

## IAFP'S EUROPEAN SYMPOSIUM ON FOOD SAFETY

## PROGRAMME

Held at La Cité des Congrès de Nantes

ORGANIZED BY



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## IAFP EUROPEAN SYMPOSIUM ON FOOD SAFETY PROGRAMME AT-A-GLANCE

	Wednesday, 24 April 2019						
Wednesday		Opening	Session				
8.30 - 10.00	Auditorium 450						
Wednesday							
10.00 - 10.30 Room	Auditorium 450	Room 200	it Hall Room GH	Room I	Poster Session 1 –		
Wednesday 10.30 - 12.00	S1 – CulturOmics: The Revival of Microbiological Culture!!!	S2 – Microbiological Hygiene and Food Safety in Primary Production and Processing of Fresh Produce – from Science to Easily Understandable Recommendations for Farmers and Suppliers	S3 – The Survival and Control of Foodborne Pathogens in Low-moisture Foods	Technical Session 1 – Meat and Poultry, Antimicrobials and Microbial Food Spoilage	Applied Laboratory Methods; Communication Outreach and Education;		
Wednesday	Lunch Exhibit Hall				Epidemiology; General Microbiology;		
12.00 - 13.30 Wednesday 13.30 - 15.00	S4 – Network Analysis to Better Decipher Functions and Dynamics of Food Microbial Ecosystems	S5 – Novel Modelling Approaches of Microbiological Spoilage in Food	S6 – Clarity through Chaos: International Perspectives on Food Safety after Recent High Profile Foodborne Outbreaks	Technical Session 2 – Applied Laboratory Methods	Microbial Food Spoilage; Novel Laboratory Methods; Produce; Risk Assessment		
Wednesday 15.00 - 15.30		Coffee/Networking Break Exhibit Hall					
Wednesday 15.30 - 17.00	RT1 – Environmental Monitoring – Friend or Foe?	S7 – Getting Ahead of Food Fraud	S8 – Challenges in Campylobacter Detection and Accurate Quantification	Technical Session 3 – Novel Laboratory Methods, Sanitation and Seafood			
Wednesday 17.00 - 18.00		Exhibit Hal	l Reception				
17.00 - 18.00		Thursday	, 25 April 2019				
Room	Room 200	Club Atlantique	Room GH	Room I			
Thursday 8.30 - 10.00	S9 – Close-up of Consumer Kitchen Practices – Can Socio(microbio)logy Aid Food Safety at Home?	S10 – Food Safety Emerging Risk Identification with Novel Computational Methods	S11 – Fast MALDI Typing to Drive Decision Making and Source Tracking	Technical Session 4 – Communication and Outreach, Food Toxicology, Pathogens and Risk Assessment			
Thursday 10.00 - 10.30							
Thursday 10.30 - 12.00	S12 – Actions Speak Louder Than Words: Ongoing Efforts for Global Harmonization in Standardization in Food Microbiology	S13 – Food Safety Culture: The Proof is in the Science	S14 – Applications of Microbial Profiling: The Present and the Future	Technical Session 5 – Dairy and Other Food Commodities and Pathogens	Poster Session 2 – Antimicrobials; Beverages and Water; Dairy and Other Food		
Thursday 12.00 - 13.30	Lunch Foc						
Thursday 13.30 - 15.00	RT2 – The Use of Chemicals in Food Hygiene and Linkage to Microbial Resistance	S15 – Hepatitis E Virus, an Emergent Foodborne Pathogen? Public Health Implications	S16 – Challenge Testing for Listeria monocytogenes : Requirements, Needs, Difficulties and Developments	Technical Session 6 – Pathogens, Epidemiology and General Microbiology	Meat and Poultry; Pathogens; Sanitation; Seafood		
Thursday 15.00 - 15.30							
Thursday 15.30 - 17.00	RT3 – Foodborne Viruses: Detection, Risk Assessment, and Control Options in Food Processing	S17 – Water Re-Use in Food Processing Industry – It's Inevitable!	S18 – Fungal Spores in Food; Implication of Natural Heterogeneity on Food Quality	Technical Session 7 – Pathogens			
Room	Room 200	Friday, 2 Club Atlantique	26 April 2019 Room GH	Room I			
Friday 8.30 - 10.00	S19 – Ongoing Research Activities in Risk-Benefit Assessment of Food	S20 – Norovirus, Glycans and Oysters: The Perfect Association?	S21 – Beef Decontamination Treatments in Slaughter Plants: Do They Improve Product Safety?	Technical Session 8 – Risk Assessment			
Friday 10.00 - 10.30							
Friday 10.30 - 12.00	S22 – How Has Metagenomics Been Useful to Food Safety Research and What Does Its Application to Public Health Hold?	S23 – Campylobacter, Health Impact, Performance Objectives and Effectiveness of Sampling Plans	S24 – Insects in Poultry Feed: Regulatory Framework, Poultry Gut Microbiota and Consumer Acceptability	Technical Session 9 – Applied Laboratory Methods, Microbial Food Spoilage and Pathogens			
Friday 12.15 - 13.45	Closing Session Room 200						
Friday 13.45 - 14.30	Farewell Refreshments						



### **China International Food Safety & Quality Conference**

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# PROGRAMME

24–26 April 2019 – Nantes, France

### Wednesday, 24 April – 8.30–10.00



# WELCOME TO THE IAFP EUROPEAN SYMPOSIUM

## 7.30 – 17.00 Registration Open

- 7.30 8.30 Morning Coffee
- 10.00 18.00 Exhibit Hours

#### OS Opening Session Auditorium 450 Chairs: Daniele Sohier and Helen Taylor

- 8.30 Introduction to IAFP DAVID THARP, Executive Director, International Association for Food Protection, Des Moines, IA, USA
- 8.40 Introduction to IAFP's European Symposium TIMOTHY JACKSON, Driscoll's, Watsonville, CA, US
- 8.50 Programme Notes and Recognition of Organising Committee DANIELE SOHIER, Bruker, Bremen, Germany

- 9.00 Food Safety Risk Assessment Policies and Procedures at the U.S. Food and Drug Administration (FDA), European Food Safety Authority (EFSA) and French Agency for Food, Environmental and Occupational Health & Safety (ANSES) MOEZ SANAA, ANSES, Maisons, France
- 9.30 Update on Monitoring Activities of European Union Reference Laboratory for *Listeria monocytogenes* Multi-EU Country Outbreak Investigation and Integration in the EU/EEA-wide Monitoring Organization BENJAMIN FÉLIX, ANSES, Laboratory for Food Safety, University of Paris-Est, Maisons-Alfort, France
- 10.00 Networking Coffee Break in the Exhibit Area

## Wednesday, 24 April – 10.30–12.00

#### Poster Session 1 – Applied Laboratory Methods; Communication Outreach and Education; Epidemiology; General Microbiology; Microbial Food Spoilage; Novel Laboratory Methods; Produce; Risk Assessment

Authors present during scheduled breaks

- S1 CulturOmics: The Revival of Microbiological Culture Auditorium 450 Organizer: Patrice Arbault Convenors: Patrice Arbault and Daniele Sohier Sponsored by the IAFP Foundation
- 10.30 Analysis and Interpretation of NGS Data Sets for CulturOmics HENK DEN BAKKER, Center for Food Safety, University of Georgia, Griffin, GA, USA
- 11.00 CulturOmics: The Revival of Microbiological Culture FLORENCE POSTOLLEC, ADRIA – UMT ACTIA19.03 ALTER'IX, Quimper, France and NOÉMIE LUBEM, UBO UNIVERSITY – UMT14. 01SPORE RISK, Quimper, France
- 11.30 How Can CulturOmics Complement Meta-genomics to Investigate the Microbial Biodiversity in Food Microbiology? LUCA COCOLIN, University of Torino-DISAFA, Grugliasco, Italy

#### 12.00 Lunch Available in the Exhibit Area

S2 Microbiological Hygiene and Food Safety in Primary Production and Processing of Fresh Produce – From Science to Easily Understandable Recommendations for Farmers and Suppliers Room 200

#### Organizers: Matteo Campagnoli and Sophie Zuber Convenor: Liesbeth Jacxsens

- 10.30 Regulatory Perspectives of Hygiene in Fresh Produce Production: Challenge of Microbiological Quality of Water KRIS DE SMET, European Commission, Brussels, Belgium
- 11.00 Microbial Risk Mitigation in Primary Production of Produce: From Scientific Knowledge to Implementation of an "Assessment–Education–Continuous Improvement" Process FRANÇOISE JULIEN-JAVAUX, Nestlé Research, Lausanne, Switzerland
- 11.30 Effect of Disinfectants on Preventing the Cross-contamination of Pathogens in Fresh Produce Washing Water and Establishment of Operating Standards Thereof IMCA SAMPERS, Ghent University, Ghent, Belgium

#### 12.00 Lunch Available in the Exhibit Area

S3 The Survival and Control of Foodborne Pathogens in Low-moisture Foods Room GH

**Organizers and Convenors: Jeffrey Farber, Linda J. Harris and Anett Winkler** *Sponsored by the IAFP Foundation* 

- 10.30 Survival and Potential for Pathogenicity Changes for Listeria monocytogenes in Low-moisture Foods JEFFREY FARBER, University of Guelph, CRIFS, Department of Food Science, Guelph, ON, Canada
- 11.00 Survival of Enterohemmorhagic *E. coli* in Low-moisture Foods LINDA J. HARRIS, University of California-Davis, Depart-
- ment of Food Science and Technology, Davis, CA, USA11.30 The Control of Foodborne Pathogens in Low-moisture
- Food Facilities PETER MCCLURE, Mondelēz International, Birmingham, UK

#### 12.00 Lunch Available in the Exhibit Area

#### T1 Technical Session 1 – Meat and Poultry, Antimicrobials and Microbial Food Spoilage Room I

#### Convenor: Lisa O'Connor

- T1-01 RNA-based Surveillance of Meat Processing Environ-
- 10.30 Revealed Selective Pressures of Gaseous Özone on Abattoir Microbiota CRISTIAN BOTTA, Ilario Ferrocino, Maria Chiara Cavallero, Simonetta Riva, Luca Cocolin, Kalliopi Rantsiou, University of Torino-DISAFA, Grugliasco, Italy
- T1-02 Litter Treatment as a Control Strategy Against Campylo-
- 10.45 bacter in Broilers: Impact on Caecal Counts and Microbiota Composition AMANDINE THEPAULT, Xavier Roulleau, Typhaine Poezevara, Pauline Loiseau, Ségolène Quesne, Florent Souchaud, Marianne Chemaly, Muriel Guyard-Nicodème, ANSES, Ploufragan, France
- T1-03\* Mathematical Modelling of *Listeria monocytogenes* Survival
- 11.00 during Low-temperature (sous-vide) Cooking of Meat Products MARTA CLEMENTE-CARAZO, Alberto Garre, Pablo S, Fernandez, Jose Lucas Peñalver, Soto, Paula

Pablo S. Fernandez, Jose Lucas Peñalver-Soto, Paula M. Periago, Arantxa Aznar, Arturo Esnoz, Jose A. Egea, Alfredo Palop, Universidad Politecnica de Cartagena, Cartagena, Spain

- T1-04 Multi-drug Resistance Spread Among Yersinia
- 11.15 enterocolitica Isolates from Swine and Pork Production Obtained in Brazil
   Bruna Torres Furtado Martins, Juliana Libero Grossi, Ricardo Seiti Yamatogi, LUÍS AUGUSTO NERO, Universidade Federal de Viçosa, Viçosa, Brazil
- **T1-05\*** The Antibiotic Resistome of Farmed Rainbow Trouts
- 11.30 Filets Using Smartchip Real-time PCR NICOLAS HELSENS, Ségolène Calvez, Agnes Bouju-Albert, Albert Rossero, Hervé Prévost, Catherine Magras, SECALIM, INRA, Oniris, Université Bretagne Loire, Nantes, France
- T1-06\*Residential Surface Bacteria Mapping in a Cold smoked11.45Salmon Processing Environment<br/>AURELIEN MAILLET, Agnes Bouju, Steven Roblin,<br/>Pauline Vaissié, Sebastien Leuillet, Xavier Dousset,<br/>Emmanuel Jaffrès, Jerome Combrisson, Herve Prévost,<br/>UMR 1014 SECALIM, UBL, INRA, Oniris, Nantes, France

#### 12.00 Lunch Available in the Exhibit Area

#### S4 Network Analysis to Better Decipher Functions and Dynamics of Food Microbial Ecosystems

#### Auditorium 450

#### **Organizer and Convenor: Sandrine Guillou** Sponsored by the IAFP Foundation

- 13.30 Microbial Network Inference through Time and Space: Tools and Network Analysis LISA RÖTTJERS, KU Leuven, Leuven, Belgium
- 14.00 Systems Biology Protocol to Investigate Microbial Network: From Co-occurrences to Phenotypic Landscapes DAMIEN EVEILLARD, Computational Biology Group LS2N UMR 6004 CNRS, University of Nantes, Nantes, France
- 14.30 Shining Light on Networks in Food Microbiomes: Food Microbionet and the ShinyFMBN App EUGENIO PARENTE, Università degli Studi della Basilicata, Potenza, Italy

#### 15.00 Networking Coffee Break in the Exhibit Area

S5 Novel Modelling Approaches of Microbiological Spoilage in Food Room 200 Organizers: Louis Coroller

and Sandrine Guillou Convenor: Jeanne-Marie Membre Sponsored by FWO (Research Foundation – Flanders) and the IAFP Foundation

- 13.30 Prediction of Spoilage Occurrence in French Fresh Poultry Sausages: Potential Role of Potassium Lactate and Modified Atmosphere Packaging NGOC-DU LUONG, SECALIM, INRA, Oniris, Université Bretagne Loire, Nantes, France
- 14.00 Identification of Volatile Food Spoilage Indicators by Multi-variate Statistical Analysis
   LOTTA KUULIALA, Research Unit Food Microbiology and Food Preservation & Research Unit Knowledge-based
   Systems; Faculty of Bioscience Engineering; Ghent University, Ghent, Belgium
- 14.30 SorfML: The Machine Learning Web Platform for Food Microbiological Quality FADY MOHAREB, School of Water, Energy & Environment Cranfield University, Cranfield, UK

#### 15.00 Networking Coffee Break in the Exhibit Area

- S6 Clarity through Chaos: International Perspectives on Food Safety after Recent High Profile Foodborne Outbreaks Room GH
   Organizers: Kalmia Kniel, Lise Korsten and Manan Sharma Convenors: Shirley A. Micallef and Manan Sharma
- 13.30 Parasites, Viruses, and Produce from the United States: Can Anything Change? KALMIA KNIEL, University of Delaware, Newark, DE, USA

- 14.00 Frozen Vegetables Outbreak: What Have We Learnt? ANA ALLENDE, CEBAS-CSIC, Murcia, Spain
- 14.30 A Year after Polony: Emerging Food Safety Risks in a Highly Diversified Food System LISE KORSTEN, DST-NRF Centre of Excellence in Food Security, University of Pretoria, Pretoria, South Africa
- 15.00 Networking Coffee Break in the Exhibit Area

#### T2 Technical Session 2 – Applied Laboratory Methods Room I

#### Convenor: Luca Cocolin

- T2-01 Salmonella Serotyping; Comparison of the Traditional
- 13.30 Method to a Microarray-based Method and an *in silico* Platform Using Whole Genome Sequencing Data BENJAMIN DIEP, Caroline Barretto, Coralie Fournier, Aneta Karczmarek, Guido Voets, Xiangyu Deng, Adrianne Klijn, Nestlé Research, Lausanne, Switzerland
- **T2-02** Optimized Methodology for Same-day Detection of *E. coli*
- 13.45 O157 By Multiplex qPCR in Ground Meat and Leafy Greens ALEJANDRO GARRIDO-MAESTU, Sarah Azinheiro, Joana Carvalho, Pablo Fuciños, Marta Prado, International Iberian Nanotechnology Laboratory, Braga, Portugal
- T2-03\* Assessment of Bacterial Indicators on Poultry
- 14.00 Carcasses by Culture Combined MALDI-TOF MS Identification and 16s rRNA Amplicon Sequencing ZHONGJIA YU, Kurt Houf, Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium
- **T2-04** The Effect of Spices on the Growth and Dectection of
- 14.15 Salmonella and Shiga Toxin-producing Escherichia coli in a Primary Enrichment GREETJE CASTELIJN, Lotte de Lange, Menno van der Voort, The Netherlands Food and Consumer Product Safety Authority (NVWA) Laboratory Feed, Food & Consumer Product Safety, Wageningen, The Netherlands
- T2-05 A DNA Tagging Method for Improved Traceability and
- 14.30 Prevention of Fraud in the Food Supply ANTONIOS ZOGRAFOS, Quin Chou, Christopher McCormick, Lucia Cerillo, Adam Idoine, Johnathan Burleson, Laurie Clotilde, SafeTraces, Pleasanton, CA, USA
- T2-06 ISO 16140-2 (2016) Validation of Genedisc® for the
- 14.45 Detection of Shiga Toxin-producing *Escherichia coli* (STEC) from O157, O111, O26, O103 and O145 Groups Justine Baguet, Cécile Bernez, NICOLAS NGUYEN VAN LONG, Christophe Quere, Maryse Rannou, ADRIA Food Technology Institute, Quimper, France

#### 15.00 Networking Coffee Break in the Exhibit Area

## Wednesday, 24 April – 15.30–17.00

#### S7 Getting Ahead of Food Fraud Room 200 Organizers: Jesse Miller and Thomas Spengler

**Convenors: Jerome Combrission and Thomas Spengler** *Sponsored by the IAFP Foundation* 

- 15.30 The Magnitude and Impact of Honey Fraud NORBERTO GARCÍA, Apimondia, President of the Scientific Commission Beekeeping Economy and Chairman of the Working Group Adulteration of Bee Products, Buenos Aires, Argentina
- 16.00 Development of a Practical Procedure Suitable to Determine the Geographical Origin and Authenticity of Spices with 1H-NMR-Analytic (HAGen) WOLFRAM WENDLER, Arotop Food & Environment GmbH, Mainz, Germany
- 16.30 1H-NMR-Profiling as an Efficient Screening Analysis for Food Fraud Detection in Fruit Juices PETER RINKE, SGF International e.V., Head of R&D, Nieder-Olm, Germany

#### 17.00 Reception in the Exhibit Area

- S8 Challenges in Campylobacter Detection and Accurate Quantification Room GH
   Organizers and Convenors: Heidy Den Besten and Nabila Haddad Sponsored by the IAFP Foundation
- 15.30 Challenges of *Campylobacter* Detection; Effect of Strain Variability and Competitive Flora on Enrichment-based Detection Procedures MAREN LANZL, Wageningen University, Wageningen, The Netherlands
- 16.00 The Survival of *Campylobacter* in the Food Chain, or How to Enhance the Robustness of the Model Prediction Using Molecular Markers BENJAMIN DUQUÉ, UMR1014 SECALIM, INRA, Oniris, Nantes, France
- 16.30 *Campylobacter* Contamination Levels in Poultry Meat in the EU and the Efficacy of Control Measures PIETRO STELLA, European Food Safety Authority (EFSA), Parma, Italy

#### 17.00 Reception in the Exhibit Area

#### **RT1 Environmental Monitoring – Friend or Foe?** *Auditorium 450*

#### **Organizer and Convenor: Anett Winkler**

#### 15.30 Panelists:

ROY BETTS, Campden BRI, Chipping Campden, UK

FRANCOIS BOURDICHON, International Dairy Federation – Standing Committee on Microbiological Hygiene, Brussels, Belgium

JOHN HOLAH, UK:IE EHEDG & Holchem Laboratories Ltd., Bury, UK

DIRK NIKOLEISKI, Senior Food Safety Specialist, Commercial Food Sanitation, Munich, Germany

ANETT WINKLER, Cargill, Munich, Germany

#### 17.00 Reception in the Exhibit Area

#### T3 Technical Session 3 – Novel Laboratory Methods, Sanitation and Seafood Room I

#### Convenor: George-John Nychas

- T3-01\* Comparative Genomics of Persistent Listeria monocyto-
- 15.30 *genes* Clonal Complexes Suggests Genetic Exchanges within and between Seafood Processing Plants of Readyto-Eat Food Products in France FEDERICA PALMA, Thomas Brauge, Nicolas Radomski, Laurent Guillier, Graziella Midelet-Bourdin, ANSES, Laboratory for Food Safety, University of Paris-Est, Maisons-Alfort, France
- **T3-02** AquaSpark<sup>™</sup> A Novel Chemiluminescent Technology
- 15.45 Platform for Onsite Pathogen Diagnostics MARIO HUPFELD, Nadine Heinrich, Lukas Reinau, Julian Ihssen, Lars Fieseler, Nemis Technologies, Zürich, Dübendorf, Switzerland
- **T3-03** Development of a Real-time Cell Analysis (RTCA)
- 16.00 Method as a Fast and Accurate Method for Detecting Infectious Particles of Hepatitis A Virus Samuel Lebourgeois, Audrey Fraisse, Catherine Hennechart-Collette, Laurent Guillier, Sylvie Perelle, SANDRA MARTIN-LATIL, ANSES, Laboratory for Food Safety, University of Paris-Est, Maisons-Alfort, France
- T3-04 Mold Identification with MALDI-TOF MS: ID-Fungi
- 16.15 Plates and Relevance of the Reference Library for Easy and Reliable Results Semcheddine Cherrad, Markus Kostrzewa, Katharina Mucek, DANIELE SOHIER, Sebastien Vacher, Bruker, Bremen, Germany
- T3-05 Dairy Isolates of Biofilm-forming Bacillus Demonstrate
- 16.30 Enhanced Resistance to Cleaning Procedures levgeniia Ostrov, MOSHE SHEMESH, Agricultural Research Organization (ARO) The Volcani Center, Rishon LeZion, Israel
- T3-06\* Using DNA Barcoding to Investigate the Accuracy
- 16.45 of Seafood Product Labelling in Taiwan PEI-YING CHEN, Tsung-Yun Liu, Kung-Hao Liang, National Yang-Ming University, Taipei, Taiwan

#### 17.00 Reception in the Exhibit Area

Check the Program Addendum for changes to the Program.

🔳 – Symposia

– Roundtables – Technicals \*Student Award Competitor

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### 7.30 – 17.00 Registration Open

7.30 – 8.30 Morning Coffee

#### 10.00 – 16.00 Exhibit Hours

Poster Session 2 – Antimicrobials; Beverages and Water; Dairy and Other Food Commodities; Food Toxicology; Meat and Poultry; Pathogens; Sanitation; Seafood

Authors present during scheduled breaks

S9 Close Up of Consumer Kitchen Practices – Can Socio(microbio)logy Aid Food Safety at Home? Room 200

> Organizers: Solveig Langsrud and Paula Teixeira Convenor: Solveig Langsrud Sponsored by the IAFP Foundation

- 8.30 Kitchen Hygiene in Six European Countries Safe or Unsafe? SOLVEIG LANGSRUD, Nofima, Norwegian Institute of
- Food, Fisheries and Aquaculture Research, Ås, Norway
   8.50 Norovirus Removal from Salads and Bivalve Molluscs
   SUSANA CLUX ARNALL University of Parcelona
- SUSANA GUIX ARNAU, University of Barcelona, Barcelona, Spain
- 9.10 The Challenging Practice of Keeping Food Cold: Refrigeration in Everyday Domestic Life HELENE MARIA FIANE TEIGEN, Consumption Research Norway, SIFO, Oslo Metropolitan University, Oslo, Norway
- 9.30 Surprisingly (un)Safe: Runny Eggs and Poultry! PAULA TEIXEIRA, Universidade Católica Portuguesa, CBQF – Centro de Biotecnologia e Química Fina – Laboratório Associado, Escola Superior de Biotecnologia, Porto, Portugal
- 10.00 Networking Coffee Break in the Exhibit Area

#### S10 Food Safety Emerging Risk Identification with Novel Computational Methods *Club Atlantique* Organizer and Convenor: Akos Jozwiak

- 8.30 Identifying Emerging Technological Fields as Drivers of Emerging Risks with Network Analysis AKOS JOZWIAK, National Food Safety Chain, Budapest, Hungary
- 8.50 Using Bayesian Network Analysis in Identifying Emerging Risks HANS MARVIN, Wageningen University and Research, Wageningen, The Netherlands
- 9.10 Emerging Risk Identification and Scenario Analysis in the Food Chain NIELS LUCAS LUIJCKX, The Netherlands Organisation for Applied Scientific Research (TNO), Zeist, The Netherlands
- 9.30 Application of Metagenomics Data Pool in Structured Problem Solving – A Bottled Water Industry Example ZOLTAN SYPOSS, Coca-Cola HBC, Vienna, Austria

#### 10.00 Networking Coffee Break in the Exhibit Area

S11 Fast MALDI Typing to Drive Decision Making and Source Tracking Room GH

Organizer: Thomas Charrier Convenors: Patrice Arbault and Thomas Charrier Sponsored by the IAFP Foundation

- 8.30 Actions Speak Louder Than Words: MALDI-TOF MS for Realistic Analysis Workflow BENOIT GASSILLOUD, ANSES, Nancy, France
- 8.50 Fast *Salmonella* spp. Screening Using MALDI-Typing ANIL PERSAD, University of the West Indies, St. Augustine, Trinidad and Tobago
- 9.10 Identification of Cereulide-producing *Bacillus cereus* by MALDI-TOF MS SEBASTIAN ULRICH, Ludwig-Maximilians-University, Munchen, Germany
- 9.30 MALDI-based Source Tracking of *Listeria monocytogenes* to Drive Decision-making THOMAS CHARRIER, Eurofins, Nantes, France
- 10.00 Networking Coffee Break in the Exhibit Area

#### T4 Technical Session 4 – Communication and Outreach, Food Toxicology, Pathogens and Risk Assessment Room I Convenor: Kali Kniel

- T4-01 The USDA-NIFA Food Virology Collaborative (NoroCORE):
   8.30 An Example of the Use of Team Science to Address a Major Food Safety Challenge REBECCA GOULTER, Lee-Ann Jaykus, North Carolina State University, Raleigh, NC, USA
- **T4-02** Prioritization of Chemical Hazards in Food to be
- 8.45 Considered in the Canadian Food Inspection Agency Establishment-based Risk Assessment Model MOHAMED RHOUMA, Manon Racicot, Alexandre Leroux, Romina Zanabria, Sylvain Quessy, Canadian Food Inspection Agency, St-Hyacinthe, QC, Canada
- **T4-03** Safe Food for Canadians Regulations, the New Canadian
- 9.00 Food Safety Law: What are the Main Differences from the European Union Hygiene Package and The United States Food Safety Modernization Act? What are the Threats and Opportunities? CLAUDIO GALLOTTINI, Paolo Quattrocchi, ITA Corporation Canada Ltd., Montreal, QC, Canada

**T4-04**Risk Benefit Assessment of Foods: Lessons Learned from9.15a Capacity Building Experience Under the RiskBenefit4EU

Project Géraldine Boué, Paula Alvito, Roberto Brazão, Paulo Carmona, LEA JAKOBSEN, Carla Lopes, Carla Martins, Jeanne-Marie Membre, Sarogini Monteiro, Pedro Nabais, Sofie Theresa Thomsen, Duarte Torres, Silvia Viegas, Sara Pires, Ricardo Assunção, SECALIM, INRA/Oniris, Nantes, France

## Thursday, 25 April – 10.30–12.00

- T4-05 Non-targeted Identification of Food Adulterants
   9.30 Using Hand-held Near-infrared Spectrometers RONALD SARVER, Douglas MacRae, Brent Steiner, Robert Donofrio, Greg McNeil, Neogen Corporation, Lansing, MI, USA
- T4-06 Leveraging WGS Databases to Enhance Risk
- 9.45 Assessment, Attribution, and Large-scale Epidemiology Studies JANE VAN DOREN, Moez Sanaa, Regis Pouillot, Francisco Garces Vega, Errol Strain, U.S. Food and Drug Administration–CFSAN, College Park, MD, USA
- 10.00 Networking Coffee Break in the Exhibit Area
- S12 Actions Speak Louder Than Words: Ongoing Efforts for Global Harmonization in Standardization in Food Microbiology Room 200

#### Organizers and Convenors: Daniele Sohier and David Tomás Fornés Sponsored by the IAFP Foundation

- 10.30 Postcard from ISO: Stay Up-to-Date! JACQUES-ANTOINE HENNEKINNE, ANSES, Maisons-Alfort, France
- 11.00 Best of Both Worlds in Salmonella Testing KIRSTEN MOOIJMAN, RIVM, Bilthoven, The Netherlands
- 11.30 Method to Our Madness: The ISO 16140-3 Standard on Analytical Method Implementation and Verification BENJAMIN DIEP, Nestlé Research, Lausanne, Switzerland

#### 12.00 Lunch Available in the Exhibit Area

#### S13 Food Safety Culture: The Proof is in the Science

#### Club Atlantique

Organizers: Liesbeth Jacxsens and Carol Anne Wallace Convenors: Helen Taylor and Carol Anne Wallace

- 10.30 Food Safety Culture Walking the Talk (Mission and Vision)
   LONE JESPERSEN, Cultivate Food Safety, Hauterive, Switzerland
- 11.00 Key Aspects of People Systems and Adaptability for Effective Food Safety Culture (People Systems and Adaptability) LIESBETH JACXSENS, Ghent University, Ghent, Belgium
- 11.30 Achieving Consistency Based on Science and Risk Understanding – Learning the Lessons to Push Beyond the Checkbox FSMS (Consistency and Hazards and Risk Awareness) CAROL ANNE WALLACE, University of Central Lancashire, Preston, UK

#### 12.00 Lunch Available in the Exhibit Area

#### S14 Applications of Microbial Profiling: The Present and the Future Room GH Organizers: Roy Betts and Greg Jones

#### Convenors: Francesca De Filippis and Greg Jones

- 10.30 Microbial Profiling: What is It, and What Can It Do? GREG JONES, Campden BRI, Chipping Campden, UK
- 11.00 Use of Metagenomics in an Industrial Setting JULIA HEWERDINE, Dunbia – A Division of Dawn Meats, Crosshands, UK
- 11.30 Current Research in Food Microbiomes and How Industry Can Benefit FRANCESCA DE FILIPPIS, University of Naples Federico II, Portici, Italy

#### 12.00 Lunch Available in the Exhibit Area

#### T5 Technical Session 5 – Dairy and Other Food Commodities and Pathogens Room I Convenor: Panos Skandamis

- T5-01 Listeriosis in South Africa Facts and Figures
- 10.30 and What We Should be Doing about It PIETER GOUWS, Centre for Food Safety, Stellenbosch University, Stellenbosch, South Africa
- T5-02 Microbial Ecology and Food Safety of Fermented Carrot

10.45 Juice Cédric Verschueren, WANNES VAN BEECK, Sarah Lebeer, Mieke Uyttendaele, Ghent University (UGent), Faculty of Bioscience Engineering, Research Unit Food Microbiology and Food Preservation (FMFP-UGent), Ghent, Belgium

- T5-03 The Inhibitory Effect of Traditional Pomegranate Molasses
- 11.00 on *S.* Typhimurium Growth on Parsley Leaves and in Mixed Salad Vegetables DIMA FAOUR-KLINGBEIL, Ewen Todd, School of Biological and Marine Sciences, University of Plymouth, Devon, UK
- **T5-04\*** Characterizing the Diversity of Bacterial Communities
- 11.15 from Imported Date Fruits to Control Listeria monocytogenes KRISHNA S. GELDA, Valeria R. Parreira, Gisèle LaPointe, Jeffrey M. Farber, University of Guelph, CRIFS, Guelph, ON, Canada
- T5-05 Shiga Toxin-producing E. coli (STEC) Occurrence and
- 11.30 Virulence Profile Comparison from Flour Samples Obtained during Monitoring and Outbreak Situations ANNIE LOCAS, Johanna Murphy, Helen Zhang, Etsuko Yamamoto, Canadian Food Inspection Agency, Ottawa, ON, Canada
- **T5-06** Bacterial Strain Selection for the Validation of High Pressuretreated Juices

Catherine Rolfe, Nathan Anderson, Glenn Black, ALVIN LEE, Institute for Food Safety and Health, Illinois Institute of Technology, Bedford Park, IL, USA

#### 12.00 Lunch Available in the Exhibit Area

### Thursday, 25 April – 13.30–15.00

#### RT2 The Use of Chemicals in Food Hygiene and Linkage to Microbial Resistance *Room 200* Organizer and Convenor: Marie-Claude Quentin

### 13.30 Panelists: JOHN DONAGHY, Nestec Ltd., Vevey, Switzerland PHILIPPE GLASER, Institut Pasteur, Paris, France JEFFREY LEJEUNE, Food Safety and Quality Unit, AGFF, Food and Agriculture Organisation, Rome, Italy PETER MCLURE, Mondelēz, Bournville, UK PIETRO STELLA, European Food Safety Authority (EFSA), Parma, Italy

15.00 Networking Coffee Break in the Exhibit Area

#### S15 Hepatitis E Virus, an Emergent Foodborne Pathogen? Public Health Implications *Club Atlantique* Organizers and Convenors: Nerea Garcia-

Benzaquen and David Rodriguez-Lazaro Sponsored by the IAFP Foundation

- 13.30 Hepatitis E Virus Burden in Europe: What Do We Know? WIM VAN DER POEL, Wagenigen University, Lelystad, The Netherlands
- 14.00 Hepatitis E Virus in Animals, Food and Where Else? NICOLE PAVIO, ANSES, Maisons-Alfort, France
- 14.30 Detection Methods for Hepatitis E Virus What Do We Need? REIMAR JOHNE, Federal Institute for Risk Assessment, Berlin, Germany
- 15.00 Networking Coffee Break in the Exhibit Area
- S16 Challenge Testing for Listeria monocytogenes: Requirements, Needs, Difficulties and Developments Room GH Organizers: Paul in't Veld and Florence Postollec Convenor: Mariem Ellouze Sponsored by ADRIA Food Technology Institute
- 15.30 Challenge Testing and Standardisation, Recent Developments FLORENCE POSTOLLEC, ADRIA - UMT ACTIA19.03 ALTER'iX, Quimper, France

- 16.00 *Listeria monocytogenes* and Challenge Testing, A European Perspective HÉLÈNE BERGIS, ANSES, Maisons-Alfort, France
- 16.30 Conducting Challenge Tests for *Listeria monocytogenes*, a Real Challenge PAUL IN'T VELD, NVWA, Utrecht, The Netherlands
- 15.00 Networking Coffee Break in the Exhibit Area
- T6 Technical Session 6 Pathogens, Epidemiology and General Microbiology *Room I*

#### **Convenor: Anne Brisabois**

- **T6-01\*** Comparative Genomics of *Listeria monocytogenes*
- 13.30 Isolated from Fresh Produce, Meat and Clinical Cases ERIN LEWIS, J. Andrew Hudson, Nigel Cook, Jerry Barnes, Edward Haynes, Newcastle University, Newcastle, UK
- T6-02 Evaluation of Methods for Elution of HEV Particles
- 13.45 in Naturally Contaminated Sausage, Figatelli and Pig Liver CATHERINE HENNECHART-COLLETTE, Audrey Fraisse, Sylvie Perelle, Sandra Martin-Latil, ANSES, Laboratory for Food Safety, University of Paris-Est, Maisons-Alfort, France
- T6-03 Genomic Method to Highlight Epidemiological Links
- 14.00 between *Staphylococcus aureus* Strains DÉBORAH MERDA, Noémie Vingadassalon, Lyasmine Negrouche, Jacques-Antoine Hennekinne, Yacine Nia, Université Paris-Est, ANSES, Maisons-Alfort, France
- T6-04\* Bacterial Spore Inactivation Mechanisms during Low
- 14.15 Energy Electron Beam Treatment YIFAN ZHANG, Nina Huber, Ralf Moeller, Barbora Dubovcova, Georgios Akepsimaidis, Nicolas Meneses, David Drissner, Alexander Mathys, ETH Zurich, Zürich, Switzerland
- T6-05\* Expression and Prediction of Staphylococcal
- 14.30 Enterotoxin G and I LIVIA SCHWENDIMANN, Déborah Merda, Thomas Berger, Anita Kläui, Jacques-Antoine Hennekinne, Yacine Nia, Michel-Yves Mistou, Hans-Ulrich Graber, Agroscope/ Université Paris-Est, ANSES, Bern, Switzerland
- T6-06\* Influence of Osmotic Stress on Heat Resistance of Listeria
- 14.45 *monocytogenes* in Food Products INGE VAN VILSTEREN, Elisabetta Saverio, Nicholas Brian Johnson, Nestlé Research, Konolfingen, Switzerland

#### 15.00 Networking Coffee Break in the Exhibit Area

## Thursday, 25 April – 15.30–17.00

#### RT3 Foodborne Viruses: Detection, Risk Assessment, and Control Options in Food Processing Room 200

#### **Organizer and Convenor: Elias Rito** Sponsored by ILSI Europe and the IAFP Foundation

15.30 Panelists: ELISSAVET GKOGKA, Arla Innovation Centre, Aarhus, Denmark

ALVIN LEE, Institute for Food Safety and Health, Illinois Institute of Technology, Bedford Park, IL, USA

FABIENNE LOISY-HAMON, bioMérieux, Grenoble, France GLORIA SÁNCHEZ. Institute of Agrochemistry and Food

Technology (IATA-CSIC), Valencia, Spain

SOPHIE ZUBER, Nestlé Research Center, Lausanne, Switzerland

#### 17.00 End of Day

S17 Water Re-Use in Food Processing Industry – It's Inevitable! Club Atlantique Organizers: Leon Gorris and Elisabetta Lambertini

Convenors: Leon Gorris, Elisabetta Lambertini and Kang Zhou

- 15.30 JEMRA Risk-based Framework to Water Re-Use Under Development LEON GORRIS, Food Safety Expert, Nijmegen, The Netherlands and JEFFREY LEJEUNE, Food and Agriculture Organisation, Rome, Italy
- 16.00 Experiences with Water Re-Use in Dairy Operations MARTIN ANDERSEN, Claus Heggum, IN-Water ApS, Brabrand, Denmark
- 16.30 Experiences in Water Reuse in the Beverage Industry JOSEP MOLAS PAGES, Coca-Cola, Madrid, Spain

#### 17.00 End of Day

- **S18** Fungal Spores in Food; Implication of Natural Heterogeneity on Food Quality *Room GH* Organizers and Convenors: Heidy Den Besten and Jan Dijksterhuis Sponsored by the IAFP Foundation
- 15.30 Significance of Environmental Conditions during Sporulation on Physiological State and Phenotypic Heterogeneity of *Penicillium* Roqueforti Conidia NICOLAS NGUYEN VAN LONG, ADRIA Food Technology Institute, Quimper, France
- 16.00 Strain Variability in Conidial Heat Resistance of Food Spoilage Fungi TOM VAN DEN BRULE, Westerdijk Fungal Biodiversity Institute, Utrecht, The Netherlands
- 16.30 Tackling Preservative-resistant Fungal Spores SIMON AVERY, University of Nottingham, Nottingham, UK
- 17.00 End of Day

#### T7 Technical Session 7 – Pathogens Room I

#### **Convenor: Annemarie Pielaat**

- T7-01 ANSR<sup>™</sup> for Salmonella spp. Detection is More Sensitive
   15.30 Than ISO 6579 for Rapid Detection of Salmonella spp. in Food and Feed Samples
   EDAN HOSKING, Jerry Tolan, Frederic Martinez, Muriel Bernard, Preetha Biswas, Robert Donofrio, Maryse Rannou, Neogen Corporation, Lansing, MI, USA
- T7-02 Salmonella Lubbock: A New Serotype in between
- 15.45 S. Montevideo and S. Mbandaka MARIE BUGAREL, Peter Cook, Henk den Bakker, Kendra Nightingale, Guy Loneragan, Texas Tech University, Lubbock, TX, USA
- T7-03\* Investigation of Genotypic Markers as Indicators for
- 16.00 Psychrotrophic Phenotypic Behaviour of *Bacillus cereus* DOREEN METTO, Yinghua Xiao, Elissavet Gkogka, Anne Elsser-Gravesen, Marianne Hammershøj, Aarhus University, Aarhus, Denmark
- T7-04 Exploring the Global Transcriptomic Response of
- 16.15 *L. monocytogenes* to Desiccation on Stainless Steel MARTIN LAAGE KRAGH, Lisbeth Truelstrup Hansen, DTU Food, Kongens Lyngby, Denmark
- T7-05\* Tryptone, Peptone and Casamino Acids Affect Acid
- Survival and Gabae Production in Listeria monocytogenes 10403S WT CAROLINA BRUSCHI, Ranju Paudyal, Conor O'Byrne, Kimon A. G. Karatzas, University of Reading, Reading, UK
- **T7-06** The Role of *Listeria monocytogenes* Glutamate
- 16.45 Decarboxyase System in Oxidative Stress Tolerance Marcia Boura, KIMON A. G. KARATZAS, University of Reading, Reading, UK

#### 17.00 End of Day

### Friday, 26 April – 8.30–10.00

### 7.30 – 12.00 Registration Open

#### 7.30 – 8.30 Morning Coffee

**Ongoing Research Activities in Risk Benefit S19 Assessment of Food** Room 200 **Organizer and Convenor:** 

### Jeanne-Marie Membré

- 8.30 Taking Substitution into Account in Risk Benefit Assessment – Two Case Studies LEA SLETTING JAKOBSEN, Technical University of Denmark - DTU, Lyngby, Denmark
- 9.00 Risks and Benefits Associated with Red Meat Consumption in France JULIANA DE OLIVEIRA MOTA, INRA, UMR 1014 SECALIM, Nantes, France
- First Results and Lessons Learnt from RBA Application 9.30 in the Industry ANNEMARIE PIELAAT, Unilever R&D, Vlaardingen, The Netherlands

#### **10.00 Networking Coffee Break**

#### **S20** Norovirus, Glycans and Oysters: The Perfect Association? Club Atlantique **Organizers: Fabienne Hamon** and Soizick F. Le Guyader **Convenor: Fabienne Hamon**

- 8.30 Metagenomic Applied to Study Norovirus MARION KOOPMANS, Erasmus University Medical Center, Rotterdam, The Netherlands
- Glycans Interaction of Norovirus and Other Pathogens 9.00 Inside and Outside the Host JACQUES LE PENDU, Inserm-Université de Nantes, Nantes, France
- Ovsters and Norovirus: How New Technologies Will Help 9.30 to Improve Safety? SOIZICK F. LE GUYADER, Ifremer, Nantes, France

#### 10.00 Networking Coffee Break

- S21 **Beef Decontamination Treatments in Slaughter Plants: Do They Improve Product** Safety? Room GH **Organizers: Xiangin Yang** and Alexander Gill **Convenor: Alexander Gill**
- Overview of Decontaminating Treatments Currently Used 8.30 in Commercial Beef Plants GARY ACUFF, Texas A&M University, College Station, TX, USA
- 9.00 Biofilm Formation, Pathogen Prevalence, and Meat Contamination RONG WANG, U.S. Department of Agriculture-ARS, Clay Center, NE, USA

9.30 Resistance of E. coli from Beef to Decontaminating Treatments/Biocides Commonly Used in Meat Plants XIANQIN YANG, Agriculture and Agri-Food Canada, Lacombe, AB, Canada

#### 10.00 Networking Coffee Break

#### **T8 Technical Session 8 – Risk Assessment** Room I Convenor: Anett Winkler

- T8-01
- Concept of Risk-benefit Analysis Balancing the Impact 8.30 of Cumulative Exposure to Pesticides Versus Beneficial Effect on Human Health Due to Fruit and Vegetable Intake LIESBETH JACXSENS, Pieter Spanoghe, Jacob van Klaveren, Faculty of Bioscience Engineering, Ghent University, Ghent, Belgium
- T8-02 Critical Analysis of Qmra of Meats: What We Do Next? 8.45 VINCENT TESSON, Michel Federighi, Enda Cummins,
- Sandrine Guillou, Jeanne-Marie Membre, Géraldine Boué, SECALIM, INRA/Oniris, Nantes, France
- T8-03\* Modelling the Risk of Contamination of Lettuce with 9.00 Escherichia coli O157:H7 from Field to Consumption in
  - Australia HAYRIYE BOZKURT, The University of Sydney, Sydney, Australia
- T8-04\* Cross-contamination Risks Factors in Domestic Chicken-
- handling Practices among Consumers in 5 European 9.15 Countries in the Transdisciplinary Safeconsume Project PIERRINE DIDIER, Christophe Nguyen-The, Isabelle Maître, Monica Truninger, Silje Elisabeth Skuland, Helene Maria Fiane Teigen, Anca Ioana Nicolau, Augustin Octavian Mihalache, Loredana Dumitrascu, Mike Foden, Lydia Martens, UMR408 SQPOV, INRA, Avignon Université - USC1422 Grappe, Université Bretagne Loire, Ecole Supérieure d'Agricultures, Angers, France
- T8-05 Introducing Aginfra+ Based Virtual Research Supporting Scientific Collaboration and Knowledge Exchange in the 9.30 Food Safety Domain MATTHIAS FILTER, Lars Valentin, Ahmad Swaid, Thomas
- Schüler, Taras Günther, German Federal Institute for Risk Assessment, Berlin, Germany
- T8-06 Improving Raw Milk Food Safety Management by
- Quantitative Risk Assessment Approach 9.45 VALÉRIE MICHEL, Janushan Christy, Fanny Tenenhaus, ACTALIA, La Roche sur Foron, France

#### 10.00 Networking Coffee Break

#### S22 How Has Metagenomics Been Useful to Food Safety Research and What Does Its Application to Public Health Hold? Room 200

Organizers: Maria Hoffmann, Jesse Miller and Eric Stevens Convenor: Eric Stevens

Sponsored by the IAFP Foundation

- 10.30 Metagenomic Sequencing for Surveillance of Food and Waterborne Viral Diseases MARION KOOPMANS, Erasmus University Medical Center, Rotterdam, The Netherlands
- 11.00 How FDA is Using Metagenomics for Food Safety and Microbiological Methods Development PADMINI RAMACHANDRAN, Center for Food Safety and Applied Nutrition, U.S. Food and Drug Administration, College Park, MD, USA
- 11.30 Utilizing Culture-independent Genomics and 'Big Data' Computation in the Food Industry BALKUMAR MARTHI, DaQshConsulting, Vlaardingen, The Netherlands

#### 12.00 15-Minute Break

#### S23 *Campylobacter*, Health Impact, Performance Objectives and Effectiveness of Sampling Plans

#### Club Atlantique

Organizers: Jeffrey Farber, Leon Gorris and Marcel Zwietering Convenor: Leon Gorris Sponsored by ICMSF and the IAFP Foundation

10.30 Health Impact of *Campylobacter*. The Main Zoonotic Pathogen in Many Countries

- JEFFREY FARBER, University of Guelph, CRIFS, Guelph, ON, Canada
- 11.00 Establishing Performance Objectives throughout the Chicken Production Chain to Achieve Control of *Campylobacter* WAYNE ANDERSON, Food Safety Authority of Ireland, Dublin, Ireland
- 11.30 Effectiveness of a (More and More Stringent) Sampling Plan for *Campylobacter* MARCEL ZWIETERING, Wageningen University, Wageningen, The Netherlands

#### 12.00 15-Minute Break

S24 Insects in Poultry Feed: Regulatory Framework, Poultry Gut Microbiota and Consumer Acceptability Room GH

**Organizer and Convenor: Luca Cocolin** 

10.30 The Use of Insects and Insects Derivative for Animal Feeding: An Attempt of a Guideline MICHELE BLASI, Associazione Italiana Allevatori, Rome, Italy

- 11.00 Impact of Innovative Feeds in the Microbiota and a Microbiome of Poultry ILARIO FERROCINO, University of Torino-DISAFA, Grugliasco, Italy
- 11.30 Consumer Acceptability of Poultry Meat Produced with Insects
   FLAVIO PELLEGRINUZZI, Euroconsumers Cooperative Partner Group of Independent European Consumer Organizations, Luxembourg, Wageningen, The Netherlands

#### 12.00 15-Minute Break

- T9 Technical Session 9 Applied Laboratory Methods, Microbial Food Spoilage and Pathogens *Room I* Convenor: Vasilis Valdramidis
- **T9-01\*** Reagent-free Detection of Silver lons in Tap Water
- 10.30 Using Square Wave Voltammetry and Local pH Control LUIZA WASIEWSKA, Geraldine Duffy, Kaye Burgess, Alan O'Riordan, Tyndall National Institute, Cork City, Ireland
- **T9-02** A RTCA-based Assay as an Innovative Approach for
- 10.45 Thermal Inactivation Studies of Hepatitis A Virus AUDREY FRAISSE, Catherine Hennechart-Collette, Laurent Guillier, Sylvie Perelle, Sandra Martin-Latil, ANSES, Laboratory for Food Safety, University of Paris-Est, Maisons-Alfort, France
- T9-03 Characterization of Meat and Seafood Spoilage
- 11.00 Mechanisms by *Brochothrix thermosphacta* Nassima Illikoud, Marie-France Pilet, EMMANUEL JAFFRÈS, Monique Zagorec, SECALIM, INRA, Oniris, Université Bretagne Loire, Nantes, France
- **T9-04** Estimation of Microbial Spoilage of Ready-to-Eat Baby
- 11.15 Spinach Using Fourier Transform Infrared Spectroscopy Evanthia Manthou, Anastasia Bakalaki, Alexandra Lianou, Panagiotis Tsakanikas, Efstathios Panagou, GEORGE-JOHN NYCHAS, Laboratory of Microbiology and Biotechnology of Foods, Agricultural University of Athens, Athens, Greece
- **T9-05** The Effect of Carbon Dioxide as a Climatic Parameter on
- Microbial Food Contaminants and Selective Isogenic Mutants
   SHOLEEM GRIFFIN, Sanja Ivanovic, Christina Chatzitzika, Jan F. M. Van Impe, Vasilis P. Valdramidis, Faculty of Health Sciences, University of Malta, Msida, Malta
- **T9-06\*** Cardinal Parameter Meta-regression Models Describing
- 11.45 Listeria monocytogenes Growth in Broth BEATRIZ NUNES SILVA, Vasco A. P. Cadavez, José A. Teixeira, Mariem Ellouze, Ursula A. Gonzales-Barron, CEB – Centre of Biological Engineering, University of Minho, Braga, Portugal Wageningen, The Netherlands

#### 12.00 15-Minute Break

#### CS Closing Session Room 200

#### **Chairs: Daniele Sohier and Helen Taylor**

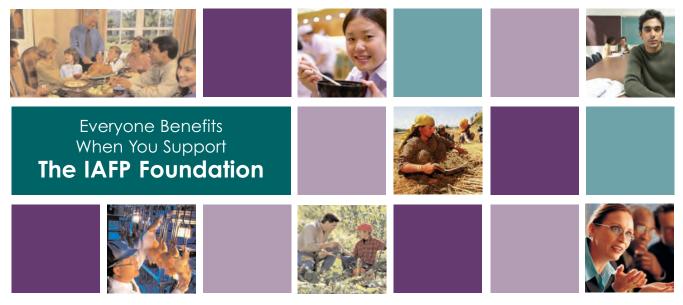
12.15 Introducing the One Health European Joint Programme (EJP) Initiative and Its ORION Project MATTHIAS FILTER, German Federal Institute for Risk Assessment, Berlin, Germany

- 12.45 How New Tools and Technologies Bring New Questions and Help to Answer Old Ones TIMOTHY JACKSON, Driscoll's, Watsonville, CA, USA
- 13.15 Awards Presentation and Concluding Remarks KALI KNIEL, University of Delaware, Department of Animal & Food Sciences, Newark, DE, USA; TIMOTHY JACKSON, Driscoll's, Food Safety, Regulatory and Social Compliance, Watsonville, CA, USA

13.30 – 14.30 Farewell Refreshments

#### Check the Program Addendum for changes to the Program.

Symposia - Roundtables - Technicals \*Student Award Competitor



For more than 30 years, the IAFP Foundation has been working hard to support the mission of the International Association for Food Protection. But we would like to do more. Much more. Food safety concerns and food defense challenges continue to grow. As a result, it is more important than ever that we provide additional programs and services to achieve our common mission of Advancing Food Safety Worldwide<sub>®</sub>. Remember, when you support the IAFP Foundation everyone benefits, including you.



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# INVITED SPEAKER BIOGRAPHIES

24–26 April 2019 – Nantes, France

## **INVITED SPEAKER BIOGRAPHIES**



#### Gary Acuff Texas A & M University, USA

Gary R. Acuff currently holds the title of Professor of Food Microbiology at Texas A&M University where he has been a faculty member for 38 years. Dr. Acuff served as Head of the Department of Animal Science at Texas A&M from 2004–2010 and was President of IAFP from 2007–2008. He has been a member of IAFP since 1982. Dr. Acuff obtained his B.S. in biology from Abilene Christian University in 1980 and his M.S. and Ph.D. in food science and technology, specializing in food microbiology, from Texas A&M University in 1982 and 1985, respectively.



#### Ana Allende CEBAS-CSIC, Spain

Ana Allende (1975) from CEBAS-CSIC (Spanish National Research Council) in Spain is a Senior Researcher with focus on quality and safety of fresh produce. She obtained her Ph.D. in Food Science and Technology at the University of Cartagena, Spain. She further pursued research as a postdoc of the Flanders Centre of Postharvest Technology, Katholieke Universiteit Leuven in Belgium and the Food Quality Lab/Environmental Microbial and Food Safety Lab at the USDA-Baltimore in USA. She has published more than 100 research articles in peer-reviewed international journals focused on the safety of fresh produce as well as the impact of pre- and post-harvest technological interventions in microbial ecology of fruits and vegetables with more than 3,400 cites. Her current H index is 34. She has built up more than eighteen years of scientific research but also management

experience by executing, initiating and guiding research projects in the area of microbial safety of fresh produce. Now she holds several positions in (inter)-national institutions including member of the BIOHAZ panel at the European Food Safety Authority (EFSA), vice-director of the CEBAS-CSIC, and member of the COST ACTION HuPlant. She is also member of three editorial boards of international journals including: (1) International Journal of Food Microbiology, (2) Postharvest Biology and Technology Journal, and (3) International Journal of Food Contamination.



#### Martin Andersen IN-Water ApS, Denmark

Martin Andersen is the Founder and Managing Partner of IN-Water ApS – a specialist consultancy on industrial water solutions. He is an expert in industrial water management and technologies related to water efficiency, wastewater treatment, water reuse and reclamation. His experience covers multiple industrial branches such as food processing, pharmaceutical, chemical, maritime, municipal, metal finishing, paper, textile, building materials and printing industry. Martin's work covers projects in Denmark, Central and Eastern Europe, Southeast Asia, South America and North America. He was the lead water technology expert in a large public-private partnership on water efficiency and reuse in the Danish dairy processing industry.



#### Wayne Anderson Food Safety Authority of Ireland, Ireland

Dr. Wayne Anderson, Director of Food Science and Standards, Food Safety Authority of Ireland. Dr. Wayne Anderson joined the Food Safety Authority of Ireland (FSAI) in 1999 from the food industry and is now Director of the Food Science and Standards. He previously served 10 years with Unilever research and a year as technical manager in a small food factory in the West of Ireland. He holds a primary degree in biochemistry and a Ph.D. in predictive microbiology. He is a member of the International Commission on Microbiological Specifications for Foods (ICMSF), a fellow of the Institute of Food Science and Technology Ireland (IFSTI) and a fellow of the Institute of Food Science and Technology UK (IFST). He has also worked with WHO/FAO on several expert consultations. Wayne has a personal interest in producing science-based food safety standards for the food industry with a particular desire to help small food businesses to produce safe food.



#### Simon Avery University of Nottingham, United Kingdom

Simon Avery is Professor of Eukaryotic Microbiology at the University of Nottingham, UK, and current President of the British Mycological Society. His interests lie in the effects of stress on organisms, with application of that knowledge for control of fungi. He has a focus on environmental toxicants and antimicrobials including preservatives, fungicides and physical approaches. He uses yeast as a eukaryotic model to enable characterization of stress-effects at the whole-cell and molecular level, including cell spore individuality. Findings are extended to fungal pathogens of plants and humans, and food spoilage fungi. Current work is funded by the BBSRC and industrial sponsors.



#### Hélène Bergis ANSES, France

Hélène Bergis is an Engineer in Food Microbiology at the Laboratory for Food Safety from the French Agency for Food Environmental and Occupational Health & Safety (ANSES) – Maisons-Alfort – France. She is a member of the EU-Reference Laboratory on *Listeria monocytogenes*; on the team in charge of shelf-life studies related to *Listeria monocytogenes* and predictive microbiology; in charge to advise National Reference Laboratories for *Listeria monocytogenes* in challenge testing, durability studies and in the use of growth modelling software. Bergis is also a member of the National Reference Laboratory for *Listeria monocytogenes*. In charge of audits related to challenge tests, of proficiency trials and training. She participates to ISO and Afnor standardisation working groups as well as a French Technical Network "Expertise in microbial determination of food products shelf life."



#### Roy Betts Campden BRI, United Kingdom

Roy Betts is Head of Microbiology at Campden BRI, an independent international food research organisation based in the UK. Roy manages a group of 45 food microbiologists, undertaking a range of industry focused food research and testing projects for a worldwide client base. He originally managed a research team at Campden BRI and concentrated on the research, development and validation of microbiological test methods. After becoming Head of Department, his interests moved to the assessment of the microbiological quality and safety of foods, advising industry on techniques and procedures to produce and market high quality safe foods. Roy has published widely in the area and is a member of the ILSI Europe Microbiological Food

Safety Task Force, the UK Food and Drink Federation Food Hygiene Sub Committee and the UK Advisory Committee on the Microbiological Safety of Foods as well as British Standards Institute and ISO committees dealing with microbiological test methods.



