the Neospectra system in growth media.

Results: The Support Vector Regression (SVR) model learned specific features (μW) >0.05 and texture (R² >0.7) which was found to be the most suitable in terms of best fit and lower cost of calibration. A cross-validation procedure of the developed model model yielded R² values (>0.7) with RMSE (±0.03) for all the organisms confirming the validity of the SVR technique. Model was applied to RTS foods including chicken soup, sauces, cake, seafood, caffeine, syrups and beans showing significant results (P<0.05).

Significance: The ML approach offers a reliable new way to determine product shelf life with less need for routine microbial enumeration.

P3-254 Extraction and Characterization of Extracellular Polymeric Substances (EPS) of E. coli O157:H7 ATCC 43388 and Listeria monocytogenes ATCC 7464 Molecular Biopolymers Grown under Different Growth Conditions Stanislav Králová
Aarhus University, Denmark

Introduction: Listeria monocytogenes and Escherichia coli can produce biopolymers as survival mechanism in food processing environments. The biocomplex contains aggregates of polymers which provide numerous benefits to cells inside the biocomplex, including protection, penetration, osmotic stress, and pathogenesis.

Purpose: To extract and characterize the extracellular polymeric substances (EPS) produced by E. coli O157:H7 ATCC 43388 and L. monocytogenes ATCC 7464 at different food related environmental conditions.

Methods: Various spectroscopic, chromatographic and microscopic techniques were used to decode the composition of the EPS harvest from different growth conditions of pH, temperature, and nutrient availability.

Results: Colorimetric analysis was used to determine the biochemical composition of the EPS biocomplex. Polymeric Bradford Protein Assay and polysaccharides (Phenol-Sulfuric Acid method). Comparison varied with different growth conditions and culture type. Fourier Transformed Infrared (FT-IR) spectra confirmed biopolymer type and FT-IR spectra confirmed the presence of proteins. The biocomplex has a high content of polymeric carbon and proteins of primary amino and aromatic compounds. Selective enzymatic treatment with Pronase, Cellulase and Pectinase revealed biofilm matrix content to the EPS.

Conclusions: The composition and structure of molecular biopolymers were significantly affected by growth conditions, which is of interest for food safety. Further investigation is needed to understand the role of molecular biopolymers in food spoilage and transmission of diseases. Understanding the composition of the biocomplex is important in initiating prevention, removal, and inactivation measures of controlling biopolymers.

P3-255 Antibacterial Properties of High Voltage Cold Atmospheric Plasma and Its Effect on Quality of Asian Sea Bass Slices

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Developing Scientist Entrant

Introduction: Plasma generated energy is utilized for the application of a neutralization system that has promising antibacterial properties. However, measurements (Western Immunoblotting (WIG), treatment (PT)) is factors determining the antibacterial efficacy of high voltage cold atmospheric pressure (HVCP). Thus, these factors should be considered and optimized.

Purpose: The effect of WIG, TT and PTT on the bacterial inhibition efficacy of in-package deleterious barrier HVCP against both spoilage and pathogenic microorganisms was investigated. The impact of HVCP on quality of Asian sea bass slices was also monitored.

Methods: Permethrin (100 µM, 1000 µM, 10000 µM) were treated with HVCP for 30 min at 12 VDC. WIG, TT and PTT influenced the bacteria inhibition of HVCP (±0.05). HVCP treated ASBS had a lower microbial load as compared to the control. The efficacy was generally increased with increasing TT. Lpid oxidation was more pronounced with coincidental decrease in unsaturated fatty acids in the slices treated with HVCP. Mysum heavy chain and actin band intensity were also decreased in Asian sea bass slices treated with HVCP for more than 30 min.

Significance: HVCP had an excellent bacterial inhibition efficiency. HVCP under appropriate conditions could reduce the microbial load of Asian sea bass slices, thus extending its shelf life and assuring safety.