#### Michele Blasi Associazione Italiana Allevatori, Italy

Dr. Michele Blasi graduated in Agriculture Science, leading the thesis on the extraction of animal fats from cheese with the supercritical  $CO_2$  method. After an experience in the wine industry as head of the commercial sector of a well-known Tuscan company, from 2002 he carried out consultancy activities in the sector of D.O. specializing in the olive and dairy sector. Since 2010 he has been collaborating with DQA srl, a certification company recognized by Mipaaft for the control of the nationals D.O. in which he holds the role of technical manager and director.



#### François Bourdichon International Dairy Federation – Standing Committee on Microbiological Hygiene, Belgium

François Bourdichon is a food safety microbiologist with 15 years of experience within different major food industry operators. Since 2010, François is delegate of France at the International Dairy Federation FIL-IDF in the standing committees dedicated to microbiology: Microbiological Hygiene (SCMH), Harmonization of Microbial Methods (SCHMM), Analytical Methods for Dairy Microorganisms (SCAMDM). François is presently the elected chair of SCMH. He represents FIL-IDF at CODEX and ISO Meetings. SCMH has a dedicated action team focused on delivering guidelines for implementation and interpretation of Process Environment Monitoring (Hygiene indicators and pathogens of concern).



#### Thomas Charrier Eurofins, France

Thomas Charrier, Ph.D. from University of Nantes in 2006 is specialized in Microbiology and Biotechnology. During his career he focused his work on the development of new method for bacterial pathogens and other bacteria detection in food or environment (Bacteriophage or Molecular methods). Within several years of experiments in R&D project management and development, he now manages the Eurofins France Microbiology's R&D team. His research focus is on new methods for flora description and strain identification or typing (NGS and MALDI-TOF MS), pathogens detection and quantification (dPCR) in food and water matrices.



#### Luca Cocolin University of Torino-DISAFA, Italy

Full Professor of Food Microbiology at the University of Torino, Italy, Dr. Luca Cocolin is an executive board member of ICFMH, editor-in-chief of the *International Journal of Food Microbiology* and academic editor of *PLOS One*. He is also a member of the editorial board of *Food Research International, Frontiers in Microbiology, Current Opinion in Food Science and Food Analytical Methods*. He is co-author of about 300 papers on national and international journals. Expert in (i) Molecular methods for the detection, quantification and characterization of foodborne pathogens; (ii) Study of the microbial ecology of foods by using culture independent and dependent methods; (iii) Bioprotection:(iv) Human microbiome.



#### Francesca De Filippis University of Naples Federico II, Italy

Francesca De Filippis is Assistant Professor of Food Microbiology at the Department of Agricultural Sciences of the University of Naples Federico II (Italy) since December 2016. She got her Ph.D. in Food Science and Technology in 2015. Her research interests span from food microbiology to the inter-connections among diet-human microbiome-health. She is expert in the study of microbial ecology in complex environments through the application of metagenomics and metatranscriptomics and in bioinformatics and biostatistics applied to high-throughput sequencing data. She deepened her bioinformatics skills through research stays in recognized international institutions (Argonne National Laboratory, USA and University College of Cork, Ireland). She

participates/participated in several National and European projects aiming at investigating microbial dynamics during food fermentation or spoilage. She is member of the Task Force on Microbiome Studies of the University of Naples Federico II since its establishment in 2017, a collective and interdisciplinary initiative that combines facilities and expertise of 110 staff scientists supporting research activities involving microbiome studies. Her research activity is condensed in > 50 publications since 2012, with an H-index of 20 and > 1000 total citations (source: Scopus).



#### Juliana De Oliveira Mota INRA, UMR 1014 SECALIM, France

Juliana De Oliveira Mota is currently in the third year of her Ph.D. thesis in France, working at SECALIM, the Food Safety and Microbiology research laboratory from the National Institute of Agronomic Research (INRA). Juliana is currently working on the development of models to assess the risk and benefits on human health when eating red meat in France. She obtained a food engineer (Master student) degree at Oniris in Nantes. While obtaining her Master's degree, she got a 3-year experience in the second major poultry company in France, Galliance, working in the production, research & development and quality departments.



#### Kris de Smet European Commission, Belgium

Kris de Smet graduated as Veterinarian Doctor in 1987. From 1988 to 1992 he was researcher at the University of Ghent (Belgium) in the Faculty of Veterinary Science. From 1992 until 2001 he was employed at the private sector. He was mainly involved in veterinary services and quality control of a poultry integration. Since 2001, he has worked as Official at the European Commission, Health and Food Safety Directorate-General. He was involved in the management of EU legislation on BSE and zoonoses (mainly *Salmonella*). Since the beginning of 2009, he coordinates the EU legislation on food hygiene and zoonoses control and the management of foodborne outbreaks. He also coordinates the EU position at the Codex Alimentarius Committee Food Hygiene.



#### Henk den Bakker Center for Food Safety, University of Georgia, USA

Henk C. den Bakker currently works as an assistant Professor in Bioinformatics at the Center for Food Safety of the University of Georgia. He received a Ph.D. in Mycology at Leiden University in The Netherlands in 2005. From 2005 to 2014, Henk worked as a Research Associate at Cornell University, on fungal population genetics lab of Dr. T. Pawlowska, and later on population genetics and genomics of foodborne pathogens and spoilage organisms in the Food Safety Laboratory of Dr. M. Wiedmann. He (co-)authored more than 50 PubMed indexed publications. Dr. Den Bakker's current research focuses on the novel field of food safety informatics.



#### Noémie Desriac LUBEM UBO University – UMT14.01SPORE RISK, Quimper, France

Noémie Desriac is Associate Professor in food microbiology at the Technological Institute of the University of Brest. She obtained a Ph.D. degree in 2013. Her Ph.D. research combined a mathematical modelling approach with gene expression quantification to integrate the bacterial fitness into risk assessment. Then, she integrated the food safety department in ADRIA technical institute as a Project Manager on food quality and safety where she was involved in various projects on bacterial sporulation, outgrowth and germination, bacterial inactivation as well as developing an integrative approach on the use of omics or molecular biomarkers in predictive microbiology. She was also in charge of the software operating system of Sym'Previus and

developed the new version available since 2016. Sym'Previus is a web-based tool to predict the microbial behavior throughout food processes and food shelf life. In 2016, she moved to the university where she teaches food microbiology, and works as a researcher in the field of quantitative description of microbial behavior in food during industrial processes and storage. She is currently involved in various projects on the use of biomarkers in predictive microbiology.



#### Benjamin Diep Nestlé Research, Switzerland

Benjamin Diep has spent 20 years in food industry focusing on Food Safety and Quality and particularly in the methodological aspect. He is currently working as a scientist at Nestlé Research in Lausanne, Switzerland. His field of expertise comprises methods development and validation (cultural and molecular based methods) for food pathogens and hygiene indicators. He is involved or lead in several Nestlé R&D and basic research projects. He also works with operation where he provides technical support and audits in order to improve the laboratories competency. He is a member of ISO 16140 WG and the co-project leader of the ISO 16140 part 3. He is also a technical reviewer for MicroVal.



#### John Donaghy Nestec Ltd., Switzerland

John Donaghy is a Corporate Food Safety Microbiologist at Nestlé, Corporate Quality Management, Switzerland. His expertise and responsibility includes global operational aspects of food safety microbiology, hygiene and allergens. He leads a team of global experts in HACCP, hygiene and thermal processing, overseeing horizontal implementation of key Nestlé food safety standards at Market and factory level. He represents Nestlé on International Commission on Microbiological Specifications of Food (ICMSF), other Codex observer groups and a number of food safety advisory stakeholder groups. Mr. Donaghy previously worked as Senior Food Safety Microbiologist in Nestlé R&D. Prior to joining Nestlé (2011), he worked as Project Leader in food safety microbiology at Agri-Food & Biosciences Institute (AFBI), N. Ireland, on projects funded by FSA (UK), Food Safety Authority Ireland, local Government Industry and European Union.



#### Benjamin Duqué UMR1014 SECALIM INRA, Oniris, France

Benjamin Duqué graduated from the Engineering School of Microbiology and Quality in France. He is now in third year of his Ph.D. in food microbiology at the University of Western Brittani in France. He is working at the SECALIM unit from INRA specialised in food safety. His research aims at predicting resistance potential of *Campylobacter jejuni* using molecular biomarkers.



#### Damien Eveillard Computational Biology Group LS2N UMR 6004 CNRS University of Nantes, France

Damien Eveillard, Ph.D., is Associate Professor at the Université de Nantes, and head of the ComBi team at LS2N. He is the co-head of the Genetics, Genomics, and Systems Biology Master's degree at Université de Nantes. After studying Oceanography at Sorbonne University, Damien Eveillard obtained his thesis in 2004, Université de Lorraine, in modeling the alternative splicing, then completed a postdoctoral fellowship at Texas A&M University. An academic member at the Université de Nantes since 2006, he specializes in the modeling of living systems by constraints (bioinformatics and systems biology). Since 2010, his research has focused on modeling microbial communities.



#### Jeffrey Farber University of Guelph, CRIFS, Canada

Dr. Jeff Farber is currently employed as a Full Professor in the Department of Food Science at the University of Guelph, in Guelph, Ontario, where he is head of the Master's Program in Food Safety and Quality Assurance (FSQA) and is also the Director of the Canadian Research Institute for Food Safety. He currently leads a team of about 10 researchers including graduate students, FSQA students, a post-doc and a research associate, who all do research on various aspects of microbial food safety. Dr. Farber previously worked at Health Canada, most recently as Director of the Bureau of Microbial Hazards, in the Food Directorate of Health Canada, where he led a group of about 60 people working in the areas of food safety research, risk assessment, policy, risk management and risk communication.

Dr. Farber has over 150 publications, plus numerous Book Chapters and has edited 4 books. He was Associate Editor of the International *Journal of Food Microbiology* for many years and has been on a number of Journal Editorial Boards. Dr. Farber has been instrumental in advancing the development of policy approaches on emerging microbial food safety issues in Canada and at a global level. Dr. Farber is a Past President of the International Association for Food Protection, and Treasurer of the International Commission on Microbiological Specifications for Foods (ICMSF). He is also a member of the Agriculture, Food and Nutrition Working Group of the New York Academy of Sciences.

Dr. Farber also has extensive experience working at the international level with organizations such as Codex Alimentarius, WHO and FAO. He has received numerous personal and team awards, the most recent being a Science and Technology award from the Canadian Meat Council. He won one of the highest awards presented to Federal Public Health Officials, the Prime Minister's Outstanding Achievement Award for his work as the lead scientist for Health Canada on the deli-meat listeriosis outbreak.



#### **Benjamin Félix** ANSES, Laboratory for Food Safety, University of Paris-Est, France

Benjamin Félix graduated from Paris VI University. He has been working for more than 11 years in the area of bacterial characterization. Félix has been working for ANSES for nine years in the *Listeria monocytogenes* Team of the unit *Salmonella* and *Listeria*, in the scope of the European Union Reference Laboratory. His working field in reference, encompasses the validation and harmonization of the molecular typing methods applied to *Lm* at European level, the method development, in particular Pulse Field Gel Electrophoresis method and whole genome sequencing methods and the setup of mean to share the data produced. His working field in researched deals with evaluation of Lm genetic diversity in food, food processing, animal and the environment, for the purpose for a better understanding of Lm genetic adaptation to food production environment.



#### Ilario Ferrocino University of Torino-DISAFA, Italy

Ilario Ferrocino's current position is Senior Lecturer level B, Department of Agricultural, Forest and Food Sciences, University of Torino, Italy. He is the author of 48 publications related to microbiology. As reported by *Scopus* (January 2018), the publications reviewed were cited 1036 times, with an index 'h' equal to 16. Area of expertise: (i) Development, optimization and application of molecular methods for the microbial detection; (ii) Study of the microbial ecology of fermented foods; (iii) Bioprotection: molecular characterization of bacteriocin production and its study in vitro and in situ; (iv) Study of the human and animal gut microbiome.



#### Matthias Filter German Federal Institute for Risk Assessment, Germany

Matthias Filter has a diploma in biochemistry. He has experience in bioinformatics, cheminformatics, software development, data mining and QMRA modeling. He has more than 20 years of experience as a project manager in public and private sector organizations. Mr. Filter's current position is senior research scientist, unit "Food Hygiene and Technology, Supply Chains and Food Defense." Federal Institute for Risk Assessment (BfR), Germany. His specific interest is the development of community resources to facilitate efficient knowledge and information exchange in the food safety domain.



#### Norberto García Apimondia, Scientific Commission Beekeeping Economy and Chairman of the Working Group Adulteration of Bee Products, Argentina

Prof. Norberto García has worked intensely during recent years creating awareness on the problem of honey adulteration through many presentations in international meetings, interviews, and publications in specialized journals. He is currently Professor of Apiculture at Universidad Nacional Del Sur, Argentina.

He is the President of the Apimondia Scientific Commission Beekeeping Economy. He also serves as President of the International Honey Exporters Organization (IHEO). García is the Chairman of the U.S. Pharmacopeia Expert Panel "Honey Quality

and Authenticity." He is also Chairman of the APIMONDIA Working Group Adulteration of Bee Products. He serves as Senior Advisor of NEXCO S.A., the main Argentine honey exporter.



#### Benoit Gassilloud ANSES, France

Benoit Gassilloud is a Water Microbiologist who graduated in 2003 as a Ph.D. at the University of Henry Poincaré in Nancy in the field of environmental viruses, and then worked for 1 year in the Laboratory of Environmental Chemistry, Physics and Microbiology (LCPME-CNRS-UMR7564) on the development of a concentration process for viral detection in bottled mineral water. Since 2005, he has been working at the French Agency for Food, Environmental and Occupational Health & Safety (ANSES) as the head of the water microbiology department, Nancy Laboratory for Hydrology. He has now more than 11 year's experience in water microbiology, dealing in particular with the detection and characterization of water-borne pathogens (bacteria, viruses and parasites). In 2013, he took the head of the MALDI-TOF Platform which is a structure that could be use by all the laboratories of the agency for identification or typing bacteria and fungi strains using this technology.



#### Elissavet Gkogka Arla Innovation Centre, Denmark

Elissavet Gkogka is an experienced food microbiologist with more than 10 years of academic and industrial experience in the areas of food safety, natural antimicrobials, predictive modeling, risk assessment and challenge testing. In her position as a research microbiologist in Arla R&D, she has been involved in numerous new product development projects, giving recommendations on product formulations and processing/packaging conditions to ensure food safety and quality throughout shelf life. Elissavet is also a member of ILSI's Microbiological Food Safety Task Force and has experience in foodborne disease epidemiology, having presented her research as a technical adviser for the World Health Organization.



#### Philippe Glaser Institut Pasteur, France

Philippe Glaser, Research Director at the Institut Pasteur, is heading the Ecology and Evolution of Antibiotic Resistance Unit. He is an internationally recognized expert in bacterial genomics and evolution. He is well known for his genomic-epidemiology studies of Group B *Streptococcus* (GBS) both in human and in animals. He has shown how the extensive use of tetracycline starting in the 1950s has been responsible for the replacement of the GBS population colonizing humans by few tetracycline resistance clones and to the emergence of neonatal GBS infection in the 1960s – 1970s both in Europe and in the U.S. With his group, he has developed a general framework for evolutionary analyses of disseminating lineages that he applied to a bovine

GBS clone endemic in Portugal and to the hypervirulent ST17 lineage. In collaboration with Thierry Naas at the Bicêtre Hospital he is currently deciphering the genetic bases for the emergence and dissemination of carbapenemase producing *Escherichia coli* and *Klebsiella pneumoniae* lineages. He is directing the Fighting Antibiotic Resistance program aiming to federate research on antibiotics and antibiotic resistance at the Institut Pasteur (IP) and in the Institut Pasteur International Network.



#### Leon Gorris Food Safety Expert, The Netherlands

Until recently, Dr. Leon Gorris was Director for Regulatory Affairs at Unilever, with responsibility for food safety globally. He joined Unilever in The Netherlands in 1997 and has been based in The Netherlands, the UK and China. Before joining Unilever, Dr. Gorris worked at one of the research institutes of the Ministry of Agriculture, Nature Management and Fisheries, The Netherlands (1990–1997). From 2002–2012, Dr. Gorris held a part-time professorship at the University of Wageningen in The Netherlands, serving as the European Chair in Food Safety Microbiology. He is Visiting Professor at three universities in Beijing

and Shanghai. Dr. Gorris is a member of the International Commission on Microbiological Specifications for Foods (ICMSF and represents ICMSF at Codex Alimentarius and in interactions with FAO and WHO. He chairs the Food Safety Committee of IUFoST (the International Union of Food Science and Technology) and has been elected to the International Academy of Food Science and Technology (IAFoST) in 2016.



#### Susana Guix Arnau University of Barcelona, Spain

Susan Guix Arnau is an Associate Professor at the Department of Genetics, Microbiology and Statistics, University of Barcelona. Her research is devoted to the study of human foodborne enteric viruses such as norovirus, hepatitis A and astrovirus, covering molecular and clinical aspects, as well as their role as food and environmental contaminants, and viral inactivation studies. She reports more than 60 peer-reviewed international publications and book chapters. She has worked as a consultant on norovirus determination in food, involved in the Thailand-EU Policy Dialogues Support Facility. She is the Secretary of the Institute of Nutrition and Food Safety of the University of Barcelona; she has been a member of the International Committee on Taxonomy of Viruses (ICTV), and the scientific committee of the Spanish Authority for Food Safety and Nutrition (AESAN).



#### Linda J. Harris University of California-Davis, Department of Food Science and Technology, USA

Dr. Linda Harris is Chair of the Department of Food Science and Technology and a Specialist in Cooperative Extension in Microbial Food Safety at the University of California, Davis. She oversees a research program on the microbial food safety of fresh fruits and vegetables and tree nuts and provides expertise on food safety microbiology throughout the food chain. She served on the board of the International Association for Food Protection from 2014–2018.



#### Jacques-Antoine Hennekinne ISO TC34/SC9 Chairman, ANSES, France

Jacques-Antoine Hennekinne is in charge of a unit working on bacteria producing toxins such as coagulase positive staphylococci, *Bacillus cereus sensu lato* and *Clostridium* at the French agency for food, environmental, occupational health and safety. His reference and research activities are in the field of these bacteria from enumeration of bacteria to toxin detection including typing. He took part in various research programs as partner or coordinator and conducted the CEN working group drafting the EN ISO 19020 for staphylococcal enterotoxin detection in food matrices. Moreover, since 2018 he is the chairman of the ISO TC34/SC9, the sub-committee dedicated to food microbiology.



#### Julia Hewerdine Dunbia – A Division of Dawn Meats, United Kingdom

Julia Hewerdine has worked in the agri-food sector for over 20 years from research bodies to manufacturing companies covering most protein species and processes. She has held diverse positions ranging from commercial to technical, culinary development to process management. Most recently her role as Group Food Safety, Quality & Animal Welfare Project Manager for Dunbia and Dawn Meats operates across the company's 22 sites in UK and ROI driving innovative science-based projects to ensure the delivery of safe and consistent quality products. Julia holds a BSc (Dual Hons) in Animal Science and Agricultural Science from Aberystwyth University and an MSc in Meat Science from Bristol University. Awarded Chartered Scientist designation from The Science Council and is also a Fellow of the Institute of Food Science and Technology.



#### John Holah UK:IE EHEDG & Holchem Laboratories Ltd., United Kingdom

John Holah is the Technical Director at Holchem Laboratories, the UK's largest supplier of food hygiene services to the food manufacturing industry. John's current responsibilities include the development of innovative cleaning and disinfection chemicals and technologies and their successful utilisation in effectively designed, engineered, validated and managed sanitation programmes. John has a passion for food safety and has been responsible for establishing many GMP/GHPs used in the food industry for the control of pathogens, particularly *Listeria, Salmonella* and *E. coli*. He has been fortunate to have worked within >500 food factories and catering establishments, in the UK, Europe, North and South America, Africa, Asia and Australia. John is an Honorary Professor of Food Safety at Cardiff Metropolitan University and was previously Head of the Food Hygiene Department at Campden BRI. He is active in the support of EHEDG, IAFP and GFSI.



#### Paul in't Veld NVWA, The Netherlands

Paul in't Veld studied Food Technology at the Agricultural University in Wageningen (The Netherlands) from 1979 to 1987 and specialised in food microbiology and food chemistry. He obtained his Ph.D. in 1998 on the topic: The development and evaluation of microbiological reference material for food microbiology. He worked, from 1987 to 1999 at the National Institute of Public Health and the Environment (RIVM). Since 1999 he has been working at The Netherlands Food and Consumer Product Safety Authority (NVWA), the competent authority in The Netherlands. His main activities at the NVWA are related to standardisation of methods in general (more specific in validation/verification of (alternative) methods as the convenor of ISO TC 24/SC9/

WG3: method validation) and support the inspectors with advice on microbiological issues, e.g., the evaluation of studies that are conducted to demonstrate the control of the growth of *Listeria monocytogenes* in ready-to-eat foods. Besides his work at the NVWA, he acts as a Technical Assessor for various accreditation bodies.



#### Timothy Jackson Driscoll's, USA

Dr. Tim Jackson is Vice President of Food Safety, Regulatory Compliance and Worker Welfare with Driscoll's Inc. in Watsonville, California. Dr. Jackson previously served as Director of Food Safety for Nestlé USA, Nestlé Canada and Nestlé Professional North America in Glendale, California, joining in 1995 as a research scientist. He worked in the Microbiology Laboratories at the Nestlé Quality Assurance Laboratory for the U.S. and Canada in Dublin, Ohio before joining the Nestlé Research Center in Lausanne, Switzerland, supporting Nestlé markets in the identification and validation of alternative methods. Dr. Jackson also served as Chief Industrial Microbiologist for Nestlé's global operations in Vevey, Switzerland. Prior to his em-

ployment with Nestlé, he was a research scientist and assistant lecturer at Texas A&M University in College Park. He has been an active IAFP Member since 2001, Dr. Jackson currently serves on the Executive Board as President. He is a member of numerous Committees and Professional Development Groups (PDGs), has chaired and served on several IAFP award selection committees, and has presented at IAFP meetings worldwide.



#### Liesbeth Jacxsens Ghent University, Belgium

Liesbeth Jacxsens, Ph.D., bio science engineering, is Professor in Food Safety Management and Risk Analysis in Agri-Food Chain at Department of Food Safety and Food Quality, Ghent University, Belgium. Her research domain encounters two research lines: food safety management and risk assessment (technical/mathematical compound of the broader framework of risk analysis related to food safety and human health impacts). The research of risk assessment is interacting with food safety management, as outcomes of risk assessment are applied as an input for the food safety management on operational level. Food safety culture is a current research topic in food safety management.



#### Lone Jespersen Cultivate Food Safety, Switzerland

Lone is a Principal at Cultivate, an organization dedicated to help food manufacturers globally make safe, great tasting food through cultural effectiveness. Lone has significant experience with food manufacturing, having previously spent eleven years with Maple Leaf Foods. Following the tragic event in 2008 when Maple Leaf products caused the loss of 23 Canadians, she led the execution of the Maple Leaf Foods, food safety strategy and its operations learning strategy. Prior to that, Lone worked for Woodbridge Foam as Engineering and Operations manager responsible for the safety and quality of automobile safety products. Lone holds a Master's in Mechanical Engineering from Syd Dansk University, Denmark, a Master's of Food Science from the University of Guelph, Canada and a Ph.D. on Culture Enabled Food Safety with Dr. Mansel Griffiths at the University of Guelph,

Canada. She has published extensively on cultural effectiveness and serves as chair of the GFSI technical working group on Food Safety Culture, a group dedicated to characterizing and quantifying food safety culture across the global food industry from farm to fork.



#### Reimar Johne Federal Institute for Risk Assessment, Germany

Reimar Johne studied veterinary medicine and currently works as a Scientist at the Federal Institute for Risk Assessment (BfR) in Berlin, Germany. Here he leads a research group and is the head of the German National Reference Laboratory for Foodborne Viruses. In addition, he is bestowed as an extraordinary Professor for Veterinary Virology at the University of Leipzig. He has participated in several research projects on zoonoses, the global food chain, virus contamination of food, and molecular characterisation of zoonotic viruses. His current research interests are hepatitis E virus transmission through the food chain, development of detection systems for viruses in food, and analysis of the zoonotic potential of enteric viruses.



#### Greg Jones Campden BRI, United Kingdom

Greg Jones is a Molecular Microbiologist by trade and has been working at Campden BRI for over 10 years. In that time he has overseen a wide range of research projects and private contract work. Research highlights include investigations into superchilling of food and the responses of *Clostridium botulinum* and *Listeria monocytogenes* to stress. Greg is also the author of a range of guidelines. Greg's current focus is on the applications of Next Generation Sequencing in Food Microbiology.



#### Ákos Jozwiak National Food Safety Chain, Hungary

Ákos Jozwiak is a Veterinarian, working for the National Food Chain Safety Office in Hungary. He is a member of the European Food Safety Authority (EFSA) Advisory Forum and member of the EFSA Emerging Risk Exchange Network. He is the head of the unit responsible for strategic and data analysis and emerging risk identification. In his research activities, the focus is on developing and applying new analysis methods for improving the effectiveness of the food chain safety controls. Within this domain, the main research areas are (1) applying computational science methods (mainly network analysis) for determining emerging risks and for improving risk-based controls; (2) epidemiological simulation of animal diseases; (3) determining the economic burden of foodborne diseases and applying health technology assessment methods for food chain safety decision making. Besides the work for the competent authority, he is involved in the education of veterinary and food engineer students.



#### Françoise Julien-Javaux Nestlé Research, Switzerland

Françoise Julien-Javaux is currently working as Senior Food Safety Microbiologist at Nestlé Research in Switzerland. She received her Ph.D. in 2000 from the University of Dijon, France, in the field of Food Microbiology. She then worked as Scientist at the Nestlé Research Center dealing with Probiotics and Lactic Acid Bacteria used in food products, before moving to Microbiological Food Safety and Quality in 2010. Since 2010, she has been focusing on generating and translating scientific data into practical recommendations to minimize the risk of microbial contamination from farm to fork. Dr. Julien-Javaux is the Reference Microbiologist in the Nestlé Early Warning System, for early identification of potential issues.



#### Kali Kniel University of Delaware, USA

Dr. Kali Kniel is a Professor in the Department of Animal and Food Sciences at the University of Delaware. She obtained her Ph.D. in Food Science from Virginia Tech in Blacksburg, Virginia and then served as a Research Microbiologist with the Animal Parasitic Diseases Laboratory at the U.S. Department of Agriculture Agricultural Research Service. Dr. Kniel leads research projects studying mechanisms of environmental persistence by zoonotic bacteria, protozoa, and viruses in pre-harvest agricultural environments focusing on water and soil amendments; as well as on the integration of food safety into educational programing across the K-12 and college curricula. She currently serves as President-Elect for the International Association for food Protection.



#### Marion Koopmans Erasmus University Medical Center, The Netherlands

Professor Marion Koopmans, DVM Ph.D. focuses on global population level impact of rapidly spreading zoonotic virus infections, with special emphasis on foodborne transmission. Her research focuses on unravelling the modes of transmission of viruses among animals and between animals and humans, and the use of pathogenic genomic information to unravel these pathways and to signal changes in transmission or disease impact. She is scientific coordinator of COMPARE, a large H2020 funded project (20 MEuro), exploring the potential uses of next generation sequencing techniques for outbreak detection and tracking (www. compare-europe.eu), and co-PI in the FP7 funded PREPARE project (www.prepare-europe.eu) aimed at building a pan-European

operational network for rapid and large-scale European clinical research in response to infectious disease outbreaks with epidemic potential. She is director of the WHO collaborating centre for emerging infectious diseases at Erasmus, and Scientific Director "Emerging infectious diseases" of The Netherlands Centre for One Health (www.ncoh.nl). She has received the Infectious disease award of the Dutch Association for Infectious Diseases and is the recipient of the Stevin Premium 2018. She has co-authored >500 papers that have been cited > 20.000 times.



### Lise Korsten

### University of Pretoria, South Africa

Professor Lise Korsten is currently the Co-Director within the Department of Science and Technology, Centre of Excellence Food Security. She is also responsible for the food safety and regulatory control programmes within the DST Centre of Excellence Food Security. She is an Editor of Crop Protection and is chairing the International Society for Plant Pathology Task Force on Global Food Security. Prof. Korsten has focussed her research mainly on complementary fields of postharvest technology and food safety as related to international trade in fresh produce. She has been able to establish research teams in food safety, postharvest pathology, biocontrol and mushroom and fruit health. As a team, they have been able to develop several innova-

tive technologies to reduce diseases and prevent product contamination. The value of her research programmes can best be illustrated by sustainable industry financial support. She has been able to attract extensive national and international long-term funding and has an extensive international network focusing on the plant microbiome. Prof. Korsten has developed one of the first biocontrol products in South Africa in 1992 that was patented, registered and commercialised and has since expanded her interest to microbial adaptations for disease control.



#### Lotta Kuuliala Ghent University, Belgium

Lotta Kuuliala is a post-doctoral Researcher with a joint affiliation at Research Unit Food Microbiology and Food Preservation (FMFP) and Research Unit Knowledge-based Systems (KERMIT) at Ghent University (UGent; Ghent, Belgium). The main focus of her current research activities is on data-driven food quality characterization within the context of smart packaging technology development. She holds an MSc in Technology (2012; chemistry) from Tampere University of Technology (TUT; Tampere, Finland) and a joint DSc in Technology and Ph.D. in Applied Biological Sciences (2018; materials science and food science) from TUT and UGent.



#### Solveig Langsrud Nofima, Norwegian Institute of Food, Fisheries and Aquaculture Research, Norway

Solveig Langsrud, Ph.D., is a Senior Scientist in the Department of Food Safety and Quality at Nofima, Norway, where she has been working for 25 years. Dr. Langsrud holds a B.S. in Biotechnology from Norwegian University of Science and Technology and a Ph.D. from the Norwegian University of Life Sciences, specializing in Food Microbiology. Her research has centred on how microbial communities in food production environments survive, adapt and affect food safety and quality. Recent activities have been transdiciplinary research on how consumer behaviors affect risk of foodborne illness. Dr. Langsrud is currently the Coordinator of a H2020 RiA on consumer food safety (www.safeconsume.eu) with 32 partners from 14 countries.



#### Maren Lanzl Wageningen University, The Netherlands

Maren Lanzl successfully completed her Bachelor of Science in the study "Food, Nutrition and Hygiene" at Albstadt-Sigmaringen University, Germany. She then completed her Master's of Science at Wageningen University, The Netherlands in the field of "Applied Food Safety." She conducted her MSc thesis at the Laboratory of Food Microbiology of Wageningen University focussing on the detection of *Campylobacter* spp. during enrichment. Since June of 2017, Maren works as a Ph.D. candidate at the Laboratory of Food Microbiology of Wageningen University on the topic "Rapid and Reliable Enrichment-based Detection of Foodborne Pathogens," focusing on *Campylobacter* spp.



#### Soizick Le Guyader IFREMER, Laboratoire de Microbiologie, France

Soizick Le Guyader has been a researcher in virology at the Ifremer Institute (Nantes, France) since 1992 where she has undertaken and managed a portfolio of research focused on human enteric viruses in the environment, with a special interest in norovirus. Following one year in the laboratory of M.K. Estes (BCM, Houston), she commenced a range of different research projects focused primarily on norovirus in sewage and shellfish, and also trace back investigations/analysis following norovirus outbreaks. The Ifremer Laboratory has significant expertise in detecting and localizing viruses within shellfish tissues and was the first to characterize the specific ligands for norovirus in oysters.



#### Jacques Le Pendu INSERM-Université de nantes, France

Dr. Jacques Le Pendu completed his Ph.D. thesis in 1984 in Paris and moved to Nantes in 1989 to start his own research group. He is currently Research Director at Inserm. His research interests have mainly been concerned with the biology of terminal glycosylations, most specifically those that constitute the so-called histo-blood group antigens (HBGAs). After working on the impact of these glycosylations in the field of cancer his interest became focused on deciphering interactions between enteric pathogens and HBGAs and their biological consequences. These include the effects of HBGA's polymorphisms on virus transmission, host-pathogen co-evolution and vaccine efficacy.



### Alvin Lee

### Institute for Food Safety and Health, Illinois Institute of Technology, USA

Dr. Alvin Lee is a Microbiologist and Virologist with more than 15 years research experience with a Ph.D. from RMIT University. Dr. Lee currently leads IFSH Center for Processing Innovation and co-leads the joint IFSH/FDA Microbiology Research Platform on food safety and defense related projects. He leads the Prevention and Control CORE of NoroCORE, a USDA-NIFA Food Virology Collaborative based at North Carolina State University and the IFSH Juice and Beverage Safety Task Force. Current research support includes funding from USDA, U.S. FDA and various industry contracts. Dr. Lee is an instructor for food microbiology in the Illinois Institute of Technology's Master's of Science program and has mentored more than 30 graduate students

and post-doctoral fellows. He is currently an active member of the International Association for Food Protection, American Society for Microbiology and Institute of Food Technologists.



#### Jeffrey LeJeune Food Safety and Quality Unit, AGFF, Food and Agriculture Organisation, Italy

Dr. Jeffrey LeJeune currently serves as Food Safety and Quality Officer, in the Food Safety and Quality Unit of the Food and Agriculture Organization of the United Nations (FAO) and the FAO Secretary for the Joint Expert Meeting on Microbial Risk Assessment (JEMRA). For over 20 years prior to joining FAO, Professor LeJeune worked in academia conducting research on the molecular and ecological mechanisms involved with the evolution, dissemination, and survival of zoonotic pathogens, notably Shiga toxin-producing *E. coli* and antimicrobial resistant bacteria. His veterinary training was completed in Canada, at the University of Prince Edward Island and his Ph.D., combined with specialty training in a veterinary diagnostics, was completed at Washington State University. He is board certified by the American College of Veterinary Microbiologist and also by the American College of Veterinary Preventive Medicine.



#### Fabienne Loisy-Hamon bioMérieux, France

Dr. Fabienne Loisy-Hamon has worked in virus foodborne safety issue for over 20 years. Following a Ph.D. in molecular virology, she founded in 2005 the private company Ceeram. This pioneer company was dedicated to help industrials to better understand and manage virus issue in food. She served for 10 years as a Chief Scientific Officer for Ceeram and develops a complete range of diagnostic solutions for virus detection in food and environmental samples. Ceeram Company received the IAFP Food Safety Innovation Awards for its worldwide expertise on foodborne viruses. After selling the Ceeram Company to bioMérieux, she joined this international group as Manager of Molecular Biology R&D in charge of the development of complete diagnostic tools for the detection of pathogens for the industry business. She is also part of several ISO experts groups in charge of virus detection methods in food and PCR detection methods for food and feed pathogens.



#### Niels Lucas Luijckx The Netherlands Organisation for Applied Scientific Research (TNO), The Netherlands

Niels Lucas Luijckx is a Biologist/Toxicologist who has worked for over 25 years in the domain of risk analysis and food safety. He has professionally covered risk assessment, risk management and risk communication. Since 2004 he has been working at TNO as a Senior Consultant and Researcher in Risk Management Food Safety with current focus on emerging risk identification (including food fraud), risk ranking and allergen cross-contamination. He currently is also involved in occupational safety risk management. Past positions include the RIVM, Ministry of Health, Ministry of Agriculture, and Schuttelaar & partners.



#### Ngoc-Du Luong SECALIM, INRA, Oniris, Université Bretagne Loire, France

Ngoc-Du Luong is currently a second-year Ph.D. candidate in Food Microbiology at SECALIM in Nantes, France, a research unit of INRA (French National Institute for Agricultural Research) and Oniris (Nantes College of Veterinary Medicine, Food Science and Engineering). After obtaining a MSc in Modelling and Computational Biology (Lyon, France), his research interests focused on biological data analysis and the development of statistical models to describe several biological processes. His Ph.D. project aims to acquire experimental data and to develop innovative modelling approaches for predicting the spoilage occurrence of meat products by taking into account various biotic (contamination by microorganisms) or abiotic factors (preservatives, packaging).



#### Balkumar Marthi DaQshConsulting, The Netherlands

Building on a strong technical and research foundation in Microbiology, Dr. Marthi developed leading capabilities in Hygiene, Food Safety, Risk Assessment & HACCP, Biotechnology & Sustainability. Dr. Marthi has been successful in building leadership positions in Food Safety in India, where he was one of the pioneers in implementing "Safety by Design" principles in the business. He was also instrumental in influencing adoption of these international principles by the Indian Government – through training, advocacy and participation in high-level Government Committees/Working Groups. Dr. Marthi has extended this reach considerably after moving to The Netherlands. Within Unilever, he led the Global Microbiology, Biotechnology, Preservation and Risk Assessment Expertise for the Foods Business. Dr. Marthi is the Company's Chief Food Microbiologit, thus responsible

for Preservation Innovation, Food Safety and Hygiene strategy, implementation and monitoring. He has also led activities to build bridges with the external scientific world as well as Government and Civil Society to drive strategic R & D and policy agendas in Microbiology & Food Safety. He has been very active in Food Safety-linked technical challenges and represent Unilever on Dutch & EU Programme Committees and initiatives in this area.



### Hans Marvin

### Wageningen University and Research, The Netherlands

Hans Marvin is a Senior Scientist at RIKILT Wageningen University and Research, The Netherlands. His research specializations are (i) methods for emerging risk identification and early warning, (ii) effect of drivers (among others climate change) on food safety, (iii) big data and application of Bayesian Networks in prediction models for food safety and food fraud, (iv) safety of engineered nanoparticles including stakeholders analysis (among others consumer perception), and (v) development of decision support systems. On these topics he has organized and chaired numerous workshops worldwide and is author and co-author of > 50 peer-reviewed scientific publications.



#### Peter McClure Mondelēz International, United Kingdom

Peter McClure gained his BSc and Ph.D. from Cardiff University and then joined the Institute of Food Research in 1985 in the UK, to work in the areas predictive modelling and microbiological food safety. He worked for Unilever for over 20 years, most recently in the Safety and Environmental Assurance Centre, as the Science Lead for Microbiological Safety. In 2014, he joined Mondelēz International as the Section Manager for Food Safety for Europe and was recently appointed as Global Food Safety Principal Scientist for Microbiology. He is responsible for overseeing microbiology-related matters linked to the food safety programme rolled out across the globe for Mondelēz, Peter is a member of the International Commission on Microbiological

Specifications for Foods, and the Advisory Committee on the Microbiological Safety of Food in the UK. He is a Co-Editor of *Foodborne Pathogens* (Woodhead Publishing) and *Food Microbiology* (Royal Society of Chemistry) and is a Visiting Professor at Leeds University.



#### Fady Mohareb Cranfield University, United Kingdom

Dr. Fady Mohareb is Senior Lecturer and Head of the Bioinformatics Group in AgriFood at Cranfield University, United Kingdom. He has over 12 years of experience in the application of cloud computing, machine learning and data science for food quality and safety. He is also leading the application of next generation sequencing informatics to unravel hidden patterns in crop genomics and transcriptomics.



#### Josep Molas Pages Coca-Cola, Spain

Josep Molas Pages is a Water Specialist with more than 30 years of experience in the food and drink industry. He has focused his activity in the bottled water industry and water as ingredient for foodstuffs where he has worked in manufacture, quality assurance, food safety and environment, lab management, product development, regulatory and scientific affairs and work with external stakeholders and technical associations. He is a member of technical committees of the European Federation of Bottled Waters, Belgian Federation of Beverages, Spanish Association of bottled Waters, the EU Water Technology Platform WssTP, and of EHEDG technical group. He has participated in the revision of Codex Standards and Code of Practices

related to Mineral Waters, and was expert reviewer of the ILSI guideline for water recovery and reuse in beverage production and food processing. Recently has participated in the revision of EHEDG standard for safe and hygienic treatment, storage and distribution of water in food facilities. He has a wide knowledge of hydrogeology, chemistry and microbiology of water, water treatments, and water management practices in the food industry. He developed an international career within Groupe Danone and since 2005 in The Coca-Cola Company with the actual role of Director, Product Water.



#### Kirsten Mooijman Convenor of the ISO 6579 standard, EURL on *Salmonella* spp., RIVM, The Netherlands

Since 1986, Kirsten Mooijman has worked as a Microbiological Researcher at the RIVM in several European projects, like the development and certification of reference materials for water and food microbiology, validation of microbiological methods. Since 2003, she has headed the European Union Reference Laboratory (EURL) for *Salmonella*, as well as the Dutch National Reference Laboratory (NRL) for *Salmonella*. She is an active member of ISO/TC34/SC9 and CEN/TC275/WG6 and convenor/ project leader of working groups under ISO and CEN, e.g., for drafting and revising EN ISO documents for detection, enumertion and (sero)typing of *Salmonella* (EN ISO 6579 parts 1 to 4).



#### Nicolas Nguyen Van Long ADRIA Food Technology Institute, France

Nicolas Nguyen Van Long was trained as a Food Safety and Quality Manager and obtained his Ph.D. in 2017 on the physiology of fungal spoilers and predictive modelling at Université de Bretagne Occidentale. Since then, he has been employed as Project Manager in ADRIA food expertise institute in Quimper where he enjoys developing tools and methods for predictive mycology in the frame of UMT 18.03 ALTER'IX. He is also involved in the ADRIA's activity of Expert Laboratory in Alternative Method Validation for Afnor, MicroVal and AOAC certification and is an active member of ISO Working Group 3 – Method Validation.



#### Dirk Nikoleiski Commercial Food Sanitation, Germany

Dirk Nikoleiski is Senior Food Safety Specialist for Commercial Food Sanitation (CFS, an Intralox Company). Before he joined CFS he gained 30 years of experience in both the manufacturing and corporate environment at Kraft Foods/Mondelēz International. He has been instrumental in continuous improvement and development of policies, standards and guidelines, as well as providing project support and training to the organization over the years. Dirk has written many articles for publications, written a book on Hygiene in Food Manufacturing and contributed chapters to various books. Dirk holds a Diploma in Food Technology from the University of Applied Science Lippe-Lemgo, Germany.



#### Eugenio Parente Università degli Studi della Basilicata, Italy

Eugenio Parente is Full Professor in Applied Microbiology at the Università degli Studi della Basilicata, Potenza, Italy since 2002. He is a member of the editorial board of the *International Journal of Food Microbiology and Food Microbiology*. His main research interests are food microbial ecology (analysis of the structure of food bacterial communities by 16S metagenomics) and food fermentations (aerobic metabolism and stress response in lactic acid bacteria). ORCID 0000-0002-5716-2348.



#### Nicole Pavio ANSES, France

Nicole Pavio is a Research Director at the French Agency for Food, Environmental and Occupational Health & Safety in the Animal Health Laboratory where she started to work on animal reservoirs of zoonotic hepatitis E virus in 2005. She was appointed by EFSA as an expert on HEV and she is member of the French BIORISK scientific expert panel for risk assessment of viruses in food. Dr. Pavio obtained her Ph.D. at the Pasteur Institute of Paris in 1996 in virology and completed a post-doctorate on hepatitis C virus at the University of Southern California, USA. Back to France she, continued to study HCV at the National institute of medical research and then switched to HEV at ANSES.



#### Flavio Pellegrinuzzi Euroconsumers Cooperative Partner Group of Independent European Consumer Organizations, Luxembourg

Flavio Pellegrinuzzi is Social Researcher and Project Officer at the Statistical Surveys Department of Altroconsumo, an Italian consumers organization that is part of Euroconsumers. His research activity is based on a consumer-centric approach and aims to analyze people experiences, attitudes, opinions and problems in order to provide guidance to the consumers in their daily life choices.



#### Anil Persad University of the West Indies, Trinidad and Tobago

Anil Persad is a Lecturer in Large Animal Medicine (Food Bias) at the School of Veterinary Medicine, University of the West Indies. He completed his DVM in 2007 from the University of the West Indies, before proceeding on a Fulbright Fellowship in 2011 to The Ohio State University where he completed the MS (2013) and Ph.D. (2016) programmes. His research focused on the epidemiology of bacterial foodborne pathogens. His career goal is to improve the standard of healthcare provided to food animals and additionally increase pre-harvest food safety in the Caribbean and Latin America.



#### Annemarie Pielaat Unilever R&D, The Netherlands

Annemarie Pielaat works as a Science Team Leader in Microbiological Risk Assessment for Unilever R&D in The Netherlands. She has a Master's in Mathematical Biology in the field of ecotoxicology from the Free University of Amsterdam. Her Ph.D. graduation was in phytopathology modelling at the Wageningen University after which she had a 2-year post-doc position in the mathematical biology group at the University of Alberta. In 2003, she joined the Dutch Institute for Public Health and the Environment (RIVM). Her main interest is in connecting people in research through setting-up biologically relevant experiments, sampling plans and subsequent statistical data analysis as input for Microbiological Risk assessment. The last few years her interest extended towards methodology development for the implementation of molecular data in microbiological risk assessment

of foodborne microorganisms, resulting in a recent (2019) publication in *Front. Microbiol.* 9:3182. doi: 10.3389/fmicb.2018.03182.



#### Florence Postollec ADRIA – UMT ACTIA19.03 ALTER'iX, France

For the past 14 years, Florence Postollec has collaborated with the Mafart Team on risks associated to foodborne sporeformers within the frame of UMT ACTIA competitive national cluster. This collaboration, based on shared Research & Developpement axis, aims at increasing knowledge and expertise to better mitigate sporeformer contaminants involved in food safety and spoilage issue. Her main interest relies in biodiversity and behaviour heterogeneity induced after stress exposure. She is involved in the developpement of applied scientific projects or services related to sporeformer hazard identification, process or shelf-life optimization in close collaboration with food industrials. She was trained as biochemist, obtained a Ph.D. degree in

bacterial interactions at the faculty of medical sciences in Groningen (NI) and gained experience in molecular microbiology when she integrated ADRIA as a post doc working on the detection and identification of sporeformers involved in food spoilage.



#### Padmini Ramachandran Center for Food Safety and Applied Nutrition, U.S. Food and Drug Administration, USA

Padmini Ramachandran is a Staff Fellow in Division of Microbiology at FDA's Office of Regulatory Science. Her research interests include using metagenomics to track foodborne pathogens, species identification of insects and plants using organelle genomes. She also specializes in data visualization and data modeling of next gen sequencing data. She also works to provide target and non target metagenomic data to describe ecologies associated with high risk crops and foods.



#### Peter Rinke SGF International e.V., Germany

Dr. Peter Rinke studied food chemistry at the Technical University Berlin and biochemistry at University Paris VI. His Ph.D. thesis about flavour compounds of hydrolysed vegetable proteins was received at Technical University Berlin. His professional experiences are composed by three years as Project Manager at CPC-Europe, Heilbronn, Germany; six years as Head of Laboratory, specialised in compositional fruit juice analytics at Eurofins, Nantes, France; and since 2000 as Technical Manager at SGF International e.V., Nieder-Olm, Germany. After an interruption of a one year mission as quality manager at Klaus Böcke Group, since 2015 he keeps the position Head of R&D at SGF. He is driving research and development in fruit juice analytics and industrial food fraud mitigation.



#### Lisa Röttjers Laboratory of Molecular Bacteriology, Belgium

Lisa Röttjers is Ph.D. student of Karoline Faust. Although she was originally trained as a (experimental) Synthetic Biologist, she always found the emergence of specific properties in biological systems interesting. Her current work focuses on emergence of hub species and clusters in microbial communities.



#### Imca Sampers Ghent University, Belgium

Imca Sampers is Associate Professor at Ghent University and is responsible for the preparation, organisation and supervision of practical and theoretical courses in the framework of the Master's of Science in the bio-industrial sciences: circular bioprocess technology (specifically food process technology, microbiology, quality systems in the food industry). She also participates in scientific and public services related to the training programme and participates in applied research projects in the field of food, water and technology with a focus on microbial and chemical quality, both at national and international level. She is Project Leader in EU projects such as VEG-i-TEC and Project Leader of several other industrial (subsidised) projects (for the

food industry) and as such also supervisor of scientific researchers and Ph.D. students. She is the Lead Author and co-author of several publications and actively participated in (inter)national congresses and symposia.



#### Moez Sanaa ANSES, France

Moez Sanaa, DVM, Ph.D., is currently leading the Food Safety Risk Assessment Unit, French Agency for Food, Environmental and Occupational Health & Safety (ANSES). He has intensive activities on risk assessment research and innovation as well as important international activities including his role as a member of EFSA scientific expert panels (Animal Health Animal Welfare panel 2006–2012 and Biological Hazards panel 2012–2018) and involvement in different building capacities and training programs in risk assessment and surveillance related to Food Safety (he trained nearly 1000 scientists from more than 30 different countries). Doctor Sanaa has worked within the U.S. FDA CSFAN Office of Analytics and Outbreak in the Division of Risk

and Decision Analysis (November 2017 to December 2018).



#### Gloria Sánchez Institute of Agrochemistry and Food Technology (IATA-CSIC), Spain

Dr. Gloria Sánchez is a Senior Scientist at the Institute of Agrochemistry and Food Technology (IATA-CSIC) where she leads the activities on food virus safety. Her research is focused on developing molecular methods for human enteric virus detection in food and water, and evaluating the effectiveness of food processing on such viruses, mainly noroviruses and hepatitis A and E viruses. She has published more than 80 scientific papers and book chapters in international journals and publishers. She was the head of the Enteric Virus Group at Nestlé Research Center (Switzerland) for 5 years. In the field of foodborne viruses, she collaborated on international cooperation projects supported by the European Committee for Standardization (CEN) and the program for European

cooperation in science and technology (COST), and currently, participates in research projects, courses and teaching.



#### Lea Sletting Jakobsen Technical University of Denmark – DTU, Denmark

Lea Sletting Jakobsen is a Postdoctoral Fellow at the National Food Institute, Technical University of Denmark (DTU). She has a Master's degree in Food Science and Technology from the University of Copenhagen and a Ph.D. from DTU Food. Lea's main research area is quantitative health impact assessment of food. She has in her Ph.D. and as postdoc developed models for estimating the disease burden of food-associated diseases with a focus on chemical exposures as well as models for risk benefit assessments of foods considering both nutritional and chemical risk factors.



#### Daniele Sohier Bruker, Germany

Daniele Sohier has recently moved to Bruker Daltonics (Ge) (www.bruker.com) to coordinate development programs in industrial microbiology. She has spent more than 15 years at Adria Food Technology Institute (Fr), at first to implement molecular microbiology and then to manage the overall activities in food microbiology, combining both R&D and expertise projects for food and diagnostic companies. She is involved in ISO standardization and certification schemes of alternative methods, and has presented over 90 communications on food microbiology in journals and at symposia. She is the chair of the Organising Committee for IAFP's European Symposium on Food Safety.



#### Pietro Stella European Food Safety Authority (EFSA), Italy

Pietro Stella is employed at EFSA, Unit on Biological Hazards and Contaminants (BIOCONTAM), as Team Leader of Biological Hazards (BIOHAZ). Originally from Italy, he is a veterinarian, with an MSc on veterinary epidemiology and public health. He has worked as a practitioner and in the meat sector. He joined EFSA in 2007 and since then he provided support to the BIOHAZ Panel, coordinating its working groups dealing with topics related to food safety, microbiological risk assessment, transmissible spongiform encephalopathies and antimicrobial resistance. He coordinates the activities of the BIOHAZ Team and BIOHAZ Panel.



#### Zoltan Syposs Coca-Cola HBC, Austria

Zoltan Syposs graduated at the Corvinus University of Budapest as Msc. Horticultural & Food Engineer. During his tenure with Det Norske Veritas Certification Body, he played a key leadership role in the establishment of the DNV Food/Agri Sector Product & System Certification. In 2000, he joined The Coca-Cola Company as Region Quality Manager. His career development included various Leadership roles in Quality, Manufacturing and Supply Chain operations across the Coca-Cola Hellenic territories. Currently, he is a member of the CCHBC Group SC Senior Leadership Team and holds a strategic position as Group QSE and R&D Director, responsible for 28 country operations. He has completed his research and Ph.D. studies in Microbiologi-

cal Risk Assessment at the Corvinus University of Budapest between 1999–2004. Since 2016 he has been serving as an Associate Professor at the Szent Istvan University in Budapest at the Department Food Safety and Microbiology supporting the development of educational programs as well as bringing Academia and Industry closer together.



#### Helene Maria Fiane Teigen Consumption Research Norway, SIFO, Oslo Metropolitan University, Norway

Helene Maria Fiane Teigen is currently a Ph.D. candidate in consumer studies at Oslo Metropolitan University. Her educational background is in media studies; however, for the past two years, she has worked at Consumption Research Norway at OsloMet and has participated in several projects. Her main contribution has been to the EU funded-project SafeConsumE about food safety, where she, among other things, has participated in the qualitative fieldwork of identifying consumers' practices related to food safety.



#### Paula Teixeira Universidade Católica Portuguesa, CBQF – Centro de Biotecnologia e Química Fina – Laboratório Associado, Escola Superior de Biotecnologia, Portugal

Paula Teixeira is a graduate of Escola Superior de Biotecnologia – Universidade Católica Portuguesa (ESB-UCP) with a B.Sc. in Food Engineering and a Ph.D. in Biotechnology. Associate Prof. with Aggregation at ESB-UCP and lead of the CBQF research group Food and Nutrition, Paula Teixeira has participated in and has led national and international projects in the area of Food Microbiology/Food Safety, has been author/co-author of several book chapters and more than 150 articles in peer-reviewed journals. She has supervised several externally funded research fellowships, as well as 17 and 39 successfully concluded Ph.D. and Master's thesis, respectively. She has been Associate Editor of *Frontiers in Food Microbiology*, Member of the Editorial Board of *International Journal of Food Microbiology and Food Microbiology*.



#### Sebastian Ulrich Ludwig-Maximilians-University, Germany

Sebastian Ulrich received his degree at LMU Munich. He has been a Veterinarian since 2013. Since 2016, he has been the Chair of Food Safety at LMU Munich. In 2018 he became Veterinarian Specialist for Meat Safety and the Habilitation on the Characterization of *Stachybotrys* spp. 2013–2019. He was recently promoted to Senior Scientist and Chair of Food Safety Veterinary faculty LMU Munich, Academic Officer at the Chair of Microbiology and Mycology Veterinary faculty LMU Munich.



#### Tom Van den Brule Westerdijk Fungal Biodiversity Institute, The Netherlands

Tom Van den Brule obtained his Master's degree in Environmental Biology at Utrecht University. During his internships in the Molecular Microbiology group of Prof. Dr. Wösten and the Microbiology group of Unilever R&D, he became interested in fungal food microbiology. Currently he works as a Ph.D. student in the TiFN project 'Heterogeneity in spores of food spoilage fungi.' TiFN is a platform that connects industry with academia. In this project, spore variability is related to differences in stress resistance. Expertise of various institutes in the fields of predictive food microbiology, molecular biology and genetics is used.



#### Wim Van der Poel Wageningen University, The Netherlands

Prof. Wim H.M. Van der Poel, DVM, Ph.D., is Senior Scientist at Wageningen Bioveterinary Research and special Professor of 'Emerging and Zoonotic viruses' at Wageningen University. He is a Principal Investigator within The Netherlands Centre of One Health (NCOH), member of the Project Management Board of the European Joint Program One Health (EJP One Health) and Coordinator of the EPIZONE European Research Group, the network on epizootic animal diseases research. The research work of Prof. Van der Poel involves at least three main areas: New and emerging viruses, Foodborne and Zoonotic viruses, icluding Hepatitis E virus, and 'Global One Health'.



#### Carol Anne Wallace University of Central Lancashire, United Kingdom

Carol Wallace is Professor of Food Safety Management Systems and Co-Director of the International Institute of Nutritional Sciences and Applied Food Safety Studies at the University of Central Lancashire. Carol's background is in microbiology, HAC-CP and applied food safety. Her career spans more than 30 years in the food industry and food safety education; including 20 years in food manufacturing, retailing and consultancy prior to joining academia. Carol is a world authority on the HACCP system and has active research interests in food safety culture. She is Chair of Salus, the Food Safety Culture Science Group, an international academic research network.



#### Rong Wang U.S. Department of Agriculture-ARS, USA

Rong Wang joined USDA in 2010 as a Microbiologist. His research focuses on bacterial biofilms, sanitizer effectiveness, and molecular mechanisms associated with strong biofilm formation and high sanitization tolerance. He also investigated multispecies biofilms and bacterial stress tolerance. His recent studies suggested that biofilm formation and sanitizer tolerance might play roles in product contamination at meat plants, such as "High Event Period" contamination, and mixed biofilms by certain environmental microorganisms might protect pathogens against sanitization and increase pathogen prevalence. These studies may help develop strategies for the industry to prevent product contamination and enhance meat safety.



#### Wolfram Wendler Arotop Food & Environment GmbH, Germany

Wolfram Wendler was born in Solingen, Germany. He received his training as a chemical laboratory technician and chemo technician. His focus is the study of food chemistry in Wuppertal; Construction of a laboratory in the baking industry; Quality management in the confectionery industry; Ph.D. at the University of Marburg; Development of residue analysis for a Mannheim laboratory; since 2012 at 'arotop food and environmen GmbH' as deputy institute director; from 2016 scientific director of the research project 'HaGen' of the Federal Ministry of Food and Agriculture.



#### Anett Winkler Cargill, Germany

Anett Winkler joined Kraft Jacobs Suchard in December 1998 to head up the research microbiology laboratory in Munich. Later, Anett concentrated on chocolate, biscuits and other low-moisture foods including supplier developments and approvals. She also consolidated the scientific basis for microbiological process controls in low-moisture foods by performing validation studies for nut and cocoa processing. Following a regional role for Microbiology in the Eastern European, Middle East and African Region, she was globally designing food safety programs, rolling out training modules related to food safety and further supporting supplier development. Anett was also the Global Expert for thermal processing within Mondelez International. In

October 2017, Anett moved to a new position as EMEA Regional Food Microbiologist Lead at Cargill.



#### Xianqin Yang Agriculture and Agri-Food Canada, Canada

Dr. Xianqin Yang is a Research Scientist of Agriculture and Agri-Food Canada (AAFC) stationed in Lacombe, Alberta. She has over 12 years of experience of research on various aspects of meat microbiology. After obtaining a Ph.D. in microbiology from the University of Waterloo in 2007, Dr. Yang worked for three years as a Visiting Fellow at the Lacombe Research Centre before joining AAFC as a Research Scientist. Her current research focuses on reduction of microbiological contamination on meat during production and distribution processes, including tracking and control source of contamination and mechanisms of survival and persistence of enteric pathogens.



#### Sophie Zuber Nestlé Research Centre, Switzerland

Sophie Zuber works as Food Safety Microbiologist at the Nestlé Research Centre, based in Lausanne, Switzerland. She received her Ph.D. in microbiology from the Department of Genetics, University of Melbourne, Australia. In her current position, her principal responsibilities include providing scientific advice and guidance on possible risks of viruses in the food chain and developing risk management strategies in this field. In this context, Dr. Zuber has published peer-reviewed publications focusing on the effects of treatments used in food processing on viruses.



#### Marcel Zwietering Wageningen University, The Netherlands

Marcel Zwietering studied biotechnology at Wageningen University and after his Ph.D. in 1993 worked in the Food Process Engineering group as Assistant and Associate Professor. From 1998 – 2002 he worked for the research lab of Danone in France. Since January 2003, he is a Professor in Food Microbiology at Wageningen University. Marcel is Editor of the *International Journal of Food Microbiology* and member of the International Commission on Microbiological Specifications for Foods (ICMSF). Prof. Dr. M.H. Zwietering Personal page: http://www.wageningenur.nl/en/Persons/Marcel-Zwietering.htm Laboratory: http://www.fhm. wur.nl.

# **PAST MEETINGS AND LOCATIONS**

2005	Prague, Czech Republic	2012	Warsaw, Poland
2006	Barcelona, Spain	2013	Marseille, France
2007	Rome, Italy	2014	Budapest, Hungary
2008	Lisbon, Portugal	2015	Cardiff, Wales
2009	Berlin, Germany	2016	Athens, Greece
2010	Dublin, Ireland	2017	Brussels, Belgium
2011	Ede, The Netherlands	2018	Stockholm, Sweden



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# SYMPOSIUM ABSTRACTS

24–26 April 2019 – Nantes, France

# **SYMPOSIUM ABSTRACTS**

#### **Opening Session**

Food Safety Risk Assessment Policies and Procedures at the U.S.-Food and Drug Administration (FDA), European Food Safety Authority (EFSA) and French Agency for Food, Environmental and Occupational Health & Safety (ANSES) MOEZ SANAA, ANSES, Maisons, France

This presentation will cover aspects related to the positioning of microbial and chemical risk assessment in the paradigm of risk analysis as applied in three main food safety organizations. Risk assessment integrates diverse types of data into mathematical models to simulate or describe situations representing food contamination or interventions at any stage in the food's production, supply, and consumption chain. Different approaches of risk assessment, top-down or bottom-up allow the estimation of public health impacts resulting from each situation and by the way the possibility to predict/compare effectiveness of different possible interventions, rank foodborne risks and suggested management options or interventions. Risk Assessment policies and procedures applied at the US-FDA, EFSA and ANSES may differ on certain aspects such as the formalization of the risk assessment requests, the use of internal and/or external experts and the validation procedures of the scientific reports or opinions. However, in all three organizations the end goal is the same. It is about strengthening our ability to inform the decision-makers by providing them conclusions based on the best available scientific data. In the event of uncertainty related to the lack of data or the poor quality of the available data, EFSA and ANSES have made a lot of effort and put in place procedures that to make the risk assessment process as transparent as possible. The concern for transparency and harmonization on the part of the Europeans seems to some to be an obstacle to the creativity that remains necessary for any risk assessment activity. A compromise between an effort of harmonization and the possibility of innovation is essential

#### Update on Monitoring Activities of European Union Reference Laboratory for *Listeria monocytogenes*: Multi-EU Country Outbreak Investigation and Integration in the EU/EEA-wide Monitoring Organization

**BENJAMIN FÉLIX**, ANSES, University of Paris-Est, Maisons-Alfort, France

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E. Leinonen<sup>7</sup>, M. Simola<sup>7</sup>, Z. Sréterné Lancz<sup>8</sup>, O. Szalay<sup>9</sup>, M. Torresi<sup>10</sup>,
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Since 2006, the European Union Reference Laboratory for *Listeria monocytogenes* (EURL *Lm*), hosted by the French Agency for Food, Environmental and Occupational Health & Safety (ANSES) and with financial support of European Commission (DG SANTE), manages a network of 41 National Reference Laboratories (NRLs) in 32 European countries. Most of these NRLs are in charge of non-human strain typing in their countries.

EURL *Lm* contributes to the EU/EEA-wide surveillance database system, which is set up by the European Centre for Disease Prevention and Control (ECDC)<sup>1</sup> and the European Food Safety Authority (EFSA). This database currently collects Pulsed Field Gel Electrophoresis (PFGE) data on human and non-human strains of *Lm*, Shiga toxin -producing *Escherichia coli* and *Salmonella*. EURL *Lm* is part of the steering committee for the joint database and in charge of the curation and cluster analysis for the non-human *Lm* strains. For three years, the EURL *Lm* has been requesting NRLs to centralize and compare the typing data (PFGE and recently whole genome sequencing (WGS)) of their non-human strains to identify outbreak contamination sources.

Since 2017, EURL *Lm* contributed to the investigation of nine multi-EU country foodborne outbreaks in collaboration with the NRLs, ECDC and EFSA. Among these nine outbreaks, three investigations enabled to trace back the contamination source <sup>2</sup>, four did not lead to source identification<sup>5</sup>, and two are still under investigation.

In one of these outbreaks, the national competent authorities identified first matches between human outbreak reference strains and food strains by core genome Multi-locus Sequence Typing (cg-MLST). Further comparison by PFGE either by the NRLs or in the ECDC-EFSA database<sup>1</sup> confirmed an ongoing multi-country outbreak. Source identification triggered large recalls of food products and extensive control measures in the processing plant, which gradually led to the reduction of outbreak-related human cases<sup>2</sup>.

In the frame of these EU-wide multi-country investigations, EURL *Lm* has provided support to eight NRLs for WGS typing of 116 strains. The ECDC-EFSA database proved to be useful but EU will benefit substantially from integration of WGS data in the future.

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# S1 CulturOmics: The Revival of Microbiological Culture

In the past decade, NGS studies have become widespread thanks to the second generation and low-cost sequencing platforms using short read lengths. Metagenomics was originally designed to fill in the gaps left by culture-based methods, such as detecting minority communities in microbiota or communities that cannot recover on the current panel of culture media. But finally, it was quickly clear that a bias is as well observed. The extraction protocols and primers might be more suitable to detect certain groups of microorganisms than other ones, and basically the approach fails as well in detecting low bacterial loads.

But Great News! The OMICS family is happy to announce the new-borne CulturOmics... Or it might be basically the rebirth of culture in microbiology. CulturOmics is definitely complementing metagenomics for investigating the biodiversity and evolution of microbiomes, expanding the described species. CulturOmics combines extensive culture conditions and MALDI-TOF MS to easily and quickly identify the isolates, followed rDNA sequencing of the non-identified colonies.

The culture-independent metagenomics has strongly modified the concepts of microbial diversity and species richness. Besides, it has changed the way of thinking microbiology. The advantages and limitations of metagenomics studies on food microbiomes will be summarized, before getting into to the substances of CulturOmics.

The pioneering CulturOmics studies started more than 5 years ago, in order to investigate the human gut microbiota. The microbial repertoire of this microbiota has been dramatically increased. The approach, the proof of concept will be first introduced, followed by illustrations on human gut microbiota.

# Analysis and Interpretation of NGS Data Sets for CulturOmics

HENK DEN BAKKER, Center for Food Safety, University of Georgia, Griffin, GA, USA

The introduction of automated high-throughput sequencing technologies has greatly improved our understanding of the role microbiomes in health, ecosystems and increasingly in food safety and food production systems. Additionally, these studies have unveiled a large diversity of microbial live that had previously gone unnoticed and until the introduction of CulturOmics approaches was largely assumed to be unculturable. NGS sequencing approaches used to study microbiomes can be largely categorized into two groups; (i) amplicon-based approaches and (ii) shotgun DNA sequencing approaches. Amplicon-based approaches consist of PCR amplification of a conserved region of a large taxonomic group within the microbiome, e.g., 16S rRNA for prokaryotes or ITS rRNA for eukaryotes. Shotgun sequencing consists of sequencing all DNA from a given sample is sequenced. The amount of information that can be derived from each class of microbiome data differs tremendously. While amplicon data can mainly be used to infer phylogenetic diversity, shotgun metagenomic data can additionally be used to reconstruct microbial genomes, infer metabolic pathways of individual members of a microbial community and many more interesting aspects. Both types of data can be used to estimate the 'microbial dark matter' in a sample, and both taxonomic and genomic information obtained from the bioinformatics analysis of NGS data can be used to improve the efficiency of a potential CulturOmics workflow.

#### CulturOmics: The Revival of Microbiological Culture

NOÉMIE DESRIAC, LUBEM UBO University -UMT14.01SPORE RISK, Quimper, France, France and FLORENCE POSTOLLEC, ADRIA–UMT ACTIA19.03 ALTER'IX, Quimper, France

Microorganisms have been around for billions of years and have adapted to nearly every environment in earth. They account for most of the diversity encountered on our planet. Since Pasteur and Koch, the systematic inventory of microbial community was based on the detection and description of microorganisms after a culture step to obtain visible colonies. More recently, metagenomics studies are performed to investigate, microbial communities within an ecosystem. These studies are unravelling our understanding of microbiology communities by using uncultured-based techniques, however, genomics technologies are facing the difficulty to access and easily detect minority population in complex ecosystem such as the gut microbiota which consists of ~1012 bacteria per gram of stool. The polyphasic approach of CulturOmics is initiating the rebirth of culture in microbiology. In this context the pioneering CulturOmics studies started to investigate the gut microbiota and have permitted the isolation of hundreds of new bacterial species from the gut in about 5 years. Furthermore, the complementarity between metagenomics and CulturOmics was demonstrated, because only 15% of species detected were detected simultaneously by the 2 techniques. This talk will first introduce the concept of CulturOmics, followed by the illustrations on human gut microbiota but as well discussion on the potential of this new approach in food microbiology.

## How Can CulturOmics Complement Metagenomics to Investigate the Microbial Biodiversity in Food Microbiology? LUCA COCOLIN, University of Torino-DISAFA,

Grugliasco, Italy

With the advent of culture-independent molecular approaches, the study of the ecology of foods has changed dramatically. From colonies on the plates, we are now studying nucleic acid sequences for both identifying the microbial ecological structure (metataxonomics) and unraveling the potential metabolic functions of the microorganisms (metagenomics). By using these techniques, we can deeply investigate complex microbial ecosystems and understand how the different microbial populations are interacting and contributing to fermentation and spoilage processes. The challenge that we are facing now is how to exploit the knowledge achieved through culture-independent approaches to improve the quality and safety of our foods.

Another aspect relates to the fact that due to speed in the advancement of the technology (e.g., sequencing), we have somehow forgotten about the importance of isolating those populations and strains, which are responsible for the most important activities in a given microbial ecosystem. It is well known that a large proportion of the microbial cells are in a viable not culturable state (VBNC) and it becomes pivotal to improve the culturing methodologies in order to allow microorgansims to grow on the plates.

In this talk, we describe a CulturOmic approach to investigate microbiota and microbiome of fermented sausages. Following a shotgun metagenomic approach, combined with culturing, isolation, identification and characterization of isolated populations, we aim at describing in detail the complexity of this fermentation process. It is essential to underline how collected data must be integrated and analyzed with innovative approaches to achieve the objective of understanding food microbial complexity and diversity.

# S2 Microbiological Hygiene and Food Safety in Primary Production and Processing of Fresh Produce – From Science to Easily Understandable Recommendations for Farmers and Suppliers

Fresh produce such as berries and leafy greens have been linked to microbiological outbreaks in different places of the world resulting from produce contamination with pathogenic bacteria, viruses or parasites. In some of these outbreaks, contamination was traced back to inappropriate practices at farm level, highlighting the need for mitigation strategies at primary production and processing. In this symposium we propose to discuss mitigation strategies at farm level and at primary processing such as during washing which is often applied to high risk produce. We aim at showing the importance of an "assessment-education-continuous improvement" process through practical training materials and assessment tools targeting each actor of the fresh produce supply chain, based on scientific knowledge.

The development of these training and assessment tools requires expert scientific knowledge to understand the criticality of each element and the capability to translate scientific knowledge into recommendations. Recommendations need to be rolled-out in a comprehensive way to food safety authorities/auditors/assessors and to growers/growers' associations to ensure that (i) they understand the impact of those strategies on the safety of the produce they are growing and/or (ii) they know how to implement these strategies in a practical, sustainable and cost-effective way. As each farm has a distinct combination of environmental risk factors and each grower has a distinct level of knowledge, it is key to develop adapted recommendations which are (i) structured by risk factors, (ii) expressed or translated to be easily understandable, (iii) precise enough to tell the growers how to achieve what is requested and (iv) feasible in terms of implementation.

This session will bring together actors from academia and the agro-food chain (industry) who will illustrate this process with practical examples and will hopefully lead to further discussions and initiatives between the scientific community and the industry.

## Regulatory Perspectives of Hygiene in Fresh Produce Production: Challenge of Microbiological Quality of Water

**KRIS DE SMET**, European Commission, Brussels, Belgium

Requirements on hygiene of food are laid down in Regulations ensuring a high level of consumers' protection anywhere in the EU and fair competition in a single market. Except for some specific microbiological criteria, hygiene rules in Regulation (EC) No 852/2004 on food of non-animal origin, including fresh produce, are general and include good hygiene practices at any stage of production, and procedures based on the HACCP principles except for primary production. Binding microbiological criteria for water used in the production of fresh produce don't exist, but measures must be in place to control potential contamination of water by biological hazards.

The need to reconsider rules on the hygiene of products of non-animal origin was triggered by the STEC O104:H4 outbreak in 2011. Apart from an opinion on risks in sprouts, the European Food Safety Authority (EFSA) provided six additional opinions on risks posed by pathogens in food of non-animal origin: one containing a risk ranking of food/ pathogen combinations, the others providing risk assessment on specific food/pathogen combinations of a high public health concern.

The opinions confirmed good hygiene practices at all stages as the primary objective for food safety. Prevention of contact with faeces by, among others, all kinds of water supply, was indicated as the main mitigation option for reducing the risk of contamination with pathogens such as *Salmonella* or Norovirus.

To address the EFSA recommendations and facilitate the application of the general hygiene rules, the European Commission adopted in 2017 a guidance document addressing microbiological risks in fresh fruit and vegetables at primary production. The guidance document pays specific attention to the use of water in fresh fruit and vegetables production. Such guidance includes a practical way for the risk assessment of the water by the use of a matrix and/or decision tree.

#### Microbial Risk Mitigation in Primary Production of Produce: From Scientific Knowledge to Implementation of an "Assessment-Education-Continuous Improvement" Process

FRANÇOISE JULIEN-JAVAUX, Sophie Zuber, Nestlé Research, Lausanne, Switzerland

Within a robust food safety management system, safety must be assured through the entire food supply chain and therefore must start at primary production, in the growing field. Microbiological contamination in primary production is linked to several risk factors or "contamination routes" that can be structured in different ways, including: growing field and adjacent land; animals; manure-based soil amendments; agricultural water; hygiene and human health; worker harvesting practices; equipment, premises and transportations. Therefore, Good Agricultural Practices (GAPs) must be in place during farm activities, to mitigate the risk posed by each of these routes. As each farm has a distinct combination of environmental risk factors and each grower has a distinct level of knowledge, it is key to develop GAPs/ recommendations which are (i) structured by risk factors, (ii) expressed or translated to be easily understandable, (iii) precise enough to tell the growers how to achieve what is requested and (iv) feasible in terms of implementation. In this presentation, we will discuss the development of such recommendations, focusing on microbial risk mitigation strategies for agricultural water, with an industry perspective. Agricultural water has been shown to be responsible for several produce contaminations and outbreaks in different regions of the world, linked to different types of water sources and of water applications. Development of recommendations and standards to manage the use of agricultural water is key to mitigate this risk. Finally, we will show, through a practical example on berries, the importance of implementing an "assessment-education-continuous improvement" process with the commitment from all stakeholders, hence developing a strong food safety culture, which is key to ensure safe produce.

#### Effect of Disinfectants on Preventing the Cross-Contamination of Pathogens in Fresh Produce Washing Water and Establishment of Operating Standards Thereof

IMCA SAMPERS, Ghent University, Ghent, Belgium

The worldwide concern of (fresh) water scarcity requires an effective and efficient water management in the food industry, pressing the need to optimise water use whilst ensuring (food) safety and wholesomeness of its products. The reuse (or recycling) of water within the food processing plant, either directly (with or without in situ water disinfection) or after reconditioning, is one of the main strategies to reduce water consumption and wastewater generation. As such, the associated accumulation of microorganisms when reusing the water must be taken into account to prevent process water becoming a source of contamination. The elimination or sufficient reduction of pathogens is therefore of utmost importance while reintroducing water into a food production process. Different water treatment technologies (chemically and/or physically) are however eligible although each has its specific (dis)advantages. In addition, these process waters are characterised with high levels of COD concentrations, dissolved and/or suspended solids, nutrients (such as ammonia) and minerals which may all have an impact on the efficiency of the used technologies. For example, chemical disinfection uses oxidising agents such as chlorine, potentially resulting in the formation of unwanted disinfection by-products due to its reaction with the organic matter. Therefore a, preferably on-line, monitoring system is needed to ensure both quality and safety of all (re)used waters. Sensors are already on the market (to measure physico-chemical parameters and/or the disinfectant residue), but it has been demonstrated that validation and verification must take place for each specific fresh produce operational system/ produce type.

# S3 The Survival and Control of Foodborne Pathogens in Low-moisture Foods

Low-moisture foods (LMFs) have been defined as those food products with a water activity  $(a_w)$  less than 0.85 and are generally considered less susceptible to microbial spoilage and the growth of foodborne pathogens. However, in recent years, outbreaks linked to LMFs have increased, with Salmonella spp., Bacillus cereus, Cronobacter sakazakii, Clostridium spp., Escherichia coli O157:H7, non-O157 E. coli, and Staphylococcus aureus being the principal pathogens involved. Because of the new concerns raised as a result of recent outbreaks, new approaches need to be developed to control foodborne pathogens in LMFs. This symposium will highlight some recent research examining the survival and potential for virulence changes of pathogenic E. coli and Listeria monocytogenes in LMFs. In addition, we will get a food company perspective on how best to control foodborne pathogens in a low-moisture food establishment. Additional research is needed to study the survival, pathogenicity and inactivation of foodborne pathogens in a wide variety of LMFs.

#### Survival and Potential for Pathogenicity Changes for *Listeria monocytogenes* in Low-moisture Foods

JEFFREY FARBER, University of Guelph, CRIFS, Department of Food Science, Guelph, ON, Canada

Low-moisture foods (LMFs) are characterized by a water activity (a,,) below 0.85 and are emerging as novel vehicles for foodborne illness. Although the growth of bacterial pathogens is inhibited by low  $a_w$ , they have been shown to survive/persist for long periods of time in some LMFs. This presents a public health concern, especially when LMFs are consumed without undergoing any microbial inactivation steps. The main purpose of our study is to assess the survival of Listeria monocytogenes on artificially inoculated LMFs. Foods were inoculated with a 4-strain cocktail of Lm at an initial concentration of 8  $\log_{10}$  CFU/g by immersion (pistachios) or misting (chocolate liquor, corn flakes). They were then dried at 30°C, and stored at both 23°C, 30-35% relative humidity (RH) and 4°C, 30–35% RH. For dried fruits (raisins, apples, strawberries) we used a dry-inoculum, and research was done to determine the best carrier for the recovery and enumeration of L. monocytogenes. Bacterial counts were done on tryptic soy agar with 0.6% (w/v) yeast extract, and/ or Oxford agar. Analysis of significant differences in survival was determined by using a two-way repeated measures ANOVA. In general, the organism survived best at both temperatures in pistachios and chocolate liquor and worst in dried apples. Monthly sampling of LMFs was done for up to a year. A new multiplex PCR method was developed to track and differentiate between the 4 L. monocytogenes serotypes (1/2a, 1/2b, 3a, and 4b) used to inoculate the LMFs. The raisins and pistachios appeared to contain the most diverse microbiomes. As the presence of any L. monocytogenes on ready-to-eat foods can potentially lead to a food recall,

research regarding the survival of foodborne pathogens on LMFs is important to understand the environmental factors underlying pathogens survival and is also important for predictive modeling used in health risk assessments.

# Survival of Enterohemmorhagic *E. coli* in Low-moisture Foods

LINDA J. HARRIS, University of California-Davis, Davis, CA, USA

Enterohemorrhagic Escherichia coli (EHEC) gastroenteritis have been associated with consumption of tree nuts and a number of other low-moisture foods (LMF). However, information on the behavior of these organisms in LMF and dry food production and processing environments is limited when compared to Salmonella. Where data are available, prevalence in raw LMF is lower than Salmonella. A range of desiccation tolerance is observed among EHEC strains; some strains are capable of surviving for long periods of time especially under dry and cool conditions. Thermal tolerance of EHEC is enhanced in LMF; however, where data are available, thermal resistance is not greater than that of Salmonella. EHEC should be considered in food safety plans for LMF. A greater understanding of the ecology of EHEC in different LMFs and additional information on the impact of a range of processing treatments is needed.

# The Control of Foodborne Pathogens in Low-moisture Food Facilities

PETER MCCLURE, Mondelez International, Birmingham, United Kingdom

Reduced water activity remains one of the key parameters used in food manufacturing for control of microorganisms in foods. Despite this, low-moisture foods continue to be associated with product recalls and foodborne outbreaks caused by infectious agents such as Salmonella. In this presentation, the main challenges related to the microbiological safety of low-moisture foods will be considered with the aim of highlighting the importance of understanding the ecology of low-moisture food manufacturing facilities. Particular attention is paid to sources of contamination (e.g., raw materials and manufacturing environment), potential controls that may be applied during processing, and the importance of good manufacturing practices including cleaning and disinfection, and verification procedures. Examples of intervention steps (e.g., CCPs) that take account of the low-moisture content of foods will be provided together with the factors that play a role in determining the survival characteristics of target microorganisms. In addition, prerequisites such as good manufacturing practices and hygiene, which are equally important in the manufacture of low-moisture foods, will be considered with a focus on pathogen environmental monitoring and practical aspects for control and corrective actions. Risks from wet versus dry cleaning and use of appropriate microbiological testing (e.g., appropriate hygiene indicators) will also be covered. Some of the tools and approaches that can be used in routine and investigational sampling will be described that can help determine sources of persistent and transient bacterial strains, and lead to direct and root-cause identification.

# S4 Network Analysis to Better Decipher Functions and Dynamics of Food Microbial Ecosystems

High-throughput DNA-RNA sequencing has increasingly been used to characterise microbial communities in various environments including foods. Going beyond abundance patterns in microbial communities represents a necessary step to understand the functional activities and dynamics of microbial ecosystems. Predicting and controlling the microbial communities is a challenge that microbial ecologists are currently facing to address environmental and health issues in marine, soil, wastewater, plant, human microbial environments, and more recently in foods. Systems modelling approach has recently emerged to infer the microbial community structure and functions from experimental metagenomic data. In this context, network analysis has been applied in many different environments including ocean, soil and human gut to decipher the structure of microbial ecosystems. However, applications in the food domain are still limited. Major advances are expected from the development of such approaches to better control spoilage and safety of food products.

The symposium will focus on network analysis and its further use in food microbial ecosystems.

The first talk will give an overview of tools and methods dedicated to microbial network inference, their strengths and weaknesses and the contributions these techniques can make towards a better understanding of microbial ecology. The second conference will focus on the applications of the systems biology protocol to understand phenotypic landscapes in various fields. The last talk will give the current state of the art of microbial association networks in food ecosystems and the promising perspectives they may offer for the future in this field.

#### Microbial Network Inference through Time and Space: Tools and Network Analysis

LISA RÖTTJERS, KU Leuven, Leuven, Belgium

Microbial association networks are valuable tools for the visualization of microbial communities. However, construction of these networks is sensitive to properties of microbial abundance data, and their interpretation can be far from straightforward. Three aspects of microbial associations networks are essential for construction of a biologically relevant network: preprocessing, network inference and network analysis. Preprocessing can help mitigate some of the effects of compositionality and sparsity, while appropriate network inference tools further prevent identification of spurious associations. Hence, the talk will include a brief overview of network inference methods and their applicability. Finally, inferred networks can be analyzed to identify emergent properties. Two properties of interest are hub species and clusters. The theoretical basis of these properties will be addressed and their analysis will be discussed. This introduction on microbial association networks will give investigators ideas on tool choice and network analysis, while providing information on pitfalls of network inference.

#### Systems Biology Protocol to Investigate Microbial Network: From Co-occurrences to Phenotypic Landscapes

**DAMIEN EVEILLARD,** Computational Biology Group LS2N UMR 6004 CNRS University of Nantes, Nantes, France

Recent progress in metagenomics has promoted a change of paradigm to investigate microbial ecosystems. These ecosystems are today analyzed by emphasizing either their gene content or "who is there and who is not" from a taxonomical viewpoint. However, understanding the interactions between microbial communities and their environment well enough to be able to predict diversity from physicochemical parameters is a fundamental pursuit of microbial ecology that still eludes us. Such a task must be achieved by dedicated computational approaches or modelings, as inspired by Systems Biology. Nevertheless, direct application of standard cellular systems biology approaches is a complicated task, because (i) communities are complex, (ii) most are described qualitatively, and (iii) quantitative understanding of the way communities interacts with their surroundings remains incomplete.

Within this seminar, we will illustrate how systems biology approaches must be adapted to overcome these points in different manners. First, we will present a network analysis. Here we use environmental and metagenomic data gathered during the Tara Oceans expedition to improve understanding of a biological process such as the carbon export. Second, we will describe how to integrate different omics knowledge. Such integration will emphasize putative functional units at the community level. Finally, we will illustrate quantitative modeling from this network. Constraint-based modeling will be used to predict microbial community structure and its behaviors based on genome-scale knowledge.

#### Shining Light on Networks in Food Microbiomes: FoodMicrobionet and the ShinyFMBN App

**EUGENIO PARENTE**, Annamaria Ricciardi, Teresa Zotta, *Università degli Studi della Basilicata, Potenza, Italy* 

The amount of data on food microbiome composition obtained by 16S metagenomics has been rising steadily in the last 15 years. Raw sequence for some studies are publicly available from databases such as NCBI Sequence Read Archive and EMBL European Nucleotide Archive and, more recently, platforms such as MGnify (https://www.ebi. ac.uk/metagenomics/) and QIITA (https://qiita.ucsd.edu/) allow complex workflows including data deposit, retrieval and processing. However, retrieval and integration of data for the purpose of metanalysis is still difficult.

We will present a new version of FoodMicrobionet, a database for the exploration of food bacterial communities. The database, available as an app built with the Shiny package of R, includes data from 44 studies and 2,234 samples. FoodMicrobionet is the largest collection of data on food bacterial communities and due to the structure of sample metadata, based on the European Food Safety Agency FoodEx2 classification, makes extraction, comparison and re-analysis of data comparatively easy. The interactive interface allows exploration of study and sample metadata, access to external resources (on line versions of the published studies, sequence data on NCBI SRA, taxonomic databases), filtering of samples on the basis of a number of criteria, aggregation of samples and bacterial taxa and export of data (OTU tables, edge tables) and metadata (study, samples, taxa) in a variety of formats ready for statistical and graphical analysis. A few examples of workflows (visualization of data as bipartite networks, estimation of microbial association networks) are provided.

# S5 Novel Modelling Approaches of Microbiological Spoilage in Food

One third of the food produced in the world for human consumption (approximately 1.3 billion tonnes) gets lost or wasted every year and represents an important economic issues (FAO, 2017). Among the reason of losses, spoilage by microorganisms contaminating the food matrices during production and storage is a major one. Growth of undesired microorganisms may cause organoleptic spoilage of food, leading to defects in texture, colour, odour, etc. Food companies usually use several preservation techniques (modified atmosphere packaging, additive uses, etc.) to counter the effects of bacterial spoilage.

This symposium aims to present innovative modelling approaches in the food spoilage area.

In the first talk, the identification of potential volatile food spoilage indicators by multivariate statistical analysis will be presented. Along with increasing knowledge regarding food quality and spoilage, this approach facilitates and accelerates the development of intelligent packaging technologies.

The second talk will address the use of Bayesian models to describe organoleptic spoilage dynamics in foods depending on industrial processing conditions, and microbiological and physicochemical factors (initial bacterial load, pH dynamics, gas composition of storage atmosphere, etc.), will be presented. These models should open new perspectives in the understanding of food spoilage and provide the food manufacturers a decision-making tool to act early in the production chain. Finally, the last talk will give an overview and a short demonstration on SorfML (www.sorfml.com). SorfML is a web-platform able to automate the procedure of identifying the best machine learning method for comparing data from several analytical techniques, to predict the counts of microorganisms responsible of food spoilage regardless of the packaging system applied. SorfML users can securely upload raw experimental data and apply various machine learning classification and regression modelling algorithms (e.g., SVM, Neural Network, Random forests) in order to identify the best method to predict food microbiological quality.

## Prediction of Spoilage Occurrence in French Fresh Poultry Sausages: Potential Role of Potassium Lactate and Modified Atmosphere Packaging

**NGOC-DU LUONG,** *SECALIM, INRA, Oniris, Université Bretagne Loire, Nantes, France* Louis Coroller<sup>2</sup>, Jeanne-Marie Membre<sup>1</sup>, Sandrine Guillou<sup>1</sup> and the RedLosses Consortium<sup>3</sup>.

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Spoilage occurrence in meat products has generally been highlighted in the literature by the assessment of various indicators mainly resulting from the contamination by microorganisms and their subsequent activities during storage. To better control food spoilage, it is necessary to go beyond the assessment and move further to the prediction of relevant spoilage indicators over time by suitably adjusting predictive mathematical models on experimental data.

The dataset used in this study was collected from experiments on French fresh pork and poultry sausages under different industrial processing conditions. Sausages were made with different formulations of potassium lactate and conditioned under different modified atmosphere packaging (MAP). Several spoilage-related responses were measured during storage, including characterisation of off-odour by trained panelists, pH values, colour measurement as well as other physicochemical and microbial responses. Exploratory analyses on the overall dataset were first performed to identify relevant indicators which could be modelled to predict spoilage occurrence, or used for model parametrisation.

Then, the possible use of a Bayesian approach to predict one relevant spoilage indicator depending on Lactate and MAP, will be illustrated through a simple example. Bayesian inference is a model parameter estimation technique combining experimental data and prior expertise knowledge for calibrating mathematical models. Prior information on parameters was collected from preliminary studies as well as expert opinion. The inference performed using numerical simulations appeared to be an appropriate method providing posterior distributions of parameters given the dataset. Within the food microbiology framework, such approach could be useful to improve the calibration of predictive models of spoilage occurrence, by using the considerable expertise available in laboratories and/or in the literature.

## Identification of Volatile Food Spoilage Indicators by Multivariate Statistical Analysis

LOTTA KUULIALA, Ghent University, Ghent, Belgium

The quantification of spoilage-related volatile organic compounds (VOCs) has widely been recognized as one of the most promising approaches in food quality monitoring, particularly when considering seafood or other highly perishable products packed under modified atmospheres. Accumulation of VOCs in the package headspace as a consequence of microbial metabolism and/or (bio)chemical reactions frequently leads to the generation of offensive offodors, eventually causing consumer rejection. By monitoring the concentrations of these compounds over storage time, real-time information about quality changes can be obtained. However, the success of this approach depends on our capacity to recognize the most characteristic and useful volatile spoilage indicators under different packaging and storage conditions. Because of the complex nature of food spoilage and the specific requirements set by the intended application(s), advanced statistical methods are typically needed for this task.

In this talk, different multivariate statistical techniques for identifying volatile food spoilage indicators are presented, with special emphasis on Partial Least Squares (PLS) Regression and Latent Dirichlet Allocation (LDA). The general principles of these two methods and the development of identification criteria on their basis will be demonstrated with examples from various seafood studies. Finally, the applicability, benefits and limitations of these methods under different scenarios will be discussed. In conclusion, the talk highlights the significance of advanced data analysis when the ultimate aim is not only to address complex biological problems, but also to support the development of food quality monitoring and novel packaging technologies.

#### SorfML: The Machine Learning Web Platform for Food Microbiological Quality FADY MOHAREB, Cranfield University, Cranfield,

United Kingdom

Ensuring top quality and maximum safety to maintain the consumer's confidence is one of the main challenges facing the food industry nowadays. As conventional laboratory and microbiological tests cannot provide a 100% inspection, non-invasive approaches based on vibrational spectroscopy, hyper- and multi-spectral imagining started gaining popularity as rapid and efficient methods for assessing food quality, safety and authentication. Due to the multi-dimensional nature of the data generated from such analyses, the output needs to be coupled with a suitable statistical approach or machine-learning algorithms before the results can be interpreted. In order to overcome this challenge, the cloud-enabled platform SorfML (www.sorfml.com) was developed to provide a framework for interpreting the output from such platforms into a freshness, safety or authentication profile; allowing therefore such platforms to be applied on-line or at-line throughout the food production chain. In this talk, the main functionalities of sorfML will be outlined; as well as a short live demo highlighting the potential uses of this application.

# S6 Clarity through Chaos: International Perspectives on Food Safety after Recent High Profile Foodborne Outbreaks

The year 2018 was marked by high profile foodborne illness outbreaks which killed and sickened and killed individuals across Europe, in South Africa and in the United States. Listeria monocytogenes in polony in, E. coli O157:H7 and Cyclospora associated with leafy greens provide an opportunity to learn lessons to improve food safety and understand potential underestimated, under-evaluated risks in global food safety systems. This symposium will address the potential upcoming challenges in responding to these outbreaks from three different international perspectives. This will include identifying challenges in growing and processing environments, and unique regional consumer preferences which can impact the risk associated with a specific commodity. The regional and local factors which influence the attention paid to some of these emerging topics will be discussed, along with commodity-specific microbial standards which may not capture the true risk associated with specific commodities. The speakers will examine how industry practices and current research are influencing the understanding of these complex scenarios. Three distinguished and internationally recognized speakers will be addressing the issues from their unique perspectives.

# Parasites, Viruses, and Produce from the United States: Can Anything Change?

KALI KNIEL, University of Delaware, Newark, DE, USA

Detection of viruses and protozoa in foods continues to challenge the food industry, while outbreaks of foodborne illness associated with these organisms continue to occur. Foodborne illness attributed to protozoa has been on the rise in the United States over the past four decades. The Centers for Disease Control and Prevention estimates 232,705 cases of foodborne illness attributed to protozoa annually and a substantial number of outbreaks associated with produce. Similarly, the Centers for Disease Control and Prevent estimates just over five million cases annually in the United States of virus-associated foodborne illness. Awareness and surveillance of protozoa and viruses increase, along with the need for novel interventions to prevent and manage contamination. Raw agricultural commodities may be more at risk for contamination as they are more often consumed raw; however, determining the origin of contamination, which may be from humans or animals depending on the microorganism, offers its own set of challenges. This talk will discuss these challenges along with issues of persistence and transmission in light of recent outbreaks and case studies.

# Frozen Vegetables Outbreak: What Have We Learnt?

ANA ALLENDE, CEBAS-CSIC, Murcia, Spain

It is well known that L. monocytogenes is widespread in soil, water, sewage, and decaying vegetation and this is why it is usually found in fruits and vegetables. Last year, Europe suffered the first multi-state outbreak associated with frozen vegetables. In the past, multi-state outbreaks associated to frozen vegetables and L. monocytogenes were only relevant in the U.S., but since last year, Europe is also aware about the problems associated with this food/pathogen combination. At the end of 2017, beginning of 2018, several member states reported human cases associated to an outbreak caused by Listeria monocytogenes serogroup IVb, sequence type (ST) 6. The outbreak was linked to frozen corn at the beginning of 2018, but despite the recall, cases continue to be reported and action against the production plant in Hungary were not taken until July 2018. Due to this outbreak, 47 fell ill and 5 of these people died. There are many lessons to be learnt from this outbreak, including consumer awareness of the risks of consuming frozen vegetables that are not cooked and also improvements on sampling and testing in the processing plants of frozen vegetables to reduce the risk implementing the "Seek and Destroy Process" which has seen very positive results in U.S.

#### A Year after Polony: Emerging Food Safety Risks in a Highly Diversified Food System

**LISE KORSTEN**, DST-NRF Centre of Excellence in Food Security, Department of Plant and Soil Sciences, University of Pretoria, Pretoria, South Africa

A year after the world's worst listeriosis outbreak, South Africa is still trying to deal with the impact of the disease and the loss in public trust. The country was ill prepared for the outbreak and this was clearly reflected in the incoherent way the public and private sector dealt with the challenge. Listeriosis only became a notifiable disease on the 05<sup>th</sup> December 2017 when the Minister of Health announced the outbreak. By then, 61 deaths and 550 cases were already noted by health authorities. It took another 90 days before it was offi-

cially announced that the outbreak was linked to cold meats i.e., polony, a local favourite amongst all income groups. This was followed by a recall of several cold meat products which caused havoc in the food industry. The pathogen, Listeria monocytogenes was characterised by multilocus sequence typing using the National Institute of Communicable Diseases whole genome sequencing (WGS) platform. The majority of the clinical isolates (91%) were identified as sequence type 6 and the rest belonged to 11 other sequence types not yet described. In our study that was done before, during and after the outbreak we found three isolates that were directly linked to the ST6 cluster. The other 12 isolates belonged to other sequence types that is not linked with the South African outbreak. The WGS work was done in close collaboration with the U.S. Food and Drug Administration, GenomeTRACR programme. This paper provides a comparative perspective how industry practices and current research are influencing the understanding of complex scenarios.

# S7 Getting Ahead of Food Fraud

Cases of food adulteration hit the headlines worldwide on a regular basis. Mislabeling of content or origin, substitution of high value ingredients by low-cost imitates, and addition of fillers or even potentially harmful components are typical fraudulent practices. High-priced food products, but also those with high volume of sales, are preferential targets of malpractices.

As new fraudulent food manipulations can elude detection by classical and targeted analytics, there is an increasing need for new and non-targeted techniques for food authenticity control.

Regulation bodies are working on setting up rules to coordinate action against "fraudulent practices" in the food supply chain. One of the keys for success is certainly method standardization and recognition.

The first presentation, using honey as an example, will show that fraud mechanisms are responsible for the injection of a very important volume of diluted and/or non-conforming product to the market. The overall result is a threat to food safety, food security and ecological sustainability.

The second presentation will focus on the utilization of New Generation Sequencing for food authenticity control.

The last presentation will show how 1H-NMR, as a non targeted approach, is being applied to overcome the complexity of food fraud in fruit juices.

# The Magnitude and Impact of Honey Fraud

**NORBERTO GARCÍA,** *Apimondia, President of the Scientific Commission Beekeeping Economy and Chairman of the Working Group Adulteration of Bee Products, Buenos Aires, Argentina* 

Global beekeeping is currently affected by many adverse factors that threaten its sustainability. Honey fraud has become a phenomenon that is nearly out of control. According to the U.S. Pharmacopeia's Food Fraud Database, honey ranks as one of the most favorite food targets for adulteration.

In order to better understand the magnitude of the problem, we must remember that honey is the best-known product of bees but surely not the most important one. Bees, through their pollination work, are essential for the maintenance of the planet's biodiversity, and absolutely necessary for the pollination of many crops that represent 35% of all our food.

Statistical information of word honey trade will be presented in order to describe current tendencies, regional peculiarities, and most visible abnormalities. The use of statistical information is a valuable tool for authorities to investigate and combat more efficiently the scourge of honey adulteration, which takes diverse forms and magnitudes depending on the source countries and the import markets.

As long as honey fraud persists, the well-being and stability of the world beekeeping industry remains in jeopardy. The sustainability of beekeeping requires the joint effort of beekeepers, honest traders, scientists, private and public laboratories, and national authorities. The protection of honey purity is not only a problem of food safety and food defense, but it is mainly a problem of food security, thus concerning the capacity of countries to provide their own food.

## Development of a Practical Procedure Suitable to Determine the Geographical Origin and Authenticity of Spices with 1H-NMR-Analytic (HAGen)

WOLFRAM WENDLER, Arotop Food & Environment GmbH, Mainz, Germany

The lecture shows the current state of development of an H-NMR-method for the authenticity (origin and adulteration) of herbs and spices. The challenge was to extract the most important information of the material from the solid substances. For this purpose, extraction methods with different polar solvents have been developed. Furthermore, the most important ingredients are to be quantified. Meanwhile, it is possible to determine different origins, adulterants and ingredients.

The project started in April 2016 and was funded by the Federal Ministry of Food and Agriculture.

#### 1H-NMR-Profiling as an Efficient Screening Analysis for Food Fraud Detection in Fruit Juices

PETER RINKE, SGF International e.V., Nieder-Olm, Germany

The Voluntary Control System of SGF International e.V. is a well-established and worldwide unique system to successfully combat food fraud in the fruit and vegetable juice industry. System audits and product checks are combined to assure fair competition while the control system includes effective measures for the implementation of corrective actions.

For this purpose a high number of control samples must be screened and a large scope of possible types of frauds must be covered by control analyses. Furthermore only databases refined for different origins allow correct analytical result interpretation and a good detection of non-authentic products. Supply chains for most fruit juices are characterised through an important diversity of different attributes like origin of raw material, cultivars and varieties, production techniques and seasonal climatic influences. All these parameters are influencing the chemical profile of a juice and must be covered by reference materials. Thus, to achieve control schedule and database update a high number of control samples and of authentic reference samples must be analysed.

Due to its high throughput capacity, NMR Juice screening became a central tool in the applied analytical strategy. Its untargeted capacity to identify deviations and to quantify important parameters allows the selection of suspicious samples in a first step. Then analyses are selected to confirm frauds with targeted methods that are the most suitable for any individual case. This two-step approach is economic and allows the enhancement of control density as long as enough authentic reference samples are available and used to build and update statistical models

## S8 Challenges in *Campylobacter* Detection and Accurate Quantification

*Campylobacter* remains the most frequently reported agent of zoonosis in the European Union, with more than 240,000 confirmed human cases of campylobacteriosis in 2016. 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 presence on chicken fillet depends on its ability to survive the scalding (i.e., heating step), chilling and subsequent storage steps. Strain variability 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 concentrations allowing subsequent detection. These enrichment-based detection methods are in practice not perfectly selective and sensitive.

This symposium focuses on the challenges to detect *Campylobacter* along the food chain and to evaluate accurately its survival capacity. It will highlight the advances to predict the robustness of *Campylobacter* to survive 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 point out Quantitative Microbiological Risk Assessment tools used to evaluate the impact of campylobacteriosis and to rank selected intervention strategies in the farm-to-fork continuum on efficacy for *Campylobacter* reduction.

#### Challenges of *Campylobacter* Detection; Effect of Strain Variability and Competitive Flora on Enrichment-based Detection Procedures

**MAREN LANZL**, Wageningen University, Wageningen, The Netherlands

**Aim:** Testing foods for the presence of *Campylobacter* spp. is crucial to ensure food safety. While advances in rapid detection techniques have tried to shorten lengthy procedures, an enrichment step is still necessary to recover damaged cells and reach detectable levels. However, recovery kinetics during enrichment are modestly understood and insight in the recovery mechanisms of *Campylobacter* spp. is is important to reduce the time-consuming enrichment step without forfeiting reliable detection.

**Method:** 13 *C. jejuni* and 10 *C. coli* isolates from different food-relevant sources were subjected to freezing (-20°C) and refrigeration (+4°C) and subsequently enriched following ISO 10272-1:2017, procedure A. Growth kinetics were determined by plate counting and the lag and growth rate were estimated using the Baranyi-model. Kinetic data were used for scenario analysis of the growth behaviour during enrichment.

**Results:** Freezing reduced cell concentrations by 1.5 ± 0.3 log CFU/ml, while refrigeration did not affect cell counts. The *Campylobacter* strains differed significantly in their ability to recover from cold stress during enrichment in Bolton broth, but species type was not a determining factor. In most cases, the recovery times were longer after freeze stress than after refrigeration. Scenario analysis demonstrated that variability in lag duration and growth rate did not result in false-negative detection outcomes for both species.

**Conclusion:** Freeze stress had a higher impact on viability and recovery time than refrigeration. In monoculture, all 23 strains clearly reached the detection limit within 48 hours as stated in ISO 10272-1:2017. False-negative results in food testing are therefore more likely caused by other factors like competitive microbiota.

#### The Survival of *Campylobacter* in the Food Chain, or How to Enhance the Robustness of the Model Prediction Using Molecular Markers

**BENJAMIN DUQUÉ**<sup>1</sup>, UMR1014 SECALIM, INRA, Oniris, Nantes, France

Nabila Haddad<sup>1</sup>, Sandrine Rezé<sup>1</sup>, Albert Rossero<sup>1</sup>, Jeanne-Marie Membré<sup>1</sup>, Sandrine Guillou<sup>1</sup> and the Biomics consortium<sup>2</sup> <sup>1</sup>SECALIM, INRA, Oniris, Université Bretagne Loire, Nantes, 44307, France

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Campylobacter is a foodborne pathogen highly prevalent in poultry and the primary cause of enteritis in humans. Campylobacter contamination on chicken fillet depends on its ability to survive slaughter, i.e., scalding and chilling steps, and storage.

Recent developments of "omics" technologies have provided new insights in the study of microbiological responses. The use of transcriptomic data from foodborne pathogens under application of different stresses could enable the identification of biological markers, i.e., biomarkers of specific resistance features of the pathogen. Until now, Quantitative Microbiological Exposure Assessment (QMEA) models have been mainly built without considering cell history (e.g., conditions subsequently encountered during food processing). The use of stress biomarkers could help to refine QMEA models by considering strain variability and cell history, and hence enable to increase robustness of model prediction.

In this context, three *C. jejuni* strains were submitted to consecutive heat (46°, 50° or 54°C for 4 min) and cold (-4° or 3°C for 2 h) stresses. Cultures were then stored at 6°C during seven days under modified atmospheres (70%  $O_2$  / 30%  $CO_2$  or 50%  $CO_2$  / 50%  $N_2$ ). Viable counts of *C. jejuni* were enumerated to determine the viability loss resulting from the application of each step.

In parallel, expression of selected genes by RT-qPCR was assessed following both heat and cold stressing steps. A strategy to select a list of 40 genes as potential candidates was set up based on a decisional tree; regulators and genes involved in different metabolic pathways (e.g., general stress and oxidative stress response) were included.

The identification of biomarkers will be based on the differential expression of genes, positively or negatively correlated with the viability loss observed during the storage step. To do so, statistical tools will be used to establish correlation between phenotypic and transcriptomic responses and point out stress biomarkers to predict the pathogen behavior while considering the conditions it has previously encountered.

## *Campylobacter* Contamination Levels in Poultry Meat in the EU and the Efficacy of Control Measures

**PIETRO STELLA**, European Food Safety Authority (EFSA), Parma, Italy

In 2017, Campylobacter was the most commonly reported gastrointestinal bacterial pathogen in humans in the EU and has been so since 2005. The highest Campylobacter occurrence was observed in fresh meat from broilers (37%) followed by that from turkeys (32%). About three quarters of the 1,201 fresh broiler meat and meat product samples were positive for C. jejuni; most of the remainder were C. coli positive (EFSA and ECDC, 2018). A scientific opinion from EFSA's Panel on Biological Hazards (BIOHAZ Panel) in 2011 assessed control options for Campylobacter in broiler meat production. Public health (PH) benefits from control in primary broiler production were expected to be greater than control later in the chain. Strict implementation of biosecurity in primary production and Good Manufacturing Practices/ Hazard Analysis Critical Control Points during slaughter may reduce colonization of broilers with Campylobacter and contamination of carcasses. The effects cannot be quantified

because they depend on many interrelated local factors. Other on-farm control options, such as fly screens, restriction of slaughter age, or discontinued thinning, as well as measures to be applied later in the chain, were quantitatively assessed with regard to the estimated reduction of human campylobacteriosis cases. Also the PH risk reduction and compliance of batches when establishing microbiological criteria with critical limits of *Campylobacter* concentrations of neck and breast skin were assessed (EFSA BIOHAZ Panel, 2011). On request of the European Commission, the EFSA BIOHAZ panel is currently carrying out an update of this scientific opinion, focusing on control measures at primary production.

EFSA and ECDC, 2018. The European Union summary report on trends and sources of zoonoses, zoonotic agents and foodborne outbreaks in 2017. *EFSA Journal* 2018;16(12):5500.

EFSA BIOHAZ Panel, 2011. Scientific Opinion on *Campylobacter* in broiler meat production: control options and performance objectives and/or targets at different stages of the food chain. *EFSA Journal* 2011;9(4):2105.

# S9 Close Up of Consumer Kitchen Practices – Can Socio(microbio)logy Aid Food Safety at Home?

It has been estimated that about 23 million European citizens suffer from foodborne disease yearly. The weakest part in the food chain is often the household, where people fails do adopt practices regarded as safe. Existing strategies for risk mitigation by changing consumer behaviour seem to fall short as about 40% of outbreaks are still tracked back to the home. Can traditional boundaries between scientific disciplines partly be blamed for this persistent societal challenge?

The symposium will bring together scientists from natural and social sciences to demonstrate how a merged scientific methodology based on Theory of Practices (sociology) and HACCP can give an understanding of why people keep to practices scientists and authorities regard as unsafe. Several examples on how consumers can or cannot be expected to protect themselves against foodborne illness will be discussed: Is it possible to remove Norovirus from lettuce by rinsing? Can an egg with a running yolk be safe? Will providing thermometers be a solution for undercooking or undercooling? Can kitchen cloth safety be monitored using your nose? Insights in risky and safe kitchen practices from a large study of consumers from six European countries will presented together with results from laboratory studies on how consumer practices affect Salmonella, Campylobacter, Listeria and Norovirus.

# Kitchen Hygiene in Six European Countries – Safe or Unsafe?

**SOLVEIG LANGSRUD**, Trond Møretrø, Anette Wold Aasli, Charlotte Nilsen and Tove Maugesten, *Nofima, Norwegian Institute of Food, Fisheries and Aquaculture Research, Ås, Norway* 

It is generally acknowledged that poor hygiene in the home is contributing to foodborne infections. Several studies demonstrate that pathogens can be transferred from contaminated food into the mouth via the hands, kitchen surfaces and washing utensils. Both case-control studies and consumer surveys have indicated that some consumers fail to adapt basic hygienic practices that could protect them from foodborne illness. One weakness of such studies, although covering large groups of consumers, is that self reporting will only cover simple, conscious behaviours. If conducted carefully, walk-along interview techniques can reveal complex patterns and unconscious habitual actions as well as new factors explaining lack of proper hygiene. Results from a transdisciplinary study, where sociologists and microbiologists studied chicken and salad meal preparation in a total of 90 households across Europe, will be presented. The habit of washing hands thoroughly before and during preparation varied much between countries. and high occurrence of pathogens on food in a country was not necessarily associated with high consumer awareness or better hand hygiene. Utensils for cleaning (sponges, cloths, brushes),

contained high bacterial numbers in all countries, but seldom pathogens. Pathogens died off rapidly in cleaning utensils compared to other bacteria, especially in brushes, which may be recommended as a safer alternative than sponges for washing up. In conclusion, the hygienic practices varied greatly and seemed to depend on several factors, as understanding of risk, convenience, habits and the infrastructure of the kitchen. Therefore, improving hygiene in Europe would require different approaches for different countries and consumer groups, including food safety information, education and basic infrastructure, such as better access to running water.

## Norovirus Removal from Salads and Bivalve Molluscs

**SUSANA GUIX ARNAU**, Eduard Anfruns, Marilisa Bottaro, Cristina Fuentes, Aurora Sabrià, Rosa Pinto and Albert Bosch

University of Barcelona, Barcelona, Spain

Norovirus (NoV) is a foremost cause of domestically acquired foodborne acute gastroenteritis and outbreaks. Despite industrial efforts to control NoV contamination of foods, its prevalence in foodstuffs at retail is significant. NoV infections are often associated to the consumption of contaminated produce, fresh and frozen berries, raw/undercooked bivalve mollusks and products contaminated during handling by infected individuals. Although certain consumer behaviors may increase the risk of transmission, interventions aiming at changing/implementing consumer habits may be considered as opportunities for risk mitigation. Decontamination of fruits and produce by washing with disinfectants and thorough cooking of bivalves are two of the most critical options performed by final consumers which could significantly contribute to mitigate the risk of infection.

The effectiveness of chemical sanitizers in inactivating genogroup I and II NoV strains on Ready-to-Eat salads were measured by viability RTqPCR assays including the use of propidium monoazide (PMA). Addition of sodium hypochlorite or peracetic acid increased reductions on virus levels by over  $2.25 \pm 0.56$  and  $2.00 \pm 0.28 \log_{10}$ , for genogroup I and II, respectively, as compared to washing with water alone. Chlorine dioxide showed lower disinfection efficiency.

Inactivation experiments on spiked clams treated in water at 90°C for 8.5 ± 1 min (ensuring that meat reached 90°C for 90 seconds, which is recommended as a safe virucidal treatment) showed  $\log_{10}$  reductions of 2.96 ± 0.79 and 2.65 ± 0.56 for genogroup I and II, respectively. The use of PMA resulted in an additional 0.6  $\log_{10}$  reduction for genogroup II.

Our results may be translated into innovative educational, social or even technological tools targeting consumers with the objective of mitigating the risk of NoV transmission.

# Surprisingly (un)Safe: Runny Eggs and Poultry!

**PAULA TEIXEIRA**, Vania Ferreira, Norton Komora and Marta Carvalho, Universidade Católica Portuguesa, CBQF – Centro de Biotecnologia e Química Fina – Laboratório Associado, Escola Superior de Biotecnologia, Porto, Portugal

According to the World Health Organization Campylobacter is the most common cause of bacterial foodborne disease and non-typhoidal Salmonella is a major cause of foodborne deaths worldwide. Campylobacteriosis have been often associated with poultry; 'eggs and egg products' are commonly implicated in outbreaks of salmonellosis. As a significant number of the outbreaks occur in domestic settings it can be anticipated that consumers may have an important role in the prevention of foodborne illness. And concerned consumers can easily find advice on the Internet or in newspapers. But are these provided advice scientifically sound? In SafeConsume we have evaluated the impact of some consumer practices on microbial spread, growth and survival. Surprising results were found. For example, we are advised to consume hard-boiled eggs to be sure to avoid Salmonella. For those who prefer eggs like this, they may be safe (if cross-contamination is prevented). But what SafeConsume

found was that runny eggs can also be safe ... stove turned on less than five minutes may be enough, depending on the level of water and on the heating/cooling regimes. Safe, runny eggs, and energy saved! Regarding poultry preparation, advice about how to monitor chicken doneness vary between different authorities and other risk communication actors, e.g., clear juices, minimum temperature of 70 °C or more, using of a thermometer to ensure combined time-temperature regimes. USDA is advising on different minimum core temperature of at least 73.4°C for poultry, which would ensure a reduction of pathogenic bacteria of more than 6 logarithmic cycles. But what SafeConsume found was that the colour of the meat juice would not be a safe indicator of poultry doneness, the core temperature is not the most important indicator for safety and that consumer advices should focus on proper heat treatment of the surfaces rather than the interior.

### The Challenging Practice of Keeping Food Cold: Refrigeration in Everyday Domestic Life

**HELENE MARIA FIANE TEIGEN** and Silje Elisabeth Skuland, *Consumption Research Norway, SIFO, Oslo Metropolitan University, Oslo, Norway* 

The World Health Organization names proper cooling as one of five keys to safer food, stating the importance of keeping food at the right temperature to prevent growth of microorganisms. Still, few studies have paid attention to how people manage refrigeration in their everyday life and how it affects food safety. The aim of the study is thus to investigate domestic fridge practices in order to provide insight about how everyday challenges prevent safe food handling. We conceptualize domestic refrigeration as a social practice, which includes certain foods, appliances, ideas about temperature and skills on when and how to cool food, and how long food can be kept uncool. The paper draws upon household observations and interviews of 15 Norwegian young families, elderly couples and young single men, employing observational methods such as walk-a-long interviews. Norwegians are among the most trusting consumers in Europe and put great trust to both the food industry and food authorities (Kjærnes et al., 2007). Meanwhile, Norwegian food culture is characterized by many cold meals during the day (breakfast, lunch and supper) including a high variety ready-to-eat cold cuts, smoked salmon and cheeses, not to mention the traditional raw salted and fermented fish product, Rakfisk. Keeping food cold may appear simple and strait forward given the domestic fridge. However, in this paper it is argued that keeping food cold is a highly complex, routinized and skill-depended activity, which is done and redone repeatedly tacitly throughout the food day. It involves the spatial and temporal organization of various foods from retail to fork, relying upon a socio-technical infrastructure of the wider food industry, the technical characteristics of the fridge, the food and its packing - all managed and handled within the social context of the household and family life.