Poster}

Journal of Food Protection Supplement
Author and Presenter Index

Abbas, Nasser, University of Sadat City (P1-53)
Abbott, Amanda, Delaware State University (T9-10)
Abd, Shirin, Eurofins (P1-107*)
Abdelhamid, Ahmed, The Ohio State University (P2-57)
Abdelmajid, Naser, U.S. Department of Agriculture – FSIS (P3-224, T4-12)
Abdul, Roshan Aara, Center for Applied Food Security and Biotechnology (CASFab), Central University of Technology (P3-237)
Abe, Hiroki, Hokkaido University (P1-154, T10-09*)
Abed, sawsan, University of Florida (T5-07)
Abley, Melanie, U.S. Department of Agriculture – FSIS (RT7*, RT19*)
Abnavi, Mohammadreza, Cleveland State University (T3-09*)
Aboubakr, Hamada, University of Minnesota (T6-01*)
Abraham, Ann, U.S. Food and Drug Administration, Gulf Coast Seafood Laboratory (T9-11)
Achar, Premila, Kennesaw State University (P1-219)
Achek, Rachid, High National Veterinary School (P1-147, P1-277, P1-270)
Acheson, David, The Acheson Group (RT2*)
Ackerman, Luke, U.S. Food and Drug Administration (S53*)
Acuff, Jennifer, Virginia Tech (P1-17*, P1-26)
Adams, Carly-Rain, University of Nebraska-Lincoln (P3-232)
Adator, Emelia, University of Manitoba (P1-179*)
Adegunwa, Mojisola, Federal University of Agriculture (P3-156*)
Adell, Aiko, Universidad Andres Bello (P2-75, P3-155*)
Adeniyi, Ayodeji, Texas Tech University (P3-95*, P3-198)
Adetunji, Victoria, University of Ibadan (P1-274)
Adeyemi, Damilare, Kyungpook National University (P2-68)
Adeyemo, Ismail Adewuya, University of Ibadan (P1-52, P2-20)
Adeyemo, olanike, University of Ibadan (P2-97)
Adhikari, Achyut, Louisiana State University AgCenter (P1-215, P2-166, P2-173, P2-210*, P2-157)
Adhikari, Jayashan, Tennessee State University (P2-60, P1-99*)
Adhikari, Koushik, The University of Georgia (P2-10, P2-112)
Aditya, Anand, University of Nebraska-Lincoln (P3-231)
Aditya, Arpita, University of Maryland (P2-06*)
Adler, jeremy, Ecolab Inc. (T9-05)
Adiziet, frederick, University for Development Studies (P2-78)
Aertszen, abram, K.U. Leuven (T6-08)
Agarwal, Sharif, University of Arizona (P2-112)
Aghaye, Oluwaseun, U.S. Food and Drug Administration (P3-80*, P3-79*)
Agholosu, Anthony Amison, University for Development Studies (P2-78)
Agga, Getahun, U.S. Department of Agriculture (P2-249, P2-21)
Aggarwal, Alisha, Illinois Institute of Technology (P1-207)
Agin, James, Q Laboratories, Inc. (P3-21, P3-22)
Aguayo-Acosta, alberto, Departamento de Microbiología e Inmunología, Facultad de Ciencias Biológicas, Universidad Autónoma de Nuevo León (P1-05*)
Aguilas, viviana, Institute for Food Safety and Health (P3-84)
Aguilar borba, monique, Tree Fruit Research Commission (P2-215)
Ahmad, imran, Florida International University (P3-253*)
Ahmad, nurul ha wa, Michigan State University (P1-07*)
Ahmed, Asfhaque, U.S. Food and Drug Administration (P2-109)
Ahn, Seolhee, Changwon National University (P2-129)
Ahn, Soohyun, University of Florida (P3-120, T5-07*)
Ai, Yuehan, The Ohio State University (P1-218*)
Airikkka, suvi, thermo Fisher Scientific (P3-50)
Ajayi, Feyisola, Federal University Gashua, Nigeria (P3-248*)
Akanni, Gabriel, University of Pretoria (P1-125*)
Akassou, mouina, FoodChek Laboratories Inc. (P3-72)
Akinleye, Tunde, Consumer Reports (S28*)
Akins-Lewenthal, Deann, Conagra Brands (P1-248, P1-143)
Akintola, ruth, National Veterinary Research Institute (P1-48)
Akter, sharmin, Jessore University of Science and Technology (T9-08)
Al-Mosawi, ahmed, thermo Fisher Scientific (P3-50)
Alaa El Din, hadeer, University of Sadat City (P1-53)
Alamu, emmanuel, International Institute of Tropical Agriculture (P3-156)
Alarape, selim, University of Ibadan (P2-97*)
Alavi, Amir, U.S. Food and Drug Administration (P2-109)
Alberti, Enrica, IITA Corporation (T8-05)
Aldrich, charles, Kansas State University (P2-13, T4-07)
Alhaji, nma, niger state Ministry of Livestock and Fisheries (P1-274)
Ali, laila, U.S. Food and Drug Administration – CFSA (P3-99)
Aljasir, Sulaiman, University of Connecticut (P2-246*)
Allard, marc, U.S. Food and Drug Administration – CFSA (S72*, P3-165, P1-180*, P3-166, P2-227)
Allard, Sarah, Maryland Institute for Applied Environmental Health, University of Maryland (P1-258, P2-160*, P2-89, P2-95, T1-04, P3-167, P3-168, T1-02)
Allebach, Jan, Purdue University (P3-93)
Allen, Jennifer, Oregon State University (P2-238)
Allende, Ana, CEBAS-CSIC (P2-159, S12*)
Alles, Susan, Neogen Corporation (P3-20)
Allison, abimbola, Tennessee State University (P1-255, P1-96*, P1-99, P1-95*)
Alred, Adam, Clear Labs (P3-87)
Allué guardia, anna, South Texas Center for Emerging Infectious Diseases (STCEID), University of Texas at San Antonio (P1-53)
Almeida, adelaida, University of Aveiro (P3-173)
Almeida, Danielle, 3M (P3-66, P3-57)
Almeida, Giselle, University of Arkansas (P1-202)
Almuhaidib, esam, University of Maryland Eastern Shore (T9-10)
Alonso, silvia, International Livestock Research Institute (P2-249)
Alshaibani, Dhafer, University of Maine (P2-247*)
Alvarado martinez, zubi, University of Maryland (P2-06)
Alvarado-Martinez, Zubi, University of Maryland (P2-52*)
Alvarenga, Veronica Ortiz, Federal University of Minas Gerais (P1-100, P3-151)
Alwan, nsreen, modern University for business & Science (P1-66, P1-76)
Alzahrani, Abdulhakeem, University of Guelph (P2-191)
Amalaradhouj, Mary Anne, University of Connecticut (P2-183)
Amanuma, Hiroshi, National Institute of Health Sciences (P1-273, P1-272)
Ameen, Saliu, University of Nigeria, Nsukka (P1-273, P1-272)
Ameen, Saliu, University of Nigeria, Nsukka (P1-273, P1-272)
Ameen, Saliu, University of Nigeria, Nsukka (P1-273, P1-272)
Ameen, Saliu, University of Nigeria, Nsukka (P1-273, P1-272)
Award Competitors