# S10 Food Safety Emerging Risk Identification with Novel Computational Methods

Emerging risks are risks posed by new hazards or by known hazards but with a significantly increased exposure. A timely and reliable identification of those risks is in the center of food safety decision making. The nature of the emerging issues implies that many data and information sources should be parsed in a short timeframe and a complex system of drivers should be considered during the emerging risk identification process. Moreover, the process tries to anticipate new risks from a very noisy environment.

As a follow-up to last year's symposium on emerging risks, this symposium aims for presenting various novel computational methods as solutions for this process.

The first presentation will present the possible uses of Bayesian Network Analysis for emerging risk identification. Bayesian networks are a class of probabilistic models originating from the Bayesian statistics and decision theory combined with graph theory, and are able to model dependencies between different variables, manage non-linear interactions and integrate different kinds of information.

The second presentation will present a possibility of using citation network of patents for identifying emerging technological fields. Patents cross-refer each other and this links the patents into a network. This network can be analysed and patents could be classified into clusters based on their similarity. The evolution of clusters in time indicates new, emerging technological fields acting as drivers to emerging technological food safety risks.

The third presentation will summarize the outcomes of a pilot project at the European Food Safety Authority where Artificial Intelligence (Natural Language Processing) was used in identifying emerging risks from scientific publication sources.

Using computational science tools in the emerging risk identification process is a new and trending area of risk assessment and risk management. All the presentations will shed light on the user needs and advantages and challenges of using such methods.

# Using Bayesian Network Analysis in Identifying Emerging Risks

HANS MARVIN, Wageningen University and Research, Wageningen, The Netherlands

Emerging risks as defined by the European Food Safety Authority (EFSA) are risks resulting from a newly identified hazard to which a significant exposure may occur, or from an unexpected new or increased significant exposure and/or susceptibility to a known hazard. It is apparent that the development of emerging risks is influenced by many factors from inside and outside the food supply chain. A system approach is needed that takes all of these factors into account in its complex interactions and that makes use of the huge amount of available data. In such a system approach, it is clear that big data technologies and tools such as artificial intelligence and machine learning are needed, including a safe and powerful infrastructure allowing handling of big data and ensuring interoperability. We have explored Bayesian Networks (BNs) as a system approach to predict known and emerging food safety risks. Data-driven BNs in combination with expert elicitation resulted in models having a high prediction accuracy for unexpected increased exposure to known hazards. BN models can demonstrate the impact of influencing factors on the development of food safety risks and are therefore useful in scenario analysis. However, to identify unknown hazards or risks further development is needed.

#### Identifying Emerging Technological Fields as Drivers of Emerging Risks with Network Analysis

**AKOS JOZWIAK**, Tekla Engelhardt, Zsusza Farkas, Kata Kerekes, Szilveszter Csorba and József Baranyi, *National Food Chain Safety, Budapest, Hungary* 

Network analysis is a computational scientific methodology for visualizing and analysis of complex structures. The identification of emerging risks in practice means that many data and information sources should be parsed in a relatively short timeframe and a complex system of drivers should be considered during this process. We use network analysis methods for detecting those complex patterns in the food safety emerging issues identification.

In the presentation, two network analysis approaches will be detailed: 1) using citation network of patents for identifying clusters and 2) using text mining and network analysis of news from media monitoring sites.

Patents cross-refer each other and this links the patents into a network. This network can be analysed and patents could be classified into clusters based on their similarity. The evolution of clusters in time indicates new, emerging technological fields acting as drivers to emerging technological food safety risks. News presents a very up-to-date, however a very noisy source of information on food-safety related issues. MediSys is a media monitoring system of the European Commission, providing event-based surveillance to rapidly identify potential public health threats using information from media reports. The texts of the news could be organized into a network of co-occurring keywords. The analysis and visualization of the network changes over time give an insight into trending topics which could serve as inputs into the emerging risk identification process.

#### Emerging Risk Identification and Scenario Analysis in the Food Chain

**NIELS LUCAS LUIJCKX** and Fred J. van de Brug, *The Netherlands Organisation for Applied Scientific Research (TNO), Zeist, The Netherlands* 

Food scares, incidents and risks, happen on an almost daily basis and modern communication technology allows easy and quick spread of the news, whatever the actual value. It is almost impossible to obtain or retrieve the relevant and interesting information in the current overload, especially as one expert or one organization. But, modern technology also permits to select and filter the right information and often at an early enough stage to prevent damage, to prepare for an incident or to reduce the commercial or societal effects.

To paraphrase, we should know the knowns and try to look for the unknowns. We learn from hindsight and want to apply our knowledge into foresight. The core is to be as sure as possible to know what is there and to make scenarios for future development. Something that regulators, authorities, policy makers and companies should do.

TNO, The Netherlands, developed a support system for the identification of early signals and research that feed scenarios for risk management. It is called ERIS and commercially applied as well as tested in many settings as a tool to help information management and risk management. In this presentation ERIS will be explained and examples given.

The next step, under current development, is to apply the early signals in a scenario analysis model for the food chain. Such a model will eventually evolve into a predictive model. Some very preliminary ideas will be presented to feed a discussion.

Modern information and communication technology can be a risk to companies and society, it is also a challenge and even more an opportunity. An opportunity to scan the horizon and beyond.

#### Application of Metagenomics Data Pool in Structured Problem Solving a Bottled Water Industry Example

ZOLTAN SYPOSS, Coca-Cola HBC, Vienna, Austria

With the increasing volume and complexity of food production and international trade, analysis needs of such complex systems have become more important than ever. Besides, an increasing amount of data is produced every day: different control activities, sensors, laboratory tests produce a lot of unstructured, but very valuable data.

The need for handling, analysis, and interpretation of large, interrelated datasets, together with the rapid development of information technology tools, have resulted in newly emerging data-related scientific fields. Their common characteristic is that with the use of computational science tools such rules or patterns could be identified which would otherwise be very challenging or impossible using smaller datasets.

The presentation will show a practical bottling industry example of using laboratory metagenomics data in the problem identification and decision-making process, focusing on the importance of the interpretation of the results as well. The case study also shows a need for bridging data scientists and food scientists for an effective utilization of data analysis methods in the food safety domain

# S11 Fast MALDI Typing to Drive Decision Making and Source Tracking

MALDI is definitely not competing with WGS; MALDI is definitely completing WGS. Fully dedicated to routine testing, the first application is the identification of microbial isolates. The generated mass spectra are then compared to a reference library to provide the identification results of the tested colonies. However, the generated mass spectra can be easily compared to each other's for typing purpose. This usually implies the development of standardized and specific protocols to get pure proteins extracts. Using relevant bioinformatics tools, it is then possible to easily identify biomarkers specific to certain phenotype, or track cross-contaminations in real time.

Pioneering studies for pathogen typing will be presented. The developed methodologies enable fast screening in routine testing laboratories.

The 2,600 different *Salmonella* serovars are routinely differentiated by the time-consuming serotyping. The potential of MALDI-TOF MS for rapid screening of the 6 *Salmonella* serotypes has been investigated, in order to reduce sample numbers that have to be subsequently analysed using conventional slide agglutination techniques.

The Bacillus cereus group is genetically highly homogenous consisting of 9 species. A small percentage of *B. cereus* strains are able to produce the heat stable emetic toxin. While the emetic strains are currently identified using laborious techniques, MALDI-TOF MS provides more than 99.0 % correct identification rate by evaluating two specific biomarkers.

Source tracking of *Listeria monocytogenes* is a key point in process hygiene and risk management. Proteome fingerprinting with MALDI-TOF MS shows a discriminatory level equivalent to PFGE, with a restricted handling-time. With less than 8 hours from pure cultures to dendrogram, the developed approach benefits to the decision-makers in their daily routine.

## Actions Speak Louder Than Words: MALDI-TOF MS for Realistic Analysis Workflow

BENOIT GASSILLOUD, ANSES, Nancy, France

Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF SM) can identify cultured micro organism faster and more precisely compared with phenotypic and biochemical methods. Expended custom reference spectra databases and their processing with appropriate computer program allow also now the typing of several species. Since 2013, Anses has promoted the evaluation of this technology and the development of exhaustive MSP spectra for the identification of different pathogenic bacteria and fungi. Results obtain and experiences acquire on MALDI-TOF SM during these 5 years have conduct to propose practical recommendations for diagnostic application that we will be expose during this presentation.

# Fast *Salmonella* spp. Screening Using MALDI-Typing

ANIL PERSAD, University of the West Indies, St. Augustine, Trinidad and Tobago, Trinidad and Tobago

Matrix Assisted Laser Desorption Ionization Time of Flight Mass Spectrometry (MALDI-TOF), offers a rapid, reproducible method for bacterial identification with a high sensitivity and specificity and at minimal cost. The purpose of this study was to determine the feasibility using MALDI-TOF to differentiate between six *Salmonella* serovars recovered from experimental microcosms inoculated with known strains of *Salmonella*. Following the establishment of a MALDI-TOF reference library for this project, the identity of 843 *Salmonella* isolates recovered from experimental microcosms was assessed using both MALDI-TOF and conventional methods (serotyping /PCR). All 843 isolates were identified as being *Salmonella* species. Overall, 803/843 (95%) of these isolates were identified similarly using the two different methods. Positive percent agreement at the serovar level ranged from 79 – 100% and negative percent agreement for all serovars was greater than 98%. Cohen's kappa ranged from 0.85 to 0.98 for different the serovars. This study demonstrates that MALDI-TOF is a viable alternative for the rapid identification and differentiation of *Salmonella* serovars

# Identification of Cereulide-producing Bacillus cereus by MALDI-TOF MS

**SEBASTIAN ULRICH**, *Ludwig-Maximilians-University*, *Munchen*, *Germany* 

The Bacillus (B.) cereus group consists of nine recognized species which are present worldwide. *B. cereus* play an important role in food-borne diseases by producing different toxins. Yet, only a small percentage of *B. cereus* strains are able to produce the heat stable cereulide, the causative agent of emetic food poisoning.

Currently, only time-consuming cell bioassays, molecular methods and tandem mass spectrometry are available for this purpose. Thus, the aim of the present study was to establish a fast and reliable method for the differentiation between emetic/non-emetic strains by MALDI-TOF MS. Selected strains/isolates of the *B. cereus* group as well as other *Bacillus* spp. (total n = 121) were cultured on sheep blood agar for 48 h before analysis.

Subsequently, the cultures were directly analyzed by MALDI-TOF MS without prior extraction steps. The samples were measured in the mass range of m/z 800 - 1,800 Da. A differentiation between emetic/non-emetic isolates was possible with a rate of correct identification of 99.1% by means of the evaluation of two specific biomarkers (m/z 1171 and 1187 Da).

#### MALDI-based Source Tracking of Listeria monocytogenes to Drive Decision-Making THOMAS CHARRIER, Eurofins, Nantes, France

Listeria monocytogenes is one of the most famous food pathogens that could impact food processes and final products. In the case of final product contamination, the industrial main objective is to rapidly identify contamination source in order to eliminate it. Usual gold standards for pathogens source tracking, like pulse field Gel electrophoresis or WGS, are time-consuming, are cost-effective and sometimes difficult to interpret. MALDI-TOF technology offers fast and accurate bacteria identification and sub-typing. By improving strains pre-treatment, spectra acquisition, and spectra analysis, many variable areas with and relevant biomarkers in Listeria monocytogenes mass spectra have been identified and associate to different MALDI-Types for this species (initial study conducted on 150 different strains). Results obtained are easy to interprate, repeatable and with an equilvalent discriminatory power as PFGE method (Simpson Index). MALDI-TOF strain sub-typing represent today a fast and robust intermediate method for pathogen rapid source tracking.

# S12 Actions Speak Louder Than Words: Ongoing Efforts for Global Harmonization in Standardization in Food Microbiology

The Vienna agreement was already a big step in standardization in the framework of microbiology of the food chain: most of standards developed by ISO are adopted by CEN, and inversely. Nowadays, the European Regulation EC 2073/2005 recognizes fully the ISO and CEN standards. This greatly facilitates food testing, risk management, and of course export/import controls to fulfil the European regulation.

The ISO sub-committee TC34/SC9 on food microbiology has already published 78 standards and is currently coordinating the development of 26 standards. Gathering 31 participating countries and 34 observing countries worldwide, efforts are constantly done to improve global harmonization and recognition of the protocols such as pathogen and quality indicators testing, challenge testing, method validation and verification, strain typing...

Stay up to date, and attend a yearly update on the currently developed and revised standards. Highlights on challenge-testing, method validation, *Clostridium* spp. and *Shigella* spp. analyses will be done to get access to the very last news. This is more than helpful when dealing with a regulated market!

It is well known that some regions of the worlds are running enrichment or plate incubation of at  $35^{\circ}C \pm 1^{\circ}C$ , and other at  $37^{\circ}C \pm 1^{\circ}C$ . What a brain teaser for the FBOs when dealing with export/import controls! Actions speak louder than words: a study has been recently conducted by several laboratories worldwide to harmonize the incubation temperature for *Salmonella* spp. testing. Take a look at the interesting outcomes.

When applying for method accreditation, routine testing laboratories are expected to run a method verification study. There are probably as many study designs as there are laboratories. There are great expectations regarding the currently developed ISO 16140-part 3: this part of the ISO 16140 series will provide clear guidance on how the implement and verify a method, alternative or reference, in routine testing.

#### Postcard from ISO: Stay Up to Date!

JACQUES-ANTOINE HENNEKINNE, ANSES, Maisons-Alfort, France

The main goal of standardization is to develop best practices in food microbiology laboratory and promote their use throughout the world, using a consensus-driven process that balances the viewpoints of industry, government and the end-user laboratories.

For this purpose the ISO TC34/SC9 already published 78 Standards and is currently under development process for 27 new and/or revised standards.

Among the 24 working groups of the SC9, some of them focus their work on conventional microbiology whereas others deal with typing as well as toxin detection methods.

Thanks to examples on spore forming bacteria (clostridies spp.), this presentation will focus on the importance to develop and standardize methods for typing and toxin detection.

#### Best of Both Worlds in Salmonella Testing

**KIRSTEN MOOIJMAN,** *Convenor of the ISO 6579 standard, EURL on Salmonella spp., RIVM, Bilthoven, The Netherlands* 

For incubation of microorganisms at 'human body' temperature, slightly different temperatures are used in different parts of the world: 37°C is generally used in Europe, while 35°C is generally used in the USA. The reasons for the differences in incubation temperatures are unclear. For harmonization of world-wide use of microbiological methods, it is important to harmonize the incubation temperatures as well. This has drawn the attention of the ISO subcommittee of 'Microbiology of the food chain' (ISO/TC34/SC9). A protocol was drafted by this subcommittee to test 35°C versus 37°C incubation of selective enrichment medium Muller-Kauffmann tetrathionate-novobiocin (MKTTn) broth and isolation media for the detection of Salmonella (ISO 6579-1). Members of the ISO subcommittee were requested to apply the protocol, preferably to natural ('routine') samples with high amounts of background flora. Study results were received from 9 laboratories from 6 different countries, representing 850 test results of which 236 were positive for Salmonella. Results confirmed for the presence or absence of Salmonella were analyzed and interpreted following the information for the sensitivity study according to ISO 16140-2. Additionally, the laboratories reported whether high, medium or low amounts of background flora were found at both temperatures. From the studies, it was concluded that the differences in the number of samples positive for Salmonella when testing ISO 6579-1 with MKTTn/isolation media -37°C and with MKTTn/isolation media -35°C, were within the acceptability limits. Additionally,

the reported differences in amounts of background flora after incubation at the two temperatures were small. Hence, for incubation of selective media for detection of *Salmonella* a range of 34–38°C can be used.

#### Method to Our Madness: The ISO 16140-3 Standard on Analytical Method Implementation and Verification

BENJAMIN DIEP, Nestlé Research, Lausanne, Switzerland

When selecting a new microbiological method, a laboratory shall consider the validation status of this method, not only to meet the requirements of the food safety authorities and accreditation bodies, but also to make sure the selected method is fitting-for-purpose. The European regulation 2073/2005 and the related amendment 2019/229 recognize the use of national/international standardized reference methods, or the use of rapid alternative methods validated against the appropriate reference method according to the technical rules of the ISO 16140-2:2016 standard. In addition, according to the ISO 17025 standard that provides the requirements for accreditation, the laboratory has to demonstrate its competence to run a method before using this method in routine. But how to proceed, what is the study design to be run, what are the requirements to meet?

Until now, there was no harmonized protocol available to verify the method before its implementation in a laboratory. The ISO 1640-3 has been developed during the past years to describe the technical rules for the verification of reference methods and validated alternative methods; this standard is now at the final stage before publication. The ISO 16140-3 applies to qualitative, quantitative and confirmation methods. It consists in two steps:

- implementation verification to demonstrate that the laboratory is able to perform the method correctly
- (food) item verification to demonstrate that the laboratory is capable of testing the (food) items it claims
- Performance characteristics and acceptance criteria are defined, taking into consideration the practical feasibility, that gives a clear guidance for any laboratory that perform method verification.

# S13 Food Safety Culture: The Proof is in the Science

Food safety culture required a major leap in scientific research, linking social science with food sciences, embracing food safety practices on a more fundamental, cultural level. The Global Food Safety Initiative (GFSI) position paper, A Culture of Food Safety, provides guidance to scheme owners about incorporating cultural aspects into GFSI benchmarked standards, and to businesses and stakeholders on maturing organizations' food safety cultures. It reflects and bundles viewpoints and approaches of scientists and practitioners into 5 dimensions: Mission and Vision, People, Consistency, Adaptability and Hazards and Risk Awareness. This symposium presents latest research from leading scientists participating in the GFSI-linked Science Group, Salus. It provides a holistic approach, incorporating all dimensions of food safety culture, and showing how scientific research is giving rigour to culture application for food businesses.

Presenters will discuss research relevant to each of the GFSI dimensions. Commencing with the importance of leaders' food safety culture mindset in setting direction and aligning expectations and messaging towards stakeholders (*Mission and Vision*), speakers will demonstrate essential characteristics of the dimensions *Consistency*, achieving standards and remaining within the context of a strong shared mission of food safety culture, and *Adaptability*, being able to adjust and respond to changing influences and conditions. The dimension *People*, the critical component of a food safety culture will be covered from multiple perspectives. People's behavior, practices and decision making, and their education/learning and empowerment strongly contribute to the safety of our food products, helping them understand their important role and motivating them in fostering food safety. The symposium will close with examination of cultural failures in food outbreaks revealing the lessons that we must learn (*Hazards and Risk Awareness*). The audience will take away key messages on how to focus within the cultural dimensions at their businesses and build a stronger food safety culture.

# Food Safety Culture – Walking the Talk (Mission and Vision)

LONE JESPERSEN, Cultivate Food Safety, Hauterive, Switzerland, Switzerland

This presenation aims to provide an overview of the GFSI diemnsions for a culture of food safety as published by GFSI in 2017. The presentation will briefly dive into each and provide examples of how companies can continue to improve their food safety performance through its culture of food safety. The presentation will also share data from five global companies and the maturity of their food safety cultures as well as degree to which they walk the food safety talk every day. Futhre the presentation will challenge participants to make a link between cost of running a food safety management system and the maturity of its food safety culture.

# Key Aspects of People Systems and Adaptability for Effective Food Safety Culture (People Systems and Adaptability)

LIESBETH JACXSENS, Elien De Boeck, P. Vlerick, Department of Food Technology, Food Safety and Health, Faculty of Bioscience Engineering, Ghent University, Ghent, Belgium

Introduction: A key characteristic of people is the fact that they have perceptions and can perceive and interpret things according to their own 'thought world,' which can be considered as the first 'level' of culture, i.e., the 'artifacts' as described by Schein (2017). This top of the iceberg of culture can be considered as one of the observable, and therefore measurable features of culture.

Materials and methods: In this research, perceptions of employees (and managers) throughout all hierarchical layers about the value of food safety in the organization are considered as the 'food safety climate.' Food safety climate is defined as employees' (shared) perception of leadership, communication, commitment, resources and risk awareness concerning food safety and hygiene within their current work organization. A self-assessment tool was developed and validated.

**Results**: Use of this tool in case studies in food industries led to important insights about the first two components: leadership and communication. 'Leadership' appeared to be one of the most important dimensions of food safety climate. For example, do leaders strive for continuous improvement of food safety and hygiene, resulting into adaptability towards new situations? Another important dimension to consider is 'communication.' Is food safety message clearly communicated in the company? As it is the individual employee who will or will not take (correct) decisions or comply with food safety and hygiene procedures, a deeper exploration of this individual employee and personal characteristics was performed.

**Conclusion**: A conceptual model was proposed by which the importance of food safety behavior, knowledge, motivation, burnout and job stress in food safety management was demonstrated.

Schein, E. H. (2017). *Organizational culture and leadership* (5th edition ed.). Hoboken, New Jersey: John Wiley and Sons, Inc.

#### Achieving Consistency Based on Science and Risk Understanding – Learning the Lessons to Push Beyond the Checkbox FSMS (Consistency and Hazards and Risk Awareness)

**CAROL ANNE WALLACE,** University of Central Lancashire, Preston, United Kingdom

This presentation aims to address two dimensions of the Global Food Safety Initiative position paper, namely Consistency and Hazards and Risk Awareness.

Hazards and Risk Awareness can be thought of as the dimension that makes food safety culture specific to food safety rather than broader organisational culture. Fundamental knowledge, expertise and experience around food safety hazards, assessment of risk and identification and implementation of appropriate control measures is essential in all food businesses. By examining cultural failures in food outbreaks and incidents the presentation will explore the lessons we must learn such that we can verify that businesses have the necessary level of hazard and risk awareness and that this is applied effectively through employee engagement.

Consistency relates to the alignment of food safety priorities with requirements around people, technology, resources and processes such that there is consistent application of the food safety programme for effective food safety performance. Consistency needs to flow through all food safety related decisions, actions and behaviours at all levels of the organisation and this reinforces a culture of food safety. However, all too often, consistency within businesses is related to compliance with existing standards and procedures, and the food safety management system (FSMS) becomes part of a checklist with items checked off without really considering if they align with food safety priorities or how measurements can influence or provide information about food safety culture. Working together with effective hazard and risk awareness, consistency can ensure that the right things are measured and evidenced, and that personnel are accountable for design and improvement of approaches related to food safety priorities.

# S14 Applications of Microbial Profiling: The Present and the Future

Recent advances in sequencing technology have allowed the food microbiologist to explore the microbiome of food products and food production environments in unprecedented detail. This technique has now moved from the academic field to be applied in a practical manner by the food industry. The first theme of this session will be to explore how the science of metagenomics is being used currently by the food industry as well as to highlight some areas in which it could easily be deployed. The second theme of the session is the future of metagenomics for the food industry, as envisioned by a thought-leader in the field.

The first speaker will introduce the topic briefly, then present a series of case-studies of the practical application of the technique. These case-studies are the result of a twoyear research project looking to demonstrate the value of this methodology to the food industry.

The second speaker comes from an industrial background and will detail their experience of metagenomics being used to investigate an issue that was otherwise intractable through the use of traditional microbiological investigation.

The third speaker is from an academic background, and is a thought-leader in the area of metagenomics in foods. They will explore some of their current research as well as providing some insight into the direction of travel the food industry needs to take if the full potential of metagenomics is to be realised.

The relevance of metagenomics to the food industry is increasing, and it is hoped that this session will demonstrate that relevance as well as providing a guide to future developments.

# Microbial Profiling: What is It, and What Can It Do?

**GREG JONES,** Campden BRI, Chipping Campden, United Kingdom

Microbial Profiling is best described as the application of metagenomics techniques to describe a microbial population. In the food industry, the predominant form this takes is via sequencing heterogeneous amplicon pools targeted at the gene coding for the 16S ribosomal RNA subunit to generate bacterial profiles. The applications of this technique are becoming increasingly apparent to the food industry, and this talk will describe some practical applications of Microbial Profiling. The results presented here are based on a 2-year research project performed at Campden BRI to explore the practical applications. The areas that will be explored will be: Shelf life of iceberg lettuce and shelf life of beefburgers; profiling of Italian hard cheese; spoilage investigation; origin of retail chicken samples and the impact of 'superchilling' on product microflora. These case-studies were designed to assess the feasibility of using profiling to advance our understanding of complex food microbiota and deliver real benefit to the food industry.

# Use of Metagenomics in an Industrial Setting

JULIA HEWERDINE, Dunbia – A Division of Dawn Meats, Crosshands, United Kingdom

Traditional microbiological culture methods mean you only find what you are looking for. This method of operation works well in many circumstances when risk assessment has taken place to identify bacteria of interest or there is a need to check if controlling process have done what is expected. However sometimes this is not enough. Sometimes you don't know what you are looking for. Sometimes the complete picture is needed.

Using 16-S metagenomics to identify all bacteria living or dead is a really useful tool in exploring food safety in real-life situations because of the complete picture it can provide. This talk reviews the practical application of 16-s metagenomics in a red meat boning hall where the method was utilised to understand the root cause of an anomaly in TVC data. The rationale behind selecting metagenomics is explored, along with the methodology of sampling, the metagenomic results and suggested conclusions.

#### **Current Research in Food Microbiomes** and How Industry Can Benefit

**FRANCESCA DE FILIPPIS**, University of Naples Federico II, Portici, Italy

The use of high-throughput sequencing (HTS) technologies strongly changes our way to approach to the study of microbial ecology. In recent years, they were used to support food industry in guality management, risk assessment and research and development. HTS-based monitoring of spoilage microorganisms in food-processing environment and mapping of the possible contamination routes is useful to prevent microbial spread along the processing chain and consequently the transition to the final product. The resident microbiota in food-processing plants can be the source of spoiling or pathogenic bacteria, that can proliferate during storage to unacceptable levels, compromising food quality and safety. Nevertheless, depending on the type of manufacturing considered, food processing microbiota may sometimes play a positive role. Indeed, cheese manufacturing plants often harbour lactic acid bacteria, beneficially involved in the fermentative and ripening processes. In addition, the use of whole metagenomics, although still limited in food industry, may be useful to understand the genomic mechanisms leading to spoilage or to the production of positive features during food production. In the future, the food processing industry will surely benefit by the integration of sequencing-based technologies for management and improvement of food quality and safety.

# S15 Hepatitis E Virus, an Emergent Foodborne Pathogen? Public Health Implications

Hepatitis E virus (HEV) is one of the main causes of viral acute hepatitis in humans worldwide. The origin of HEV infection in humans in industrialized countries is linked to direct contact with infected animals or to consumption of raw or undercooked meat and liver, particularly of pig origin. Likewise, the presence of HEV has been described also in other livestock animals, fruits and vegetables, and mollusks.

New epidemiological evidences are posing HEV as a relevant emerging foodborne zoonotic disease in Europe. Indeed, over the last 10 years the reported cases have been increased in 10 fold times, even though that is considered and underdiagnosed and underreported disease. Most cases are associated with the consumption of raw or undercooked meat and liver pork products. Taking into account that pork meat is the major type of meat produced in EU/EEA countries and that European pork meat exportation balance is very positive, the implications at European level in both on the market and consumers could be extremely serious, particularly after the press coverage in UE and particularly in UK linking pig products with cases of HEV infection. Thus, not only Animal and Human Health professionals and porcine producers are concerned about HEV, but also general pop ulation.

Consequently, EFSA has published recently a scientific report that highlights the high increase of HE cases in Europe and emphases the need for more research efforts on the epidemiology, diagnosis and control of HEV. In this symposium, we will present the more relevant advances inepidemiology, diagnosis and control of this virus.

#### Hepatitis E Virus Burden in Europe: What Do We Know?

WIM VAN DER POEL, Wageningen University, Lelystad, The Netherlands

Hepatitis E virus (HEV), family Hepeviridae, is a main cause of epidemic hepatitis in developing countries and sporadic and cluster cases of hepatitis in industrialized countries. Genotypes 3 and 4 of the virus have zoonotic potential and cause single cases of hepatitis throughout the world. Both of these genotypes have a main reservoir in domestic swine and this leads to contaminations in the food chain. The virus may be transmitted to humans by pork products and different types of foods like shellfish, fruits, vegetables, and water as environmental routes may be involved.

There is an increasing number of reported cases in humans especially in industrialized countries and there is a high potential for transboundary spread of zoonotic genotypes of the virus through the transport of pigs, pig products and by-products. Bloodborne transmission of the virus has been reported with a significant medical concern.

HEV is a relatively stable non-enveloped RNA virus, and may remain infectious at temperatures used in some cooking regimes. Best feasible inactivation methods include heating at plus 71°C, chlorine treatment and UV light. Focal points for control along the food chain depend on the type of food and the stage in the food production process. There is a need for a sensitive and broadly applicable method to test food items for HEV infectivity.

Besides control options at the pork retail level and by the consumer it is important to try to reduce HEV in primary production and the whole swine reservoir. Raising the level of biosecurity on swine farms is indicated, so farms should review and tighten their biosecurity protocols and be especially diligent about visitors and supplies, feed ingredients, food items, etc.

In immunocompromised and liver disease patients, HEV infections can be very serious and fatal. Ribavirin monotherapy currently is the treatment of choice for patients chronically infected with HEV; however, this is not always successful. Therefore, in a number of European countries it has been decided to test blood donations for HEV positivity.

# Hepatitis E Virus in Animals, Food and Where Else?

NICOLE PAVIO, ANSES, Maisons-Alfort, France

In Western countries, the concept of zoonotic hepatitis E has emerged two decades ago following the discovery of animal strains of hepatitis E virus (HEV). In particular, HEV strains circulating in domestic and wild pigs are genetically very similar to strains identified in human cases. Contamination of pork products (liver, sausages, pies...) by HEV is now well described and the consumption of raw or undercooked contaminated meat or meat products are known to transmit. Hence, HEV can be a zoonotic foodborne pathogen. Recent advances in the identification of HEV strains in other animal species (rabbit, camel, rat...) and their detection in human have confirmed that several HEV members of the Hepeviridae family can be zoonotic. Thus, other food products such as meat or milk from other animal species (camel) may also represent significant risks. Possible HEV zoonotic reservoirs in ovine or bovine species are still debated. Further investigations are needed to screen for the presence of infectious HEV in larger variety of animal species and derived food products.

HEV from infected human or animal faeces can be released in effluents (waste water treatment plans, slurry ...) leading to contamination of the environment. Hence, food of non-animal origin consumed raw, such as crops, berries and bivalve molluscs, could vehicle HEV. Environmental contamination may also lead to accidental water borne Hepatitis E in developed countries.

An exhaustive understanding of HEV animal reservoirs and transmission routes is essential to control and prevent efficiently this disease with increasing public health concern.

#### Detection Methods for Hepatitis E Virus – What Do We Need?

**REIMAR JOHNE**, Federal Institute for Risk Assessment, Berlin, Germany

The hepatitis E virus (HEV) genotype 3 has its major reservoir in domestic pigs and wild boars, and the foodborne transmission route is considered most important for human infections. However, many questions on specific transmission pathways, high risk food products and safe treatments of food are hampered by the lack of sensitive and standardized detection methods for HEV and its infectivity.

Several protocols for HEV detection based on molecular techniques like real-time RT-PCR are available. However, most of them have not been validated or standardized for their use with food products. Low-virus concentrations and the presence of PCR inhibitors are often challenging for method development in food virology. Recently, a detection method for HEV RNA in meat products has been optimized for liver sausages and raw sausages, and validated in an interlaboratory ring trial. An international standardization of HEV detection methods should be aimed in order to allow reliable HEV testing of food and comparison of results. In addition, typing protocols should be harmonized.

Although molecular methods may enable reliable, HEV genome detection, they cannot distinguish between infectious and inactivated virus. HEV infectivity determination is still hampered by the lack of appropriate methods. Limited infectivity studies have been done by experimental inoculation into pigs; however, this system is generally restricted. Cell culture systems may overcome these problems, but robust and sensitive systems are only rarely described. Novel cell culture-adapted HEV strains, which show more efficient and reproducible growth, may be used in future HEV inactivation and disinfection studies. In addition, alternative methods, such as molecular capsid integrity assays, should be developed for HEV and validated against infectivity assays.

# S16 Challenge Testing for *Listeria mono-cytogenes*: Requirements, Needs, Difficulties and Developments

*Listeria monocytogenes* in ready-to-eat (RTE) foods in Europe is becoming increasingly important due to the increase of cases of illness and deaths due to listeriosis. Since 2005 there are legal requirements for *L. monocytogenes* asking RTE producers to carry out studies on the growth capabilities of *L. monocytogenes* in their foods.

These studies can include challenge tests for which there are guidance documents form the EU and EURL (EU Reference Laboratory) available to help the Food Business Operators and laboratories performing these challenge tests. Since 2014 ISO and CEN are also working on drafting a standard on challenge testing (ISO 20976-1) which will be published in 2019.

In this symposium more information will be given on the developed ISO standard on challenge testing and the link between this document and the existing EU-documents. Also difficulties in performing challenge test and the interpretation of results will be discussed. These question will be highlighted in this symposium that is targeting both FBOs that have to submit studies as well as and laboratories that conduct challenge tests and authorities evaluating them.

#### Challenge Testing and Standardisation, Recent Developments

FLORENCE POSTOLLEC, ADRIA – UMT AC-TIA19.03 ALTER'IX, Quimper, France

Under the general principles of the Codex Alimentarius on food hygiene, it is the responsibility of Food Business Operators to control microbiological hazards in foods and to manage microbial risks. Challenge test is one of the recognized approaches used to validate control measures within the HACCP system, as well as to assess microbiological safety and quality of food, food production processes, food storage conditions and food preparation recommendations for consumers. In agreement with already available and valuable guidance document, the aim of ISO20976-1:2019 standard is to provide general requirements and guidelines for conducting challenge tests on food and feed products.

Within the frame of the International Organization for Standardization (ISO), members from all over the world collaborate to create internationally recognized documents providing requirements, specifications, guidelines or characteristics that can be used consistently to ensure that products, processes and services are fit for their purpose. The working group ISO/TC34/SC09/WG19 comprises experts from the food industry, food technology institute, food testing laboratory, research center and regulatory bodies. WG19 developed a standardised protocol to conduct challenge tests to study growth potential, lag time and maximum growth rate (ISO 20976-1:2019) which has a link with European legislation (Regulation 2073/2005) on microbiological criteria for foodstuffs. At present WG19 is working towards consensus on two additional standards: challenge tests to study inactivation potential and kinetics parameters (ISO 20976-2) and determination and use of cardinal values in predictive microbiology (project ISO 23691).

# Listeria monocytogenes and Challenge Testing, a European Perspective

HÉLÈNE BERGIS, ANSES, Maisons-Alfort, France

Regulation (EC) No 2073/2005 on microbiological criteria for foodstuffs sets out specific food safety criteria for *L. monocytogenes* in ready-to-eat (RTE) foods. A quantitative limit of 100 *Listeria monocytogenes*/g for RTE foods placed on the market during their shelf life has been laid down. According to article 3.2 of the Regulation, Food Business Operators (FBO) producing RTE foods, shall conduct studies, in accordance with Annex II, in order to investigate compliance with the criteria throughout the shelf life.

Beside Regulation (EC), guidance documents were drafted for:

- Food business operators in order to guide them in identifying the monocytogenes risk in their RTE foods "Guidance document on Listeria monocytogenes shelf-life studies for ready to eat foods, under Regulation (EC) No. 2073/2005 of 15 November 2005 on microbiological criteria for foodstuffs," DG-Sanco 1628/2008 ver. 9.3, (DG-SANCO Guidance document).
- Laboratories to help them in implementing shelf-life studies with detailed and practical information on challenge tests and durability studies. "EURL Lm Technical guidance document for conducting shelf-life studies on *Listeria monocytogenes* in ready to eat foods," version 3 – 6 June 2014).
- National Competent Authorities (CAs), NRLs and other organisations involved in assessing whether laboratories are competent to conduct shelf-
- life studies related to *Listeria monocytogenes*. "EURL Lm guidance document to evaluate the competence of laboratories implementing challenge tests and durability studies related to *Listeria monocytogenes* in RTE foods." Version 2 – 3 May 2018.

# Conducting Challenge Tests for *Listeria monocytogenes*, a Real Challenge

PAUL IN'T VELD, NVWA, Utrecht, The Netherlands

Despite a recently published standard on challenge testing and a European technical guidance document it is not easy to conduct a challenge test properly. To be able to conduct a challenge test, different kind of expertise is needed coming from different parties involved. The first is the Food Business Operator (FBO) who has the knowledge on the composition, production process and intrinsic factors of the specific ready-to-eat food. On the other site there is a laboratory that has the analytical knowledge. In addition, a third party might be involved with knowledge on how to set up a challenge test, e.g., how to spike the food in such a way that is representative of the possible contamination route of Listeria in the product. This is just a example on what is needed even before a challenge test is started. In practice many foods are involved as the ready-to-eat foods are a very diverse group of products, such as pre-packed sandwich, smoked salmon or sliced delicatessen meat.

In this presentation the steps that need to be taken to conduct a proper challenge test will be presented together with examples on what can go wrong. At the end both the FBO and the responsible authorities have to agree that the work done is truly representative for the food (or group of foods) that has been challenged. This should lead to greater confidence in the control of *Listeria monocytogenes* in readyto-eat food and thus leading to safer food.

## S17 Water Re-Use in Food Processing Industry – It's Inevitable!

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. Amongst the possible other sources of water available to the food industry could be water that is recovered from food or from particular 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 dealt with. There is as yet little science and operational best practice for responsible water re-use in key segments of the food industry. Certainly, when deciding whether a re-use water source can be utilized for a particular food application and whether or not 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 risk to the safety of the consumer that eats the food product. Essentially, a risk-based approach needs to drive matching re-use water sources/treatments to possible applications. Risks need to be carefully assessed and

managed in the specific context of the particular food facility and risk-mitigation measures must be managed within that facility's food safety management system (e.g., GHP/HAC-CP). As more food industry sectors in more countries start to explore water re-use and develop new solutions, innovative methods and scientific data, there is an opportunity to reduce the learning curve by sharing practical experiences. This symposium will introduce risk-based approaches and practical experiences.

# JEMRA Risk-based Framework to Water Re-Use Under Development

**LEON GORRIS**<sup>1</sup> and **JEFFREY LEJEUNE**<sup>2</sup> <sup>1</sup>Food Safety Expert, Nijmegen, The Netherlands <sup>2</sup>Food Safety and Quality Unit, AGFF, Food and Agriculture Organisation, Rome, Italy

Water is a major input in food from primary production through all stages in the value chain to consumption. Water can contact food directly or indirectly as it is used as a food ingredient or for maintenance of hygiene and sanitation and in food production and processing operations. Water is a diminishing resource globally and not all food producers and processors have access to safe water sources. Water has to be used conservatively and it is possible to reuse water if it does not present a health risk for consumers.

Through their JEMRA expert resource group, The Food and Agriculture Organisation (FAO) and the World Health Organisation (WHO) has started to compile guidance on the safe reuse of processing water as well as on the safe use of water in the primary production of produce and, fishery products.

The safest option in all applications may be using of water of potable or drinking water quality. However, this is often not a feasible, practical or responsible solution and other types of water could be fit for some purposes provided they do not compromise the safety of the product for the consumer.

As part of the guidance, a framework was developed that may help users to decide on the safe, fitness-for-purpose use of water sources from within food business operations. The framework considers the health risks of food at consumption and addresses the context for water use at a particular step and location in the food value chain.

While the framework informs users of the general principles for fit-for-purpose water re-use, there is a high level of diversity and variability in food products, water/food/microbe interactions, microbial hazards and factors influencing their presence and control at different stages along the supply chain, and the end use of food products that have to be considered in operationalising the framework.

# Experiences with Water Re-Use in Dairy Operations

MARTIN ANDERSEN<sup>1</sup> and Claus Heggum<sup>2</sup> <sup>1</sup>IN-Water ApS, Brabrand, Denmark <sup>2</sup>Danish Agriculture & Food Council, Aarhus N. Denmark

Dairy operations consume high amounts of drinking water resulting in similar high amounts of waste water discharge. Reuse and recycling of water as well as claiming water from milk, whey and other dairy liquids often constitute good business cases. Means to claiming safe milk water (or "cow water") is currently gaining interest among dairy processers world-wide, mostly in Northern Europe, California and Australia where systems are in place to reduce the need for external water supplies.

The legal barriers to reuse and reclamation are subject to review by competent authorities, including Codex Alimentarius, which is expected to lead replacing the approach of mandatory use of drinking water quality in food production with the concept of fit-for-purpose quality of water. Seizing these new opportunities requires properly documented safe and suitable best practices that do not negatively impact on the quality of milk and milk products.

The presentation will address the experiences of the Danish dairy sector resulting from a private-public partnership development project "Water Efficient Dairies" which are made operational through a best Code of Practice on Reclaim and Reuse of Water. The Code will provide 14 ready-to-implement scenarios – most of them tested, documented and implemented in the Danish dairy processing industry.

The presentation will address best technology for dairying, the control of operations and of the hazards involved and means to maintain microbial control.

#### Experiences in Water Reuse in the Beverage Industry

JOSEP MOLAS PAGES, Coca-Cola, Madrid, Spain

Food and beverage producers use water in their facilities for several purposes. Water can be used for cleaning and rinsing purposes, toilet and handwashing facilities, watering, etc. However, the most critical use is in cases where the water may come in indirect contact with a foodstuff or be used as an ingredient in the final product.

In the European Union, the water used in food facilities shall meet the quality criteria in the Drinking Water Directive 98/83 EC, unless the authority is satisfied with the quality of the water that cannot affect the wholesomeness of the finished foodstuff.

Water reuse in the food and beverage industry can be done applying HACCP principles and taking into consideration the intended use of the reused water. A critical aspect in food and beverage processes is the microbiology, but also traces of chemicals and by-products that may be present due to different processing.

Reuse of water implies decision trees to ensure that the final product and health of the workers and consumers are not affected. A fit-for-purpose reuse needs to be established. Water origins need to be carefully assessed and reconditioning evaluated accordingly to ensure compliance with applicable regulations and food law.

In a world where fresh water resources are stressed, the reuse of water shall be considered and practiced where feasible.

## S18 Fungal Spores in Food; Implication of Natural Heterogeneity on Food Quality

Fungal spores are resistant and therefore widespread in food ingredients and foods. Efficacy of control of spores by the food processing industry is challenged by diversity between and within strains with respect to their spore robustness and ability to rapidly germinate. Food processing and food characteristics can impose significant selection forces on spores, and thereby selecting for the most robust spores and/or the best grower. This symposium will focus on diversity of fungal spores in food and the implication on controlling food quality. We will address these issues with experts in the spore research field, discussing heterogeneity in (heat) resistance of fungal strains. Also we will discuss the effect of (a)biotic factors on heterogeneity in formation and outgrowth of individual fungal spores, thereby highlighting the relevance of these aspects in the actual risk evaluation and control of spores to protect food.

## Significance of Environmental Conditions during Sporulation on Physiological State and Phenotypic Heterogeneity of *Penicillium roqueforti* Conidia

NICOLAS NGUYEN VAN LONG, ADRIA Food Technology Institute, Quimper, France

The control of fungal spoilers in the food processing industry addresses economic and sanitary issues. In this context, the consideration of phenotypic heterogeneity among asexual spores (conidia) populations is relevant towards decision tools such as predictive mycology. The present work illustrates how environmental conditions occurring during the sporulation can affect the physiological state of *Penicillium roqueforti* conidia and phenotypic heterogeneity within conidial population. Using a strain isolated from blue cheese, conidia were produced at 7 different environmental conditions including optimal and suboptimal levels of temperature, pH and water activity ( $a_w$ ). Following a monofactorial study design, the germination kinetics of these conidia were modelled in different temperature, pH and  $a_w$  conditions. Model provided kinetic parameters related to the ability to germinate within a population ( $P_{max}$ ), the median germination time (r) and estimate of heterogeneity among individual germination times (d).

Conidia produced at suboptimal temperature (5°C) and  $a_w$  (0.900  $a_w$ ) germinated significantly earlier (up to 48 h earlier) than those produced at optimal levels (20°C and 0.980  $a_w$  respectively) regardless of germination conditions. An effect observed on *d* parameter suggested that environmental conditions during sporulation could affect variability among conidia. Further investigations of intracellular trehalose and mannitol assessments suggested that earlier germination might be related to delayed conidial maturation, even though no ultra-structural evidences were observed by transmission electron microscopy (TEM) analysis.

This study suggested a significant relation between maturation process, physiological state of conidia and individual germination behavior; altogether affected by environmental conditions. Such heterogeneity is still poorly taken into account when designing experiments for food spoilage prevention as well as predictive mycology tools.

#### Strain Variability in Conidial Heat Resistance of Food Spoilage Fungi TOM VAN DEN BRULE, Westerdijk Fungal

Biodiversity Institute, Utrecht, The Netherlands

Fungal food spoilage often begins with contamination by spores. To prevent spoilage of processed foods and drinks, industry often challenges their products with the worst-case spoilage scenario. Interspecies and intraspecies variability of food spoilage fungi should be taken into account when defining the worst case presented by fungal spores. Among other types of spores, food spoilage fungi produce airborne conidia. These asexual spores vary in resistance properties among species, but also within species. Recent studies showed that growth conditions can alter heat resistance and germination time of conidia. This presentation focusses on strain variability in heat resistance of conidia. Conidial heat resistance was measured of various Paecilomyces variotii strains and variation was quantified by methods adapted from bacterial spore research. The most heat resistant conidia measured to date were only produced by distinct strains of Paecilomyces variotii. Furthermore, physiological properties as internal compatible solute composition and spore size appeared to be related to strain variability in conidial heat resistance. This work emphasizes the importance of quantifying strain variability in predictive food microbiology to realistically predict heat inactivation of fungal spores.

# Tackling Preservative-resistant Fungal Spores

**SIMON AVERY,** University of Nottingham, Nottingham, United Kingdom

Like populations of single cells, fungal spore populations display marked phenotypic heterogeneity. Phenotypic heterogeneity describes differences in a measurable phenotype between individuals, despite genetic uniformity of the population. Such heterogeneity has implications for control of food-spoilage and other foodborne fungi, as measures like food preservatives may only impair a portion of the fungal population. Indeed, heterogeneity is evident in the responses of spoilage-fungi to weak acid preservatives like sorbic acid. Heterogeneous resistance of spoilage fungi to sorbic acid appears to be rooted primarily in the spore stage, originating prior to the onset of germination that occurs in nutrient-rich conditions. A number of factors may underlie such sporespore heterogeneity. Heterogeneity is also apparent with low-residue potential food protection measures, like treatment with electrolysed water or use of materials that resist spore attachment. Effective combination approaches would be needed to target the diverse spore subpopulations likely to be hyper-resistant to different food protection measures.

## S19 Ongoing Research Activities in Risk-Benefit Assessment of Food

Risk-Benefit Assessment and multi-risks assessment can be used to evaluate the consequences of changes in food intake, i.e., changes in food ingredients, processing, formulation, packaging, consumer behavior, etc. These consequences can be expressed in terms of safety (public health) and quality (spoilage). Risk-Benefit assessment is based on nutrition, toxicology and microbiology as well as modelling and multi-criteria decision analysis.

In the last decade, risk-benefit assessment has been deployed by food agencies and research institutes with applications on various domains. In particular, DTU has worked on method development with focus on food substitutions while INRA has worked on infant formula. Based on their experience, they are currently running challenging projects such as the followings:

Dietary advices include choosing whole grain products over refined carbohydrates as well as increasing fish and decreasing red meat consumption. These advices all indicate substitution of one food with another. The overall health impact of substitutions (brown for white rice and fish for red meat) has been evaluated in a common health metric, the number of years lived with disability (DALY).

Red meat consumption receives a lot of attention nowadays. The risk of foodborne outbreaks, cardiovascular fatalities and colorectal cancer cases were compared with the beneficial effect of anemia reduction. All these health effects were expressed in the same unit, the DALY. Scenarios were built to identify the optimum quantity of red meat consumption per age class and gender.

In parallel, multi-risks assessment has gained interest in the industry to evaluate the application of clean label preserving techniques in relation to the potential growth of spoilage organisms.

Results from these three on-going activities will be presented; moreover, the similarities and differences in the methodology carried out in the three studies will be highlighted to help the audience to seize this new research area.

#### Taking Substitution into Account in Risk Benefit Assessment – Two Case Studies LEA SLETTING JAKOBSEN, National Food Institute

Technical University of Denmark - DTU, Lyngby, Denmark

Maarten Nauta, Sara Pires, Sofie Theresa Thomsen, National Food Institute Technical University of Denmark – DTU, Lyngby, Denmark

Risk-benefit assessment of foods (RBA) may be applied to estimate the overall health impact associated with a change in intake of a particular food. This change in intake is usually the change from the current intake to an alternative intake. Up until now, most RBAs have ignored substitution, that is the fact that a change in intake of one food product usually results in a concomitant change in intake of other foods, which in turn may impact health risks and benefits. In this presentation, we show different approaches to account for substitution between foods in two cases: 1) an RBA on substituting red meat by fish, and 2) an RBA of substituting white rice by brown rice. In both case studies, overall health impacts are estimated in Disability Adjusted Life Years (DALYs). We discuss the impact of accounting for substitution of foods in RBAs and its implication on the formulation of food based dietary guidelines, as well as identified future challenges

# Risks and Benefits Associated with Red Meat Consumption in France

JULIANA DE OLIVEIRA MOTA<sup>1</sup>, SECALIM, INRA, Oniris, Université Bretagne Loire, Nantes, France Patrick Tounian<sup>2</sup>, Géraldine Boué<sup>1</sup>, Fabrice Pierre<sup>3</sup>, Sandrine Guillou<sup>1</sup>, Jeanne Marie Membré<sup>1</sup>

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The consumption of red meat has become a public health concern, notably due to its link with foodborne illnesses, cardiovascular disease and colorectal cancer. However, red meat also has beneficial effects by the nutritional contributions, in particular iron, which contributes to the decrease of iron deficiency and, subsequently, anemia.

The aim of the study was to quantify the risks and benefits associated with red meat consumption, and finally to express them in disability adjusted life years, to enable a comparison of the impact of various health effects on the output.

Probabilistic risk-benefit assessment models were built to quantify risks and benefits from red meat consumption, per age, class and gender. The effects of variability and uncertainty of the models' outputs were also characterized.

Current consumption of red meat may induce 19 [95% CI = 8-33] DALYs per 100,000 people per year for colorectal cancer and 21 [95% CI = 12-32] DALYs per 100,000 people per year for cardiovascular disease mortality. Foodborne illnesses attributable to red meat were responsible of 44.5 [95% CI = 29.6-65.4] DALYs per 100,000 people per year. In return, consumption of red meat could decrease up to 12.5 [95% CI = 8.1-16.7] DALYs per 100,000 people per year.

This study may help authorities in settings recommendations and also help consumers to grasp consequence of their food habits.

# First Results and Lessons Learnt from RBA Application in the Industry

**ANNEMARIE PIELAAT,** Unilever R&D, Vlaardingen, The Netherlands

Securing product quality needs continuous attention in Unilever's strategy to improving nutritional value and reducing the environmental footprint. The development of on-trend sustainable innovations is inseparably connected to this strategy. Science based knowledge and technology development for natural food preservation is a consumer-relevant focus that contributes to Unilever's overall scope. Yet, from a microbiological perspective, natural food preservation is inherent to product development towards boundary conditions for microbial growth. This gives rise to the need for risk benefit approaches that can reveal the impact of a change in the product (e.g., salt reduction) or process (e.g., mild heat treatment) at an operational level on the microbiological safety and stability of a product. In this symposium we will reflect on where Risk-Benefit approaches could contribute to Unilever's sustainable innovation strategy and illustrate with a case study on assessing the benefits of salt reduction and the associated microbiological risk in intermediate moisture foods.

# S20 Norovirus, Glycans and Oysters: The Perfect Association?

Norovirus (NoV) was ranked among the top foodborne disease causes by a recent global burden of disease estimate from the World Health Organization Food Epidemiology Reference group (FERG). Regrding foodborne alerts reported through the European Union Rapid Alert System for Food and Feed (RASFF), NoV accounted for 92% of alerts with bivalve shellfish implicated in 85% of theses, mainly oysters. While the use of ISO 15216 method will help to prevent marketing contaminated oysters the major question is how the detection of viral genome translates to human health risk. NoV is a group of highly diverse viruses and very resistant in environmental conditions. One specificity of NoV is that they bind to a set of glycan motifs that dictates human sensitivity or resistance to human infection and some selection by oysters. Furthermore, the binding of NoV to some enteric bacteria may impact virus distribution and persistence in coastal environment.

Recent research has provided novel tools such as the major breakthrough with the development of cell culture systems that allow measurement of NoV infectivity. The other important breakthrough are the novel catch-all virus detection assays, building from the Next Generation Sequencing (NGS) genomics revolution, that have recently become available and allow profiling of all viral matter in food and environmental samples, thus providing a rich source of data that can contribute to source tracking and monitoring of (sources of) contamination for any viruses (traceability method).

These new methods will help to better characterize the contaminations with the aim to find new solutions to decrease the risk for consumer.

#### **Metagenomic Applied to Study Norovirus**

MARION KOOPMANS: Erasmus University Medical Center, Rotterdam, The Netherlands

Genome sequencing has been used extensively in research in the past 25 years to understand how viruses spread, to assess the contribution of food in their transmission, and to characterise sources of outbreaks. With the rapid development of lower cost sequencing platforms, the implementation of these technologies for real-time surveillance has been stated as a priority by national and supranational public health organisations. Challenges to address are the lack of standardisation in the field, the fast evolution of technologies, and the need for increasingly complex bioinformatic analyses, with its own issues regarding standardisation. There is an increasing push for real-time open sharing of newly generated pathogen genome sequencing data on the one hand, but the need for some level of protection to safeguard against false positives which could erroneously point at a food source and recalls. Despite these hurdles, it is clear that the technological revolution brings interesting opportunities to the field, that - when properly implemented- will help to improve our understanding of foodborne pathogen transmission. This presentation will review lessons learned on the use of pathogen genomics in the norovirus field, and discuss recent developments based on the COMPARE project.

#### Glycans Interaction of Norovirus and Other Pathogens Inside and Outside the Host

#### JACQUES LE PENDU, Inserm-Université de Nantes, Nantes, France

Noroviruses (NoVs), and type A rotaviruses (RVAs) represent the most common cause of gastroenteritis in humans. Despite their complete lack of phylogenetic relationship, strains of these two viruses present similar recognition properties for glycans. They attach to terminal glycosylation motifs called histo-blood group antigens (HBGAs). These are mainly expressed in the gut epithelium and present genetic polymorphisms between humans. Either volunteers' studies and/or analyses of outbreaks demonstrated that for both NoVs and RVAs, the HBGA polymorphism restricts infection to individuals who present HBGA motifs matching the glycan motifs recognized by a given virus strain.

Since the HBGA motifs recognized by various strains are expressed at variable frequencies among humans, strains that bind to HBGAs expressed in large fractions of the populations can circulate broadly and become epidemiologically dominant, whilst strains that attach to HBGAs motifs less frequently encountered cause sporadic outbreaks only. An additional impact of the HBGAs polymorphism is also seen across human populations since allelic frequencies of the glycosyltransferases genes that govern HBGA synthesis vary considerably across human populations. As a result, the circulation of several virus strains is restricted to certain geographical areas, as particularly well illustrated in the case of RVAs. HBGA motifs are also expressed on the cell wall of edible plants such as lettuce or in the digestive tract of shellfish like oyster. Specific virus attachment to these HBGA-like motifs appears to facilitate their bioaccumulation and contribute to the risk of transmission via contaminated food items.

## Oysters and Norovirus: How New Technologies Will Help to Improve Safety?

**SOIZICK F. LE GUYADER,** *Ifremer, Laboratoire de Microbiologie, Nantes, France* 

Oysters are known as vector for human pathogens and despite regulation based on enteric bacteria they are still implicated in viral outbreaks. In Europe, norovirus in BMS are still among the most frequently recognised causes of foodborne illness among all virus/food commodity combinations. Significative advances have been made with the publication of an ISO method that will help to quantify norovirus concentrations and thus to prevent marketing contaminated oysters. However, the major question is how the detection of viral genome translates to human health risk, and how we can identify rapidly to limit the spread of other viruses.

The diversity and the low copy numbers of the viral pathogens found in oyster complicate identification, containment and mitigation of foodborne outbreaks. Pathogen sequencing is used for confirmation of virus presence and for tracking and tracing, but without taking the source of contamination into account, and enteric bacteria that may be present. The development of the novel catch-all virus detection assays, building from the Next Generation Sequencing (NGS) genomics recently become available, allow profiling of all viral matter in food samples. The development of technology such as NGS will provide more detailed information on the full range of strains present in samples. Obtaining more accurate information on strain diversity and quantification will be valuable for molecular epidemiology studies and management.

# S21 Beef Decontamination Treatments in Slaughter Plants: Do They Improve Product Safety?

Shiga toxin-producing Escherichia coli (STEC), particularly the serotype O157, are significant human pathogens, with cattle identified as an important source. To reduce beef contamination with STEC O157, many meat plants in North America have implemented HACCP based systems. These often incorporate decontaminating treatments, including carcass pasteurization with hot water or steam, and washing carcasses with organic acids. A significant body of research has been produced investigating novel decontamination technologies and assessing the efficacy of approved decontaminating treatments in commercial meat packing plants. However, some researchers have raised concerns that the use of decontaminating treatments in meat plants may increase the risk to consumers, by selecting for pathogen strains that are resistant to exiting hurdle technologies. Much of the information on resistance of E. coli has been derived from experiments in which a small number of bacterial strains were repeatedly exposed to sub-lethal concentrations of biocides. It may not be appropriate to extrapolate these findings into the setting of a meat processing plant.

The objective of this session is provide a forum to put into perspective the use of decontamination treatments in beef processing for the reduction of STEC. Recent findings to be discussed include whether cattle are the only source of *E. coli* O157 (STEC) on beef, whether there is evidence that the adoption of decontamination treatments by beef processors has impacted rates of STEC exposure and illness, and whether the use of decontamination treatments is associated with increased levels of resistance in *E. coli* to physico-chemical stress.

#### Overview of Decontaminating Treatments Currently Used in Commercial Beef Plants GARY ACUFF, Texas A&M University, College Station. TX. USA

To reduce the presence of enteric pathogens on beef, all beef processing facilities in North America have implemented HACCP-based systems and most have implemented multiple intervention steps designed to decontaminate carcasses, cuts and trim. Treatments often include carcass surface treatments with hot water or steam and spraying of carcasses, cuts and trim with various forms of organic acids or antimicrobials. Significant published research has investigated current decontamination systems, as well as novel decontamination technologies, and validation studies designed to assess the efficacy of decontamination treatments have been conducted in laboratories and within commercial meat packing plants. This presentation will review the current use of decontamination treatments in beef processing in North America for the reduction of enteric pathogens and will discuss multiple hurdle approaches to pathogen control.

#### **Biofilm Formation, Pathogen Prevalence, and Meat Contamination**

**RONG WANG**, U.S. Department of Agriculture-ARS, Clay Center, NE, USA

Biofilm formation by foodborne pathogens poses a serious threat to meat safety because biofilm cells are more tolerant to sanitization. In addition, the meat processing plants may harbor a wide variety of environmental microorganisms. A large portion of such mixed microbial community could persist in the plants as multispecies biofilms, therefore, mixed biofilm formation by environmental microorganisms with integrated foodborne pathogens could potentially provide an ecological niche for these pathogens to better colonize and obtain a higher survival capability against routine sanitization/cleaning procedures, as a result, increase pathogen prevalence and the risk of meat contamination. Our research revealed a potential role of biofilm formation and sanitizer tolerance in "High Event Period" beef trim contamination by E. coli O157:H7 and Salmonella enterica, the two major pathogens of concern in the meat industry. Furthermore, the potential contribution of mixed biofilm formation with environmental microflora in meat plants to pathogen sanitizer tolerance was investigated, and results showed that the pathogens obtained significantly enhanced tolerance by forming mixed biofilms with certain environmental microorganisms. The mechanisms underlying such protective effect appeared to be biofilm structure - and/or species - related, as certain specific bacterial species present in the environment might provide protections to the pathogens in mixed biofilms, leading to a high pathogen prevalence. These studies that focus on environmental bacterial community, pathogen integration, and the resultant sanitizer tolerance may help identify contamination sources and develop novel strategies to improve meat safety.

#### Resistance of *E. coli* from Beef to Decontaminating Treatments/Biocides Commonly Used in Meat Plants

XIANQIN YANG, Agriculture and Agri-Food Canada, Lacombe, Alberta, Canada

Shiga toxin-producing *Escherichia coli* (STEC), particularly STEC O157, are significant human pathogens, with cattle being identified as an important source. The infectious dose of STEC O157 is believed to be extremely low, such that exposure to a single cell may still represent a significant risk for potential infection. To reduce the contamination of beef with STEC O157, many beef processing plants in North America have employed various antimicrobial interventions, particularly during the carcass dressing process. Consequently, the level of *E. coli* on beef has been greatly reduced. In addition to decontaminating treatments for meat, biocides are also regularly applied to meat processing equipment as cleaning or sanitizing agents. Some concerns have been raised that these treatments may lead to increased level of resistance in *E. coli* and/or its persistence in meat processing environment. We investigated the response of E. coli originating from beef processing plants to commonly used decontaminating treatments and biocides for sanitation using a population based approach. No statistically significant difference in reductions by heat, acid or alkaline treatment was observed between E. coli populations originating from a plant where multiple antimicrobial interventions are routinely used and from a plant where no antimicrobial interventions are used. When two populations of E. coli, one persisting and the other transient, were assessed for their response to two sanitizers and one cleaning agent commonly used at food processing plants, the minimal inhibitory concentration and the minimal bactericidal concentration for the persisting group were not significantly higher than those for the transient group. Thus, the application of these treatments at beef processing plants does not seem to lead to elevated resistance in E. coli.

# S22 How Has Metagenomics Been Useful to Food Safety Research and What Does Its Application to Public Health Hold?

Food science finds itself at a new and exciting interface of microbiology, nutrition, and immunology. The full understanding of the microbial load in our food and the impact of the food matrix itself is of vital importance to public health. The promise of culture-independent next generation sequencing (NGS) technologies - or metagenomics - have fueled a renaissance in our understanding of foods and the impact on our own human microbiome. Both targeted and target-independent metagenomic data has been used to describe microbial and viral ecologies from soils to water to foods, helping us in our understanding of the foods we consume and the complex microbial ecologies along the farm to fork continuum – important both for food safety from pathogens and for issues of nutrition, allergy, and immunology. Metagenomic approaches in agro-ecologies have helped to identify critical control points and important drivers of phyllosphere microbiota. Food microbiome data will contribute to improved understandings of important ecologies for questions surrounding pathogenicity, food quality (spoilage), and nutrition. These data are advancing recommendations for data based Good Agricultural Practices and FSMA regulations The convenience and affordability of NGS technologies, improved bioinformatic pipelines, and converging reference databases will further rapidly increase the uptake of this technological application. Important questions remain to ensure that we harness the full potential of this technology. In this symposium, we propose to explore how metagenomics will help us build better risk assessment tools and consequently, better and more effective food safety management systems.

Speakers will cover the whole breadth of these issues; from rapidly and unambiguously identifying contaminants and pathogens such as bacteria and viruses and to understanding the community behavior and ecology of spoilage microbes or pathogens. This symposium will also examine and discuss the regulatory impact of the application of these technologies.

#### Metagenomic Sequencing for Surveillance of Food and Waterborne Viral Diseases

MARION KOOPMANS: Erasmus University Medical Center, Rotterdam, The Netherlands No abstract provided.

#### How FDA is Using Metagenomics for Food Safety and Microbiological Methods Development

**PADMINI RAMACHANDRAN**: Center for Food Safety and Applied Nutrition, U.S. Food and Drug Administration, College Park, MD, USA

Efficient recovery of pathogens from implicated foods is first and foremost in public health endeavors to understand source and route of contamination events via genomic and

epidemiological approaches. Relationships between endemic microbiota of foods in traditional enrichments are complex and likely vary by source microbiome. Using shotgun sequencing to characterize these dynamics contributes to an optimization of pathogen recovery methods.

The microbiology and chemistry associated with foods has a direct impact on human health. Understanding how pathogens become associated with surfaces of foods has clear implications for biological food safety and understanding levels of pesticides and heavy metals that persist in phytobiomes has tremendous significance to chemical and nutritional food safety. Significant data gaps remain in both areas. In certain cases, it can still take weeks to identify the source of foodborne illness outbreaks associated with foods. One data gap in our understanding of the microbiology associated with the surfaces of foods surrounds the origin of the microbes that persist there. Using shotgun metagenomics to track, trace and identify the source and efforts put forth by U.S. FDA will be discussed.

#### Utilizing Culture-independent Genomics and 'Big Data' Computation in the Food Industry

BALKUMAR MARTHI, DaQshConsulting, Visakhapatnam, India

Genomics and omic technologies offer the potential to develop deep insights into microbial community behaviour and to also follow how microorganisms evolve over time. The rapidity and high throughput potential of these technologies allow us to develop massive data sets to help understand behaviour. The resultant "Big Data" and the subsequent capability of profiling microorganisms and their communities to hitherto unachievable detail provides us the power to develop very effective strategies for the control of microbial contaminants and pathogens in the Food Industry.

On the one hand, genomics and omic technologies foster the development of mechanism-based preservation systems, leading to better, more effective and safer combinatorial preservative systems. Based on natural material, this allows for more robust formulation and process design. On the other hand, validated rapid technologies (based on Whole Genome Sequencing, for example) would allow practitioners to detect and contain contaminants much faster and with significantly lesser impacts. In addition, using metagenomic techniques to understand microbial ecology in the food system may help in predicting behaviour of contaminants in the environment and provide the means for their control.

In this talk, I will elaborate on the use of "culture-independent" genomics and the resultant "Big Data" in the Food Industry and discuss the potential impacts going forward. I will also discuss industry examples where this approach has been incorporated into food safety systems, and, finally, will look at how the food safety regulatory and compliance scenario would/should develop to ensure that these cutting-edge technologies are utilised to their fullest.

# S23 Campylobacter, Health Impact, Performance Objectives and Effectiveness of Sampling Plans

Campylobacter is the zoonotic bacterium with the largest public heath impact in many countries. Since the organism does not grow in the food chain, main points of control are in primary production, during slaughter and in the food preparation area. Absolute control is not possible at these stages, therefore balanced and targeted control measures should be implemented along the chain. The Codex/ICMSF approach of setting a food safety objective and linking it to performance objectives, process criteria, and microbiological criteria along the chain is instrumental in reaching a public health objective in a structured and flexible manner. Until recently, microbial criteria in chicken meat were not used, but since 2018 a process hygiene criterion has become mandatory in the European Union (EU). Notably, this is a dynamic criterion of which the stringency will increase in the next 7 years (m = 1000 CFU/g, n = 50, c = 20 from 1.1.2018and with c = 15 from 1.1.2020 and c = 10 from 1.1.2025). In this symposium the health impact of *Campylobacter* and its epidemiology will be described, potential control along the food chain by making use of performance criteria will be exemplified and the effectiveness of the current and future sampling plans in the EU will be discussed. The symposium is of relevance for governments, academia as well as for food industry.

### Health Impact of *Campylobacter*: The Main Zoonotic Pathogen in Many Countries

**JEFFREY FARBER**, University of Guelph, CRIFS, Guelph, ON, Canada

Campylobacteriosis is the leading or close to the leading cause of bacterial foodborne illness in many countries around the world. Although Campylobacter jejuni/coli are most often involved, other species such as C. lari and C. upsaliensis have also been isolated from patients with diarrhoeal disease, but are reported much less frequently. In this talk, a number of areas of importance regarding the public health implications of this pathogen in foods will be covered. Awareness of the public health implications of Campylobacter infections have evolved for over more than a century. Although it is well recognized that most cases of human campylobacteriosis are sporadic, outbreaks do occur and have different epidemiological characteristics from sporadic infections. A discussion of the global outbreaks of campylobacteriosis and their causative foods will be undertaken. This will include what new food vehicles have been involved in outbreaks, along with future trends in the epidemiology of campylobacteriosis. An examination will also be done of the global incidence of this pathogen. Infants tend to have the highest age-specific disease rates and there is also a notable high disease rate among young adults. Although Campy lobacter infections are generally mild, disease can be fatal among very young children (especially less than 2 years old), the elderly and immunosuppressed individuals. The factors associated with sequelae of Campylobacter spp. will be discussed, as well as the cost-of-illness and disease burden of the organism. In addition, an overview will be given of some of the potential risk factors for infection such as global warming, community socioeconomic factors and animal feeding operations. The issue of whether antimicrobial resistance can or has played a role in the epidemiology of campylobacteriosis, will also be explored.

# Establishing Performance Objectives throughout the Chicken Production Chain to Achieve Control of Campy-lobacter

WAYNE ANDERSON, Food Safety Authority of Ireland, Dublin, Ireland

The Codex Alimentarius Commission recognises several risk management metrics applicable to microbiological controls including food safety objectives (FSO), performance objectives (PO), performance criterion (PC) and microbiological criterion (MC) (CAC, 2007). The ability to set FSOs in a way that links directly to improvements in public health requires the establishment or inference of an Appropriate Level of Protection (ALOP). Hence, FSOs should be set by Government, but these can be operationalised through PO<sub>s</sub> set by authorities and/or industry allowing for flexible control measures to meet these targets (ICMSF, 2018).

Campylobacter causes the greatest burden of foodborne zoonotic disease in Ireland and the EU. Chicken meat has been considered a significant contributor to this disease burden (EFSA, 2010). The industrial production of raw chicken meat from start of slaughter to end of retail is relatively linear and lends itself to the establishment of PO<sub>s</sub> at various steps in the production chain. Therefore, it is possible to illustrate how the use of risk management metrics might be applied to control Campylobacter on raw chicken meat and how different control approaches might achieve these PO<sub>s</sub>. For illustration purposes, an ALOP can be inferred from U.S. campylobacteriosis statistics in humans in combination with attribution data from chicken meat. A public health goal may be set to reduce this disease burden and a related FSO calculated using an appropriate dose response model. To meet the FSO, a PO, can be calculated at the end of retail by estimating the reduction in Campylobacter achieved during the preparation and cooking of chicken meat. Similarly, a PO can be established at the end of slaughter to meet the PO, depending on the retail storage conditions of the meat (frozen or chilled) which follow. Finally, a PO, can be estimated after catching, transport and lairage of live chickens, the magnitude of which is affected by the subsequent slaughter process applied to the birds and the retail storage process applied to the meat. MCs can be established to verify that any of the POs are being met at a particular point in the chain. Hence, a flexible and verifiable control approach to Campylobacter in chicken meat chain is entirely possible. References

CAC. (2007). Principles and guidelines for the conduct of microbiological risk management (MRM) CAC/GL 63-2007.

ICMSF. (2018) Evaluation risks and establishing food safety objectives and performance objectives. In Microorganisms in Foods 7, microbiological testing in food safety management 2<sup>nd</sup> Ed. Springer. ISBN 978-3-3-319-68458-1.

EFSA. (2010) Scientific opinion of the quantification of the risk posed by broiler meat to human campylobacteriosis in the EU. EFSA Journal 8(1):1437.

#### Effectiveness of a (More and More Stringent) Sampling Plan for Campylobacter MARCEL ZWIETERING, Wageningen University,

MARCEL ZWIETERING, Wageningen University, Wageningen, The Netherlands

Until recently, there were no microbial criteria for *Campylobacter* in chicken meat, but since 2018 a process hygiene criterion has become mandatory in the European Union (EU) (EC, 2017). Notably, this is a dynamic criterion of which the stringency will increase in the next 7 years (m = 1000 CFU/g, n = 50, c = 20 from 1.1.2018 and with c = 15 from 1.1.2020 and c = 10 from 1.1.2025). The performance of the current and future sampling plans in the EU will be discussed also in relation to baseline data of various countries, based on the ICMSF spreadsheet (Legan et al. (2001) and Van Schothorst et al. (2009).

#### References

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## S24 Insects in Poultry Feed: Regulatory Framework, Poultry Gut Microbiota and Consumer Acceptability

This symposium tackles various aspects connected to the introduction of insects and insect derivatives in poultry feed. Specifically: the regulatory framework, the impact of those ingredients in the physiology and regulation of poultry gut microbiota/microbiome and, finally, the consumer acceptability.

The results presented have been generated by the Innopoultry project, which aims at improving/resolving critical issues related to the poultry food chain of the 21st century. More specifically, poultry meat has been identified as one of the primary sources of foodborne pathogens for humans. Moreover, due to the past improper use of antibiotics as a growth promoter, poultry meat is still considered among the primary source and vehicles of antibiotic resistance. The Innopoultry strategy focused on primary production: considering the feed as an essential tool to exploit, it proposed to adopt alternative approaches in poultry feeding, able to control and reduce food safety risks. The inclusion of alternative ingredients (insect proteins and oils) in the diet and the adoption of innovative strategies in animal feeding (e.g., addition of probiotics) it is foreseen to positively impact the poultry gut microbiota/microbiome and promote the reduction of foodborne pathogens and antibiotic-resistant bacteria. The project address also the consumer acceptability of such innovative ingredients and the regulatory framework for their use.

The Innopoultry project is funded by the EIT Food, the Europe's leading food innovation initiative, with the aim to create a sustainable and future-proof food sector. The initiative is made up of a consortium of key industry players, start-ups, research centers and universities from across Europe. EIT Food final objective is to develop new knowledge, technology-based products, and services that will deploy a healthier lifestyle for all European citizens.

#### The Use of Insects and Insects Derivative for Animal Feeding: An Attempt of a Guideline

**MICHELE BLASI,** Associazione Italiana Allevatori, Rome, Italy

The use of insect to feed farm animal in general and poultry or fish principally is a promising alternative to the conventional source of protein both for their nutritional properties and for the positive impact on the sustainability of the whole production chain. Insect are, in fact, part of the natural diet of chicken and fish.

Arthropod exoskeleton contains chitin, a large, structural polysaccharide made from chains of modified glucose which acts as a probiotic and stimulator of the immune system. Also insects are valuable source of minerals as iron and zinc, omega fatty acid and aminoacids.

Insects are efficient feed-converter and can be raised on many organic matters including waste and require less water and less space if compared to other protein sources. Considering both that the environmental sustainability of the animal production is a hot topics and the potential benefit of raising insect also as a contribute to the circular economy in animal production, as partner of the Innopoultry project founded within the EIT food Connect, we have developed the first guideline for the responsible use of insect source protein in feeding poultry.

The guideline is tailored to farmers and outline the major advantages in introducing insect as a source of high quality protein as well as insect oil as source of sutured fatty acid.

The guideline also provides also information on the best practice for the insect fodder conservation and some basic principles on their correct use in the diet formulation.

Besides the already rich literature on the best methods to rear insect, our guideline offer the very first attempt of a supporting tools for their use in animal production chain. We hope that this first edition of the guideline will be followed by several more complete versions, proving the interest of farmers for this new opportunity.

# Impact of Innovative Feeds in the Microbiota and Microbiome of Poultry

#### **ILARIO FERROCINO,** University of Torino-DISAFA, Grugliasco, Italy

The inclusion of alternative ingredients in poultry feed is foreseen to impact poultry gut microbiota. We evaluated the effects of dietary insect oil inclusion (i) and of probiotic dietary inclusion (ii) on performance, gut microbiota and histomorphology of broiler chickens. (i) A basal diet containing only soybean oil as added fat was formulated and served as control group (C). It was tested against three experimental diets where the soybean oil was partially substituted by insect oil (IO) or one of two types of modified insect oil (lauric acid enriched oil – MIO). (ii) A multi-strain probiotic formulation (*Lactobacillus plantarum* and *L. pentosus*) was daily supplemented to broilers.

Dietary insect oil or probiotic inclusion did not significantly influence bird performance, thus suggesting that these alternative ingredients allow broilers to maintain the required high growth standards. The identification of unaffected histopathological alterations is also indicative that insect oil or probiotic do not impair the overall health status of the birds.

A shift in the gut microbiota composition was observed as a function of the diets. In particular, the use of MIO reduced the presence of *Clostridium* and *Corynebacterium*, which can frequently cause infections in poultry. The multistrain probiotic inclusion in broilers clearly increased the abundance of several taxa (*Blautia, Faecalibacterium* and *Lachnospiraceae*) recognized as short chain fatty acids (SC-FAs)-producing genera. The positive modulation of microbiota observed in both insect oils- and probiotic-fed broilers is particularly relevant, since the use of these alternative ingredients could promote a healthy status of the broiler's gut.

#### Consumer Acceptability of Poultry Meat Produced with Insects

**FLAVIO PELLEGRINUZZI**, Euroconsumers Cooperative Partner Group of Independent European Consumer Organizations, Luxermburg, Luxembourg

**Purpose:** Investigate the consumers' willingness to eat poultry meat from chicken raised introducing insects' derivatives in their diet.

**Methods:** a general population representative survey has been done in Belgium, Italy, Portugal and Spain within persons 18 to 74 years old. The fieldwork was conducted in May 2018, through both CATI and CAWI interviews. A total number of 3.979 responses were considered for the analysis (Belgium 1.024, Italy 1.018, Portugal 1.000, Spain 937). To respect the distribution of the general population original results have been weighted for each country by educational level, gender, region where the respondent lives and age.

**Main results:** Relevant differences have been observed among countries, the percentages of people willing to eat meat from chickens fed (also) with insects' derivatives varies from 70% (Belgium) to 31% (Italy). In all the countries women, people above 55 years of age and people with primary education are less willing to eat this kind of meat. A psychological barrier to eat insects (even if indirectly) is the main restraint. Nevertheless, in each of the four countries the majority of consumers would be willing to eat this kind of poultry meat if it was proven that including insects in the chickens' diet improves the animal health.

**Significance:** Our study shows consumers' resistance to the introduction of insects' derivatives in the poultry diet. On the other hand, it highlights the importance to prove and communicate the benefits this diet can induce in terms of sustainability and health.

## **Closing Session**

## Introducing the One Health European Joint Programme (EJP) Initiative and Its ORION Project

**MATTHIAS FILTER,** German Federal Institute for Risk Assessment, Berlin, Germany

*Introduction:* The One Health European Joint Programme (OHEJP) is a H2020 funded project carried by 39 food, veterinary and public health institutes from 19 EU member states and the Med-Vet-Net-Association. The project started in January 2018, runs for 5 years and has an overall budget of 90 Million Euro including 50% co-funds.

**Purpose:** The main objective of this programme is to share knowledge to enhance the prevention, detection and control of zoonoses and antimicrobial resistance. For this OHEJP initiates transdisciplinary cooperation and development activities between medical, veterinary and food institutes based on a strategic research agenda that is built on programmes supported by national authorities and by ECDC and EFSA.

*Methods:* The OHEJP supports implementation of the 'One Health' concept on national and international level through dedicated joint research projects (JRPs) and joint integrative projects (JIPs) addressing current needs in the area of cross-disciplinary harmonization of approaches, methodologies, formats and procedures for the assessment and management of foodborne hazards, emerging threats and AMR across Europe. It also implements Doctoral Programmes, Annual Scientific Meetings, Summer Schools and Continuous Professional Development Modules.

**Results:** The work carried out so far in the 11 JRP and 2 JIPs proof the need and relevance of the OHEJP objecttives. On the example of the EJP ORION project it will be illustrated how each of the individual projects executed under the OHEJP contribute with specific solutions serving the overall EJP objectives.

**Significance:** The OHEJP will contribute to the development of a sustainable European One Health Framework taking into account the initiatives taken by stakeholders like WHO, EFSA, ECDC, JPI AMR, COMPARE and EFFORT. Moreover, through the existing links with the Programme Owners (national or regional authorities and policy makers) national interests are taken account of in OHEJP.

#### How New Tools and Technologies Bring New Questions and Help to Answer Old Ones

#### TIMOTHY JACKSON, Driscoll's, Watsonville, CA, USA

Advances in analytical methodology bring greater precision to our understanding of food safety risk, help to answer vexing questions, and bring to light new issues that need to be addressed by food safety managers. Collaborative initiatives and multi-institutional projects bring context to food safety issues and help to identify new solutions to existing programs. New approaches to data management can help food safety managers design new or more effective solutions to the challenges we face. Food Safety managers need to be open to new approaches, track the experiences of early adopters, and be open to new approaches and ideas to protect products and consumers.

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Abstract Submission Deadline March 6<sup>th</sup>, 2020

Abstract Notification April 10th, 2020

Early Bird Registration Deadline May 10<sup>th</sup>, 2020





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# **ROUNDTABLE ABSTRACTS**

24–26 April 2019 – Nantes, France

# **ROUNDTABLE ABSTRACTS**

# RT1 Environmental Monitoring – Friend or Foe?

#### Panelists:

ROY BETTS: CAMPDEN BRI, Chipping Campden, United Kingdom

FRANCOIS BOURDICHON: International Dairy Federation - Standing Committee on Microbiological Hygiene, Brussels, Belgium

JOHN HOLAH, UK: IE EHEDG & Holchem Laboratories Ltd., Bury, United Kingdom

DIRK NIKOLEISKI, Commercial Food Sanitation, Munich, Germany

ANETT WINKLER, Cargill, Munich, Germany

Over the last years Process Environment Monitoring (PEM) gained a lot of attention from both the industry and regulators. PEM offers a proactive approach to identify possible microbiological contamination of the food product, where finished product testing is too reactive and not fit for purpose to detect low level of contamination.

Well thought out and valid PEM systems are of great value and can be a great friend to us, while poorly designed systems where little thought is given to design, handling results and corrective actions, are of little value and are our foe. Monitoring in areas, where neither adequate zoning, GMP nor cleaning practices are implemented, would not provide meaningful information – apart from confirming the before-mentioned measures being vital for a clean process environment.

Many documents (e.g., published by Codex Alimentarius, FDA, GMA, BRC, ISO 22000) underline its importance, provide general guidance and even require PEM as part of Food Safety management. However, PEM will only form an integral part of food safety systems when it is set up adequately – assuring that the environment won't contribute to contamination of products. Even more, an adequate monitoring of the environment can provide early warnings of potential product contamination.

This roundtable will focus on the practical aspects of PEM to ask what we can learn from the past – incidents with both good and bad examples of PEM practice will be considered.

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 based on risk? How useful are indicators? How to interpret results? Are there adequate corrective actions defined? How can these be verified?

#### RT2 The Use of Chemicals in Food Hygiene and Linkage to Microbial Resistance

#### Panelists:

JOHN DONAGHY: Nestec Ltd., Vevey, Switzerland PHILIPPE GLASER: Institut Pasteur, Paris, France JEFFREY LEJEUNE: Food Safety and Quality Unit, AGFF, Food And Agriculture Organisation, Rome, Italy

PETER MCCLURE: Mondelēz, Bournville, United Kingdom

PIETRO STELLA: European Food Safety Authority (Efsa), Parma, Italy

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 good reasons: to protect animal health, support good food hygiene practice, and ultimately ensure 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 the FAO will talk about the tripartite global action plan agreed with WHO and OIE to protect public health against AMR, and the role of the food supply chain in this mission. You will also hear from regulators on the difficulty of their role as policy makers, and from food industry representatives on their experience about how the wrong decision on this critical topic can have serious consequences. Members of the recently-run GFSI Technical Working Group on Chemicals in Food Hygiene have agreed to come and share the outcome of their 18-month work programme on how to ensure consumer protection through the appropriate application of sanitizers, disinfectants and cleaning agents from farm to fork. Ultimately, we look at balancing risks and benefits of these agents' use whilst facilitating the global food trade.

## RT3 Foodborne Viruses: Detection, Risk Assessment, and Control Options in Food Processing

#### Panelists:

ELISSAVET GKOGKA, Arla Innovation Centre, Aarhus, Denmark

ALVIN LEE, Institute for Food Safety and Health, Illinois Institute of Technology, Bedford Park, IL, USA

FABIENNE LOISY-HAMON, BioMérieux, Grenoble, France

GLORIA SÁNCHEZ, Institute of Agrochemistry And Food Technology (Iata-Csic), Valencia, Spain

SOPHIE ZUBER, Nestlé Research Center, Lausanne

Foodborne viruses were recognized among the top rated food safety priorities and have become a greater concern to the food industry over the past few years. Although control measures for viruses throughout the food chain are required there are still gaps in knowledge and understanding of viral detection and control strategies for the food industry with respect to the effectiveness of these controls and how to properly validate their performance. Research effort needs to be undertaken to understand the ecology, behaviour and transmission of foodborne viruses from the farm and to the consumer.

This roundtable will discuss the current state of the science on epidemiology, public health burden and risk assessment for viruses in food processing environments. Current technologies developed for viral detection and control as well future perspectives on the application, along with suggestions on how the food industry could implement effective control strategies and management options for viruses in foods will be introduced and discussed.



# TECHNICAL ABSTRACTS

24–26 April 2019 – Nantes, France

# **TECHNICAL ABSTRACTS**

#### \* Student Award Competitor

# WEDNESDAY - 10.30 - 12.00

# T1 Technical Session 1 – Meat and Poultry, Antimicrobials and Microbial Food Spoilage

#### T1-01 RNA-based Surveillance of Meat Processing Environments Revealed Selective Pressures of Gaseous Ozone on Abattoir Microbiota

**CRISTIAN BOTTA**<sup>1</sup>, Ilario Ferrocino<sup>1</sup>, Maria Chiara Cavallero<sup>2</sup>, Simonetta Riva<sup>3</sup>, Luca Cocolin<sup>1</sup> and Kalliopi Rantsiou<sup>1</sup>

<sup>1</sup>University of Torino-DISAFA, Grugliasco, Italy, <sup>2</sup>M.I.A.C. S.c.p.A – Polo AGRIFOOD, Dronero, Italy, <sup>3</sup>Veterinary Food Safety, Salmour, Cuneo, Italy

Introduction: Microbial contamination of beef cuts mainly occurs during the deboning and cutting phases, where meat contact surfaces of the processing rooms can concurrently act as reservoirs and donors of psychrotrophic spoilage bacteria. Cleaning and sanitization of these environments is a challenging task for meat industries that are constantly seeking new, eco-friendly and economical disinfection processes, such as gaseous ozone. This oxidative gas has proven to be an effective disinfectant for several bacterial species. **Purpose:** However, its effect on abattoir resident microbiota still needs to be investigated *in situ*.

**Methods:** Herein, the RNA-based amplicon target sequencing of the 16S gene was applied, in parallel with targeted microbial counts, to monitor the effects of gaseous ozone on the potentially active microbiota of meat processing environments. A total of 278 environmental swabs were collected from two abattoirs before cleaning (BC), after cleaning/sanitization (ACS) and after three overnight ozone treatments (AOT) at 4, 20 and 40 parts per million (ppm).

**Results:** The results showed a shift in the microbiota composition in parallel with the reduction of total microbial counts after the cleaning step, with a significant increase of  $\alpha$ -diversity and a decrease of Firmicutes, which constituted the transient, animal-derived microbiota of the abattoirs. Afterward, the four ppm ozonisation did not significantly diminish the ACS counts, whereas between the two effective treatments, the ozonisation at 40 ppm mainly affected *Proteobacteria* and determined a significant variation of  $\beta$ -diversity between the ACS and AOT communities. The amplicon sequencing data highlighted inactivation of *Pseudomonaceae* and *Brochothrix* by the highest ozone concentrations, while *Staphylococcaceae* showed a remarkable survival capability.

Significance: Our outcomes suggest a synergistic activity between gaseous ozone and routine cleaning procedures since the highconcentration ozonisation acted preferentially on abattoir microbiota originating from water/soil and some of the major meat spoilage organisms.

#### T1-02 Litter Treatment as a Control Strategy Against Campylobacter in Broilers: Impact on Caecal Counts and Microbiota Composition

**AMANDINE THEPAULT**<sup>1</sup>, Xavier Roulleau<sup>2</sup>, Typhaine Poezevara<sup>1</sup>, Pauline Loiseau<sup>3</sup>, Ségolène Quesne<sup>1</sup>, Florent Souchaud<sup>1</sup>, Marianne Chemaly<sup>1</sup> and Muriel Guyard-Nicodème<sup>1</sup>

<sup>1</sup>ANSES, Ploufragan, France, <sup>2</sup>Dietaxion, Le Loroux Bottereau, France, <sup>3</sup>Terrena, Ancenis, France

**Introduction:** Campylobacteriosis is the main bacterial foodborne zoonosis in Europe. Source attribution studies highlighted that poultry, highly contaminated by *Campylobacter*, constitute the major source of human infection. Since 2018, a European process hygiene criterion for *Campylobacter* in broiler carcasses has been implemented, leading to stepped up efforts to reduce *Campylobacter* at the farm.

**Purpose:** This work aims to test a commercial product (COBIOTEX 410 Absorbant), a complex of positive bacteria and drying compounds, to reduce *Campylobacter* in broilers and explore its impact on their caecal microbiota.

Methods: Ross PM3 broilers were contaminated with *Campylobacter jejuni* at day 18. In one group, no treatment was used while in the second group the litter treatment was applied weekly during the whole rearing period (day one to day 35). *Campylobacter* loads were assessed in caecal contents following the decimal dilution method at day 22 and day 35. The statistical analysis was performed using the Mann-Whitney U test. Sequencing of the V3-V4 region of the 16S RNA gene was performed using Illumina MiSeq technology. Resulting sequences were analyzed using the FROGS pipeline to comparatively explore the bacterial communities associated with each experimental condition.

**Results:** The comparison between treated and untreated groups did not reveal *Campylobacter* reduction in broilers at day 22 and day 35 (*P*>0.05). The investigation of caecal microbiota highlighted the predominance of *Firmicutes* and *Bacteroidetes* in both groups followed by *Proteobacteria*. The bacterial richness did not differ between the two groups with an average of 202±58 and 222±48 operational taxonomic units, respectively. However, the richness significantly increased with age (*P*=0.000). Treatment, age and their combination have an effect on community structure, explaining around 60% of the beta diversity variance.

**Significance:** This trial constitutes a basis for broader investigations of the cause and effect relationships between *Campylobacter* colonization, litter treatment and microbiota composition.

# T1-03\* Mathematical Modelling of *Listeria mono-cytogenes* Survival during Low-temperature (*sous-vide*) Cooking of Meat Products

**MARTA CLEMENTE-CARAZO**<sup>1</sup>, Alberto Garre<sup>1</sup>, Pablo S. Fernandez<sup>1</sup>, Jose Lucas Peñalver-Soto<sup>1</sup>, Paula M. Periago<sup>1</sup>, Arantxa Aznar<sup>1</sup>, Arturo Esnoz<sup>1</sup>, Jose A. Egea<sup>2</sup> and Alfredo Palop<sup>1</sup>

<sup>1</sup>Universidad Politecnica de Cartagena, Cartagena, Spain, <sup>2</sup>CEBAS-CSIC, Murcia, Spain

Introduction: Sous vide cooking is a novel way of preparing meat products that has gained popularity during the last few years. It is based on the application of low temperatures (much lower than the boiling temperature of water). The reduction of the cooking temperature can enable the survival of bacterial pathogens. Therefore, it is a potential risk for food safety.

**Purpose:** The development of an experimental protocol to replicate low-temperature cooking of meat products and, based on it, the development of predictive models applicable to food safety studies. **Methods:** An experimental technique to simulate the inactivation of *Listeria monocytogenes* during *sous vide* cooking of meat products was developed. Briefly, 10 g samples of food matrix (minced chicken, pork or beef meat) were inoculated with *Listeria monocytogenes* and vacuum sealed in a bag. After homogenising, they were introduced into a water bath simulating isothermal cooking at different low temperatures (53.6, 56, 59.5, and 62°C). Experimental data were analysed using the Geeraerd inactivation model.

**Results:** Significant differences are observed in the model parameters obtained from each heating medium, evidencing the influence of the food matrix on the inactivation kinetics. Among the food matrices tested, the chicken meat was the most protective for *L. monocytogenes,* having the longest shoulder length and highest *D*-value.

**Significance:** Predictive models have been developed to describe the simulate the inactivation of *L. monocytogenes* in meat products during low-temperature cooking. This model can potentially be incorporated in quantitative microbial risk assessment studies to evaluate the risk associated with this novel type of product.

#### T1-04 Multidrug Resistance Spread Among Yersinia enterocolitica Isolates from Swine and Pork Production Obtained in Brazil

Bruna Torres Furtado Martins, Juliana Libero Grossi, Ricardo Seiti Yamatogi and **LUÍS AUGUSTO NERO** *Universidade Federal de Viçosa, Viçosa, Brazil* 

**Introduction:** Yersinia enterocolitica is the causative agent of yersiniosis, gastroenteritis highly associated with pork products. As swine are considered natural reservoirs of Y. enterocolitica, all stressing and selective factors during swine production can lead to effects on this pathogen. So, antibiotic therapies can promptly lead to resistance in Y. enterocolitica, a major worldwide concern. **Purpose:** This study aimed to evaluate the antibiotic resistance of Y. enterocolitica isolated from swine, environmental and human clinical samples from Brazil.

Methods: Y. enterocolitica isolates (n=75) were subjected to PCR targeting multidrug resistance (MDR) related genes (emrD, marC, yfhD). Isolates were subjected to disk diffusion testing to assess their resistance to 14 antibiotics (11 classes). Based on resistance and PFGE profiles (after Xbal macro-restriction), 25 isolates were selected and subjected to susceptibility assay based on minimal inhibitory concentrations (MIC) of eight antibiotics (seven classes). Results: The majority of isolates (71, 94.7%) presented simultaneously all MDR related genes, while two presented emrDmarC, one marC-yfhD, and one emrD. Based on disk diffusion assay, isolates were predominantly resistant to ampicillin (73, 97.3%) and sulfamethoxazole (60, 80.0%), followed by nalidixic acid (30, 45.3%), trimethoprim (24, 32.0%), chloramphenicol (24, 32.0%) and tetracycline (19, 25.3%); less than five percent of the isolates presented resistance other tested antibiotics. Based on MIC, resistance was recorded mainly to ampicillin (24, 96.0%), amoxicillin (22, 88.0%), and cefoxitin (20, 80.0%), followed by sulfa & trimethoprim (five, 20.0%) and tetracycline (four, 16.0%); less than 10% of the isolates were resistant to other tested antibiotics. Significance: Isolates obtained from pigs and the environment were remarkably more resistant than isolates from clinical samples. MDR is a common characteristic of Y. enterocolitica isolates from pigs and pork, highlighting its relevance as a public health hazard.

#### T1-05\* The Antibiotic Resistome of Farmed Rainbow Trout Filets Using Smartchip Real-time PCR

NICOLAS HELSENS<sup>1</sup>, Ségolène Calvez<sup>2</sup>, Agnes Bouju-Albert<sup>1</sup>, Albert Rossero<sup>1</sup>, Hervé Prévost<sup>1</sup> and Catherine Magras<sup>1</sup>

<sup>1</sup>SECALIM, INRA, Oniris, Université Bretagne Loire, Nantes, France, <sup>2</sup>BIOEPAR, INRA, Oniris, Nantes, France

Introduction: The role of food in the routes of transmission of resistant bacteria and antimicrobial resistance genes (ARG) is yet to be explored. There can be a risk of ARG presence on farmed fish filets because of their environmental exposures (animal farms, human activities, aqueous environment). Consequently, it is essential to determine the farmed fish antibiotic resistome. To analyse fresh fish filet resistomes, where bacterial load is known to be low, a highly efficient method like high-throughput qPCR arrays, able to detect and quantify hundreds of selected ARGs in a single run, can be useful. **Purpose:** The objective was to evaluate the significance of Smartchip RT-PCR technology to analyse the antibiotic resistome profile of rainbow trout filets.

Methods: The analyses were performed on both fresh and spoiled rainbow trout filets. The filets were inoculated with antimicrobialresistant bacterial strains (n=6) at various concentrations (10<sup>4</sup> to 10<sup>8</sup> CFU/g) to assess the capacity to detect a specific gene among the microbial communities of the filet. The ARG were detected and quantified using a 245 primer pair set. The set was chosen after bibliography analyses and In Silico verification. The amplification was realised thanks to the Smartchip Real-Time PCR technology (Takara). Results: The ARG detection threshold was determined to be 3.52 log CFU/g. Some ARG were detected at C, values around 25 on fresh filets (tetL, tetB, sul1, qacEA1). In spoiled filets, the quantification of some ARG was enhanced (Ct around 18 to 20). Other genes such as tetS, tetB, strB, mexE, mexF were only detectable in spoiled filets. Significance: The Smartchip real-time PCR technology allows detecting ARG at different presence levels in the bacterial communities of fish filets. It may be interesting to use this tool to investigate and describe the resistome patterns of rainbow trout filets.

#### T1-06\* Residential Surface Bacteria Mapping in a Coldsmoked Salmon Processing Environment

AURELIEN MAILLET<sup>1</sup>, Agnes Bouju<sup>1</sup>, Steven Roblin<sup>2</sup>, Pauline Vaissié<sup>2</sup>, Sebastien Leuillet<sup>2</sup>, Xavier Dousset<sup>1</sup>, Emmanuel Jaffrès<sup>1</sup>, Jerome Combrisson<sup>2</sup> and Herve Prévost<sup>1</sup> <sup>1</sup>UMR 1014 SECALIM, UBL, INRA, Oniris, Nantes, France, <sup>2</sup>Biofortis Mérieux NutriSciences, Nantes, France **Introduction:** Microbial communities play an essential role in food safety and quality. In the food processing environment, residential bacteria remain on surfaces after sanitization procedures and are a major source of product contamination and spoilage. Residential bacteria characterization should be an important concern. The relationship between environmental microbiota and product quality can be appreciated by 16S metabarcoding.

Purpose: The aim of this study is to evaluate the processing environment impact on cold-smoked salmon quality by the development of an innovative methodology to track the bacterial environmental source of contamination. A polyphasic approach based on culture-dependent and 16S metabarcoding was used.
Methods: Samples from surfaces of a cold-smoked salmon processing environment and from products during the process and storage all along the shelf life were collected. Bacteria were plate counted and identified by MALDI-TOF MS or full 16S rDNA sequencing. DNA was extracted directly from samples and analyzed by V3-V4 16S metabarcoding.

**Results:** The residential bacterial community was identified and characterized as homogeneously spread within the processing environment. A core microbiota between products and the environment was identified and was mainly composed of environmental spoilage bacteria: *Brochothrix thermosphacta, Carnobacterium maltaromaticum, Photobacterium phosphoreum, Serratia liquefaciens, Staphylococcus equorum, Psychrobacter* spp., and *Pseudomonas* spp. *B*-diversity and network analysis allowed us to highlight environmental bacterial source hotspots and to solve contamination routes. These results suggest that the environment can impact product quality and safety.

Significance: Microbial ecology knowledge in a complex ecosystem such as a processing environment could be useful to characterize microbial reservoirs, improve targeted hygiene procedures, and lead to a better product quality all along its shelf life.

# WEDNESDAY - 13.30 - 15.00

# T2 Technical Session 2 – Applied Laboratory Methods

#### T2-01 Salmonella Serotyping: Comparison of the Traditional Method to a Microarray-based Method and an In Silico Platform Using Whole Genome Sequencing Data

**BENJAMIN DIEP**<sup>1</sup>, Caroline Barretto<sup>1</sup>, Coralie Fournier<sup>1</sup>, Aneta Karczmarek<sup>2</sup>, Guido Voets<sup>2</sup>, Xiangyu Deng<sup>3</sup> and Adrianne Klijn<sup>1</sup>

<sup>1</sup>Nestlé Research, Lausanne, Switzerland, <sup>2</sup>Check-Points, Binnenhaven, The Netherlands, <sup>3</sup>University of Georgia, Center for Food Safety, Griffin, GA, USA

**Introduction:** While *Salmonella* molecular subtyping is used to investigate outbreaks and for source tracking, serotyping still remains the first step to characterize a *Salmonella* isolate. Determination of *Salmonella* serovars is traditionally performed with the Kauffmann-White-Le Minor (KWL) method, but several alternative solutions are available.

Purpose: Here, we compared a microarray-based method, the Check & Trace Salmonella kit (Check-Points B.V., The Netherlands) and the In Silico platform SeqSero (University of Georgia, US) to the KWL method.

**Methods:** The microarray-based method targets proprietary specific genetic markers. The In Silico platform targets the gene sequences encoding the individual O and H antigens. Strains (*n*=100) were provided by The Netherlands Institute for Public Health and the Environment.

**Results:** The microarray-based method was able to identify correctly 98% of the isolates. One strain was not assigned a serovar but was allocated a unique genovar code, and one strain did not match with the KWL result. The method was initially validated for the 100 most commonly encountered serovars, however the continuous expansion of the database would allow identifying more serovars. The In Silico platform presented 98%, 34% showed a multiple prediction including the expected serovar. One strain was not identified and one strain was discordant with KWL result.

Significance: Our study indicates that a microarray-based method offers a transition to an ease of use method with a short turnaround-time. An In Silico platform could be a long-term solution if whole genome sequencing (WGS) will be used routinely by all laboratories. However, in order to be cost efficient, it must be considered as a WGS "full package" solution, which includes pathogen characterization, pathogen source tracking and other research applications such as virulence and antibiotic resistance gene determination.

#### \* Student Award Competitor

#### T2-02 Optimized Methodology for Same-day Detection of *E. coli* O157 By Multiplex qPCR in Ground Meat and Leafy Greens

ALEJANDRO GARRIDO-MAESTU, Sarah Azinheiro, Joana Carvalho, Pablo Fuciños and Marta Prado International Iberian Nanotechnology Laboratory, Braga, Portugal

**Introduction:** *E. coli* O157 is the most commonly identified serotype among Shiga toxin-producing *E. coli*. Reference methodologies take several days, as they require selective enrichment, molecular detection of the bacteria, and plate confirmation. Alternative methods also need long enrichments to increase the bacterial concentration to detectable numbers, resulting in "next-day detection" methodologies. **Purpose**: The aim of the current study was to develop and evaluate a methodology based on real-time PCR, capable of detecting *E. coli* O157 in one working day.

Methods: Two different types of food matrices were selected for spiking experiments, ground meat and leafy greens. The inoculated samples were diluted in mTSB and incubated for three hours with constant agitation at 37°C. Upon completion, the supernatant was recovered, centrifuged to concentrate the microorganisms, and the pellet was treated with a protease mixture to degrade food tissue that might be present. This was followed by another centrifugation, and the pellets were washed with PBS. The clean pellets were subjected to DNA extraction with Chelex 100/proteinase K, and final thermal lysis (99°C for 10 min). The recovered DNA was analyzed by multiplex qPCR targeting the fbE gene, and an internal amplification control. The overall method could be completed within five hours. Results: The described methodology allowed detection below five CFU/25 g, regardless of the food matrix selected, in one working day. The relative sensitivity, specificity, accuracy, positive and negative predictive values were higher than 90%, and the Cohen's k gave a value of one, indicating that the results obtained were in "almost complete concordance" with the expected ones. The performance parameters fulfill the requirements of the NorVal regulation for the validation of alternative methods.

Significance: The described methodology will allow a higher throughput in testing laboratories, with a shorter turnaround time resulting in higher food safety.

#### T2-03\* Assessment of Bacterial Indicators on Poultry Carcasses by Culture Combined MALDI-TOF MS Identification and 16s rRNA Amplicon Sequencing

#### ZHONGJIA YU1 and Kurt Houf2

<sup>1</sup>Department of Veterinary Public Health and Food Safety, Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium, <sup>2</sup>Ghent University, Merelbeke, Belgium

Introduction: The examination of bacterial contamination on food products is still largely performed by standardized culture methods, while culture-independent methods have only been performed more recently. However, knowledge of the diversity of bacteria present, as well as the impact on the outcome of several analytical steps remains unclear.

**Purpose:** To evaluate the performance of ISO methods generally applied in the microbiological quality examination of poultry carcasses compared to a culture-independent approach, and assess the impact of incubation at 7°C and 30°C on total aerobic bacterial count and diversity.

**Methods:** Breast skin of 16 chicken carcasses was analyzed for general bacterial parameters: total aerobic and anaerobic bacteria, *E. coli*, lactic acid bacteria and presumptive *Pseudomonas* using normalized culture methods combined with MALDI-TOF MS identification, and 16S rRNA amplicon sequencing.

Results: No significant impact of incubation temperature on the total aerobic bacteria level was detected, limiting the usefulness of additional psychrophilic examination. With ISO culture methods, other bacteria phenotypically similar to Pseudomonas were identified on selective CFC plates. Escherichia coli and Staphylococcus spp. were commonly present on MRS plates along with lactic acid bacteria. Application of 16S rRNA amplicon sequencing revealed a higher bacterial diversity regardless of prior cultivation, but different DNA extraction kits applied have a significant impact on the bacterial population detected in non-cultured samples (P<0.05) Significance: The study demonstrated that determination of total aerobic bacteria based on ISO 4833 is suggested to determine both the bacterial contamination level and the diversity. ISO culture methods for Pseudomonas and lactic acid bacteria require further colony confirmation. When a culture-independent method is applied, the impact of the selected DNA extraction kit and the detection of nonviable bacteria should be taken into account to interpret the outcome.

#### T2-04 The Effect of Spices on the Growth and Detection of *Salmonella* and Shiga Toxinproducing *Escherichia coli* in a Primary Enrichment

**GREETJE CASTELIJN**, Lotte de Lange and Menno van der Voort

The Netherlands Food and Consumer Product Safety Authority (NVWA) Laboratory Feed, Food & Consumer product safety, Wageningen, The Netherlands

**Introduction:** The first step of most detection methods for foodborne pathogens is primary enrichment. The growth of foodborne pathogens in primary enrichment can be influenced by the food product that is present in the primary enrichment as well.

**Purpose:** The effect of 46 spices on the growth of *Salmonella* and Shiga toxin-producing *Escherichia coli* (STEC) in a primary enrichment was analyzed.

**Methods:** Salmonella and STEC were cultured in different primary enrichment media in the presence of different spices. After incubation of the primary enrichments for 18 h at 37°C, the growth of Salmonella and STEC was evaluated by different isolation media and real-time PCR.

Results: The results indicated that allspice, badiane, cinnamon, clove, garlic, mustard seed, sesame, ginger and onion had an antimicrobial effect on the growth of Salmonella and STEC in buffered peptone water (BPW), which is the medium generally used for primary enrichments as described in the ISO methods. Additionally, allspice, badiane, cinnamon and clove also had an inhibitory effect on real-time PCR used to screen enrichments for the presence of Salmonella or STEC. Therefore several spices influenced the Salmonella and STEC detection methods, resulting in false negative results. Therefore we explored whether the antimicrobial effect of the growth-inhibiting spices could be countered by replacing BPW with tryptic soy broth (TSB), adding K<sub>2</sub>SO<sub>3</sub>, or increasing the dilution factor of the spices in the primary enrichment. This revealed that for most spices the growth inhibiting effect could be countered by using TSB+K<sub>2</sub>SO<sub>3</sub> instead of BPW, or by increasing the dilution factor. Furthermore, it was shown that the PCR-inhibiting compounds originating from some spices could be removed by the use of a different DNA extraction method, and that different PCR DNA polymerases differ in their sensitivity to these components. Significance: In conclusion, the results presented in this study can be used to improve the detection methods for Salmonella and STEC in the presence of spices.

#### T2-05 A DNA Tagging Method for Improved Traceability and Prevention of Fraud in the Food Supply

ANTONIOS ZOGRAFOS<sup>1</sup>, Quin Chou<sup>1</sup>, Christopher McCormick<sup>1</sup>, Lucia Cerillo<sup>1</sup>, Adam Idoine<sup>1</sup>, Johnathan Burleson<sup>1</sup> and Laurie Clotilde<sup>2</sup> <sup>1</sup>SafeTraces, Pleasanton, CA, <sup>2</sup>ScanX Technologies, Palo Alto, CA

**Introduction:** Adulteration and fraud present major threats to food safety and supply, costing the industry \$30-40 billion annually. Present traceability methods are generally insufficient to secure supply chains as they are tied to the packaging, not the content. Even modern blockchain solutions cannot close the gap between the physical product and the digital record.

**Purpose:** We evaluated a new method that applies DNA taggants directly to the food, not the packaging. These taggants act as invisible barcodes and are used to identify detailed source information and in many cases confirm purity, reducing or eliminating the likelihood of fraud or adulteration of the food.

**Methods:** Short DNA sequences, encapsulated in food grade materials, were delivered as barcodes using two commercial systems, the miniDART and the DART 3000, which can be easily retrofitted into processing equipment. Thirty-two unique DNA sequences were used for the combinatorial tagging allowing up to 232 barcodes to be formed. Unique barcodes can be delivered every three seconds, enabling tagging of food products during automated bag or bottle filling. A multiplex, 15-minute, qPCR assay was used to recover the barcodes.

**Results:** DNA barcodes were applied to a number of commodities, including apples, soybeans, canola seed and oil, caviar, ammonium nitrate fertilizer and others. In all cases, the DNA barcodes remained stable for the life of the product and were reliably recovered. Stability of certain formulation exceeded two years. Recovery success was measured using Youden's J statistic, which was as high as 0.995. Dilution as low as 15% was detected. Sensory and chemical tests revealed no effect on the taste or shelf life of the products.

**Significance:** This method enables defense against adulteration and fraud in the food supply chain for bulk and high-value commodities at a cost estimated as low as 0.25% of the value of the product.

#### T2-06 ISO 16140-2 (2016) Validation of Genedisc for the Detection of Shiga Toxin-producing *Escherichia coli* from O157, O111, O26, O103 and O145 Groups

Justine Baguet, Cécile Bernez, **NICOLAS NGUYEN VAN LONG**, Christophe Quere and Maryse Rannou *ADRIA Food Technology Institute, Quimper, France* 

**Introduction:** Shiga toxin-producing *Escherichia coli* (STEC) is a serious foodborne hazard of increasing concern to food safety authorities due to significant outbreaks, mainly involving the top five serogroups (O157, O111, O26, O103 and O145). Whereas current methods for detection of *E. coli* O157:H7 are well developed, alternative methods validated against ISO/TS 13136:2012 for these five serogroups are still lacking.

**Purpose:** For the first time, an independent study compared an alternative method for the detection of STEC from the top five serogroups in raw dairy products, vegetables and raw ground beef to the ISO/TS 13136:2012 (reference method) according to the ISO 16140-2:2016 for NF-Validation (Afnor).

**Methods:** The alternative method is a combination of different realtime PCR assays (GeneDisc, Pall Genedisc Technologies) after enrichment of 25 g portions diluted 1:10 in buffered peptone water supplemented with 10 mg/l acriflavin (for raw dairy products) or prewarmed (for ground beef). Samples were incubated at 37  $\pm$  1°C 16 to 20 h (dairy) or 41.5  $\pm$  1°C 8 to 20h (ground beef). DNA extractions were performed on 50µl of enrichment before PCR screening. The alternative method was compared to ISO/TS 13136:2012 in order to evaluate its sensitivity, relative level of detection (RLOD), inclusivity and exclusivity.

**Results:** Overall, 231 samples were analysed with both methods, providing 105 to 109 positive results depending on the assay combination. Depending on enrichment protocol and assay combination, the sensitivity of the alternative method ranged from 83.8 to 85.3%; it was from 66.1 to 68.8% for the reference method. Among the five tested matrix/strain pairs, RLOD values from 0.761 to 1.249 indicated equivalent levels of detection for both methods. The 50 tested target-strains were detected and no cross-reaction was observed with the 30 tested non-target strains.

**Significance:** Equivalent performances but better practicability were observed for the alternative method, for which the present study is the first step towards validation and compliance with European food safety regulations.

# WEDNESDAY - 15.30 - 17.00

# T3 Technical Session 3 – Novel Laboratory Methods, Sanitation and Seafood

T3-01\* Comparative Genomics of Persistent *Listeria monocytogenes* Clonal Complexes Suggests Genetic Exchanges within and between Seafood Processing Plants of Ready-to-Eat Food Products in France

> FEDERICA PALMA<sup>1</sup>, Thomas Brauge<sup>2</sup>, Nicolas Radomski<sup>1</sup>, Laurent Guillier<sup>1</sup> and Graziella Midelet-Bourdin<sup>2</sup> <sup>1</sup>ANSES, Laboratory for Food Safety, University of Paris-Est, Maisons-Alfort, France, <sup>2</sup>ANSES, Laboratory for Food Safety, Boulogne sur Mer, France

> **Introduction:** In efforts to control and eradicate *L. monocytogenes* in the food chain, its ability to adapt to environmental stressors in food-processing environments continues to pose a serious challenge to the food industry and public health authorities.

**Purpose:** The aim of this study was to characterize the genomic diversity of persistent *L. monocytogenes* isolates collected from French seafood factories to determine the molecular basis of their long-term survival.

**Methods:** Based on comparative genomics, we investigated the genetic relatedness and the presence of genetic determinants contributing to the persistence of 94 *L. monocytogenes* strains repeatedly isolated over two to six years from food and working surfaces of three seafood-processing facilities (A, B, C).

**Results:** Five clonal complexes (CCs) were detected (7, 101, 121, 155, and 204), showing limited SNP distance between isolates of a single CC, with the highest genetic diversity for CC121 (pairwise SNP distances' first and third quartiles=five and 51). Except for a few strains isolated at the beginning of the sampling timeframe, mobile genetic elements such as plasmids, prophages and transposons were highly conserved within each CC and harbored genes related to biocide resistance (e.g., arsenic, cadmium and benzalkonium chloride). Moreover, entire or partial plasmids/prophages were

shared between CCs; a "mobilizable" pLM80-like plasmid was almost identical in CC204, CC155 and CC7 strains isolated from different facilities over the same period. A unique set of prophages characterized CC7/CC121 and CC101/CC155 strains from plant A and B, respectively. Furthermore, part of a *comK* prophage was shared by CC7, CC101 and CC155 strains.

**Significance:** These results suggest that inter- and intrafacility exchanges of genetic material conferring adaptive and survival advantages may occur between *L. monocytogenes* CCs colonizing specific niches and/or harbourage sites where sanitation failed. Investigating the evolution and transfer of phage-derived elements could support traceback of strains with long-term survival potential and implementation of effective operational strategies to prevent contamination of RTE foods.