Tabashsum, Zajeba, University of Maryland (T4-02)
Takeoka, Kohei, Hokkaido University (P1-154)
Thomas, Merlyn, University of Georgia (P1-153)
Thomas-Popo, Emalie, Iowa State University (P1-102)
Tikekar, Rohan, University of Maryland (P1-206)
Trudelle, Danielle, The University of Tennessee (P2-65)
Vega, Daniel, Kansas State University (P1-203)
Vengarai Jagannahad, Badrinath, University of Kentucky (P2-64)
Verma, Tushar, University of Nebraska-Lincoln (P1-15)
Wadhawan, Kirty, University of Wisconsin-Madison, Department of Pathobiological Sciences (P2-254)
Wang, Hongye, Clemson University (P1-266)
Wang, Kaidi, Food, Nutrition and Health Program, Faculty of Land and Food Systems, The University of British Columbia (P1-236)
Wang, Peien, Department of Food Science and Technology, The University of Georgia (P2-214)
Wang, Qingyang, University of Maryland (P1-108)
Wang, Wenqian, University of Arkansas, Department of Poultry Science (T5-09)
Weeraratne, Pahasara, Oklahoma State University (P2-67)
Wei, Xinyao, University of Nebraska-Lincoln (P1-22)

West, Molly, The University of Tennessee (T1-05)
White, Chanelle, University of Maryland Eastern Shore (P3-168)
Wong, Catherine, Food, Nutrition and Health, University of British Columbia (P2-176)
Wu, Blyu, University of Hawaii at Manoa (P3-97, P3-96)
Wu, Sophia Tongyu, Purdue University (P1-126)
Wu, Xueyang, University of Guelph (P2-191)
Xu, jie, Washington State University (P1-202)
Xu, Yumin, The Ohio State University (P3-234)
Yan, Jia, University of California, Davis, Food Science and Technology Dept., (P3-153)
Yan, Runan, The Pennsylvania State University (P1-199)
Yang, Ren, Washington State University (T11-02, T11-01)
Yao, Shiyun, University of Delaware (T3-06)
Yeom, Wooric, Department of Biotechnology, College of Life Sciences and Biotechnology, Korea University (P2-116)
Yi, Jiyeon, University of California-Davis (T2-03)
Zhao, Luyao, Department of Food Science and Technology, The University of Georgia (P2-240)
Zhong, Zeyan, McGill University (T6-05)

Undergraduate Student Award Competitors

Arbon, Jeremy, Brigham Young University (P2-245)
Byun, Suyeun, U.S. Department of Agriculture (P2-194)
Chen, Han, Purdue University (P1-90)
Cobar, Joshua, Louisiana State University (P3-177)
Craig, Jackson, University of Tennessee (P2-126)
Gomez, Carly, Michigan State University (P1-145)
Hodges, Jack, University of Houston (P2-135)
Johnson, Erica, University of West Alabama (P2-40)
Kelly, Alyssa, University of Delaware (P2-204)
Ladner, Taylor, Mississippi State University (P2-46)
Patregnani, Emma, U.S. Food and Drug Administration – CFSAN, Office of Applied Research and Safety Assessment (P3-100)
Walker, Kayla, University of West Alabama (P1-262)