#### T3-02 AquaSpark, a Novel Chemiluminescent Technology Platform for Onsite Pathogen Diagnostics

MARIO HUPFELD<sup>1</sup>, Nadine Heinrich<sup>2</sup>, Lukas Reinau<sup>2</sup>, Julian Ihssen<sup>3</sup> and Lars Fieseler<sup>2</sup>

<sup>1</sup>Nemis Technologies, Zürich, Dübendorf, Switzerland, <sup>2</sup>ZHAW, Waedenswil, Switzerland, <sup>3</sup>Biosynth AG, Staad, Switzerland

Introduction: The detection of microorganisms in food, clinical diagnostics, and water and environmental samples remains challenging despite recent advances in molecular biology techniques. Growing demand for local and sustainable production contrasts with the requirements of laboratory analysis, which excludes many current technologies from becoming effective manufacturing-site screening tools.

**Purpose:** We evaluated the novel AquaSpark technology for the detection of foodborne pathogens as well as for clinical applications. **Methods:** Detection molecules tested were composed of a light-emitting part (modified dioxetanes) combined with tailored enzyme labile groups (ELG). For example, a caprylate moiety was used as ELG for *Salmonella* spp. detection. Target bacteria were inoculated at low levels (10 CFU/ml), enriched by incubation, and detection molecules were added after determined time intervals. A selective enrichment broth with specific phage components was designed and successfully tested to inhibit disturbances by competitive microflora such as *Citrobacter*.

**Results:** Results could be obtained as early as six hours after inoculation. More than 60 serovars of *Salmonella* spp. and over 30 strains of *L. monocytogenes* were tested and detected within the same day as inoculation. Competitive microflora could be inhibited in co-culture experiments and allowed target bacteria to be detected even if the starting inoculation number of the ladder was two orders of magnitude lower. Further proof-of-concept detection molecules for colliforms, carbapenemases, *E. coli, Listeria* spp. and *Staphylococcus aureus* will be presented.

Significance: Investigated phenotypic detection molecules release light in response to specific bacterial enzymatic activity. The simplicity of the assay allows safe tests on site even for pathogenic organisms such as *Salmonella* spp. without the need for a full-sized laboratory.

#### T3-03 Development of a Real-time Cell Analysis Method as a Fast and Accurate Method for Detecting Infectious Particles of Hepatitis A Virus

Samuel Lebourgeois, Audrey Fraisse, Catherine Hennechart-Collette, Laurent Guillier, Sylvie Perelle and **SANDRA** MARTIN-LATIL

ANSES, Laboratory for Food Safety, University of Paris-Est, Maisons-Alfort, France

Introduction: Hepatitis A virus (HAV) has been increasingly reported as the cause of foodborne disease outbreaks. To date, a standard method is currently available for the detection of HAV genomes in foodstuffs by RT-qPCR (ISO 15216). Despite its usefulness in the investigation of foodborne viruses, this molecular method is unable to discriminate between non-infectious and infectious virus. Recently, an innovative real-time cell analysis (RTCA) technology based on cellular impedance measurements has been used to monitor the cytopathic effects (CPE) induced by several viruses.

**Purpose:** The aim of this study was to evaluate cell impedance as a measure of HAV-induced CPE. **Methods:** The cellular impedance of FRhK-4 cells uninfected or

Methods: The cellular impedance of FRRK-4 cells uninfected or infected with the adapted strain of HAV (HM175/18f) was measured in real-time using the xCELLigence system.

**Results:** Kinetics of cell impedance showed that HAV induced a decrease in cell index (CI) whilst CI remained constant in uninfected cells. The HAV-induced CI drop was correlated with the onset of HAV-induced cell death as shown by using a cell viability assay and appeared earlier than visual observation under a microscope. FRhK-4 cells were further infected with serial tenfold dilutions of HAV.

#### \* Student Award Competitor

The kinetic curves showed that the Cl drop in HAV-infected cells was delayed with the dilution of viral inoculum and that the sensitivity of the RTCA assay was 10<sup>1</sup>PFU per well. In addition, an inverse linear relationship was established over a range of five log between the concentration of HAV and the time to reach 50% of Cl decrease (TCl<sub>sp</sub>).

Significance: The innovative RTCA-based titration method for HAV could be used for high-throughput screening of inactivation treatments or antiviral molecules against HAV. Therefore this innovative method will be helpful in managing the viral risk in food virology.

#### T3-04 Mold Identification with MALDI-TOF MS: ID-Fungi Plates and Relevance of the Reference Library for Easy and Reliable Results

Semcheddine Cherrad<sup>1</sup>, Markus Kostrzewa<sup>2</sup>, Katharina Mucek<sup>2</sup>, **DANIELE SOHIER**<sup>2</sup> and Sebastien Vacher<sup>1</sup> <sup>1</sup>Conidia, Quincieux, France, <sup>2</sup>Bruker, Bremen, Germany

Introduction: MALDI-TOF MS is now perceived as one of the most promising alternatives for the identification of spoiler and technological molds, solving currently encountered issues: lack of robustness of phenotypic procedures and very few experts operating in the field. But in some cases, direct harvesting of molds from agar plates is difficult. Liquid cultivation is then prescribed, postponing the time-to-result.

Purpose: ID-Fungi Plates contain a membrane filter to facilitate mold harvesting. The workflow combining this new plate format with MALDI-TOF MS was evaluated by two laboratories on strains from various genera (70 strains at one lab and 90 strains at the other). Methods: The handling and reliability were compared to direct harvesting from classical Sabouraud plates using the MALDI Biotyper automate. The MALDI Biotyper Reference Library for fungal identification, which covers the most important taxa in food mycology with 58 genera and 151 species, was used.

**Results:** The required biological material for MALDI-TOF analysis is obtained in only 24 hours of cultivation when using the ID-Fungi Plates. These ID-Fungi Plates enable collection of the biomass in less than 10 seconds for almost 90% of the tested strains, while more than one minute is usually required for 60% of the strains. More than 80% of the strains were easily identified at the genus or species level using the ID-Fungi Plates, while only 60% were identified using the Sabouraud plates.

Significance: The ID-Fungi Plate with the filter membrane improves the practicability and accuracy of MALDI-TOF MS for mold identification. Results could be easily obtained in one hour starting from isolates on ID-Fungi Plates. The need for liquid cultivation with longer time-to-result is significantly reduced and required only for some very specific molds that are usually not encountered in food mycology. The Reference Library should be constantly updated with species of relevance in food mycology.

#### T3-05 Dairy Isolates of Biofilm-forming *Bacillus* Demonstrate Enhanced Resistance to Cleaning Procedures

#### levgeniia Ostrov and MOSHE SHEMESH

Department of Food Quality and Safety, Agricultural Research Organization (ARO) The Volcani Center, Rishon LeZion, Israel

**Introduction:** Biofilm-forming bacteria are often involved in hygiene problems during food processing, and therefore present a major microbiological challenge in the field of food quality and safety. The main strategy to ensure the optimal hygiene level in food processing facilities is regular cleaning and disinfection regimes. However, these regimes show varying efficacy in eliminating biofilm bacteria, since biofilm formation can facilitate bacterial survival in unfavorable conditions.

**Purpose:** Investigating the role of biofilms formed by *Bacillus* species and their resistance to cleaning operations.

**Methods:** Identification of new milk-derived *Bacillus* strains, obtained from the dairy-associated environment, was performed based on colony morphology as well as whole-genome sequencing and bioinformatics analysis. The biofilm-formation potential was evaluated through characterizing submerged, pellicle and bundle biofilm formation as well as genome comparison analysis. Susceptibility of *Bacillus* species to industrial cleaning procedures was detected using a specially designed model system which resembles the conditions of cleaning-in-place regimes.

**Results:** Dairy-associated *Bacillus* strains were characterized by the formation of robust biofilms during growth in milk. Moreover, genome comparison analysis revealed notable differences in putative biofilm-associated determinants between the dairy and non-dairy *Bacillus* isolates, which correlated with biofilm phenotype. Furthermore, the dairy-associated *Bacillus* isolates demonstrate higher resistance to cleaning procedures compared to non-dairy *Bacillus*. Also, our observations indicate enhanced resistance of the poly- $\gamma$ -glutamic acid overproducing strain, which generates high amounts of proteinaceous extracellular matrix, to the cleaning procedures. We, therefore suggest that the enhanced resistance of the dairy *Bacillus* isolates can be attributed to robust biofilm formation.

Significance: Findings of the study underline the importance of evaluating the efficacy of commercial cleaning agents for biofilm-forming bacteria, which are relevant to industrial conditions. Consequently, we believe that this study can facilitate assessing and improving industrial cleaning procedures.

#### T3-06\* Using DNA Barcoding to Investigate the Accuracy of Seafood Product Labelling in Taiwan

**PEI-YING CHEN**, Tsung-Yun Liu and Kung-Hao Liang National Yang-Ming University, Taipei, Taiwan

Introduction: As in many far eastern countries, seafood is one of the major types of food frequently consumed by people in Taiwan. With higher attention to food nutritional value, seafood consumption has dramatically increased nowadays. Food fraud has become a concerning issue recently, and seafood mislabeling has been considered a fraud problem.

**Purpose:** Since food technology has become more advanced, morphological identification is no longer the usual method to authenticate. Due to the lack of extensive surveys of seafood identification, the present study aimed to evaluate seafood product label correctness in Taiwan, including fresh, frozen and processed products.

**Methods:** This study employed DNA barcoding approaches to investigate seafood label accuracy in Taiwan. Firstly, we designed PCR primers for the amplification of the mitochondrial cytochrome c oxidase subunit I (*Coi*) gene. Later, we collected 41 fish samples, including tuna, swordfish, salmon, yellowtail, snapper and cod, from hypermarkets, chain Japanese restaurants, supermarkets, and fast food restaurants. *Coi* gene sequences were then compared with those in the nucleotide collection hosted by the United States National Center of Biotechnology Information (NCBI), as well as those in the Barcode of Life Data System (BOLD).

**Results:** Among the 41 samples, eight samples (19.5%) were mislabeled. Processed products had the highest mislabeling rate of four out of nine (44.4%). In the mislabeled products, snapper appeared to have the highest mislabeling rate of five out of six (83.3%).

Significance: With this high mislabeling rate, it is necessary to pay more attention to seafood products and manage their labels, especially for snapper products. Since it is not compulsory to label scientific names on the package, it might be difficult to govern seafood industry integrity. In order to manage seafood products and reduce the mislabeling rate, DNA barcoding appears to be a very useful technique.

### THURSDAY - 8.30 - 10.00

# T4 Technical Session 4 – Communication and Outreach, Food Toxicology, Pathogens and Risk Assessment

#### T4-01 The USDA-NIFA Food Virology Collaborative (NoroCORE): An Example of the Use of Team Science to Address a Major Food Safety Challenge

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Introduction: Human norovirus (HNV) is the leading cause of foodborne disease. The NoroCORE project (Norovirus Collaborative for Outreach, Research, and Education, also called the Food Virology Collaborative), was established in 2011 through a \$25 million award from the USDA National Institute of Food and Agriculture (NIFA). **Purpose:** NoroCORE's purpose was to develop improved knowledge, skills, and capacity to study foodborne viruses across the farm-to-fork continuum.

**Methods:** The project was organized into six "core" areas: (i) Molecular Virology; (ii) Detection; (iii) Epidemiology and Risk Analysis; (iv) Prevention and Control; (v) Extension and Outreach; and (vi) Capacity Building. By project's end, over 20 academic and government groups had received funding to collaboratively address myriad projects, and more than 50 stakeholder organizations were actively engaged throughout the project.

**Results:** Some key NoroCORE outputs include (i) development of an *in vitro* method to cultivate HNV; (ii) comprehensive comparison of the relevance of various cultivable HNV surrogates; (iii) identification of various ligands to facilitate HNV capture and detection; (iv) NorOPTIMAL, an agent-based quantitative risk assessment model; (v) a comprehensive comparison of the anti-HNV efficacy of traditional and emerging sanitizers and disinfectants; (vi) various digital and online educational materials targeting key stakeholder groups; (vii) production of editable guidelines for vomit/fecal cleanup in food service establishments; and (viii) an innovative social media campaign to facilitate education and information exchange. Scientific and professional capacity were built as evidenced by the establishment of a publicly available food virology literature database and formal reagent exchange, as well as the training of over 50 graduate students.

Significance: Through collaborative activities and extensive stakeholder involvement, the NoroCORE project has resulted in a greater appreciation for the role of viruses in foodborne disease and rapidly moved the scientific understanding of foodborne viruses forward. The purpose of this presentation is to provide details about NoroCORE activities outputs and demonstrate the power of team science.

#### T4-02 Prioritization of Chemical Hazards in Food to be Considered in the Canadian Food Inspection Agency Establishment-based Risk Assessment Model

**MOHAMED RHOUMA**<sup>1</sup>, Manon Racicot<sup>1</sup>, Alexandre Leroux<sup>2</sup>, Romina Zanabria<sup>2</sup> and Sylvain Quessy<sup>3</sup>

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**Introduction:** The Canadian Food Inspection Agency (CFIA) has developed a quantitative risk assessment model for food establishments that quantifies the impact of microbiological hazards. Chemical hazards are also important to be considered by this model but, as limited information is available, there is a need to develop a prioritization method to target the most important chemical contaminants in food.

**Purpose:** To prioritize chemical contaminants in food consumed by Canadians in order to identify the most significant ones that should be considered in the CFIA model.

**Methods:** Data on chemical contaminants assessed through the CFIA's National Chemical Residue Monitoring Program between 2013 and 2017 were used in this study. The risk matrix approach, which combines the probability of occurrence and the severity, was used for the prioritization of chemical contaminants. The scores of the probability of occurrence were categorized based on the prevalence of historical violations of the Canadian maximum regulatory limits. The scores of severity were categorized using a toxicological decision tree. Chemical contaminants were plotted in a 4×4 risk matrix based on both scores.

**Results:** Overall, 74 chemical contaminants were included in this analysis. Out of 60,148 food samples taken from 2013 to 2017, including both domestic and imported products, 2,947 were in violation (4.9%). The probability of occurrence was categorized as severe, high, medium or low for four, 17, 19, and 34 chemical contaminants respectively. Among contaminants with a severe and high probability, the severity was found to be severe, high, medium and low for two, four, seven, and 8 eight contaminants respectively. Based on this risk matrix approach, nitrofurans, thiouracil, arsenic and cadmium are proposed to be the highest priority compounds in foods in Canada.

**Significance:** This prioritization method allowed the identification of the most significant chemical contaminants to be included in the CFIA risk assessment model to help with the risk-based allocation of inspection resources.

#### T4-03 Safe Food for Canadians Regulations, the New Canadian Food Safety Law: What are the Main Differences from the European Union Hygiene Package and the United States Food Safety Modernization Act? What are the Threats and Opportunities?

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**Introduction:** The Safe Food for Canadians Regulation (SFCR) became effective January 15, 2019, introducing a new approach to food safety by the Canadian Food Inspection Agency. In this study, we assess the EU awareness of SFCR.

**Purpose:** Why the SFCR? what are the main differences between SFCR, the United States Food Safety Modernization Act (FSMA), and the EU Hygiene package? What are the threats and opportunities? **Methods:** We studied the n. 16 Part and n. 8 Division of SFCR and compared it with FSMA and its seven rules, and the EU Hygiene Package and its four main regulations. Then we developed a matrix chart to identify the similarities and differences.

Results: On October 24, 2018, the Canadian Food Inspection Agency officially presented to Canadian food industries at the 9th Annual Food Regulatory & Quality Assurance Summit in Toronto. The main differences with FSMA are different denominations of Preventive Controls (PC), a clear request of hygienic design development inside the PC, the licence for importer built more clearly and built in a clearer and stronger way compared to the FSVP Rule. More similarity for the structure with the EU Hygiene Package (all in one rule: animal food origin, vegetables and fruit, processed and manufactured food, beverages and wine), but many differences about PC, the Hazard Analysis, the inclusion of Hygienic Design and Food Safety Culture. No food safety exists if the wrong behaviour doesn't change into the right behaviour if the paper doesn't become awareness. The opportunity? The opportunity is global business growth. Significance: The pillars of the new North American Food Safety is the Food Safety Culture, the Preventive Controls and the Hygienic Design. Those Pillars are spreading the world food safety systems. While much voluntary food standard exists, the revolution approach is coming from mandatory national law.

#### T4-04 Risk Benefit Assessment of Foods: Lessons Learned from a Capacity Building Experience Under the RiskBenefit4EU Project

Géraldine Boué<sup>1</sup>, Paula Alvito<sup>2</sup>, Roberto Brazão<sup>2</sup>, Paulo Carmona<sup>3</sup>, **LEA JAKOBSEN**<sup>4</sup>, Carla Lopes<sup>5</sup>, Carla Martins<sup>2</sup>, Jeanne-Marie Membre<sup>1</sup>, Sarogini Monteiro<sup>6</sup>, Pedro Nabais<sup>3</sup>, Sofie Theresa Thomsen<sup>4</sup>, Duarte Torres<sup>5</sup>, Silvia Viegas<sup>2</sup>, Sara Pires<sup>4</sup> and Ricardo Assunção<sup>2</sup>

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Introduction: Risk-benefit assessment of foods (RBA) has emerged recently to estimate the overall impact of food, food ingredients and diets on human health. Significant methodological progress has been made and its value is now recognized to support the decision-making process in public health to prevent food-associated diseases and promote wellbeing in populations.

**Purpose:** At this time, few research groups have experience in RBA. RiskBenefit4EU project (RB4EU) was funded by the European Food Safety Authority (EFSA) to promote and disseminate the use of RBA by developing a harmonized RBA framework. The presentation will introduce the strategy developed to capacitate a new team to perform an RBA of foods and lessons learned from this experience.

#### \* Student Award Competitor

**Methods:** Activities of the capacity building experience were designed by experienced researchers in RBA to transfer methodologies to a new multidisciplinary team comprised of experts in risk assessment in toxicology, microbiology or nutrition, epidemiology, dietary assessment or data analysis.

**Results:** Short courses were organized in two weeks of training, using a learning-by-doing process, to:

 (i) build a common language within the team by harmonizing important concepts: hazard, health effect, adverse health effect, beneficial health effect, risk, benefit, health and health impact;
 (ii) learn basics used in RBA, including risk assessment in toxicology, microbiology, and nutrition, epidemiology, data analysis, modeling, statistics and uncertainty analysis;

(iii) become familiar with the stepwise RBA approach with the explanation of key steps that were illustrated with RBA examples previously performed;

(iv) initiate an RBA case study, performed by the new RBA team, to practice and answer a specific public health question. Significance: The RB4EU project organized the first training in RBA. The strategy developed, with the materials and method used, can now be re-used to capacitate other new teams in RBA and can be considered as a robust basis to build on.

#### T4-05 Non-targeted Identification of Food Adulterants Using Hand-held Near-infrared Spectrometers

**RONALD SARVER**, Douglas MacRae, Brent Steiner, Robert Donofrio and Greg McNeil

Neogen Corporation, Lansing, MI, USA

Introduction: Non-targeted methods to identify food ingredients and products that have been accidentally or intentionally contaminated are important to maintain a safe food supply. Miniaturization of near-infrared spectrophotometers, the ability to interface these detectors to smartphones, and cloud-based access to chemometric and artificial intelligence software are increasing the use of handheld spectrophotometric methods for applications in food safety and traceability.

**Purpose:** The purpose of this work is to present chemometric methods developed using hand-held near-infrared spectrophotometers for non-targeted identification of adulterants in various food ingredients and products.

**Methods:** Near-infrared spectral data were collected on five samples of several herbs and spices, with triplicate spectra for each sampling. Spectra of common adulterants were also collected, including corn starch, monosodium glutamate, brick dust, sawdust, olive leaves, and prickly pear seeds, as well as mixtures of spices with these adulterants. Near-infrared spectra were preprocessed and analyzed using principal component analysis to classify the spectra and determine the presence of the contaminants. Calibration matrices were constructed and cross-validated using spectra that were collected independently of those used to construct the calibrations. To evaluate the utility of developing PCA methods for other commodities, additional near-infrared spectral data were collected on ground coffee, chicory, coffee and chicory mixtures, bovine milk, and mixtures of milk and water, urea and chalk.

**Results:** Principal component analysis of near-infrared spectral data were used to detect the presence of 10% or greater of the contaminants tested in turmeric, black pepper, mustard seed, chili powder, cinnamon, coffee, coriander seed, cayenne, oregano, paprika and saffron. Testing of other commodities showed principal component analysis methods were able to different ground coffee from chicory and mixtures of milk and water, urea or chalk. **Significance:** The principal component analysis methods that were developed enable rapid non-targeted detection of contaminants in food ingredients and products using inexpensive hand-held near-infrared spectrophotometers bluetooth interfaced to smartphones.

#### T4-06 Leveraging WGS Databases to Enhance Risk Assessment, Attribution, and Large-scale Epidemiology Studies

JANE VAN DOREN<sup>1</sup>, Moez Sanaa<sup>1</sup>, Regis Pouillot<sup>1</sup>, Francisco Garces Vega<sup>1</sup> and Errol Strain<sup>2</sup> <sup>1</sup>U.S. Food and Drug Administration–CFSAN, College Park, MD, USA <sup>2</sup>Center for Food Safety and Applied Nutrition, U.S. Food and Drug Administration, College Park, MD, USA

**Introduction:** Food safety risk assessments, attribution, and largescale epidemiological investigations have the potential to provide better and new types of information when whole genome sequence (WGS) data are effectively integrated.

**Purpose:** To create, apply, and make freely available a set of tools appropriate for risk assessment, attribution, and large-scale epidemiologic studies for analysis and visualization of whole genome sequencing datasets provided by National Center for Biotechnology Information (NCBI) Pathogen Detection, which is supported by the GenomeTrakr and PulseNet networks.

Methods: Salmonella enterica and Listeria WGS and associated metadata provided by NCBI Pathogen Detection database were curated and organized by implementing controlled vocabularies for each field and aligning food source names to the Interagency Food Safety Analytics Collaboration Food Categorization scheme. Network analysis methods were used to characterize and visualize relationships among isolates based on SNP distance, including relationships among food, environment and clinical isolates. Additional relationships were described using a minimum spanning tree, clustering tree, circular and Sankey plots, geographical maps, and epidemiological (time) curves. The interactive interface was developed to facilitate queries and hypothesis generation. Results: The application is currently populated with NCBI Salmonella enterica and Listeria monocytogenes data sets (160,000 isolates in the database as of 2018). Through the categorization scheme, it is possible to determine numbers of clinical strains genetically close to foods at a variety of levels (romaine lettuce, leafy greens, or produce). For example, 113 Salmonella strains isolated from leafy green vegetables are closer than 12 SNPs from 930 clinical strains. Study of 34 connected components clarifies the global relationships among strains. The application is freely available (https://fda-riskmodels. foodrisk.org/genomegraphr/).

Significance: The new user-friendly open-source web application GenomeGraphR leverages whole genome sequencing data collected primarily for use in outbreak investigations and transforms it to provide a resource for research and hypothesis generation by risk assessors, attribution scientists, and epidemiologists.

## THURSDAY - 10.30 - 12.00

## T5 Technical Session 5 – Dairy and Other Food Commodities and Pathogens

# T5-01 Listeriosis in South Africa: Facts, Figures and What We Should be Doing about It

#### PIETER GOUWS

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Introduction: During 2017 and 2018, South Africa experienced the largest outbreak of listeriosis ever recorded in the world. Purpose: To understand the organisms implicated in the outbreak. Methods: A total of 636 clinical Listeria monocytogenes isolates have been sequenced by the National Institute for Communicable Diseases (NICD). Genomic DNA from each isolate was extracted using the QIAamp DNA mini kit (Qiagen, Germany), and paired-end libraries were prepared using the Nextera XT DNA library kit, followed by 2×300-bp sequencing on a MiSeq platform (Illumina, Inc., USA). Results: A total of 1,060 laboratory-confirmed cases have been reported during this time: 443 (42%) in neonates, followed by 32% in the age group 15 to 49 years, and a total of 216 deaths. The case mortality rate in the South African outbreak was 27% and is comparable to other outbreaks in the world. Prior to 2017, an average of 70 confirmed cases per year were reported in South Africa. Of the 636 clinical isolates, 91% belong to the sequence type 6 (ST6) and the other isolates belong to 16 different sequence types. Implicated products were recalled on 4 March 2018, and a total of 87 cases were reported since 5 March 2018. Of these isolates, 48 were sequenced and 33 were ST6, and the remainder belong to seven other sequence types (ST1, ST2, ST5, ST7, ST039, ST554). Of the food and environmental isolates, only 13% were ST 6 while 87% belong to 19 different sequence types.

**Significance:** This outbreak did provide an opportunity to evaluate the readiness of the food sector and state to deal with this complex issue. Food safety requires effective regulation, capacity and transparency. Shortcomings were evident in the extensive delay between the first reported cases in January 2017, the announcement of the outbreak in December 2017 and the possible source being identified in March 2018.

#### T5-02 Microbial Ecology and Food Safety of Fermented Carrot Juice

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Introduction: Food fermentation is an age-old preservation method. Not only at the household level but also in Michelin-star restaurants, vegetable fermentations are regaining popularity. Therefore it is of importance to provide evidence on the safety and robustness of these fermentation processes. Purpose: The study evaluates the survival-suppressing qualities of spontaneous carrot juice fermentation at 20°C on three foodborne pathogens and the impact of the presence of these pathogens on the microbial community.

Methods: Listeria monocytogenes, Salmonella Typhimurium and Escherichia coli O157 (10<sup>3</sup> CFU/ml each) were inoculated into freshly juiced carrots, simulating initial contamination. The pathogen development and pH were monitored, and the microbial community characterised by 16S amplicon sequencing. After 30 days, the fermented carrot juice was mixed with fresh cucumber juice for organoleptic reasons and stored under refrigeration (7.5°C). During the mixing step post-contamination was simulated by adding fresh pathogens

Results: The fermentation process had a suppressive effect on all three pathogens, especially on Listeria monocytogenes, which was absent (<10 CFU/ml) after three days of fermentation (pH 4.6). The other two pathogens could not be detected after 15 days (pH 4.0). The presence of pathogens did not affect the pH drop or the microbial community. L. plantarum dominated among the lactic acid bacteria after 15 days of fermentation. During post-contamination, in the mix of 30-day fermented carrot juice with fresh cucumber juice (pH 4.4), both L. monocytogenes and E. coli died off during refrigeration whereas Salmonella maintained constant.

Significance: Spontaneous fermentation of carrot juice, if the fermentation process is longer than 15 days, enables at least fourlog reduction of L. monocytogenes, Salmonella and Escherichia coli O157. Fermented carrot juice (alone or mixed with fresh cucumber juice) was shown to be a robust and stable microbial environment not supporting the growth of pathogens.

#### T5-03 The Inhibitory Effect of Traditional Pomegranate Molasses on S. Typhimurium Growth on Parsley Leaves and in Mixed Salad Vegetables

#### DIMA FAOUR-KLINGBEIL<sup>1</sup> and Ewen Todd<sup>2</sup>

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Introduction: Fresh-cut vegetables implicated in food poisoning are well documented; at the same time, the washing methods in restaurants or home settings are reportedly not effective enough to mitigate the risks of pathogens found in vegetables. **Purpose:** Pomegranate (PG) molasses is an essential condiment that

is commonly used in Middle Eastern cuisine in local and international gastronomic markets. There is scarce information on the inhibitory effect of PG molasses on Salmonella growth under In Situ conditions and in a food matrix.

Methods: PG molasses in dilution ratios of 1:1 and 1:7, combined with sodium chloride (two percent, v/v), vinegar (2.4%, v/v), and in a dressing mix, was tested against Salmonella enterica serovar Typhimurium LT2 on parsley and salad vegetables. Results: The results showed significant log reductions in Salmonella on parsley treated with PG solutions for 15, 30, and 60 min, reaching a level of 2.55 log CFU/g. The addition of sodium chloride (two percent, v/v) and vinegar (2.4%, v/v) to PG molasses did not exert a synergistic or antagonistic effect on its antibacterial activity. Additionally, the application of PG molasses dressing on salads contaminated with low (three log CFU/g) and high (six log CFU/g) inoculum levels resulted in two to three log reduction independent of temperature (P<.05) compared with 0.5 to one-log reduction for thorough washing alone which may damage the leaf surfaces. Significance: This study shows that PG molasses has greater efficacy than chlorine (200 ppm) against S. Typhimurium and achieved comparable to greater log reduction values than organic acids. The storage temperature and food matrix did not alter the potency of PG. The present work showed that promoting the use of PG as a natural additive and condiment to RTE vegetables offers great potential to effectively reduce the risk of Salmonella contamination and improve microbial safety.

#### T5-04\* Characterizing the Diversity of Bacterial **Communities from Imported Date Fruits** to Control Listeria monocytogenes

#### KRISHNA S. GELDA, Valeria R. Parreira, Gisèle LaPointe and Jeffrev M. Farber

University of Guelph, CRIFS, Department of Food Science, Guelph, ON, Canada

Introduction: Illnesses caused by foodborne pathogens continue to remain a major health and economic concern worldwide. Listeria monocytogenes is one of the most important of these pathogens due to its case-fatality rate being the highest of any foodborne pathogen. Purpose: This project investigates the culturable microbiome of imported, RTE date fruits and how well bacteria isolated from date fruits could control/inhibit the growth of L. monocytogenes. Date fruits were selected over other imported RTE foods tested (such as pollen, seaweed, seeds, and pistachios) based on the diversity of their microbiome and the ability of the associated microbiota to show inhibitory growth against L. monocytogenes.

Methods: Dates from five geographical regions were obtained, including China, Iran, Palestine, Saudi Arabia, and Tunisia. Bacterial strains were isolated from these dates using tryptic soy agar enriched with five percent defibrinated sheep blood. The isolates were then screened using a growth inhibition plate assay against L. monocytogenes and the inhibitor strains detected were identified using 16S rRNA sequencing.

Results: A large diversity of bacteria was isolated from the dates, iwth a total of 191 isolates belonging to 91 different phenotypes being observed. From the inhibition plate assay, 35 isolates were found to produce a zone of inhibition around *L. monocytogenes*, with zone sizes ranging from 0.5 to 5.7 mm. These inhibitor strains were identified as Bacillus spp. Further screening narrowed down the list of inhibitor strains to three to five to be pursued for their antimicrobial activity and whole genome sequencing.

Significance: The results from this research could lead to the discovery of either novel antimicrobial metabolites or beneficial bacteria that could be added to foods to inactivate and/or control the growth of L. monocytogenes. This research was conducted at CRIFS (Canadian Research Institute for Food Safety) and has been funded by CFREF (Canada First Research Excellence Fund).

#### T5-05 Shiga Toxin-producing E. coli (STEC) Occurrence and Virulence Profile Comparison from Flour Samples Obtained during Monitoring and Outbreak Situations

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Introduction: Between 2016 and 2017, Canada experienced two novel outbreaks of Shiga toxin-producing *E. coli* O121 (STEC) illnesses linked to wheat flour.

Purpose: (i) Gather baseline data on the occurrence of STEC and other microorganisms in flour. (ii) Compare occurrence and virulence profiles of STEC isolated from samples obtained during the outbreak investigations (biased sampling) versus monitoring samples (unbiased sampling).

Methods: Outbreak samples: Samples (n=114) were obtained from retail locations or cases' homes as part of the outbreaks and were tested for STEC. Monitoring samples: Samples (n=350) were taken from retail locations across Canada (May 1st, 2018 to March 31st, 2019) and tested for STEC, *Salmonella, Listeria monocytogenes*, and indicator organisms. All samples were analyzed using published methods and isolates were characterized by WGS

Results: Fifteen of one hundred fourteen (13.2%; 95% CI 7.6 to 20.8) of the outbreak samples were found to contain STEC. So far, six of two hundred fifty-two (2.4%; 95% CI 0.9 to 5.1) of the monitoring samples were found to contain STEC. Various STEC serotypes were identified in both sets of samples (08, 088, 0103, 0121, 0159, 0187). The 0121 isolates associated with the outbreak samples possessed the combination of stx2a and eae virulence markers. Significance: The results suggest that STEC of human health concern could be present in flour. The differences in the data sets do not allow for a direct comparison of the occurrence; however, a somewhat higher occurrence was observed in the outbreak samples versus the monitoring samples, which was as expected. The virulence profile of isolates recovered from outbreak samples included the O121 serotype with virulence markers considered to be associated with the potential to cause severe illness. The profiles for isolates recovered from monitoring samples presented various combinations of markers that historically have not been associated with severe illness. These results can inform surveillance design, controls, and risk management decisions linked to STEC in flour.

#### T5-06 **Bacterial Strain Selection for the Validation of High Pressure-treated Juices**

Catherine Rolfe<sup>1</sup>, Nathan Anderson<sup>2</sup>, Glenn Black<sup>2</sup> and **ALVIN LEE**<sup>1</sup>

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Introduction: High pressure processed (HPP) juices are required to demonstrate efficacy as part of process control, e.g., a five-log reduction of pertinent microorganisms. However, there is no common approach for bacterial strain selection and preparation for validation studies, nor is there consensus on HPP process parameters that may affect overall efficacy.

Purpose: The study was to compare HPP inactivation of matrix-adapted *Escherichia coli* O157:H7, *Listeria monocytogenes*, and *Salmonella* isolates in buffer and apple juice and their

appropriateness in process validation. **Methods:** Individual bacterial strains were grown using three different growth conditions: neutral, cold-adapted, and acid-adapted. Approximately six log CFU/mL of the matrix-adapted bacterial strains were inoculated into HCI-adjusted buffered peptone water (BPW) at pH 3.5±0.1 and store-purchased apple juice at pH 3.5±0.1. Samples were treated at sublethal pressures of 500 MPa for E. coli O157:H7 and 200 MPa for Salmonella and L. monocytogenes (180 s, 4°C initial temperature). Analyses were conducted at zero, 24, and 48 h (4°C storage) following HPP using non-selective media.

#### \* Student Award Competitor

**Results:** *E. coli* O157:H7 exhibited barotolerance compared to *Salmonella* spp. and *L. monocytogenes*. In BPW neutral growth conditions, *E. coli* O157:H7 strain TW14359 demonstrated resistance (<3-log reduction) and *E. coli* O157:H7 strain SEA13B88 was most sensitive. Acid-adapted *L. monocytogenes* strains CDC and ScottA were most sensitive in BPW. *Salmonella* isolates grown in neutral and acid-adapted conditions had comparable results (<1.5-log reduction), while cold-adapted S. Cubana and S. Montevideo showed resistance compared to other strains. Bacterial inactivation in apple juice showed similar strain inactivation to BPW but incremental barotolerance was observed. Progressive loss of viability occurred from all post-HPP storage samples.

Significance: These results suggest HPP is an effective inactivation method for *E. coli* O157:H7, *L. monocytogenes* and *Salmonella;* however, matrix composition, bacterial strain selection and preparation methods and process parameters can influence validation outcomes.

#### THURSDAY — 13.30 – 15.00

T6 Technical Session 6 – Pathogens, Epidemiology and General Microbiology

#### T6-01\* Comparative Genomics of *Listeria monocytogenes* Isolated from Fresh Produce, Meat and Clinical Cases

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Introduction: Listeria monocytogenes is a gram-positive, facultative anaerobe that commonly occurs in the natural environment. L. monocytogenes accounted for 57% of deaths caused by foodborne disease in the European Union in 2015, with contaminated food being the main route of infection, and has high morbidity and mortality rates. Whole genome sequencing of bacteria is increasingly being applied in the context of source tracking of L. monocytogenes within the food supply chain. This data gives a wealth of information, including the genetic variability of isolates within food and outbreak environments, and the presence of genes of interest.

Purpose: The purpose of this study was to examine the whole genome sequence of 128 isolates of *L. monocytogenes* isolated from fresh produce, meat products and clinical cases.

**Methods:** *L. monocytogenes* was isolated from fresh produce (n=48) and sequenced on the MiSeq. Meat (n=34) and clinical (n=46) isolates' whole genome sequences were downloaded from the NCBI Sequence Read Archive. All were bioinformatically analysed using Nullarbor.

Results: The results showed all *L. monocytogenes* analysed were from lineages I and II. Isolates from fresh produce, meat and clinical samples did not cluster phylogenetically based on sample type. The results confirmed the antimicrobial resistance (AMR) gene *fosX* is ubiquitous amongst *L. monocytogenes* isolates. The genes *brcB* and *brcC* were found in four isolates, two of vegetable origin, one of meat origin and one of clinical origin. The virulome showed no genes present in clinical samples that were not also present in meat and vegetable isolates.

**Significance:** These results suggest isolates that can lead to human cases of listeriosis can be isolated from both meat and fresh produce, and that currently, *L. monocytogenes* is not a key route of transmission for AMR genes within the food chain, due to the low levels of AMR associated genes found in all sample types.

#### T6-02 Evaluation of Methods for Elution of HEV Particles in Naturally Contaminated Sausage, Figatelli and Pig Liver

CATHERINE HENNECHART-COLLETTE, Audrey Fraisse, Sylvie Perelle and Sandra Martin-Latil

ANSES, Laboratory for Food Safety, University of Paris-Est, Maisons-Alfort, France

**Introduction:** Foodborne transmission of HEV is a growing public health concern in industrialised countries, where the disease is mainly autochthonous, caused by zoonotic HEV of genotype three or four. To date, there is no standardised protocol for the detection of HEV in meat products. Virus extraction is a crucial step and requires efficient grinding for releasing HEV from food of animal origin.

Purpose: The aim of this study was to evaluate six methods for their efficiency in releasing HEV viral particles from figatelli, pig liver sausages and liver samples that previously tested positive for the presence of HEV.

**Methods:** One liver, one figatelli and one sausage that tested positive for the presence of HEV genomes were selected for this study because of their high HEV contamination levels. Three parameters

were assessed for virus elution (weight of food sample, the ratio of weight of sample to elution volume, and homogenisation method). Then, HEV genomes were extracted and quantified by RT-qPCR. **Results:** The quantification of HEV (in log<sub>10</sub> HEV genomes per gram) ranged from 8.79 to 9.70 (sausage), 6.92 to 7.82 (figatelli) and 8.64 to 8.96 (liver), whichever elution method was used. A one-way ANOVA analysis showed that the elution method was a significant factor in improving HEV recovery from figatelli and sausages. Two crucial parameters were the weight to volume ratio of elution buffer (three g/15 ml or 1:5) and the type of mechanical cell disruption for figatelli and pig liver sausages (FastPrep-24 homogenizer). **Significance:** To our knowledge, this study is the first to evaluate several methods for elution of HEV particles from naturally contaminated pig liver products, and may be extended for quantifying other viral genomes from food of animal origin. This approach may improve the risk assessment of HEV in food virology.

#### T6-03 Genomic Method to Highlight Epidemiological Links between *Staphylococcus aureus* Strains

DÉBORAH MERDA, Noémie Vingadassalon, Lyasmine Negrouche, Jacques-Antoine Hennekinne and Yacine Nia *Université Paris-Est, ANSES, Maisons-Alfort, France* 

Introduction: Staphylococcus aureus is involved in several foodborne outbreaks. Pathogenic strains secrete more than 25 types of enterotoxins while only five can be detected by immunoenzymatic assays according to the European official method. In Europe, 380 cases were observed in 2017 (EFSA, 2018). Whole genome sequencing (WGS) may allow detection of all types of enterotoxin genes and aid in investigation of outbreaks in terms of contamination sources, particularly using SNP analysis. **Purpose**: The objective of this work was to implement a typing method to identify whether strains are epidemiologically related.

Purpose: The objective of this work was to implement a typing method to identify whether strains are epidemiologically related. Methods: First, reference genomes for SNP analysis was established using a Bayesian analysis of genetic structuration with the R package "adegenet" on the whole *S. aureus* species. For this, 143 genomes representing *S. aureus* diversity, sequenced by the NextSeq Illumina method, were used. Another panel of strains from foodborne outbreaks was studied using the developed reference genomes. A collection of 144 genomes with strains isolated from the same outbreaks (occurring in Europe between 2005 and 2018) and epidemiologically unrelated strains were used. A phylogenetic inference was performed in order to assign strains to a genetic group obtained previously. In addition, SNP analysis was performed with the reference genome of the genetic group in order to establish epidemiological correlation.

**Results:** Six genetic groups were identified within the *S. aureus* species and the strains involved in foodborne outbreaks were found in all genetic groups. Within each group, strains from the same outbreak present a few SNPs in common with the unrelated strains. So, unlike the *S. aureus* strains found in the clinical environment, it was difficult to determine a threshold of SNPs to discriminate related and unrelated strains in a foodborne outbreak. **Significance:** This first work concerning the WGS of *S. aureus* in

**Significance:** This first work concerning the WGS of *S. aureus* in food highlights the importance of epidemiological metadata when investigating outbreaks due to *S. aureus*.

#### T6-04\* Bacterial Spore Inactivation Mechanisms during Low Energy Electron Beam Treatment

**YIFAN ZHANG**<sup>1</sup>, Nina Huber<sup>1</sup>, Ralf Moeller<sup>2</sup>, Barbora Dubovcova<sup>3</sup>, Georgios Akepsimaidis<sup>3</sup>, Nicolas Meneses<sup>3</sup>, David Drissner<sup>4</sup> and Alexander Mathys<sup>1</sup> <sup>1</sup>ETH Zurich, Zürich, Switzerland, <sup>2</sup>German Aerospace Center, Cologne, Germany, <sup>3</sup>Bühler AG, Uzwil, Switzerland, <sup>4</sup>Albstadt-Sigmaringen University, Sigmaringen, Germany

Introduction: Bacterial spores are the main target of sterilization due to their extreme resistance, and food industries often eliminate them with intensive thermal processing, leading to significant quality losses. Low energy electron beam (LEEB,  $\leq$ 300 keV) has been investigated as an alternative non-thermal microbial decontamination approach for foods due to its minimal impact on food quality and easy implementation in existing processing lines. However, only limited research has been conducted on spore inactivation efficiency by LEEB technology and the inactivation mechanisms are not well understood.

**Purpose:** Investigation of spore inactivation mechanisms by LEEB and contribution to the understanding and application of this emerging technology for enhancing food safety and quality. **Methods:** Spores of *Bacillus subtilis* and four isogenic mutants

**Methods:** Spores of *Bacillus subtilis* and four isogenic mutants lacking different DNA repair mechanisms were treated with a LEEB system (EBLab-200, Switzerland) at energy levels of 80 keV and 200 keV with doses from 1.9 to 9.7 kGy. Average (n=6) *D*-values (the dose required for one-log reduction at a given energy level) were obtained. Differences between average *D*-values were analyzed using Student's *t*-test with a significance level of 0.05.

**Results:** The inactivation curves were log-linear. *D*-values were not significantly different between 80 keV and 200 keV, indicating that inactivation mechanisms are not energy-dependent within the tested range. All DNA repair-deficient mutants showed significantly lower resistance towards LEEB treatment compared to the wildtype, indicating DNA is a target for LEEB treatment. Mutants that are deficient in DNA repair by homologous recombination and nonhomologous end joining showed the lowest resistance, indicating DNA double-strand breakage is one of the main inactivation mechanisms during LEEB treatment.

Significance: This research provides a better understanding of spore inactivation by LEEB treatment and contributes to its application as a non-thermal microbial decontamination approach to delivering safe and high-quality foods.

#### T6-05\* Expression and Prediction of Staphylococcal Enterotoxins G and I

LIVIA SCHWENDIMANN<sup>1</sup>, Déborah Merda<sup>2</sup>, Thomas Berger<sup>3</sup>, Anita Kläui<sup>3</sup>, Jacques-Antoine Hennekinne<sup>2</sup>, Yacine Nia<sup>2</sup>, Michel-Yves Mistou<sup>2</sup> and Hans-Ulrich Graber<sup>3</sup> <sup>1</sup>Agroscope/Université Paris-Est, ANSES, Bern, Switzerland, <sup>2</sup>Université Paris-Est, ANSES, Maisons-Alfort, France, <sup>3</sup>Agroscope, Bern, Switzerland

Introduction: According to the European food safety authority, there are around 300 outbreaks caused by staphylococcal enterotoxin annually. Most of them are described as weak evidence outbreaks. The reason for the lack of information about their occurrence could be that currently only five out of 26 enterotoxins can be analyzed using commercially available kits. Consequently, the presence of the so-called "new enterotoxins" cannot be determined. A group of these new enterotoxin genes (*seg, sei, sem, seo, and seu*) is located on the enterotoxin gene cluster (*egc*) which is part of the staphylococcal beta island (vSa $\beta$ ). These enterotoxins have been described as being involved in staphylococcal food poisoning outbreaks.

genome data can be used for the prediction of staphylococcal egc enterotoxin expression, particularly SEG and SEI.

Methods: The whole genome sequence of 80 *Staphylococcus aureus* strains from different origins (food poisoning outbreaks, humans and animals) was investigated applying bioinformatic methods. Staphylococcal enterotoxin G and I expression was tested In Vitro using an internal sandwich ELISA method.

**Results:** Strains could be perfectly allocated to 14 different vSa $\beta$  types. Furthermore, the type of vSa $\beta$  correlated at 100% with the clonal complex of the strains. The enterotoxins G and I were detected in culture in 75% of the strains. The amount of SEG and SEI produced clearly correlated between the vSa $\beta$ -type and the clonal complex of the strain.

Significance: The present results clearly demonstrate that the In Vitro production of SEG and SEI can be predicted based on the vSa $\beta$ -type and the clonal complex of a strain. This information will enhance the ability to better understand the involvement of the *egc* enterotoxins in staphylococcal food poisoning outbreaks.

#### T6-06\* Influence of Osmotic Stress on Heat Resistance of *Listeria monocytogenes* in Food Products

**INGE VAN VILSTEREN**, Elisabetta Saverio and Nicholas Brian Johnson *Nestlé Research, Konolfingen, Switzerland* 

Introduction: Listeria monocytogenes is a foodborne pathogen which can be found in many raw ingredients. Food products are often stored during food processing, where, due to temperature and other environmental conditions, cells can adapt and become more resistant to additional inimical environments whilst processing, such as during heat treatments.

**Purpose:** In this study, the aim was to investigate the transient changes in heat resistance of *L. monocytogenes* when pre-exposed to environmental stress (i.e., mild osmotic shock) in a dairy-based food matrix.

**Methods:** *L. monocytogenes* EGDe culture was grown and exposed to mild stress, by inoculating a late exponential phase cell culture into a reference food matrix prepared at different total solids (between 10 and 50%) for periods of 0, 10, 20 or 30 minutes. Experiments were also carried out in a reference broth. Thereafter, lethal heat stress was applied at 60, 62.5 and 65°C. Survival rates were measured by plate count, and the data used to calculate D- and z-values. These were then used as the basis to develop a secondary model which was tested using different heat inactivation conditions and food matrices. **Results:** Kinetic parameters based on food matrices and developed for the predictive model (i.e., log<sub>10</sub> D-and z-values were found to be correlated with the water activity resulting from the TS% of the food matrices. A significant increase was found in heat resistance of *L. monocytogenes* as total solids in the matrix increased. The increase in heat resistance was transient and maintained only for 10–20 minutes.

Significance: The study resulted in a predictive model, based on realistic food matrix systems and food processing conditions. The subsequent model provides opportunities to reduce pasteurization temperatures, and therefore reduce costs and deleterious quality impacts.

#### THURSDAY - 15.30 - 17.00

#### T7 Technical Session 7 – Pathogens

#### T7-01 ANSR is More Sensitive than ISO 6579 for Rapid Detection of *Salmonella* spp. in Food and Feed Samples

**EDAN HOSKING**<sup>1</sup>, Jerry Tolan<sup>1</sup>, Frederic Martinez<sup>1</sup>, Muriel Bernard<sup>2</sup>, Preetha Biswas<sup>1</sup>, Robert Donofrio<sup>1</sup> and Maryse Rannou<sup>2</sup>

<sup>1</sup>Neogen Corporation, Lansing, MI, <sup>2</sup>ADRIA Food Technology Institute, Quimper, France

Introduction: A rapid assay for detection of *Salmonella* spp. has been developed using the nicking enzyme amplification reaction technology (NEAR). This isothermal nucleic acid assay (ANSR) produces results in under 45 minutes following sample enrichment. A molecular beacon probe generates a fluorescent signal which is measured in real time using a simple incubator/fluorescence reader. **Purpose**: A validation study was performed according to EN ISO 16140-2:2016 for an extension of our previous validation to cover all human food and feed products.

**Methods:** Four hundred twelve samples were analyzed by ADRIA Developpement to determine the sensitivity of the ISO 6579-1 standard and the candidate method. The confirmation procedure used was RVS broth, XLD and ASAP agar plates, and latex testing. **Results:** The study was conducted under very challenging conditions. This is demonstrated by the observed deviations, which confirm that the contamination levels of the samples were very low, giving sampling heterogeneity within this unpaired study. The method, after supplemented buffered peptone water (BPW) enrichment, showed a significantly higher sensitivity (SE=90.2%) than the standard method (SE=80.3%). A paired study with BPW enrichment showed no significant difference between the two methods. The relative level of detection (RLOD) of the method and the reference methods for five food and one feed sample categories were evaluated with various strain/matrix pairs. According to the ISO 16140-2 protocol, three contamination levels were used (five uninoculated, 20 low level, and five at a slightly higher level). For all the tested matrix/strains, the RLOD is lower than the acceptability limits defined in the validation standard, demonstrating similar RLOD results. Pure cultures of 102 target strains and 30 non-target strains were tested giving expected positive or negative results, respectively.

Significance: The method was successfully certified by NF Validation and demonstrated that it is more sensitive than the ISO 6579 method.

## T7-02 Salmonella Lubbock: A New Serotype in between S. Montevideo and S. Mbandaka

MARIE BUGAREL<sup>1</sup>, Peter Cook<sup>2</sup>, Henk den Bakker<sup>3</sup>, Kendra Nightingale<sup>1</sup> and Guy Loneragan<sup>4</sup> <sup>1</sup>Texas Tech University, Lubbock, TX, USA, <sup>2</sup>CDC, Atlanta, GA, USA <sup>3</sup>Center for Food Safety, University of Georgia, Griffin, GA, USA <sup>4</sup>Texas Tech University, Department of Animal and Food Sciences, Lubbock, TX, USA

Introduction: In cattle, *Salmonella* is commonly recovered from peripheral lymph nodes of asymptomatic animals. Serotypes Montevideo, Mbandaka and the newly described Lubbock are frequently recovered from this niche. *Salmonella* Lubbock evolved from a *Salmonella* Mbandaka progenitor strain that performed two different recombination events with *Salmonella* Montevideo leading to the acquisition of the *fliC* operon and thus to the emergence of two *Salmonella* Lubbock lineages.

Purpose: Our objective was to determine phenotypic and genotypic aspects of *Salmonella* Lubbock in comparison to *Salmonella* Montevideo and *Salmonella* Mbandaka.

Methods: Whole genome sequencing was done using short-read sequencing by synthesis technology of 30 Salmonella Montevideo, 18 Salmonella Mbandaka and 32 Salmonella Lubbock. Raw sequencing data were assembled using SPAdes and analyzed using pipelines available on the Center for Genomic Epidemiology website. A subset of strains belonging to each serotype was investigated phenotypically, including invasion and survival in bovine macrophages, and cytotoxicity on bovine macrophages and Caenorhabditis elegans. Results: Salmonella Lubbock harbors a significantly longer genome than Salmonella Montevideo and Salmonella Mbandaka, especially linked to the presence of a larger number of genes associated with mobile elements. Phenotypically, Salmonella Lubbock behaves similarly to Salmonella Mbandaka but not Salmonella Montevideo while infecting bovine-derived macrophages, including the increase of intracellular concentration and lower cytotoxicity. At the opposite, Salmonella Lubbock, together with Salmonella Montevideo, presents greater toxicity for nematodes, but not Salmonella Mbandaka. Significance: This study provided new knowledge of various phenotypic and genotypic characteristics of Salmonella Lubbock in order to better understand routes of transmission and survival of Salmonella in cattle.

#### T7-03\* Investigation of Genotypic Markers as Indicators for Psychrotrophic Phenotypic Behaviour of *Bacillus cereus*

**DOREEN METTO**<sup>1</sup>, Yinghua Xiao<sup>2</sup>, Elissavet Gkogka<sup>2</sup>, Anne Elsser-Gravesen<sup>3</sup> and Marianne Hammershøj<sup>1</sup> <sup>1</sup>Aarhus University, Aarhus, Denmark, <sup>2</sup>Arla Innovation Centre, Aarhus, Denmark, <sup>3</sup>ISI Food Protection ApS, Aarhus, Denmark

Introduction: Bacillus cereus is a pathogen of importance in refrigerated processed foods of extended durability (REPFEDs) as some strains can grow at refrigeration temperatures. Proper control strategies for *B. cereus* psychrotrophic strains are suggested to be developed based on the precise phenotypic definition. However, thresholds used in previous growth tests vary from five to 14 days, whereas REPFEDs may have a shelf life of >40 days. Presence of the major cold shock protein CspA and specific psychrotrophic/ mesophilic 16S rDNA signatures were proposed and have been used for rapidly predicting psychrotrophic behavior. **Purpose:** To characterize psychrotrophic phenotypes of 62 *B. cereus* strains and their association to the presence of the proposed genetic signatures.

**Methods:** Growth at 7°C was determined by inoculating *B. cereus* strains on nutrient agar (NA), tryptone soya agar supplemented with 5% sheep blood (TSA-SB) and brain heart infusion (BHI) broth, and recorded for 56 days. Significant growth, indicated by visible colonies or turbidity, was checked every seven days. The *cspA* gene and 16S rDNA psychrotrophic and mesophilic signatures were detected following PCR and electrophoresis.

**Results:** Within 42 days of incubation at 7°C, all 62 strains grew in BHI but only 14 strains grew on NA and TSA-SB agars. Mesophilic and psychrotrophic 16S rDNA signatures were present in 60 and 59 strains respectively. *cspA* gene was present in eight strains, which grew in all media tested within seven days.

**Significance:** Presence of *cspA* indicated better adaptability to cold conditions while the psychrotrophic 16S rDNA signature predicted psychrotrophic behavior better. Presence of these two genetic markers plus apparent significant growth at 7°C in BHI broth for 21 days would confidently define a *B. cereus* psychrotrophic strain. Strains characterized as mesophilic by growth test actually grew within the shelf life of REPFEDs. The potential threat of these strains in extended shelf-life products should be evaluated and relevant control measures designed.

#### T7-04 Exploring the Global Transcriptomic Response of *L. monocytogenes* to Desiccation on Stainless Steel

MARTIN LAAGE KRAGH and Lisbeth Truelstrup Hansen DTU Food, Kongens Lyngby, Denmark

**Introduction:** The ability of *Listeria monocytogenes* to survive desiccation for extended periods on food contact surfaces remains a challenge for the food industry.

**Purpose:** The purpose of this study was to further our understanding of the bacterium's survival by investigating the global transcriptomic response of *L. monocytogenes* to desiccation (43% relative humidity (RH), 15°C) on food grade stainless steel surfaces.

**Methods:** Two strains (a food and an outbreak strain) of *L. monocytogenes* were desiccated (43% RH, 15°C) on stainless steel under conditions simulating a food processing plant. Survivor counts and RNA extracts were obtained after zero (control), six, 12, 24 and 48 hours for subsequent rRNA-depleted Illumina TrueSeq RNA library preparations and strand-specific Illumina Hiseq 2000 paired-end RNA-sequencing. Differentially expressed genes were reported as significant (P = < 0.05) if log twofold changes were >1

Significant ( $P_{adjust}$ <0.05) if log twofold changes were >1. **Results:** Both strains were reduced by 1.8 to 2.0-log CFU/cm<sup>2</sup> over 48 hours (from 7.7 log CFU/cm<sup>2</sup>), with the first log reduction occurring after six hours. The number of differentially expressed genes varied among the food (336±20) and outbreak strains (646±32). After commencement of the desiccation, gene expression remained stable over the 48 hours for both strains. A core set of 154 genes were differentially expressed ( $P_{adjust}$ <0.05) in both strains throughout the desiccation and included the downregulated *che* Y and *cheA* (two-component system involved in chemotaxis), the upregulated qoxABCD operon (*sigB* dependent quinol oxidase), and the upregulated *phdA* (general metabolism related to osmotic stress). In contrast, genes such as *inIH* (internalin H) and *Imo0781-0784* (PTS mannose system) were differentially up- or downregulated in the strains

**Significance:** The present study detected novel desiccation associated stress genes in *L. monocytogenes* and revealed strain differences. Taken together this will increase our knowledge of the bacterium's desiccation-stress response and lead to improved control in food processing plants.

#### T7-05\* Tryptone, Peptone and Casamino Acids Affect Acid Survival and GABAe Production in *Listeria monocytogenes* 10403S WT

**CAROLINA BRUSCHI**<sup>1</sup>, Ranju Paudyal<sup>1</sup>, Conor O'Byrne<sup>2</sup> and Kimon A. G. Karatzas<sup>1</sup>

<sup>1</sup>Department of Food and Nutritional Sciences, University of Reading, Reading, United Kingdom, <sup>2</sup>National University of Ireland Galway, Galway, Ireland

Introduction: *Listeria monocytogenes* is a ubiquitous grampositive foodborne pathogen responsible for severe infections in immunocompromised individuals, pregnant women, infants and the elderly. In 2017, 2,480 cases were reported with a 24% fatality rate among the elderly and overall mortality of one in ten cases in the EU. Outbreaks were related to RTE meats, dairy products, fruits, and vegetables. The glutamate decarboxylase (GAD) system is an acid-resistance mechanism defined by the production of GABA/CO<sub>2</sub> through consumption of glutamate/H\*, increasing milieu pH. Tryptone, peptone and casamino acids are sub-products of casein, a protein found in milk and red meats, and an important source of nitrogen for bacterial growth.

**Purpose:** The aim of this study is to analyse the effect of tryptone, peptone and casamino acids on acid survival and extracellular GABA (GABAe) production in *Listeria monocytogenes* 10403S WT. **Methods:** Defined media (DM) was supplemented with 6.8% tryptone, peptone or casamino acids. An overnight culture of *L. monocytogenes* 10403S WT was inoculated in supplemented DM and non-supplemented DM (control). Cultures were incubated aerobically at 37°C for 24 h at 120 RPM. Acid stress survival was evaluated by decreasing the pH to 2.5 with 1 M HCI, and samples for GABAe measurements were taken after reducing pH to 4.3. Cells were incubated aerobically at 37°C for one hour at 120 RPM. Log reduction after acid stress was calculated and GABAe production was analyzed by enzymatic reaction.

**Results:** Cells grown in supplemented DM had higher acidic survival and GABAe production than cells grown in non-supplemented DM. Tryptone, peptone and casamino acids increased survival by 1.48, 2.74 and 2.21-fold, respectively. The highest levels of GABAe (±10mM) were detected in DM supplemented with tryptone (*P*=0.0001) and peptone (*P*=0.0009) after 60 min, and ±4mM with casamino acid supplementation (*P*=0.0005).

**Significance:** Acid tolerance and GABAe production are enhanced in environments containing high levels of amino acids through increased GAD activity in *Listeria monocytogenes* 10403S WT.

#### T7-06 The Role of *Listeria monocytogenes* Glutamate Decarboxyase System in Oxidative Stress Tolerance

Marcia Boura<sup>1</sup> and **KIMON A. G. KARATZAS<sup>2</sup>** <sup>1</sup>University of Reading, Reading, United Kingdom, <sup>2</sup>Department of Food and Nutritional Sciences, University of Reading, Reading, United Kingdom

**Introduction:** The glutamate decarboxylase system is widely present in several types of organisms and is known to play an important role in bacterial acid tolerance. It converts glutamate into γ-aminobutyric acid (GABA), with the consumption of a proton, increasing the intracellular pH, which can be coupled or not with a membrane transporter that imports glutamate and exports GABA. The role of the GAD system in other functions of *L. monocytogenes* has been limited. **Purpose:** To assess for the first time the role of the *L. monocytogenes* GAD system in oxidative stress tolerance.

**Methods:** We investigated the role of three GAD genes in three different *L. monocytogenes* strains (EGD-e, 10403S and LO28) in survival and growth against  $H_2O_2$ . We also looked at disk diffusion assays for each mutant.

**Results:** Our results suggest for the first time a major role for the genes comprising the intracellular GAD system of each strain in oxidative stress resistance and growth.

Significance: Our work suggests a major synergistic effect between the inhibition of the intracellular GAD system and the oxidative stress which could be exploited further to significantly increase inactivation of *L. monocytogenes*.

#### FRIDAY - 8.30 - 10.00

#### T8 Technical Session 8 – Risk Assessment

#### T8-01 Concept of Risk-Benefit Analysis Balancing the Impact of Cumulative Exposure to Pesticides Versus Beneficial Effect on Human Health Due to Fruit and Vegetable Intake

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Introduction: The balance of risk and benefit assessment procedures indicates the net health effect of food consumption patterns. **Purpose:** A proof of concept is presented to calculate the net health effect of cumulative exposure to a hazardous components group containing 94 pesticides and 32 other contaminants for the toxicological endpoint of liver steatosis.

**Methods:** A cumulative risk assessment for steatosis from fruit and vegetable intake was done using MCRA 8.1 software, which contains a fully-fledged probabilistic methodology for both acute and chronic cumulative risk assessment (Boon et al., 2015) for Belgian consumers. Lock et al., (2005) have derived a global risk index for the increased uptake of fruit and vegetables with respect to cardiovascular diseases and some of the more common types of cancer.

Results: It can be derived that when consuming an extra portion of 80 g/day, a reduction in the risk of ischaemic heart diseases is reduced by 10% for the age cohort 15 to 29 years and 30 to 44 years. The median exposure to the included CAG is 1.26 µg/kg body weight/ day (high consumers in the pessimistic model) and 1.47 µg/kg body weight/day (optimistic model). Even most extreme consumers in Belgium do not run a risk of adverse health effects, both in optimistic and pessimistic scenarios of residue exposure assessments. The net risk-benefit of higher consumption of fruit and vegetables showed a decrease in risk relative to the total consumption pattern. This means that the benefit of consuming fruits and vegetables overrules the hazard of pesticide exposure, at least for the included cumulative assessment group of 94 pesticides involved in liver steatosis. Significance: A new concept and methodology have been introduced to assess the net impact of an increase of fruit and vegetable consumption on the health of consumers. It uses the WHO global burden of disease estimates, and the results are extrapolated to the Belgian population as a case study.

## T8-02 Critical Analysis of QMRA for Meats: What Do We Do Next?

VINCENT TESSON<sup>1</sup>, Michel Federighi<sup>1</sup>, Enda Cummins<sup>2</sup>, Sandrine Guillou<sup>1</sup>, Jeanne-Marie Membre<sup>1</sup> and Géraldine Boué<sup>1</sup>

<sup>1</sup>SECALIM, INRA/Oniris, Nantes, France, <sup>2</sup>UCD School of Biosystems and Food Engineering, Dublin, Ireland

Introduction: Each year in Europe, food consumption is associated with 23 million foodborne illnesses, including more than 5,000 deaths. Meat is one of the main sources of these diseases due to contamination by pathogenic bacteria during the farm-to-fork chain. To prevent these outbreaks, decisionmaking processes in food safety are based on Quantitative Microbiological Risk Assessment (QMRA). Within this context, and with several meat safety crises, the development of QMRAs has expanded in recent decades.

**Purpose:** The aim of the study was to carry out a critical analysis of existing QMRAs of meat and foresee future challenges. **Methods:** A critical analysis of QMRAs was performed for the three most consumed types of meat (beef, pork, and poultry) in order to give an overview of existing models. They were reviewed for required inputs and obtained outputs as well as their respective scopes to highlight steps of the farm-to-fork chain that are considered in each model (e.g., production, processing, distribution, retail and consumer phases).

**Results:** At the request of public health authorities and/of food safety agencies, QMRAs models were historically developed according to specific public health concerns related to meats consumption in different countries: Argentina, Belgium, Canada, China, Chile, Denmark, France, Ireland, Netherlands, etc. More specifically, risk assessments in beef, pork and poultry meat sectors were carried out for *Salmonella* spp., *Escherichia coli* O157:H7 and *Campylobacter* spp. Future challenges identified were (i) to consider the whole farm-to-fork continuum because most of QMRAs are focused on only one or few stages; (ii) centralise data collection processes; and (iii) harmonise models developed in order to facilitate their use and reuse in other contexts.

**Significance:** The present critical analysis can be considered as a robust basis to start a new QMRA of meats and will help the scientific community and food safety authorities to identify specific needs in terms of monitoring and research.

#### T8-03\* Modelling the Risk of Contamination of Lettuce with *Escherichia coli* O157:H7 from Field to Consumption in Australia

#### HAYRIYE BOZKURT

The University of Sydney, Sydney, Australia

Introduction: Leafy vegetables are an essential component of a healthy diet; however, they have been implicated in the majority of food safety incidents in Australia. Since there are no legislative requirements to be regulated for leafy greens growers, development and application of quantitative risk assessment models have been recognized as a strong tool to identify and minimize potential risks associated with foodborne pathogens.

**Purpose:** The objectives of this study were to develop a quantitative microbial risk assessment model to evaluate the public health risks associated with consumption of lettuce contaminated with *Escherichia coli* O157:H7 in Australia, and to evaluate the impact of irrigation water and manure quality on public health risks.

Methods: The supply chain of lettuce was modeled from in field production until consumption at home. The developed model was simulated using Latin hypercube sampling for 100,000 iterations to estimate the number of illnesses due to consumption of lettuce in Australia by using @RISK software. Sensitivity analyses were performed to capture the effect of uncertainty and variability of the different parameters used in the model on the predicted risk of illness. Results: The QMRA model predicted a mean of 158 illness cases per year in Australia. The quality of irrigation water quality is an important risk factor for preharvest contamination. The manure quantity applied, the decline in manure, the number of rain events, the time interval between manure application and planting, and the aggregation parameter were identified as factors that affect the probability of contamination. Sensitivity analyses indicated that retail and home storage temperature, initial microbial concentration, and consumption time were the most important postharvest factors affecting the predicted number of illness cases.

**Significance:** This study provides a scientific foundation for risk managers and public health professionals to assess potential risk reduction strategies in Australia.

#### T8-04\* Cross-contamination Risk Factors in Domestic Chicken-handling Practices among Consumers in Five European Countries in the Transdisciplinary SafeConsume Project

**PIERRINE DIDIER**<sup>1</sup>, Christophe Nguyen-The<sup>2</sup>, Isabelle Maître<sup>3</sup>, Monica Truninger<sup>4</sup>, Silje Elisabeth Skuland<sup>5</sup>, Helene Maria Fiane Teigen<sup>5</sup>, Anca Ioana Nicolau<sup>6</sup>, Augustin Octavian Mihalache<sup>6</sup>, Loredana Dumitrascu<sup>6</sup>, Mike Foden<sup>7</sup> and Lydia Martens<sup>7</sup>

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Introduction: Nearly 40% of foodborne outbreaks in the EU are attributable to improper food safety practices by consumers. The EU-Horizon2020 funded project SafeConsume studied domestic raw-chicken handling practices in relation to cross-contamination risks. **Purpose:** The study aims to identify and understand cross-contamination factors from chicken handling in the domestic environment by a transdisciplinary and comparative analysis in five European countries.

**Methods:** Seventy-five participants from France, Norway, Portugal, Romania and the UK were observed and interviewed in their daily lives as they bought, transported, cooked and stored chicken and vegetables. An original methodology to analyze video with ObserverXT software was developed to identify risky practices. Verbatim analyses of consumers' discourses helped us understand reasons for these practices; microbiological analyses correlate observations on cross-contamination with the potential spreading of pathogens around kitchens.

**Results:** Critical points while preparing poultry came mostly from hands, utensils and serving dishes. Regarding hands, for instance, only a third of participants washed hands after handling raw chicken

(100% in Norway, zero to 30% in other countries). Whenever they washed hands, a perception of dirtiness was associated with a feeling of greasiness on hands rather than with safety concerns (except in Norway). French consumers, for example, perceived touching the bin, handling the cat and blowing their nose as risky behaviors but none mentioned handling raw poultry, compared to a majority of Norwegian participants. The risk, however, depends on the prevalence of pathogens which varied among countries (highest in Romania and lowest in Norway).

Significance: This study shows differences between countries, resulting in safe or risky practices. These results highlight a lack of knowledge of microbiological hazards of raw meat in most of the countries observed. These critical food-handling practices can result from habits, lack of knowledge about risks factors as framed by microbiologists and health professionals but also kitchen equipment. They also point out the construction and perception of safety practices developed by consumers.

#### T8-05 Introducing Aginfra+ Based Virtual Research Supporting Scientific Collaboration and Knowledge Exchange in the Food Safety Domain

**MATTHIAS FILTER**, Lars Valentin, Ahmad Swaid, Thomas Schüler and Taras Günther

German Federal Institute for Risk Assessment, Berlin, Germany

Introduction: The AGINFRA+ project aims at providing innovative e-infrastructure services and applications to support the needs of scientific communities in the agriculture and food domains. It demonstrates how these scientific communities can exploit the potential of virtual research environments (VRE) customized by AGINFRA+ in their specific research domain.

**Purpose:** This presentation will introduce the conceptual foundation and existing VREs pilots developed by the AGINFRA+ project. It will specifically illustrate the large number of services and features that the corresponding pilot VREs offer to the RAKIP community and the DEMETER community.

Methods: VREs are web-based, distributed, open-science compliant working environments for scientists and practitioners. Within the AGINFRA+ project, eight partners collaborated on the extension of existing e-infrastructures and services to the needs of specific research communities. In particular, AGINFRA+ aggregates resources from "generalist" service providers (e.g., D4Science, EGI) as well as from community-specific ones (e.g., AGROVOC, RAKIP model repository)

**Results:** Within the AGINFRA+ project two dedicated VRE pilots were carried out to illustrate the VRE benefits to researcher and governmental agencies in the multidisciplinary field of food safety risk assessment (RAKIP VRE) and emerging risk identification (DEMETER VRE). The DEMETER VRE allows to share and execute data-analysis pipelines in a protected cloud-based environment and contains innovative data visualization and knowledge representation features. The RAKIP VRE exploits the existing Data Catalogue technology to FAIR-ify risk assessment models. This VRE also supports the harmonized information exchange format "FSK-ML" and allows to perform user-driven computational-intensive simulations online. An important new feature is the support for KNIME and Galaxy workflows.

Significance: The implementation and customization of two community-centric VREs for the food safety domain illustrate the great potential of this open e-Infrastructure. As AGINFRA+ services will become part of the European Open Science Cloud (EOSC) further improvement and long-term maintenance is likely.

Acknowledgment: This work received funding from the EU-H2020 programme under AGINFRA PLUS project (grant agreement No. 731001).

#### T8-06 Improving Raw Milk Food Safety Management by a Quantitative Risk Assessment Approach

## VALÉRIE MICHEL<sup>1</sup>, Janushan Christy<sup>1</sup> and Fanny Tenenhaus<sup>2</sup>

<sup>1</sup>ACTALIA, La Roche sur Foron, France, <sup>2</sup>Cniel, Paris, France

Introduction: Raw milk cheeses may contain pathogen contamination and lead to outbreaks. To reduce them, food business operators use different control measures, such as good hygiene practices based on hazard analysis and critical control point principles, and sampling plans from farm to fork. However, they often have difficulty evaluating the impact of these measures. **Purpose:** Firstly, the aim of the work conducted was to develop microbial quantitative risk assessment approaches in different cheese types. Then, deployment of the approach was conducted with the final objective being to enable cheese business operators to generate customized results. **Methods:** By simulating the behaviour of the targeted food pathogens all along the cheese production chain, the impact of different food control measures can be assessed. Such a predictive microbial model was developed for three types of raw milk cheeses (soft cheeses, semi-hard cheeses, blue-veined cheeses) and different microbial hazards (VTEC and *Salmonella*). Uncertainty and variability were also taken into account.

**Results:** Results are presented in a risk versus benefit approach comparing the loss of batches that were rejected (noncompliant batches) to the reduction of illness. A specific software iAQR was also developed to allow food business operators to use the developed models and customize input data in these QMRA models. For example, for soft cheese and VTEC, a combination of raw milk sorting based on *E. coli* levels and of the definition of microbial criteria (e.g., appropriate sampling plans) can lead to a significant reduction of hemolytic uremic syndrome outbreaks.

**Significance:** The approaches developed and the generated models are now displayed and used by cheese operators. It gives them a better understanding and knowledge of the impact of their everyday practices. It also enables them to base their management options on a scientific and transparent approach.

#### FRIDAY — 10.30 – 12.00

#### T9 Technical Session 9 – Applied Laboratory Methods, Microbial Food Spoilage and Pathogens

#### T9-01\* Reagent-free Detection of Silver lons in Tap Water Using Square Wave Voltammetry and Local pH Control

 $\mbox{LUIZA WASIEWSKA}^1,$  Geraldine  $\mbox{Duffy}^2,$  Kaye  $\mbox{Burgess}^3$  and Alan O'Riordan^1

<sup>1</sup>Tyndall National Institute, Cork City, Ireland, <sup>2</sup>Teagasc Food Research Centre, Ashtown, Dublin, Ireland, <sup>3</sup>Teagasc, Dublin, Ireland

Introduction: Silver is becoming more ubiquitous in a wide variety of products which has resulted in its release into the environment, particularly into the water. Depending on its chemical form, silver can be toxic, with silver ions being the most toxic form. Based on WHO (WHO, 2017), concentrations in excess of 0.1 mg/L (around one  $\mu$ M) may contribute to a disease state.

 $\label{eq:purpose:Develop} \begin{array}{l} \mbox{Purpose: Develop an electrochemical sensor for silver ions detection} \\ \mbox{able to detect at least one } \mu M \mbox{ of silver in tap water samples. In} \\ \mbox{addition, explore the use of local } pH \mbox{ control at new sensors to} \\ \mbox{eliminate the requirement for addition of reagents typically used to} \\ \mbox{lower solution } pH. \end{array}$ 

**Methods:** Using the interdigitated gold microband electrodes (Dawson et al., 2014), silver ions were first reduced by applying the negative potential at the working electrode and afterward oxidized using square wave voltammetry resulting in a peak which height was related to silver concentration. During the procedure, a constant oxidative potential was applied to the protonator electrode causing water decomposition with hydrogen ions being released. By varying this potential, the local pH surrounding an electrode could be selected between pH two and seven.

**Results:** The optimal conditions were first established in a sodium acetate solution. Afterward, the calibration was done in tap water. Using two minutes as a reduction time, the linear region was found between 0.25  $\mu$ M and two  $\mu$ M. The limit of detection was, therefore sufficient to detect one  $\mu$ M of silver in line with WHO.

**Significance:** Using the presented technique for silver detection, the toxic silver ions which may contaminate drinking water, can be detected in real samples in less than three minutes.

References: Karen Dawson, Amélie Wahl, Sean Barry, Colm Barrett, Nicolas Sassiat, Aidan J Quinn, Alan O'Riordan, Electrochimica Acta, 115, 239–246, 2014

Guidelines for drinking-water quality: fourth edition. Geneva: WHO; 2017.

#### T9-02 A Real-time Cell Analysis-based Assay as an Innovative Approach for Thermal Inactivation Studies of Hepatitis A Virus

AUDREY FRAISSE, Catherine Hennechart-Collette, Laurent Guillier, Sylvie Perelle and Sandra Martin-Latil ANSES, Laboratory for Food Safety, University of Paris-Est, Maisons-Alfort, France

Introduction: Hepatitis A virus (HAV) is mainly transmitted through the fecal-oral route, either directly by close contact with infected people or indirectly through ingestion of contaminated food and water. Controlling transmission through their removal from drinking water and foodstuffs is an important challenge in reducing the burden of viral foodborne illnesses. Purpose: The aim of the study was i) to evaluate the previously developed real-time cell analysis (RTCA) assay for thermal inactivation studies and ii) to compare inactivation kinetic curves obtained with other available detection methods (plaque forming-unit (PFU) assay) and molecular-based methods (dye-combined method (EMA-RT-qPCR) and RT-qPCR).

Methods: The impedance of FRhK-4 cells uninfected or infected with HAV suspensions that were either heat-treated (37, 50, 56, 65, 72 or 80°C) or not was measured in real-time using the xCELLigence system. In uninfected cells, CI remained constant whereas a CI drop appeared in HAV-infected cells according to the temperature and time treatment. The times to reach 50% CI decrease (TCI<sub>50</sub>) were determined to quantify the remained infectious HAV following heat-treatment from a standard curve established between the concentration of HAV and the  $TCI_{50}$ . Increasing the temperature and/ or the duration of heat treatment at constant temperature led to a delay in CI decrease until there were no longer any decreases in CI. Results: Inactivation kinetic curves obtained by using the RTCA assay followed the same profile than the ones obtained with the traditional PFU assay regardless of the heat treatment. On the contrary, kinetics profiles obtained by using the EMA-RT-qPCR were close to the ones obtained with both cell-based methods only for high temperatures (≥56°C).

Significance: As the RTCA-based titration method presents many advantages in comparison with the traditional PFU method (less fastidious, giving results faster, less expensive), it could be helpful to validate technological treatments used in food industries for better viral risk management.

#### T9-03 Characterization of Meat and Seafood Spoilage Mechanisms by *Brochothrix thermosphacta*

Nassima Illikoud<sup>1</sup>, Marie-France Pilet<sup>1</sup>, **EMMANUEL** JAFFRÈS<sup>2</sup> and Monique Zagorec<sup>1</sup>

<sup>1</sup>SECALIM, INRA, Oniris, Université Bretagne Loire, 44307, Nantes, France, <sup>2</sup>SECALIM, INRA, Oniris, Université Bretagne Loire, Nantes, France

**Introduction:** Food microbial spoilage causes considerable economic losses. *Brochothrix thermosphacta* is one of the main spoilage bacteria of meat and seafood products. It produces various metabolites responsible for off-odors, which seem to depend on strains and food type.

**Purpose:** To better understand the *B. thermosphacta* spoilage mechanisms, we assessed the genetic and phenotypic diversity among a collection of 161 strains from various origins (seafood, meat, dairy products, and environment).

meat, dairy products, and environment). **Methods:** MALDI-TOF MS, PFGE, rep-PCR, and acetoin/diacetyl production were used to classify strains into distinct groups. Four strains representing the species diversity were analyzed by comparative genomics and the volatilomes and transcriptomes of two of these strains inoculated in two model matrices (meat and shrimp juices) were compared.

Results: The strain discrimination revealed a significant diversity among the 161 strains. However, the intraspecies diversity was not related to the environment strains were isolated from. The comparative genomic analysis of the four strains representative of intraspecies diversity revealed a similar genomic content. The only differences reside in phage and plasmid content and on surface components that may contribute to their ecological adaptation. Base substitutions were observed in or upstream from genes responsible for the production of spoilage molecules. The transcriptomic study showed that the genes differentially expressed in seafood or meat matrices encoded functions important for growth and survival in these specific niches. In addition, the expression of some functions involved in spoilage was strain dependent. The volatile analysis showed that B. thermosphacta produced different molecules responsible for off-odors that vary depending on the growth matrix. Therefore, we showed a matrix effect on the strain fitness and a strain effect on the spoilage potential.

Significance: This work is a complete functional analysis which significantly improves the knowledge on the *B. thermosphacta* diversity and their food spoilage mechanisms.

#### T9-04 Estimation of Microbial Spoilage of Ready-to-Eat Baby Spinach Using Fourier Transform Infrared Spectroscopy

Evanthia Manthou<sup>1</sup>, Anastasia Bakalaki<sup>2</sup>, Alexandra Lianou<sup>1</sup>, Panagiotis Tsakanikas<sup>1</sup>, Efstathios Panagou<sup>1</sup> and **GEORGE-JOHN NYCHAS**<sup>3</sup>

<sup>1</sup>Agricultural University of Athens, Department of Food Science and Human Nutrition, Laboratory of Microbiology and Biotechnology of Foods, Athens, Greece, <sup>2</sup>AUA, Athens, Greece, <sup>3</sup>Laboratory of Microbiology and Biotechnology of Foods, Department of Food Science and Human Nutrition, Agricultural University of Athens, Athens, Greece Introduction: Minimally processed vegetables are food commodities of increasing popularity among consumers, meeting the current lifestyle preferences for nutritious and easy-to-prepare food. However, the freshness and quality of fresh produce is still an issue of concern, due to its fast evolving physicochemical and microbial deterioration in the food supply chain.

Purpose: The aim of this study was the evaluation of the microbial spoilage of RTE baby spinach stored under different temperature conditions using Fourier transform infrared spectroscopy (FTIR). Methods: Packages of fresh baby spinach were stored aerobically under isothermal (four, eight, and 12°C) and dynamic temperature conditions for a maximum time period of 10 days. At regular time intervals, duplicate samples (originating from different packages) were analyzed for the determination of the total mesophilic microbial populations, using conventional microbiological approaches, as well as FTIR spectroscopy. Two independent experimental replicates were conducted. Partial least squares regression (PLSR) was applied for establishing the correlation between spectral data and microbial counts. Due to high variability among replicate samples, the average values (spectra and microbial counts) of the duplicate samples were used for model development. The calibration and validation (i.e., prediction) of the PLSR model was performed with the data collected during storage of baby spinach at isothermal (80 samples) and dynamic temperature (27 samples) conditions, respectively. Results: The values of the slope, the correlation coefficient between predicted and measured microbial counts and the root mean square error (RMSE) of prediction were 0.89, 0.77 and 0.34, respectively. Significance: FTIR spectroscopy appears to be a promising rapid and non-destructive tool for the assessment of the microbial spoilage of ready-to-eat baby spinach.

This work has been supported by the project "PhasmaFOOD".

#### T9-05 The Effect of Carbon Dioxide as a Climatic Parameter on Microbial Food Contaminants and Selective Isogenic Mutants

SHOLEEM GRIFFIN<sup>1</sup>, Sanja Ivanovic<sup>1</sup>, Christina Chatzitzika<sup>1</sup>, Jan F. M. Van Impe<sup>2</sup> and Vasilis P. Valdramidis<sup>1</sup> <sup>1</sup>Department of Food Sciences and Nutrition, Faculty of Health Sciences, University of Malta, Msida, Malta, <sup>2</sup>Department of Chemical Engineering, BioTeC - Chemical & Biochemical Process Technology & Control, Katholieke Universiteit Leuven, Ghent, Belgium

Introduction: Increasing carbon dioxide levels are associated with a rise in global temperatures. Accumulation of CO<sub>2</sub> in the atmosphere, known as the greenhouse effect, was essential for driving life and keeping the temperature habitable on Earth. However, if left uncontrolled, increasing  $\mathrm{CO}_{_2}$  will cause irreversible changes to our planet. The enhanced greenhouse effect is causing excessive heat retention in the atmosphere, raising global temperatures, and affecting the growth of crops and the microflora associated with them. Purpose: This study aims (i) to evaluate the effect of increasing CO<sub>2</sub> concentration on bacterial and fungal responses and (ii) assess its mechanism of action with the use of knock-out bacterial mutants. Methods: Wild-type Escherichia coli and its CO<sub>2</sub> metabolism or stress response mutants dnaK, gadB, gadC, gadD and rpoS, Salmonella enterica, and Penicillium expansium were cultured in a plate reader until stationary phase at optimal temperature and  $CO_2$  levels of atmospheric, 2.5, 5 or 10% . The optical density was recorded at 600 nm and used to calculate the area under the curve to assess their growth

**Results:** Growth of *E. coli* strains increased with increasing CO<sub>2</sub> concentration. The *dnaK* mutant of *E. coli* had significantly lower growth at 2.5% CO<sub>2</sub> concentration (*P*<0.05) compared to the wild-type. All other *E. coli* mutants had significantly higher growth than the wild-type strain at 10% CO<sub>2</sub> concentration (*P*<0.001), increasing by 1.5 to twofold. *Salmonella enterica* growth also increased with CO<sub>2</sub> concentration (*P*<0.001) increasing by 0.5-fold. The growth of *P. expansium* decreased upon an increase in CO<sub>2</sub> concentration, dropping by twofold (*P*<0.001).

**Significance:** Our results show that a change in  $CO_2$  effects the growth of microorganisms. Microbial growth can increase if appropriate mutations occur in genes related to  $CO_2$  metabolism or stress response allowing these microorganisms to adapt to increasing  $CO_2$ .

#### T9-06\* Cardinal Parameter Meta-Regression Models Describing *Listeria monocytogenes* Growth in Broth

**BEATRIZ NUNES SILVA**<sup>1</sup>, Vasco A. P. Cadavez<sup>2</sup>, José A. Teixeira<sup>1</sup>, Mariem Ellouze<sup>3</sup> and Ursula A. Gonzales-Barron<sup>4</sup> <sup>1</sup>*CEB* - *Centre of Biological Engineering, University of Minho, Braga, Portugal,* <sup>2</sup>*Polytechnic Institute of Bragança, Bragança, Portugal,* <sup>3</sup>*Nestlé Research Centre, Lausanne, Switzerland,* <sup>4</sup>*School of Agriculture, Polytechnic Institute of Braganza, Braganza, Portugal* 

**Introduction:** Since *Listeria monocytogenes* is one of the most virulent foodborne pathogens, substantial research has been devoted to estimate its growth rate (GR) under different conditions of temperature, pH and water activity (a<sub>w</sub>).

**Purpose:** Thus, this study's objective was to extract all published findings on *L. monocytogenes* growth in broth, and unify them by constructing meta-regression models based on cardinal [temperature], [temperature][pH] and [temperature][pH][a<sub>w</sub>] secondary models.

Methods: Suitable primary studies were identified through an exhaustive literature search. From each of the 49 studies considered appropriate for inclusion, the following data were extracted: GR, temperature, pH, a,, type of broth, reading type (CFU or optical density) and inoculum concentration. The meta-analytical dataset comprised 2009 GR measures. Weights were assigned to studies according to the number of time points sampled to calculate GR. Results: The cardinal [temperature][pH] meta-regression suggested that higher inoculum levels (2.0 to 6.5 log CFU/g) tend to produce lower estimates (*P*<.0001) of  $T_{max}$ ,  $T_{opt}$ ,  $pH_{max}$  and  $pH_{opt}$ , although it does not seem to affect (*P*=0.909) the optimum GR ( $\mu_{opt}$ ). The cardinal [temperature] meta-regression demonstrated that reading type was not as important a source of between-study variability (R<sup>2</sup>=1.0 to 7.5%) as type of broth, which was responsible for 25 to 70% of the heterogeneity ( $\tau^2$ =11.22). Thus, the most parsimonious cardinal [temperature][pH][a, ] meta-regression model was built upon a nested random-effects structure of studies within type of broth, which produced the following cardinal estimates:  $T_{min}=-1.273^{\circ}C$ (SE=0.178),  $T_{out}=37.24^{\circ}C$  (SE=0.789),  $T_{max}=45.12^{\circ}C$  (SE=0.013), pH<sub>min</sub>=4.298 (SE=0.005), pH<sub>opt</sub>=7.090 (SE=0.073), pH<sub>max</sub>=9.510 (SE=0.117),  $a_{w min}=0.829$  (SE=0.005) and  $a_{w opt}=0.995$  (SE=0.001). The  $\mu_{opt}$  was affected by reading type (R<sup>2</sup>=15%), being higher for OD (1.224cm/s) SE=0.400 then for CELI (0.067 hand): SE=0.004) (1.324 log/h; SE=0.100) than for CFU (0.967 log/h; SE=0.094). Significance: By integrating the outcomes from numerous L. monocytogenes growth experiments, the cardinal parameters for temperature, pH and a, obtained through meta-analysis represent very accurate estimates that can be used as reference values.



# **POSTER ABSTRACTS**

24–26 April 2019 – Nantes, France

# **POSTER ABSTRACTS**

\* Student Award Competitor

#### WEDNESDAY, 24 April - 10.30 - 16.00

Poster Session 1 – Applied Laboratory Methods; Communication Outreach and Education; Epidemiology; General Microbiology; Microbial Food Spoilage; Novel Laboratory Methods; Produce; Risk Assessment

#### P1-01 Microbiological Certified Reference Materials: LENTICULE Discs and Vitroids

**MATHIAS BAEUMLISBERGER**<sup>1</sup>, Barbara Gerten<sup>2</sup> and Markus Obkircher<sup>3</sup>

<sup>1</sup>Merck KGaA, Sigma-Aldrich Production GmbH, Buchs, Switzerland, <sup>2</sup>Merck KGaA, Darmstadt, Germany, <sup>3</sup>Merck KGaA,Sigma-Aldrich Production GmbH, Buchs, Switzerland

Introduction: The use of freeze-dried control strains in order to ensure an acceptable quality of food, water, environmental microbiological laboratory samples, or prepared culture media according to EN ISO 11133:2014+Amd1:2018 is very time-consuming and expensive. Thus, ready to use microbiological certified reference materials (CRMs) should reduce costs, hands-on time, and possible error sources.

**Purpose:** To determine cost and time savings, a comparison between both reference materials of needed hands-on time for media performance testing was conducted and the results were evaluated to determine whether the use of CRMs is worthwhile for laboratories in their daily work.

**Methods:** Four hands-on time comparisons for three different media performance testing methods (pour plate, membrane filtration and spread plate method) were carried out using typical freeze-dried microbiology control strains and ready-to-use microbiological CRMs. This study included the time choosing organisms, resuscitation, and steps to prepare the test dilutions.

**Results:** The results of the comparison show that hands-on time can be reduced from an average of 65 minutes over three days to only six minutes per control strain by incorporating ready-to-use microbiological CRMs. These consist of pure bacterial and fungal cultures (provided under license by NCTC//NCPF and CECT, respectively) in a solid water-soluble matrix, which preserves them in a viable state. Time savings are achieved by reducing the time spent preparing stock cultures, recovering banked stocks, and performing pre-enrichment steps. The elimination of the pre-enrichment steps additionally decreases the probability of contamination. **Significance:** LENTICULE discs and Vitroids provide laboratories with tools to save significant time and to reduce expenditures.

#### P1-02 Propidium Monoazide qPCR Quantification of Viable *Brochothrix thermosphacta* in Coldsmoked Salmon

AGNÈS BOUJU-ALBERT, Sabrina Saltaji, Xavier Dousset, Hervé Prévost and Emmanuel Jaffrès

UMR 1014 Secalim, UBL, INRA, Oniris, Nantes, France

Introduction: Microbial food spoilage is responsible for significant economic losses. *Brochothrix thermosphacta* is one of the major bacteria involved in the spoilage of meat and seafood. This bacterium is frequently isolated from the processing plant and from raw materials to end product. Thus *B. thermosphacta* needs to be specifically quantified all along the cold-smoked salmon process. **Purpose:** The aim of this study was to develop a rapid and accurate propidium monoazide (PMA) qPCR method to quantify viable and viable but nonculturable (VBNC) *B. thermosphacta* cells in cold-smoked salmon and associated processing plants.

**Methods:** A specific qPCR was developed targeting the single-copy *rpoC* gene, combined with treatment with the cell viability dye PMA and an improved version, PMAxx. The PMA-qPCR method was optimized on heated and unheated cells in brain heart infusion broth and smoked-salmon juice, then validated on naturally contaminated cold-smoked salmon samples.

**Results:** The average efficiency of the qPCR on 100% viable *B.* thermosphacta cells was 90.62% (*n*=6) with very good correlation ( $r^2$ =0.996) and a standard deviation of 6.53. In a 100% viable cell population, no significant difference was observed between cells treated with PMA or PMAxx and untreated cells. In contrast, in a population of 0% viable cells, the results showed a significant difference (*P*<0.001) between cells treated with PMA or PMAxx and untreated cells, showing the ability of both PMA dyes to considerably suppress DNA amplification of dead cells.

**Significance:** This approach represents a novel, rapid, and specific assay to quantify viable *B. thermosphacta* cells, improving the detection of a significant spoilage microorganism in the food factory environment and the final product.

#### P1-03 AOAC-RI PTM and NF Validation of the Thermo Scientific Suretect Salmonella Species PCR Assay Using the Quantstudio 5 PCR Instrument

Jessica Williams<sup>1</sup>, Benjamin Bastin<sup>2</sup>, Ana-Maria Leonte<sup>1</sup>, Maryse Rannou<sup>3</sup>, Muriel Bernard<sup>3</sup>, Liz Harrison<sup>1</sup> and **DAVID CRABTREE**<sup>1</sup>

<sup>1</sup>Thermo Fisher Scientific, Basingstoke, United Kingdom, <sup>2</sup>Q Laboratories, Inc., Cincinnati, OH, <sup>3</sup>ADRIA Food Technology Institute, Quimper, France

Introduction: The AOAC-RI PTM program and NF Validation by AFNOR certification have previously validated the Thermo Scientific SureTect Salmonella Species PCR assay for the detection of Salmonella species from all human food products and environmental samples using the Applied Biosystems 7500 Fast PCR Instrument with Applied Biosystems RapidFinder Express version 2.0 software. Purpose: To extend the validated claims to include the use of the SureTect Salmonella Species PCR Assay on the Applied Biosystems QuantStudio 5 Real-Time PCR Instrument with Thermo Scientific RapidFinder Analysis version 1.0 Software (the alternative method). Methods: The validation studies were conducted according to the AOAC-RI PTM, NF Validation, and ISO 16140-2:2016 guidelines. For the alternative method, all samples underwent an enrichment step followed by direct lysis. Following direct lysis, PCR was run and results were automatically interpreted by the software. The reference method was conducted according to EN ISO 6579-1:2017. Results: A total of 180 (AOAC-RI PTM) and 456 (NF Validation) food and environmental samples were tested using the alternative and EN ISO 6579-1:2017. The alternative method demonstrated equivalent performance for all human food and environment samples analysed to the EN ISO 6579-1:2017 reference method during the AOAC-RI PTM and NF Validation by AFNOR certification studies. Significance: The alternative method proved to be a suitable substitute for the EN ISO 6579-1:2017 reference method for Salmonella species detection.

#### P1-04 NF Validation of the Thermo Scientific SureTect Cronobacter Species PCR Assay Using the Applied Biosystems Quantstudio 5 PCR Instrument

Ana-Maria Leonte<sup>1</sup>, Liz Harrison<sup>1</sup>, Maryse Rannou<sup>2</sup>, Muriel Bernard<sup>2</sup> and **DAVID CRABTREE**<sup>1</sup> <sup>1</sup>Thermo Fisher Scientific, Basingstoke, United Kingdom,

<sup>2</sup>ADRIA Food Technology Institute, Quimper, France

Introduction: NF Validation by AFNOR certification has previously validated the Thermo Scientific SureTect *Cronobacter* species PCR Assay for the detection of *Cronobacter* species in powdered infant formula samples (PIF) (10g and 300g) and production environmental samples using the Applied Biosystems 7500 Fast with Applied Biosystems RapidFinder Express 2.0 software in accordance with EN ISO 16140-2:2016.

**Purpose:** ADRIA Développement laboratories conducted a study to extend the validated claims to include the use of the Applied Biosystems QuantStudio 5 Real-Time PCR Instrument using the RapidFinder Analysis Software version 1.0.

Methods: Matrices of 10 g PIF with and without probiotics, 300 g PIF with and without probiotics, and production environment samples were tested during this extension study. Samples were enriched in buffered peptone water (BPW), supplemented with 6 mg/L of vancomycin depending on the sample type and size. Enrichments were incubated for 16 to 24 hours (depending on sample type and size) at 37±1°C before the lysates were prepared and 20  $\mu L$  aliquots of the lysates were transferred to SureTect PCR Tubes containing SureTect Cronobacter spp. PCR tablets. The tubes were then transferred to the QuantStudio 5 Instrument for processing. The alternative method was tested in comparison to EN ISO 22964:2017. Results: A total of 208 milk powder and environmental samples were tested using the alternative and EN ISO 22964:2017 methods. Fourteen positive deviations were observed and confirmed with culture methods. The sensitivity of the alternative method was demonstrated to be 91.7% with relative trueness of 89.4% and a false positive ratio of 6.3%. The alternative method demonstrated equivalent performance for all samples analysed in comparison to the EN ISO 22964:2017 reference method, during NF Validation by AFNOR certification studies

Significance: The alternative method proved to be a suitable substitute for the EN ISO 22964:2017 reference method for *Cronobacter* species detection.

#### P1-05 NF Validation of the Thermo Scientific Listeria Precis Enumeration Method in Accordance with EN ISO 16140-2:2016

Ana-Maria Leonte<sup>1</sup>, Francois Le Nestor<sup>2</sup>, Liz Harrison<sup>1</sup> and **DAVID CRABTREE**<sup>1</sup>

<sup>1</sup>Thermo Fisher Scientific, Basingstoke, United Kingdom, <sup>2</sup>Laboratoire Microsept, Le Lion d'Angers, France

**Introduction:** NF Validation by AFNOR certification has previously validated the Thermo Scientific<sup>™</sup> Listeria Precis<sup>™</sup> Enumeration method (alternative method) for the enumeration of *Listeria monocytogenes* from a broad range of foods and environmental samples.

**Purpose:** To assess performance in comparison to the ISO 11290-2:2017 reference method, and renew the validation in line with EN ISO 16140-2:2016.

**Methods:** The samples were enriched in Buffered Peptone Water (1-in-10 ratio) and serially diluted as described in ISO 6887:2017. The alternative method was compared to the EN ISO 11290-2:2017 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 within the EN ISO 16140-2:2016 acceptability limits for all six matrices/strain combinations. Inclusivity testing (50 *L. monocytogenes*) isolates were all successfully detected and exclusivity testing (31 non-target isolates) showed no cross-reaction. The ILS results from 10 laboratories were scatter plotted using reference method vs the alternative method; the data meets the Acceptability Limits (AL) for all levels of contamination, therefore is equivalent to 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.

#### P1-06 NF Validation of the Thermo Scientific Listeria Precis Detection Method in Accordance with EN ISO 16140-2:2016

Ana-Maria Leonte<sup>1</sup>, Maryse Rannou<sup>2</sup>, Muriel Bernard<sup>2</sup>, Liz Harrison<sup>1</sup> and **DAVID CRABTREE**<sup>1</sup>

<sup>1</sup>Thermo Fisher Scientific, Basingstoke, United Kingdom, <sup>2</sup>ADRIA Food Technology Institute, Quimper, France

**Introduction:** NF Validation by AFNOR certification has previously validated the Thermo Scientific Listeria Precis Detection method (alternative method) for the detection of *Listeria monocytogenes* from a broad range of foods and environmental samples.

**Purpose:** To assess performance in comparison to the EN ISO 11290-1/A1:2005 reference method and renew the validation in line with EN ISO 16140-2:2016.

Methods: The samples were enriched in Thermo Scientific ONE Broth-*Listeria* (1-in-10 ratio) and serially diluted as described in ISO 6887:2017. The alternative method was compared to the EN ISO 11290-1/A1:2005 method during the relative level of detection (RLOD), sensitivity study, inclusivity and exclusivity, and interlaboratory (ILS) studies.

**Results:** The RLOD study results were below the acceptability limit of 2.5 for an unpaired study design for (RLOD of 0.851 overall for all six

matrices/strain combinations). 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 each category individually and for all categories overall, as described in EN ISO 16140-2:2016. Inclusivity testing (50 *Listeria monocytogenes*) isolates were all successfully detected and exclusivity testing (31 non-target isolates) showed no cross-reactions. The ILS results from 12 laboratories were analysed and the sensitivity, specificity of the alternative and reference method fulfilled the acceptability limits of EN ISO 16140-2:2016.

**Significance:** The Listeria Precis Detection method is equivalent to the EN ISO 11290-1/A1:2005 reference method for the detection of *Listeria monocytogenes* from a broad range of foods and environmental samples.

#### P1-07 Microval Validation of the Thermo Scientific Brilliance Staph 24 Enumeration Method in Accordance with ISO 16140-2:2016

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**Introduction:** MicroVal has previously certified the Thermo Scientific Brilliance Staph 24 Enumeration method as an alternative method for the enumeration of coagulase-positive *Staphylococcus* species from a broad range of foods.

Purpose: To assess performance in comparison to the EN ISO 6881-1:1999 DAM2L2017€ and renew the validation in line with EN ISO 16140-2:2016.

Methods: The alternative method 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. The samples were enriched in buffered peptone water (1-to-10 ratio) and serially diluted as described in ISO 6887:2017. Results: The relative trueness study (83 samples) results satisfied the requirements of EN ISO 16140-2:2016. The accuracy profile tolerance intervals were within the EN ISO 16140-2:2016 acceptability limits for all five matrix/strain combinations. Forty-nine out of 51 coagulase-positive Staphylococcus strains were successfully detected during the inclusivity testing; the remaining 2 strains were not detected by the alternative or the reference method. Exclusivity testing (35 non-target isolates) showed 2 cross-reactions from both the alternative method and the reference method and a further 3 cross reactions from the reference method only. The ILS results from 11 laboratories were scatter plotted using reference method vs the alternative method; the data meets the Acceptability Limits (AL) for all levels of contamination, and therefore is equivalent to the reference method

Significance: The 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.

#### P1-08 Naked-Eye Detection of *E. coli* O157 by Recombinase Polymerase Amplification and SYBR Green I

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**Introduction:** In 2016, 6,378 confirmed cases of Shiga toxinproducing *E. coli* were reported in Europe. Out of these, 38.6% corresponded to serotype C157. Reference methodology takes several days to complete, as it requires selective enrichment, molecular detection of the bacteria, and plate confirmation. Additionally, expensive equipment is needed as well as trained personnel.

**Purpose:** Evaluate a novel methodology combining recombinase polymerase amplification (RPA) and SYBR Green I for the naked-eye detection of *E. coli* O157 in one working day.

**Methods:** Ground meat was selected as the reference food matrix for spiking experiments. After a short enrichment in m-tryptic soy broth, the supernatant was recovered and the DNA extracted with Chelex 100/proteinase K followed by thermal lysis. The detection of *E. coli* O157 was performed targeting the *rfbE* gene, adapting qPCR primers to RPA. After 20 min of amplification SYBR Green, I was added, and the results were assessed by naked eye observation under a UV lamp. The overall method was completed in less than five hours. The results were confirmed by qPCR and also by plating on cefixime tellurite sorbitol MacConkey agar.

**Results:** The described methodology obtained a LOD<sub>95</sub> of 19 CFU per 25 g, in one working day. The relative sensitivity, specificity, accuracy were higher than 90%, and the Cohen's k obtained a value

of 0.81, indicating that the results obtained were in "almost complete concordance" with the expected ones by qPCR. Discrepancies were observed with the plating approach due to high levels of interfering microorganisms.

**Significance:** The described methodology allowed to obtain reliable results in one working day without the need for complex and expensive equipment.

#### P1-09 Involvement of Staphylococcal Enterotoxins Types SEG, SEH and SEI in Foodborne Outbreaks: Myth or Reality?

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Introduction: Staphylococcal enterotoxins (SEs) are the leading cause of food poisoning outbreaks (FPO) of bacterial origin in Europe (380 outbreaks in 2017 according to EFSA data). However, most of cases were declared "suspected" due to a lack of suitable detection methods. Indeed, among 26 SEs described in the literature, only five (SEA to SEE) are detected routinely according to the European official method. Some FPO have shown a typical SE symptomatology but no enterotoxin of types SEA to SEE could be detected in food. Actually, epidemiological data highlight the frequent presence of *seg*, *seh* or *sei* genes, but no validated method is currently available to detect the related enterotoxins (SEG, SEH and SEI), which is a risk for consumers.

**Purpose**: An immunoassay test for the detection of SEG, SEH and SEI toxin types has been developed, validated and used for analysis of several samples from FPO.

**Methods**: A validation step was performed on about 20 *Staphylococcus aureus* strains carrying *seg*, *seh* or *sei* genes and on artificially contaminated foods. Then, more than 100 foods from FPO and official controls were analyzed.

**Results**: Sensitivity and specificity were evaluated at >95% in food matrices. SEG, SEH and SEI toxins were detected in cultures of the tested strains and in foods suspected in FPOs. For example, toxins SEG and SEI were detected in a cheese responsible for an FPO (50 illnesses, nine hospitalizations) while the official method gave a negative result.

Significance: An enlargement of the analytical scope of the official method to other toxins, such as SEG, SEH and SEI, is strongly recommended. Thus, public authorities could improve characterization of the SE hazard and FPO investigation and increase the number of "strong evidence" outbreaks. Moreover, this enlargement is already covered by the EN ISO 19020 Standard and the next amendment of the EC Regulation.

#### P1-10 Reducing the Number of Presumptive Positive Results in Environmental and Food Samples Using PR*Eraser* BAC*Gene*

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Introduction: Real-time PCR has several advantages over the traditional cultural detection of pathogens, including shorter time to result, less hands-on time and no detection of background flora. However, due to their high sensitivity, PCR methods detect DNA from living as well as dead pathogens. This can be problematic in food production, as presumptive positives have to be culture confirmed leading to costly delays when non-confirmable presumptive positives occur.

**Purpose:** The aim of this study was to evaluate the ability of the newly developed sample treatment to remove free DNA from samples and to assess potential adverse effects of the treatment on the viability and detection of pathogens.

**Methods:** Twelve matrices, including nine environmental samples plus ice creams and sherbet, were artificially contaminated with live bacteria and free DNA from *Salmonella* or *Listeria*. The initial test investigated the removal of free DNA from samples and the second tested the effect of the treatment on the viability of living pathogens and their detection using PCR. Three real-time PCR devices were used throughout the study and results were verified using culture confirmation according to the ISO6579-1:2017 and ISO11290:2017 method.

**Results:** The results showed that the treatment successfully removed free DNA. The treatment resulted in Cq reductions of >10 Cq for both *Salmonella* and *Listeria* across all sample types on all three cycles. Additionally, the analyses showed that there were no adverse effects of the pretreatment on the viability and detection of either of the pathogens. All results were culture confirmed. Additional tests are being performed within the frame of an AOAC certification and results are expected soon.

**Significance:** PR*Eraser* BAC*Gene* enables laboratories to reduce the number of presumptive positive results, thereby reducing time to result as well as cost.

#### P1-11 Evaluation of the Genedisc STEC Top5 Short Protocol for Same-Day Release of Raw Beef Meat Samples

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**Introduction:** For the release of raw ground beef products, manufacturers have to guarantee the absence of Shiga toxinproducing *E. coli* (STEC) in 25 g of sample. In order to limit costs linked to storage, the time to result (TTR) of the method for STEC detection should be less than one workday, i.e., eight hours. **Purpose:** As part of the ISO 16140-2 (2016) validation of GeneDisc STEC Top5 method, an independent sensitivity study compared a short protocol for raw beef samples (5 h) to the ISO/TS 13136:2012 reference method.

**Methods:** Thirty-nine raw beef samples were included in the study. Three enrichment times were tested for the alternative method: five, eight, and 20 h in buffered peptone water at 41.5°C. The test portion for DNA extraction was five milliliters after five hours of enrichment, and bacteria were concentrated by centrifugation. For a longer incubation time (eight to 20 hours), 50  $\mu$ L of enriched sample were directly transferred into a lysis tube. After mechanical lysis, the resulting DNA extract was analyzed using the STEC Top7 GeneDisc Plate.

**Results:** The results showed six negative deviations and 10 (five hours enrichment) or 11 (eight to 20 hours enrichment) positive deviations. The only difference observed was after five hours of enrichment; compared with longer incubation (eight h and 20 h), it corresponded to a seasoned beef sample which was negative by the GeneDisc method after five hours incubation (negative agreement with the reference method) and became positive after eight to 20 hours incubation (positive deviation).

Significance: This study demonstrated that the GeneDisc method for early detection of STEC in raw beef meat samples fulfills the EN ISO 16140-2:2016 requirements. Applied to the beef industry, this method enables the manufacturer to significantly decrease the storage time of fresh products before release.

#### P1-12 The Use of the BAX PCR Detection System for Pathogens in Cannabis Flowers and Edibles

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Introduction: Cannabis products were evaluated for compatibility with Hygiena's BAX System X5 *Listeria* spp., *Salmonella* spp., and *E. coli* O157:H7 kits. Stringent regulations require microbial testing of finished cannabis products. Finished products are to have less than one CFU of *Salmonella* spp. and Shiga toxin-producing *E. coli*. Upon confirmation of compatibility, the focus of the study shifted to endpoint detection of Target bacteria using the BAX System X5.

**Purpose:** To demonstrate that cannabis products are compatible with the BAX PCR system.

**Methods:** Cultures of target organisms were started in five ml of TSB and incubated at the organism's optimal growth temperature the night prior to assay. Cannabis suspensions were prepared the following day by adding 10 ml of media to one gram of cannabis product. An overnight culture was serially diluted in the cannabis suspension to attain concentrations of 10<sup>4</sup> CFU/mL, 10<sup>1</sup> CFU/mL, and 10<sup>o</sup> CFU/mL. An overnight culture was also serially diluted in brain heart infusion (*Listeria monocytogenes*) or tryptic soy broth (*Salmonella* Enteritidis and *E. coli*) to concentrations of 10<sup>4</sup> CFU/mL, 10<sup>1</sup> CFU/mL, and 10<sup>o</sup> CFU/mL. Dilutions of overnight cultures were incubated at 37°C and tested in triplicate at 24-hour time points for up to 72 hours. Enriched samples were also plated at each test time point.

**Results:** Salmonella Enteritidis and E. coli O157:H7 were detected at low levels utilizing the BAX System X5. Detection of E. coli was seen at all time points. The probability of detection of Salmonella Enteritidis declined (100% at 24 hours, 33% at 48 hours and zero percent at 72 hours) with extended incubation and Listeria monocytogenes was only detected after a 72-hour incubation period at an initial inoculum level of  $\geq$ 10<sup>2</sup> CFU/mL.

**Significance:** The use of rapid microbiological methods can be successfully applied to the growing cannabis food industry

#### P1-13 Rapid Bioluminescence Detection of Bacteria in Cannabis-Infused Foods Using Microsnap

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Introduction: Increasing use of cannabis-infused foods, especially in countries and states where the consumption is legal, has led to a need for rapid testing

**Purpose:** This study demonstrates a rapid bioluminogenic microbiology method for the detection of total viable counts, *Enterobacteriaceae* and coliforms.

**Methods:** Organisms were cultured in TSB and cannabis suspensions were prepared by adding 10 ml water to one gram of cannabis product. Cultures were then diluted in the cannabis suspension or water. One milliliter of the dilutions was added to the corresponding MicroSnap enrichment devices.

These were incubated at the test method incubation temperature. Enriched samples were tested with the corresponding detection devices hourly after 5 to 8 hours.

Results: Detection of the target bacteria panel was determined by an RLU value greater than that of the threshold value. Thresholds were set to the background signal average and three times the standard deviation. Low inocula of coliforms were detected within eight hours at 37°C. The lowest inoculum for cannabis flower and edibles indicated bacterial presence after just five and six hours. MicroSnap coliform RLU thresholds were determined to be positive at >244 RLU, >53 RLU and edibles, >8 RLU. Detection of the Enterobacteriaceae bacterial panel at ≤10 CFU/mI in all tested flower strains occurred within the 8-hour incubation period and as early as five hours. The lowest bacterial inoculum level of 102 CFU/ml in the cannabis edible was detected by six hours. RLU thresholds were determined to be >2 RLU. Detection of the aerobic bacterial panel at ≤102 CFU/mL was within the eight-hour incubation period for all three strains. The lowest bacterial concentration tested with edible was detected by seven hours of incubation. RLU thresholds were determined to be >8 RLU and >2 RLU.

Significance: The use of rapid microbiological methods can be successfully applied to the growing cannabis food industry.

#### P1-14 Validation of the Ultrasnap Surface ATP Test and Ensure Luminometer for ATP Hygiene Monitoring on Stainless Steel Surfaces

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**Introduction:** The use of rapid ATP tests allows for a quick determination of cleanliness; this study demonstrates the use of dry and wet bacteria from surfaces as a measure of the performance of the device.

Purpose: To demonstrate the use of dry and wet bacteria from surfaces as a measure of the performance of a rapid ATP test device. Methods: Cultures of Pseudomonas aeruginosa, Bacillus subtilis and Saccharomyces cerevisiae were tested. The bacteria were cultured in tryptic soy broth, yeast in Sabouraud. Cells were harvested by centrifugation, washed with buffered peptone water, and resuspended. Each organism was diluted to five contamination levels. Surfaces were tested wet and dry. Replicate samples of 0.25 mL were spread over a 4"×4" stainless steel coupon. Twenty coupons were used for each dilution. Sterile buffered peptone water was spread over 20 coupons for the background. Ten coupons were tested for each dilution while the surfaces were wet, and 10 coupons for each dilution while the surfaces were dry. The surfaces were visibly wet when sampling or stored at room temperature until visibly dry. Results: S. cerevisiae was detected at lower dilutions than the other organisms. Wet, it had a LOD of 849 CFU, and when allowed to dry, it had a LOD of 1,012 CFU, corresponding to 69 RLU and 280 RLU, respectively. B. subtilis, when wet had a LOD of 1.93×10<sup>4</sup> CFU and dry, it gave a LOD of 3.07×10<sup>4</sup> CFU, corresponding to 36 RLU and 28 RLU, respectively. *P. aeruginosa* was detectable at higher dilutions. When wet, it gave a LOD of 1.34×10<sup>5</sup> CFU, and when dry for 2 h, it gave a LOD of 7.46×10<sup>4</sup> CFU, corresponding to 37 RLU and 145 RLU, respectively.

Significance: The direct detection of bacteria from surfaces using non-specific ATP tests can be a first line defense for cleaning verification. This study validates this device with significant levels of microorganisms

#### P1-15 ISO 16140-2 (2016) Validation of the RAPID' B. cereus Method for the Enumeration of Presumptive Bacillus cereus Group in Dairy Products, Ready-to-Eat and Ready-to-Reheat Products, Cereals, Spices, Dehydrated Fruits, and Vegetables

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Introduction: The Bacillus cereus group includes gram-positive spore-forming rod-shaped bacteria, and is highly dispersed in the food production environment and potentially responsible for food poisoning. The RAPID'B. cereus plate allows specific enumeration of the B. cereus group from food products within one day. Purpose: An independent study compared this new alternative method to the EN ISO 7932:2005 (reference method) according to the ISO 16140-2:2016 for NF-Validation (Afnor). Methods: Different matrices were tested: dairy products, RTE and ready-to-reheat products, cereals, spices, dehydrated fruits, and vegetables. Two inoculation procedures were tested: spreading of 0.1 ml of suspension or decimal dilutions onto the plate (or 1 ml onto three plates), or pouring the medium on 1 ml of suspension or decimal dilutions. The plates were incubated 21 h at 30°C ± 1°C. Characteristic colonies (red colonies most often surrounded by an opaque halo) were enumerated. Storage of plates for 72 h at 5°C  $\pm$ 1°C was evaluated for spreading inoculation. The study investigated relative trueness, accuracy profile, inclusivity, and exclusivity. Results: Overall, 73 and 66 samples were analysed by spreading and pour plate techniques, respectively, providing 51 interpretable results each. Among these samples, 23.5% (spreading) and 27.5% (pour plate) were naturally contaminated. The mean difference between the alternative method and the reference method counts ranged from -0.06 to -0.14 log CFU/g. For the spreading procedure, performance remained unchanged after 72 h storage at 5°C. For the accuracy profile study, the lower and upper β-ETI was within the acceptability limits, fixed at ±0.5 log. The inclusivity and exclusivity study showed a satisfying result, highlighting that the alternative method allows the enumeration of colonies from the 7 different phylogenetic groups within the B. cereus group. Significance: The alternative method is reliable for the enumeration of the B. cereus group and offers more practicability to the user than

#### P1-16 Validation of a Real-time RT-PCR Method for Detection and Quantification of Norovirus in Shellfish

the reference method.

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Introduction: Noroviruses (NoV), members of the *Caliciviridae* family, are recognized as a leading cause of viral foodborne outbreaks in Europe. Shellfish consumption is frequently involved in foodborne outbreaks, as shellfish can actively accumulate these viruses during filter-feeding activity. Genetically, NoVs are divided into seven distinct genogroups, with genogroup I (GI) and II (GII) responsible for most of the clinical cases. As multiplication of NoV In Vitro is highly restricted, detection relies on molecular methods, such as real-time RT-PCR. The detection of NoV in shellfish is a challenge, as the shellfish matrix is known to be rich in inhibitors of RT-PCR and contamination levels are low often close to the limit of detection.

**Purpose:** The aim of the study was to characterize and validate a real-time RT-PCR method based on ISO 15216-1 for detection of NoV in shellfish.

**Methods:** The method was validated on shellfish contaminated with NoV GI and GII by bioaccumulation, mimicking as much as possible the natural contamination process. The validation approach was based on U47-600-2: 2015 standard method, including a large panel of controls.

**Results:** The results of the characterization of the real-time RT-PCR method showed its specificity, repeatability and reproducibility. The limits of detection and quantification, calculated for both genogroups, confirmed its high sensitivity.

Significance: The validated method enable robust and quality assured control method for hygiene control of shellfish, and analysis of samples implicated in outbreaks.

#### P1-17 Requirements and Guidelines for Conducting Challenge Tests of Food and Feed Products

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Introduction: It is the responsibility of food business operators and establishments who manufacture a food product to take measures to ensure that safety criteria are applicable throughout production processes, storage conditions, and food preparation recommendations dedicated to consumers. In the case of ready-toeat and/or perishable foods that support microbial growth, several practical issues may arise: How to conduct a challenge test? On how many food batches? Should one strain or a cocktail be used for inoculation? How to determine food shelf life during storage? How to determine the effect of product reformulation or processes (salting, smoking) on microbial growth?

Purpose: The aim of the International Organization for Standardization (ISO) ISO/TC34/SC09/WG19 working group is to establish general requirements and guidelines for conducting challenge tests on food and feed products. A distinction is made between studies establishing growth or inactivation of a specific microorganism/food combination.

**Methods:** Within the frame of the ISO, members from all over the world collaborate to create internationally recognized documents providing requirements, specifications, guidelines or characteristics that can be used consistently to ensure that products, processes and services are fit for their purpose.

**Results:** Our working group comprised experts from the food industry, food technology institutes, food testing laboratories, research centers and regulatory bodies. WG19 developed a standardised protocol to conduct challenge tests to study growth potential, lag time and maximum growth rate (ISO 20976-1:2019). This standard has a link with European legislation (Regulation 2073/2005) on microbiological criteria for foodstuffs. At present WG19 is working towards consensus on two additional standards: challenge tests to study inactivation potential and kinetics parameters (ISO 20976-2) and determination and use of cardinal values in predictive microbiology (project ISO 23691).

Significance: General and consensus documents on best practices for conducting challenge tests will ensure harmonization and efficiency between all stakeholders.

#### P1-18 Development of a Combined Recombinase Polymerase Amplification and Lateral Flow Assay for Detection of Emetic *Bacillus cereus*

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Introduction: Bacillus cereus is a gram-positive, spore-forming bacterium, widely distributed in many environments and often found in foods. The *B. cereus* emetic toxin, cereulide, is the cause of food poisoning when ingested by the consumer. In the case of a foodborne outbreak, the investigation relies on traditional microbiological methods, supported by immunological and molecular assays for the rapid detection of the emetic toxin and its genes. Recombinase polymerase amplification (RPA) is a novel isothermal technology that has been widely used to improve the rapidity and sensitivity of molecular diagnosis of foodborne pathogens.

**Purpose:** To develop a rapid and sensitive detection method for emetic *B. cereus* using an RPA based on the sequence of the cereulide synthesis genes (*ces*), combined with a lateral flow assay (LFA).

Methods: Cereulide-producing B. cereus (NCTC 11143) and cereulide-non-producing B. cereus (ATCC 14579) were used as reference strains. Specific RPA primers targeting ces genes were designed with Primer3Plus software according to TwistDx guidelines. For LFA visualization, the forward and reverse primers were modified with 6-FAM and biotin at the 5' end, respectively. RPA was performed with the TwistAmp Basic Kit at 37°C for 20 minutes. The amplification products were visualized both by three percent agarose gel electrophoresis (AGE) and by LFA. In the latter gold nanoparticles (GNPs) are used as signal reporters; the amplicon is captured by streptavidin printed at the test line at 3'(biotin) and bound to GNPslabeled antibody at 5'(6-FAM), while the excess of GNPs is captured by a secondary antibody printed at the control line. Results: The novel RPA gave specific amplification of the positive strain after 20 minutes. The LFA allowed detection of the positive reaction with less than six fg of DNA per reaction within five minutes. Significance: The fast reaction, simplicity, cost-effectiveness, sensitivity and specificity make the RPA-LFA an attractive diagnostic tool for routine examination of suspected B. cereus food poisonings.

#### P1-19 Detection of Mango Ingredients in Food By Loop-mediated Isothermal Amplification Assay

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Introduction: Mango is a common allergen in Taiwan. The best way to prevent an allergic reaction is to avoid the food allergen or foods containing the allergen. Therefore, a rapid, sensitive and specific assay for the detection of mango is required. A well-established nucleic acid amplification method, loop-mediated isothermal amplification (LAMP) has been used for pathogen analysis, but application to food analysis has rarely been reported. **Purpose**: The objective of this study was to develop a LAMP assay for the detection of mango in food.

Methods: The specific LAMP primers were designed to target the internal transcribed sequence one (ITS1) of the nuclear ribosomal DNA sequence regions of mango. The feasibility of the established LAMP method for the detection of mango was assessed. **Results:** Both the detection limits of LAMP and traditional PCR for detecting mango were one ng of DNA. These LAMP primers sets showed high specificity for the identification of mango and had no cross-reaction to other species of fruits, including apple, guava, orange and pear. Moreover, when a minimum of one percent mango was mixed with other fruit ingredients at different ratios, no cross-reactivity was shown during LAMP. Processed (boiled and steamed) samples could also be detected by LAMP assay. **Significance:** A rapid, sensitive and simple LAMP based method was developed for detection of mango. This method can be used not only in raw but also processed fruit products. This assay will be useful was the specified end method.

for the rapid detection of mango in food markets. This work was supported by the grant from the Ministry of Science and Technology (MOST 103-2221-E-020-038-), Taiwan.

#### P1-20 Reproducibility of MALDI-TOF MS for Pathogen Confirmation and Identification of Non-Pathogenic Bacterial Isolates: Assessment According to the ISO 16140-Part 6 Standard

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Introduction: MALDI-TOF MS is recognized as a valuable method to confirm and identify microbial isolates. Four interlaboratory studies were recently run in order to assess the reproducibility of the MALDI Biotyper.

**Purpose:** Variable conditions were tested, including instruments, operators, types of target plates, and culture media.

Methods: Twenty four to 36 isolates were evaluated per study, according to the ISO 16140-part 6. Each set of strains consisted of 16 pathogens including *Salmonella* spp., *Listeria monocytogenes, Cronobacter* spp. and *Campylobacter* spp., as well as eight to 20 relevant non-pathogenic strains. Each study was organized by an independent laboratory: two studies were run in Europe, two others in North America. Seven to 17 laboratories were involved depending on the study, with 14 to 17 collaborators. Sixteen standard formulations and chromogenics were tested. A non-selective agar was always

used as quality control. Reusable and disposable targets were tested. The appropriate ISO procedures were run in parallel. The data showing cross-contamination with the ISO protocols were excluded. **Results:** One hundred seventy sets of data and 4,652 identification results were used for interpretation. Isolated colonies were selected for analysis with the MBT. No impact of the selective culture media was observed. No influence of the tested target plates was noticed. The MALDI Biotyper showed 100% correct confirmation rate of foodborne pathogens and provided more than 99% correct identification rate at the group or species level. **Significance:** The MALDI Biotyper provides reliable and reproducible results to confirm the pathogen after a first presumptive screening step, and to identify the other bacteria. These collaborative studies demonstrate that MALDI Biotyper is easily implemented in routine testing.

#### P1-21 In the Classroom with Dietitians: Student Interest and Engagement in Food Safety

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Introduction: Food safety is of major concern to the population at risk. Dietitians have trusted health professionals for food-related messages as they primarily engage with the delivery of nutritional and therapeutic dietary counsel. However, studies with a population at risk demonstrate low awareness of the increased food safety risks due to their condition. This disconnect can severely impact the health of this group. Targeting dietetics program trainees who are about to begin their professional career will likely lead to long-term public health improvements.

**Purpose:** The purpose of the study is to describe the food safety risk awareness, knowledge and attitudes among dietetics students, and their attitudes and self-efficacy in delivering food safety messages to patients.

Methods: In a cross-sectional study design, an online version of a validated survey using Qualtrics was administered to students enrolled in an accredited DPD program in Ohio. The questionnaire was designed to collect demographic information, food safety knowledge and attitudes, and self-efficacy in food-safety message delivery.

**Results:** Of 120 students enrolled in the program, 106 future dietitians participated (90.6% age 19 to 22). Two-thirds (67.9%) of students who received food safety training (*n*=56) thought they had not received sufficient food safety training to deliver education to the clients. The majority (*n*=86; 81.1%) had heard about listeriosis, but only three (2.8%) understood associations with high-risk foods. Less than half (46.3%) knew about *Campylobacter*, but only one student was able to correctly link the pathogen to high-risk foods. The majority (67%) did not identify pregnant women as a population at risk, and only five percent knew that patients with diabetes have increased susceptibility to foodborne illness. The students considered food safety important to their practice.

Significance: The findings of this study will be used to develop novel approaches to teaching food safety to dietetics students and continuing education modules targeting specific food safety information.

#### P1-22 Using an Intervention to Improve Traceability Knowledge and Practices of Food Handlers in Wales

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**Introduction:** The BRC Global Food Safety standard requires businesses to ensure all staff are trained effectively and must conduct traceability of products and packaging. Research on the effectiveness of traceability systems is available but little research is available on the knowledge of food handlers.

Purpose: To understand and improve the knowledge and practices of food handlers regarding traceability systems.

**Methods:** Food handlers (n=54) in food and drink manufacturing businesses in Wales (n=10) participated in a short traceability course in line with BRC requirements. Food handlers' knowledge of traceability was measured before and after the training intervention. Following the intervention, (4 to 8 weeks), technical managers (n=4) were interviewed regarding training impact and effectiveness. **Results:** Following the training intervention, a statistically significant increase in food handlers' familiarity with the company's traceability procedures was reported by food handlers, increasing from 83% to 98% (P<0.05). Food handlers reported increased confidence in using traceability paperwork, from 70 to 94% (P<0.001). Additionally, awareness of non-conformances increased significantly (P<0.001) from 80 to 89%.

Food handlers' confidence in understanding the consequences of poor traceability also significantly increased, from 89 to 98% (*P*<0.05). Additionally, confidence in listing traceability documentation post-intervention increased significantly from 81 to 94% (*P*<0.001), as did their confidence in knowing what their responsibilities are towards traceability, from 81 to 98% (*P*<0.05).

Following intervention training, technical managers reported improvements in food handler familiarity with traceability procedures (67%) and improvements in paperwork completion where reported (67%). Half of the technical managers reported overall traceability systems had improved.

**Significance:** This study has designed, delivered and evaluated training on request of food manufacturing businesses based in Wales, which increased awareness, knowledge and understanding of traceability (no actual behaviours were measured). Food handlers tended to have some knowledge of traceability to carry out their role but evidence shows that a short 2-hour session on traceability could improve knowledge and confidence significantly.

#### P1-23 Investigating the Effect of Washing Raw Chicken on Cross-contamination of Kitchen Surfaces and Ready-to-eat Food Products

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Introduction: Cross-contamination has been known to occur in consumer kitchens to surfaces and RTE food products while preparing raw poultry, but the effect of chicken washing on crosscontamination is not well studied. The impact of current messaging on poultry washing has also not been evaluated.

**Purpose:** This study was conducted to determine how washing raw poultry effects the frequency and level of cross-contamination to kitchen surfaces and a lettuce salad, and determine the effectiveness of a text-based consumer food safety message on washing chicken. **Methods:** A meal consisting of chicken thighs inoculated with 10<sup>8</sup> to  $10^{10}$  PFU/g of a surrogate organism (DH5-*a* with green fluorescent protein) and a lettuce salad was prepared by participants (*n*=281) in a test kitchen. The lettuce and surfaces were sampled after preparation was completed and plated on selective media for enumeration. One hundred forty of these participants received a food safety message regarding chicken washing in their participation confirmation email (intervention group) and 139 served as a control group. Statistical analysis was completed using R software.

**Results:** Chicken washers had a significantly higher prevalence of cross-contamination across all surfaces, including the sink, than non-washers (P=0.0011). Salad lettuce was more frequently positive when the chicken was washed, but not significantly higher than non-washed. Cleaning reduced the level and prevalence (P<0.0001) of surrogate on the sink and counter for both groups. Non-washers were significantly more likely to have no detectable cross-contamination (P=0.0053) and washers were more likely to have three or more detectable instances of cross-contamination (P=0.0360). The food safety email message was also effective at changing consumer behavior.

Significance: This study provided important information on how poultry washing effects cross-contamination, which has not been thoroughly studied, and how effective a subtle food safety message can be for consumers.

#### P1-24 Method Triangulation to Assess Different Aspects of Food Safety Culture in Food Service Operations

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Introduction: Recently, a lot of attention, from both scientists and the food industry, has gone to the food safety culture in food companies. However, assessing food safety culture remains difficult. **Purpose:** The advantages and added value of applying method triangulation to gain a more comprehensive evaluation of the prevailing food safety culture in catering establishments is illustrated by means of a case study. **Methods:** Three methods are applied to assess the food safety

**Methods**: Three methods are applied to assess the food safety culture in food service operations of a Flemish university spread over different locations in the city of Ghent, but centrally managed. Each method sheds light on one of the aspects of food safety culture as defined in the food safety culture conceptual model, in which food safety culture is considered as the interplay between a technomanagerial aspect and a human aspect.

Results: Verification of CCP monitoring registrations and internal audits can provide organizations with information about, e.g., employee compliance with procedures, and problems related to infrastructure, equipment and work methodology. By assessing the food safety climate, insights can be gained about perceived leadership, communication, availability of resources, commitment and risk awareness from both staff and responsibles points of view. By triangulation of these methods, different aspects of the food safety culture at the different locations could be investigated, illustrating how single-method derived results could lead to wrong conclusions. Moreover, by combining the assessment methods case by case, locations in which the hazard of optimistic bias and complacency might exist can be identified. As such, more tailored and locationspecific strategies for improvement of food safety management and/ or food safety culture can be put in place. Significance: By combining food safety/hygiene performance

assessment methods (triangulation) different aspects of food safety culture could be investigated and the weaknesses of one method can be mitigated by the strengths of other methods.

#### P1-25 Improving the Food Safety Awareness and Knowledge of Primary School Children through Illustrative Teaching Cases

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Introduction: Education and training about basic food safety principles are emphasized as important factors contributing to the reduction of foodborne illnesses. As childhood usually coincides with the beginning of meal preparation experiences, it is recognised as a crucial time for developing food safety awareness, knowledge and skills. Children grow up and, as adults, they will continue to practice food-related behaviours at home as caregivers for family members or possibly as employees in the food business sector.

**Purpose:** The main purpose of this study was to investigate the effectiveness of a specifically tailored workshop-based educational intervention targeting barriers to controlling microbiological hazards in domestic kitchens by primary school children.

**Methods:** A cross-sectional pre-test/post-test survey with a control group was administered. A total of 1,272 respondents (enrolled in the 6<sup>th</sup> grade of primary school) participated in the study. The effectiveness of a 45 min workshop divided into four sections: i) impact of temperature on microorganisms, ii) cleaning of kitchen gear, iii) removal of bacteria with handwashing, and iv) prevention of cross-contamination was evaluated with a questionnaire developed for the target group, considering preventive measures identified by the World Health Organization.

**Results:** The results show considerable change in respondents' susceptibility to food-related risk measured through perceived severity and vulnerability. The positive effect of the workshop is most considerable (P < .001) when the perceived vulnerability to food poisoning is put into the context of the domestic environment (29% of respondents changed their opinion). The greatest impact on knowledge is observed in relation to food handling after purchase (P < .001) and critical temperatures (P < .001) with 17.9% and 14.0% improvement.

Significance: The findings might assist educators, especially home economics teachers, in primary schools in presenting food safety practices in more illustrative ways. This survey was a part of the project 'What can I do for safe food?', financially supported by the Municipality of Ljubljana, Capital of Slovenia.

#### P1-26 Factors Influencing Food Safety in Children's Co-curricular Food Preparation Classes

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Introduction: The rise of foodborne illness prevalence among children aged less than five years affirms the importance of children's food-safety education. UK national curriculum changes have resulted in limited nutrition/food-safety education in schools, however, in recent years there has been an increase in co-curricular food-preparation classes (CCFPCs) for infants/junior children. To date, little is known about food safety learning in such groups.

**Purpose:** This study aims to ascertain information delivery and potential influence and contribution CCFPCs can have on the food safety learning of young children.

**Methods:** Qualitative in-depth telephone interviews with foodpreparation class leaders/parents (n=5) were undertaken to determine the inclusion of food-safety in CCFPCs and reported/perceived adequacy of food safety information delivery. CCFPC websites were evaluated to assess food safety inclusion and class recipes (n=45) were analysed to determine ingredient/required-practice frequency.

Results: Findings suggest that CCFPCs aim to advocate safe food preparation. Indeed, all websites evaluated stated intention for promotion/learning about food safety; similarly, all class leaders reported the importance of teaching food safety in their classes. However, parents attending the classes perceived food safety was not adequately addressed and some class facilities were not suited to accommodate safe food production. Ingredient frequency analysis showed that 60% of CCFPC recipes included raw egg, 22% fruit/ vegetables and 18% ready-to-eat foods; few included raw meat/raw chicken. Food-safety behaviours particularly associated with hand hygiene and food storage were required during all CCFPCs, however, parents indicated other than being told "to wash hands at the start and end of the class, very little, if any food-safety information was given". Significance: CCFPCs may provide a valuable opportunity to convey information about safe food-handling/storage to children. However, findings indicate a disparity between intention to do so and current practice. Tailored and age-appropriate information development and food safety support regarding food-preparation class recipe selection and food safety practice are required to improve and optimise this co-curricular educational opportunity.

#### P1-27 Hierarchical Perspectives on Training Culture among Food Handlers

**EMMA SAMUEL** and Elizabeth C. Redmond ZERO2FIVE Food Industry Centre, Cardiff Metropolitan University, Cardiff, United Kingdom

Introduction: Food safety culture research emphasises the importance of quality food safety training, education, and knowledge sharing for every employee in an organisation. Often, the transient nature of food operatives, coupled with inadequate management skills, time constraints and lack of consistent training material can lead to the delivery of incorrect food safety information. Subsequently, perpetuated ill-informed behaviour can be adopted and shared across sub-cultures.

**Purpose:** This purpose of this study was to explore attitudes and perspectives towards food safety among employees in a typical small food business operation to determine potential food handler training opportunities.

**Methods:** Face to face semi-structured interviews were conducted with food handlers (n= 7) in various roles including management, chefs, bar and junior food-handling staff. The thematic analysis enabled identification of common themes associated with food safety perceptions and factors that may influence training across the dataset.

**Results:** Food safety perspectives differed between subculture roles and responsibilities. One chef indicated that watching to "make sure everything is being done right" was a priority while for another chef, "because there's always stuff going on", monitoring junior food handler behaviour was difficult. Junior serving/bar staff would receive food safety training after "6 or 7 months" implying that previous staff turnover had influenced current procedures. Kitchen staff bemoaned serving staff who left food to "sit for 15-20 minutes" before service believing that all employees should be trained in "basic standards" as "sooner or later you are going to have an issue". Head chef was viewed "like a headmaster", a positive role model who "doesn't want us getting into any bad habits".

Significance: Experience, inconsistent training and positive role models emerged as factors relevant to future training. Exploring subculture food safety attitudes may identify beneficial focused training needs which could contribute to safer food practices and enhanced food safety management; a positive step towards improving organisational food safety culture.

#### P1-28 Assessing Food Safety Culture in Food Manufacturing: A Review of Applicable Determinants and Tools

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Introduction: Comprehensive assessment of organisational cultures within the food industry and the impact on safe food production has gained momentum. During the last decade, the impetus for establishing criteria and clear definitions offering businesses an accessible, straightforward means to effectively evaluate culture has increased. However, key concept definitions, classification systems or consensus on one approach have yet to be agreed on by professionals in this research field.

**Purpose:** The purpose of this study was to identify definitions, determinants, methods and tools available for culture assessment and to assess their applicability to a multi-site food-manufacturing operation.

**Methods:** A search of online databases (2008-2018) identified food safety culture (FSC) research studies/publications; primary search terms (foods, safety, culture) were combined with secondary keywords (management, evaluation, assessment, determinants, organisation). Content analysis identified relevant articles demonstrating construct validity in food and organisational-culture domains.

**Results:** In total, 41 FSC research studies were reviewed. Twentynine percent included discussion of tools or methods developed to assess FSC components in whole or part, 12% considered five or more determinants (frequently recurring across the dataset) relating to technical, organisational, societal and cognitive elements. Nineteen percent applied a theoretical framework (e.g., theory of planned behaviour) and 10% considered national or subculture as a key evaluative component. Few (seven percent) studies offered a comprehensive assessment (triangulated multi-methods); the majority were exclusively conducted in the food sector.

Significance: The variety of methods identified in this review to accurately assess FSC elements that impact food safety outcomes reflects the complex multilayered nature of this subject field. Recent studies indicate progress, however, definitive guidance indicating determinants applicable to assessing and evaluating cultures within food manufacturing (and the extent to which each impacts food safety), may be useful to concentrate efforts on producing one tool that is accessible, manageable and relevant to food manufacturing and processing operators.

#### P1-29\* Partnering to Strengthen the Risk-Benefit Assessment within EU Using a Holistic Approach

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Introduction: Human diet may present both risks and benefits to consumers' health. A risk-benefit assessment (RBA) of foods intends to estimate the overall health impact associated with exposure (or lack of exposure) to a particular food or food component. **Purpose:** A project funded by the European Food Safety Authority to assess and integrate food risks and benefits regarding toxicology, microbiology and nutrition. The project, named "RiskBenefit4EU – partnering to strengthen the risk-benefit assessment within EU using a holistic approach" (RB4EU) integrates a multidisciplinary team from Portugal, Denmark and France. One of the specific objectives of RB4EU was to capacitate the recipient partners from Portugal on RBA.

**Methods:** and **Results:** In order to attain this objective, a capacity building strategy including theoretical and hands-on training, development of a case-study, and participation on short-term scientific missions was established.

Significance: The aim of this presentation is to present the project, its structure and its deliverables.

#### P1-30 A Qualitative and Quantitative Analysis of Consumer Food Safety Concerns in Lebanon

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Introduction: In recent years there have been considerable public health concerns regarding food safety in Lebanon; food poisoning is reportedly widespread. Enabling consumer food safety through appropriate infrastructure and education is critical to reducing health risks; however, little is known about Lebanese consumer food safety. Purpose: This study aimed to compare qualitative and quantitative research on Lebanese consumers' attitudes towards food safety risks and perceptions associated with acquiring food poisoning. Methods: Qualitative face-to-face interviews (n=43) were conducted with consumers who attended a Lebanese university Health Day to determine food safety perceptions and concerns. A quantitative selfcompleted food safety questionnaire was distributed to a convenience sample (n=97) of Lebanese consumers to determine attitudes towards food safety in Lebanon using a five-point Likert-type rating scale. Comparisons between qualitative and quantitative findings were made.

**Results:** Lebanese consumers indicated their personal risk of foodborne illness to be "*very high*" and "*greater*" when eating outside of the home; this perception corresponded with quantitative data denoting that 87% consumers agreed "food-poisoning is common in Lebanon". Interviews revealed that Lebanese consumers believed food-poisoning is more likely to occur when eating out: "*I do not eat in restaurants in Lebanon to protect my health…and avoid microbes in…food*" as opposed to the home: "*I always take care of the food produced at home*". Correspondingly, 70% were not confident that food outlets followed necessary food safety guidelines and 89% believed they were very unlikely to get food poisoning in their own home. Concern was expressed regarding electricity interruptions and unsafe food storage "*because the temperature of the fridge goes up*" and 77% agreed it is difficult to store refrigerated foods safety in Lebanon.

**Significance:** Qualitative and quantitative data collated using separate research approaches largely concurred to highlight food safety concerns unique to Lebanon, as well as the need for consumer food safety education to overcome food safety challenges and reduce the risk of foodborne illness.

#### P1-31 Food Safety Attitudes of Undergraduate Dietitians

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Introduction: Dietitians are well-placed to deliver food safety information to reduce the risk of foodborne illness in vulnerable patients. Attitudes towards food safety may influence the likelihood that trainee dietitians engage with and deliver food safety advice to patients. Understanding their attitudes towards food safety is required to enable delivery of effective food safety education to those in need. **Purpose:** The purpose of this study was to determine attitudes of trainee dietitians in Wales, Ohio, and Lebanon towards the role of the dietitian in providing food safety information. We assessed the effectiveness of current training in producing dietitians competent in food safety education delivery.

**Methods:** Questionnaires assessing attitudes towards the dietitian's role and current provision of food-safety training were completed by 210 trainee dietitians in Wales (n=78), Ohio (n=102) and Lebanon (n=30).

**Results:** Attitudes towards food safety were highly polarized. Half (48%) agreed vulnerable patients needed food safety advice; half (49%) felt the role of dietitians is solely to provide nutritional advice, not food safety information. Agreement differed between Wales (16%) Lebanon (14%) and Ohio (85%). Thirty percent thought doctors, not dietitians, were responsible for teaching vulnerable patients about food safety. Forty-five percent felt they would not be confident to provide food safety advice, only 29% felt the food safety training they received was adequate, and 48% agreed educating dietitians to inform vulnerable patient groups on the importance of food safety may reduce the risk of foodborne illness. The majority (93%) reported they would like to learn more about food safety for vulnerable populations.

Significance: While trainee dietitians want to improve food safety knowledge, the majority report inadequate training. Most are unclear about a dietitian's role in providing food safety information to vulnerable patients. Research is required to explore how trainee dietitians' food safety education can be improved to enhance their role in vulnerable patient care and in turn reduce the risk of foodborne illness.

#### P1-32 A Comparison of Hand Hygiene Compliance in High-care and High-risk Areas in a Welsh Food Manufacturing Business Using Covert Observation

## **ELLEN W. EVANS**<sup>1</sup>, Catherine Bunston<sup>2</sup> and Elizabeth C. Redmond<sup>1</sup>

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Introduction: Food handler hand hygiene (HH) is the most effective method for preventing cross-contamination and reducing foodborne illness risk among consumers. Covert observation of behavior, using CCTV cameras, is superior to overt observation or cognitive measures, which may be subject to method-influenced biases. Although previous covert observational research has assessed behaviors at food retail/catering establishments, this method is underutilized in food manufacturing/processing environments.

Purpose: To evaluate food handler compliance with company HH protocol at a producer and supplier of sweet and savory RTE products to wholesale, retail, food service and catering establishments. Methods: CCTV footage over 24 h from point-of-entry HH facilities in high-care (cake/pie production) and high-risk (sandwich/salad production) areas was reviewed and assessed using an electronic behavioral checklist to evaluate compliance with company HH protocol.

**Results:** A total of 403 occurrences of food handlers passing through the two hand hygiene areas were observed; 203 exiting production, 47 entering high-care and 153 entering high-risk. No significant differences were found in attempts, practices or compliance (*P*>0.05) in the two areas. Less than nine percent failed to attempt implementing HH prior to entering production areas. HH duration ranged from two seconds to 71 seconds with 93 to 96% of HH attempts shorter than the 20 seconds specified in company protocol. Although >99% utilized soap, only 56 to 69% wet hands first and 76 to 91% failed to push up sleeves prior to HH. Failure to rub all parts of hands was widespread (>87%) and 24 to 35% failed to apply sanitizer. Consequently, >98% of attempts were not compliant with company protocol.

Significance: Despite different food handlers working in the two separate areas of the company, extensive malpractices were observed in both that were contrary to company protocol, which may compromise food safety during production. Findings suggest the need for bespoke training to inform food handlers of identified site-specific issues to improve HH practices.

## P1-33 Prevalence of *Campylobacter* spp. on Cattle and Sheep Farms in Normandy, France

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**Introduction:** Campylobacteriosis is the first most frequently reported foodborne bacterial zoonosis. Livestock are known to be reservoirs of *Campylobacter*. Moreover, this bacterium can survive for a long time in surface waters and sewage and was detected in shellfish from shellfish growing area in Normandy.

**Purpose:** Because, cattle and sheep productions are important in the department of "La Manche", in Normandy (France), we looked at the prevalence of *Campylobacter* in these two productions.

**Methods:** A total of 14 cattle farms and of 12 sheep farms were visited from March and September 2017. Approximatively, 10 pools of feces were sampled in each farm (118 samples for cattle and 130 samples for sheep). *Campylobacter* was detected in the samples according to the EN ISO 10272 method, and the identification of the species was determined by MALDI-TOF.

**Results:** In total, 86% and 83% of the cattle and sheep farms were positive for *Campylobacter*, respectively. For cattle, *Campylobacter* was detected in 57% of the samples. 204 isolates were recovered, mostly *C. jejuni* (98%). For sheep, 31% of the samples were positive with *C. jejuni* the most prevalent species, 100 of 151 isolates (66.6%), followed by *C. coli* (33.3%).

**Significance:** This study is the first that investigated the prevalence of this foodborne pathogen in cattle and in sheep in France and in this area. The prevalence is high, and this could be a risk for contamination of shellfish by *Campylobacter*.

## P1-34 Which Typing System to Assess Diversity of Pathogenic Yersinia enterocolitica?

## Pierre Raymond, Emmanuelle Houard, Martine Denis and **EMILIE ESNAULT**

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**Introduction:** Yersiniosis is the third most frequently reported foodborne bacterial zoonosis after campylobacteriosis and salmonellosis, with an incidence of 1.82 cases per 100,000 European Union inhabitants in 2016. Pigs are the principal reservoir for a human pathogenic strain. A description of the *Yersinia enterocolitica* population in its reservoir, and accurate discriminatory techniques for typing isolates are needed for prevention, outbreak investigation, and surveillance.

**Purpose:** This study is the first which investigated the usefulness of the two reference techniques, RFLP-PFGE and multiple-locus variable number tandem repeat analysis (MLVA), to describe and to understand the genetic diversity of pathogenic *Y. enterocolitica* isolates from pigs at a slaughterhouse over time. In addition, this is the first study for France that describes by MLVA the genetic diversity of *Y. enterocolitica* isolated from pigs.

**Methods:** A total of 167 isolates were considered. They were recovered in one slaughterhouse from two previous surveys that had occurred over two years. Isolates were biotyped, and typed by PFGE using the Xbal enzyme. MLVA was tested on 82 isolates selected according to their Xbal-PFGE profiles and their prevalence in the population.

**Results:** As in the majority of the European countries, the most prevalent pathogenic biotype recovered was BT4 (92.81%), followed by BT3 (7.19%). The BT4 population was genetically more heterogeneous than the BT3 population. With PFGE, we showed that some profiles were maintained in the pig production sector in two consecutive years. With MLVA, we improved the differentiation between isolates (Simpson's index of diversity=0.962 (95% CI 0.945 to 0.979) versus 0.656 (95% CI 0.583 to 0.729)), and showed that clones recovered during both years may be genetically closely related.

**Significance:** These data indicate that MLVA, alone or in combination with PFGE, proved its effectiveness as a tool for investigating foodborne pathogenic *Y. enterocolitica* strains and for assessing their genetic diversity over time.

#### P1-35\* Investigation by Whole Genome Sequencing of the Recurrence of a Stable Genotype of *C. jejuni* Isolated from Food and Humans between 2005 and 2018

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Introduction: Campylobacter jejuni is the leading cause of bacterial gastroenteritis in Luxembourg. Integrated surveillance of this pathogen, mostly transmitted by contaminated poultry meat, was implemented at a national level since 2005. Isolates from distinct sources were collected routinely for molecular typing purposes. During the application of marker genes *porA* and *gyrA* to multilocus sequence typing (MLST), an unexpected endemic pattern in the spatiotemporal distribution of a specific genotype was observed. **Purpose:** This study aimed to analyze this specific genotype using whole genome sequencing (WGS) to investigate the relatedness of isolates belonging to this endemic genotype and to assess its possible origin and spatiotemporal emergence.

**Methods:** *C. jejuni* isolates (*n*=2,600) from various origins, including ruminants, poultry, surface waters and humans, were sequenced using WGS on Illumina Miseq or MiniSeq. Genomes were compared using a two-step strategy: a core genome MLST (cgMLST) scheme of 637 targets (Ridom SeqSphere+, Germany) classified isolates in cluster types (CT) and a specific cluster CT82 was reanalyzed using the cgMLST scheme from Oxford, based on the quality criteria of assembled genomes with at least 98% of targets found. Results: Thirty-one isolates belonging to CT82 and originating from veterinary (12), environmental (two), food (three) and human (14) sources were observed over a 12-year period with an unexpectedly stable core genome. The comparison of isolates using the Oxford scheme showed a difference of less than 10 alleles between genomes. These results suggest the circulation of a genomically stable endemic strain in Luxembourg over several years. Significance: Our study shows that certain genomically stable profiles of Campylobacter jejuni are able to infect humans regularly over long time periods and deserve further investigation for the public health risks they pose.

## P1-36 Effects of Salinity, Heating and Alcohol on the Growth of *E. coli* and *B. cereus* in Soybean Paste during Storage

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Introduction: As people are concerned about high sodium intake. several trials have been done to reduce the salinity of traditional fermented foods in Korea. The salt in these foods plays an important role in controlling pathogen growth, and thus changes in salt concentration should be tested to ensure food safety. Purpose: The objective of this study was to evaluate the effects of salinity combined with heat treatment and the addition of alcohol on the fate of E. coli and B. cereus in soybean paste during storage. Methods: Sixteen percent soybean paste (Doenjang) made by traditional methods had cooked and crushed soybeans added to reduce salinity to six, eight, 10, or 12%. A cocktail of two strains of *E. coli* and three strains of *B. cereus* spores (American Type Culture Collection) were mixed with the paste, resulting in 4 to 5 log CFU/g. Each salinity group was divided into three alcohol concentrations (zero, one, and three percent) and heat treatment (65°C, 20 min) or no heat treatment. Samples were stored at room temperature and monitored for bacterial viability for 15 days.

**Results:** At 15 days, *E. coli* was not detected in samples with 10 and 12% salinity regardless of heat or alcohol treatment. At six and eight percent salinity, *E. coli* increased by about 1 to 2 log CFU/g, then

decreased and remained at 4 to 5 log CFU/g in zero percent alcohol samples, with or without heat treatment. *E. coli* was inactivated after 15 days only in samples with alcohol. *B. cereus* remained at at 3 to 4 log CFU/g, except at 10 and 12% salinity, where no growth was detected at 15 days.

**Significance:** These results suggest that individual or combined treatments of heat and alcohol after fermentation were effective to control non-spore-forming bacteria, but paste quality should be carefully monitored to avoid lactic acid fermentation occurring due to low salt concentration.

#### P1-37\* Identifying Mechanisms of Piezotolerance through Screening of the KEIO *E. coli* Mutant Library

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Introduction: Hydrostatic pressure processing (HHP) involves the mild treatment of food by non-heat pasteurization using pressure between 400 and 600 MPa which is uniformly transmitted in the fluid. However, there is a food safety concern related to possible increased tolerance of foodborne pathogens to HHP associated with great variability in piezotolerance.

**Purpose:** Our study aims to investigate the KEIO *E. coli* mutant library for piezotolerance. However, HHP microbiological testing is lengthy, and we focused on various ways of reducing the time it takes for this analysis.

**Methods:** An important first step was the use of bubble wrap provided by the Sealed Air company, employed in the HHP treatment of *E. coli* strains. We further assessed various ways of sealing the bubbles that could also withstand the high pressures. The sealing integrity of white nail hardener, silicone, adhesive sealant and adhesive septum were tested in sealing needle holes made in cells of bubble wrap.

**Results:** We proved that bubble wrap was sterile and could be used for these experiments. The best method of sealing the bubbles was the use of adhesive sealant and the adhesive septum was finally selected for experiments due to rapidity. Out of a total of 600 strains of *E. coli* mutants assessed within 5 months, 27 were piezotolerant and 53 sensitive.

**Significance:** The work identifies a plethora of previously unknown mechanisms playing a role in piezotolerance. The work deepens our understanding of the molecular basis of piezotolerance.

#### P1-38 Assessing the Impact of Cold Atmospheric Pressure Plasma Processing on Quality and Microbiological Aspects of Red Currants via Multivariate Statistics

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Introduction: Foodborne illness outbreaks around the world have been associated with frozen and fresh berries. Therefore, due to the increasing consumption of berries over the last decades, it is paramount to study technologies that can eliminate pathogens responsible for such outbreaks. Cold atmospheric pressure plasma (CAPP) has been shown to have great potential to be used as an alternative to traditional decontamination of food methods. **Purpose:** In this context, this work was designed to evaluate the performance of CAPP treatment for decontamination of red currants (*Ribes rubrum*) and its effect on quality (TSS, pH, phenolics, ascrobic acid, anthocyanins, titratable acidity, texture and color). Moreover, the decontamination efficiency with respect to fungal and bacterial contaminants of the process gas was assessed.

**Methods:** Multivariate analysis of covariance at a significance level of five percent using the software Statistical Package for the Social Sciences (SPSS) was performed to analyze the data. Red currants were treated with CAPP in a diffuse coplanar surface barrier plasma chamber discharge plasma unit for zero (control), five, and 10 min. The berries were treated from top and bottom at a power of 300 W, using air and nitrogen as the process gas.

**Results:** Results showed that the microbial inactivation was dependent on the process gas; the highest antimicrobial efficiency was obtained with nitrogen plasma, both for the total viable count and for yeasts and moulds. Moreover, the treatment time did not have a significant effect (*P*>0.05) on the quality parameters measured, except for the texture of the red currants treated with nitrogen plasma. **Significance:** CAPP can be used to improve food safety of berries.

#### P1-39 A New Validation Method for Untargeted Next Generation Sequencing Analysis

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Introduction: Next generation sequencing (NGS) microbiota analysis applied on food samples has a large potential for controlling nonconformity (color change, unpleasant odor or taste, swelling) and preventing batch recalls. A major challenge in food testing is to simultaneously handle the large diversity of microorganisms and food products. To tackle this problem, the validation of methods is usually carried out microorganism by microorganism and food matrix by

food matrix. Carrying out such a validation process for metagenetic analysis is obviously impossible, as metagenetic analysis belongs to the untargeted approaches.

**Purpose:** No detailed approach has been proposed to validate a metagenetic-based method on a large collection of food matrices. Considering the variability of chemical composition (with associated PCR inhibitors) and the physiological conditions of bacteria, validation on a pure bacterial community seems insufficient to guarantee the quality of all future analysis. The purpose of this study is to define a validation process for high confidence analysis of the microbiome in various samples.

**Methods:** The method is to intentionally contaminate various real food matrices with a well-characterized bacterial community. The food matrix microbiota is first analyzed before any contamination. This allows the subtraction of the natural flora from the result of the matrix when contaminated.

**Results:** Most of the strains that make up the bacterial community are retrieved when mixed in cheese, meat, and fish based products. However, some strains of the community might be masked due to the initial high concentration of the natural flora or due to differential DNA yield extraction or PCR amplification bias.

Significance: Although the metagenetic analysis of food matrices has the potential to properly manage the risk of nonconformities, the costs, delays and lack of confidence in outputs limit its widespread application. By proposing ways to validate a new untargeted approach, we wish to democratize the NGS analysis.

#### P1-40 The Shelflife of Daily Fresh Bread is Markedly Enhanced after Short, Intense UV Treatments

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**Introduction:** Spoilage fungi are a major factor in food losses of bakery products. Colonization of bread often starts by means of airborne conidia, mostly one-celled fungal cells that are moderately resistant to several stressors.

**Purpose:** In this study, a surface treatment of bread by means of short, intense UV radiation was tested.

**Methods:** Fungal species isolated from spoiled bread or tortillas were selected, including *Aspergillus niger*, *Penicillium paneum*, *Penicillium echinulatum* and the xerophile fungus *Aspergillus glaucus*. Conidia of these species were harvested and used for experiments. The effect of a treatment with intense short (1 to 5 seconds) UV-pulses was tested on sprayed individual conidia on agar surfaces, on inocula of different densities on paper filters and on different concentrations of spores jet-sprayed on white and wholemeal bread. The outgrowth of fungi was followed over time.

**Results:** After UV treatment no colonies developed on treated agar plates after 4 to 5 days, except for *A. niger*, which showed reduced numbers. Controls showed up to 27 colonies. There is a clear delaying effect on germination and visible colony size of spoilage-related fungi inoculated as droplets on paper filters. Droplets of 25, 50 and 50 spores did not show any growth 5 days post-treatment in the case of *Aspergillus glaucus* and *Penicillium* species, while colonies were visible on the controls. UV-treated wholemeal or white loaves of daily fresh bread that was jet sprayed with conidia in all cases showed an increase of shelf life of one day. Additionally, the colony numbers on the loaves after 11 days had decreased from 32 to 100 to 2 to 18 after treatment. UV-treated control loaves without sprayed conidia showed fungal development at least 2 days later compared to their untreated, non-sprayed counterparts.

Significance: Increase of the shelf life of daily fresh bread is quickly realized by this short UV treatment and will lead to reduced food losses and energy waste.

## P1-41 Heat Resistance of Spoilage Alicyclobacillus spp. in Fruit-based Beverages

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Introduction: The acidothermophilic, aerobic and spore-forming bacterial genus *Alicyclobacillus* is important in the fruit juice industry because of its ability to resist commercial pasteurization and spoil juice products. Novel processing strategies need to be developed and tested on a diverse range of *Alicyclobacillus* strains. **Purpose:** This study evaluated the heat resistance of two common spoilage species, *A. acidoterrestris* and *A. cycloheptanicus*, and compared heat resistance values of common strains against guaiacol production capability.

Methods: Alicyclobacillus strains (n = 17) were selected based on the results of whole genome sequencing and spoilage guaiacol production. Spores of Alicyclobacillus were subjected to 90°C heat processing in commercial apple juice (pH 3.74) and surviving cells were enumerated on yeast-starch-glucose agar under optimal growth conditions (pH 4.0 and 45°C). D<sub>90</sub>-values were calculated and compared using the t-Test (cutoff for significance was P<0.01). Results: Statistically significant differences were observed in the heat resistance of A. acidoterrestris versus A. cycloheptanicus strains.  $D_{_{qo}}\text{-values}$  were 14.79  $\pm$  1.87 vs. 9.53  $\pm$  1.29 for A. acidoterrestris and A. cycloheptanicus respectively. Heat resistant strains were found to produce high levels of guaiacol, however guaiacol production capacity was not positively correlated with heat resistance. Significance: These findings covered a diverse sample of resilient juice spoilage strains. Testing of the more thermally resilient strains can assist the development of novel high pressure thermal processing against the occurrence of guaiacol producing Alicyclobacillus in fruit juice and fruit based beverages.

#### P1-42 Effect of Carvacrol, Eugenol and Trans-Cinnamaldehyde on Total Aerobic Plate Count in Fresh, Pasteurized and Ultrasonicated mosambi (*Citrus limitta*) Juice

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**Introduction:** Preservation of fruit juice with minimal thermal treatment and limited use of synthetic preservatives is a major challenge. There are limited published data on antimicrobial effects of natural preservatives applied in synergy with non-thermal treatments of freshly extracted fruit juice.

**Purpose**: This study was to determine the effectiveness of certain essential oil components as natural antimicrobials in mosambi (*Citrus limetta*) juice pasteurized or ultrasonicated and stored at refrigeration  $(4^{\circ}C)$  temperature.

Methods: For sample preparation, 0.2% (v/v) carvacrol, eugenol, or trans-cinnamaldehyde was mixed well with 10 ml manually extracted mosambi juice. Their antibacterial effects against naturally occurring aerobic bacteria were compared with that of benzoic acid (0.15% w/v) treatment during refrigerated (4°C) storage of three types of juice: pasteurized (60°C for two minutes), ultrasonicated (18% tween 20 at 750 W, 20 KHz, 40°C, and 40% amplitude for 10 minutes), or extracted from whole pasteurized fruit (80°C for one minute). Three replicates of all the treatments were performed, and the mean and standard deviation values were analysed using one-way ANOVA Results: All three essential oil treatments showed significant (at least three-log CFU ml-1 reduction) antibacterial activity against naturally occurring aerobic bacteria in mosambi juice during refrigerated storage. Pasteurization of the juice or whole fruit further enhanced this antibacterial activity to at least four-log CFU ml-1 reduction after three to four days of refrigerated storage. However, ultrasonication of the juice resulted in lesser (at least two-log CFU ml-1reduction) antibacterial effect in the treated samples during four days of refrigerated storage.

**Significance**: Carvacrol, eugenol or trans-cinnamaldehyde, alone or in synergy with pasteurization (thermal) or ultrasonication (nonthermal) may be effective natural preservatives in fresh mosambi juice against naturally occurring aerobic bacteria.

#### P1-43 Utilising Molecular Tools to Elucidate Microbial Community Composition in Beef Spoilage

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**Introduction:** In recent years molecular technologies have changed the approach for analysing microbial diversity, providing much greater detail than culture-based studies alone. This has particular

application for the food industry, providing a greater understanding of the microbial ecology of foods, and factors which influence it, from production to end of shelf life. However, with these technologies, new challenges also emerge. To implement a whole chain approach for analysing microbial diversity and to identify contamination routes, different samples types are required which may impact on the quality of template DNA material. In order to ensure sequencing performance, the DNA should be of very high quality, regardless of the matrix, to avoid bias on the identification of bacterial communities. **Purpose:** The objective of this study was to develop an SOP for sample preparation and DNA extraction to obtain high-quality DNA from beef abattori samples and assess the composition of the lactic acid bacteria and *Enterobacteriaceae* communities using a quantitative PCR assay.

Methods: DNA samples from beef hide and carcass swabs, environmental swabs and meat samples were utilised in a qPCR assay to elucidate the species present from two common meat spoilage groups, lactic acid bacteria (four species) and *Enterobacteriaceae* (three species). These results were then correlated with culture-based results.

**Results:** For lactic acid bacteria the culture and qPCR results correlated well, while for *Enterobacteriaceae* the qPCR results were generally higher. In addition, the groups' species composition varied between samples through shelf life, providing evidence of individual species' potential role in meat spoilage.

**Significance:** Using this SOP, a metagenomic approach will be performed to undertake a more in-depth analysis of the microbial community alterations of beef from slaughter through to retail, and assessing its correlation with spoilage. Such information can support the industry to ultimately increase product shelf life.

#### P1-44 A Generic Model System for Collecting Data to Predict Microbial Spoilage of Commercially Packaged Fresh-cut Vegetable Salads Using Only Temperature and CO<sub>2</sub> Levels as Predictor Variables

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Introduction: Modified atmosphere packaging (MAP) and storage temperature are two hurdles capable of slowing down changes of gases inside packaging and consequently extending the shelf life of fresh-cut salads. Existing predictive models are commonly constrained to the product and packaging conditions for which they were developed.

**Purpose:** To develop a vegetable-packaging model system to minimize the effect of packaging characteristics and post-harvest physiology, e.g., film permeability, headspace volume, and respiration rate, and predict only the impact of temperature and in-package gas composition on microbial spoilage of fresh produce.

**Methods:** Fifteen grams of rocket pulp (model food) were packaged (low permeability film) in different O<sub>2</sub>/CO<sub>2</sub> ratio atmospheres (0 to 20% O<sub>2</sub>, 20 to 0% CO<sub>2</sub>, 80% N<sub>2</sub>) and stored at zero to 15°C. Gas composition was monitored via a gas meter. Pseudomonad and lactic acid bacteria (LAB) growth were primarily modelled using the Baranyi model; a polynomial model was used for  $\mu_{\rm max}$  as a function of temperature and CO<sub>2</sub>. Growth models were validated using combinations of commercial fresh-cut vegetables of high respiration rate, MAPs, packaging films, and isothermal or dynamic temperature conditions.

**Results:** Variations in the package gas concentration of rocket pulp were eliminated or significantly decreased (*P*<0.05), under different MAP conditions, adequately verifying the hypothesis of the developed vegetable/packaging model system. *Pseudomonas* spp. and LAB were the most important microbial groups (dominant and of high growth dynamics, respectively). The developed polynomial model for  $ln(\mu_{max})$  of LAB and pseudomonads showed  $R^2_{ad}$  of 0.975 and 0.839, respectively. B, values were 0.93 to 1.10, indicating no substantial bias of the model as well as fail-safe predictions, while predictions and observations showed good agreement (A<sub>r</sub> 1.03 to 1.11). **Significance:** The proposed vegetable-packaging model system is a practical and easily implemented approach that may assist the fresh produce sector in predicting the shelf-life of a group of freshcut salads, i.e., high respiration rate, under different packaging and storage conditions.

## P1-45 Strain Variability in Three Strains of the Food Spoilage Fungus *Paecilomyces variotii*

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Introduction: Fungal food spoilage often begins with contamination by spores. To prevent spoilage of processed foods and drinks, the industry challenges their products with the worst-case spoilage scenario. Intraspecific variation of food spoilage fungi should be taken into account when defining the worst case scenario presented by fungal spores. **Purpose:** In this study, we explored strain variability of the important food spoilage fungus and heat-resistant mold *Paecilomyces variotii*. **Methods:** One hundred eight strains isolated from various locations and different environments were screened for conidial heat resistance. To discover the limits of this resistance, we quantified  $D_{eo}$  values for two sensitive strains and one resistant strain. In addition, we described morphological characteristics of the three strains by microscopy, measured conidial compatible solute compositions by HPLC, and measured spore size distributions by flow cytometry and Colter counter.

**Results:** Conidia of three *P. variotii* strains (DTO 032-I3, DTO 212-C5 and DTO 217-A2) showed variability in heat resistance with  $D_{e0}$ -values of 5.5, 3.7 and 22.9 minutes, respectively. To our knowledge, strain DTO 217-A2 produced the most heat-resistant conidia ever measured with a  $D_{e0}$ -value of 22.9 minutes. Conidia of this heat-resistant strain contained larger conidia with a higher concentration of trehalose compared to DTO 032-I3 and DTO 212-C5 conidia. Altogether, we found intraspecific heterogeneity between the three strains in all the experiments presented.

Significance: The heterogeneity we found in heat resistance for *P. variotii* conidia emphasizes the importance of intraspecific variation when challenging industrial processed foods and drinks.

#### P1-46 Choosing the Correct Tool for an Integrated Environmental Monitoring Program

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**Introduction:** Different methods are employed to test for surface cleanliness in the food production environment. These methods can measure the removal of microbes, ATP residues, and allergens. In some food production environments a single environmental monitoring method is used to check cleaning efficacy and verify removal of food residues

**Purpose:** The purpose of this investigation was to examine which of the commonly used environmental monitoring systems gives the most comprehensive view of the efficacy of cleaning processes in the food environment.

**Methods:** a series of different surfaces were artificially contaminated with different levels of ATP-containing food, allergens, and bacteria. These surfaces were tested for cleanliness by methods commonly used to check cleaning efficacy. ATP testing was carried out by ATP surface swabbing and a luminometer. Allergen testing was carried out with lateral flow ELISA. Microbial contamination was checked using direct contact growth media assessment.

**Results:** The results demonstrated that all the methods were effective for evaluating cleanliness. Lateral flow ELISA testing was able to detect down to 1 ppm for some allergens and ATP detected contamination at very low levels. Microbial detection was down to one CFU per area tested but some of the methods gave potentially false negative results for some contaminants.

Significance: As it has been shown that not all commonly used environmental testing methods were able to detect all remaining surface residues, it is recommended that a comprehensive environmental monitoring program contain a mixture of test methods to ensure all residues can be detected and action is taken to remove them before food production can begin.

#### P1-47 Discrimination of Infectious and Heat-treated Norovirus by Combining Platinum Compounds and Real-time PCR

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Introduction: Human noroviruses (NoV) are major agents of foodborne outbreaks. Cell culture models have been developed recently; however, due to the lack of a standardized cell culture method, real-time RT-PCR is now commonly used for the detection of NoV in foodstuffs and environmental samples. However, this approach cannot discriminate between viral nucleic acids of infectious and noninfectious viruses, thus limiting the evaluation of infectivity for public health risk assessment.

Purpose: The aim of this study was to develop a viability PCR method to discriminate between native and heat-treated virus for NoV (GI and GII) and its surrogate, murine norovirus (MNV).

**Methods:** Viability markers (monoazide dyes, platinum and palladium compounds) were assayed using native or heat-treated virus, and incubation conditions were optimized with PtCl<sub>4</sub>, the most efficient viability marker. The impact of the region of the viral genome amplified by the molecular model and the amplicon length were evaluated on inactivated MNV genomic titer. The optimal viability PCR conditions (incubation with 2.5 mM PtCl<sub>4</sub> in PBS for 10 min at 5°C) were finally applied to MNV by performing heat inactivation studies and to native and heat-treated NoV clinical strains.

**Results:** The titer reduction of heat-treated MNV assayed by viability PCR was higher with molecular models targeting the 5' NTR, ORF1 and 3' UTR regions. However, the amplicon length had no impact on the titer of the heat-treated virus. The viability PCR discriminated efficiently between native and heat-inactivated MNV at 72 and 80°C, and efficiently reduced the genomic titer of heat-treated NoV clinical strains.

Significance: This viability PCR method could be useful to study heat inactivation kinetics of NoV and MNV. It could also be evaluated for the identification of infectious enteric viruses in foodstuffs and environmental samples to provide a better assessment for viral risk management.

#### P1-48\* Towards a Point-of-Use Nanosensor System for On-farm Disease Detection

#### **FERNANDO GARRIDO DIAZ** and Alan O'Riordan *Tyndall National Institute, Cork City, Ireland*

Introduction: Farm diseases in animals and plants have increased because a variety of pathogenic viruses, bacteria, protozoa, and helminths which have developed numerous mechanisms that render them resistant to some antimicrobial agents. Disease outbreaks in high-value crops can cause significant economic damage; consequently, a method for the rapid identification of viruses is critical for the protection and prevention of costly outbreaks. **Purpose:** Design an immunosensor to detect the presence of potato virus Y and process this information with the help of an electrical device generating a response directly to the farmer.

**Methods:** On-chip electrodes were modified using different reagents in order to get a covalent attachment of a specific capture antibody and afterward, samples with the presence or absence of virus were able to be deposited on electrode surfaces. In order to detect the presence or absence of virus in those samples, cyclic voltammetry (CV) and electrochemical impedance spectroscopy (EIS) were applied to compare the limitation of electron transfer of FcCOOH to the electrodes. The decrease in current intensity and increase in impedance was monitored, in order to control the effectiveness and efficiency of the detection.

Results: Virus presence was confirmed by a decrease of the current intensity on the CV voltammogram and the increase of the charge transfer resistance and the global capacitance in EIS Nyquist, comparing to each one of the shapes obtained after the deposition of the antibody. In the case of the negative samples (without a target virus), the CV and EIS shapes were similar to the capture antibody measurements.

**Significance:** This immunosensor could be a very important tool for food safety from farm-to-fork because virus presence can be detected in minutes on the field instead of days in the laboratory.

#### P1-49 One-step *Listeria* Enrichment Broth with Improved Selectivity

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**Introduction:** In order to detect one *Listeria* cell per food sample, which is the accepted threshold of most regulatory agencies, rapid detection methods require an efficient enrichment broth. New medium formulations need to focus on the recovery of sublethally injured *Listeria* in challenging food matrices with a high level of microflora, inhibitory substances and non-optimal pH.

**Purpose:** This study aims to develop a one-step enrichment broth for the rapid recovery of *Listeria* from food with a high level of background flora.

Methods: The repair capacity and growth kinetics characteristics of sublethally injured and healthy Listeria, as well as competitor bacteria, were monitored at 30 and 35°C using the one-step broth in comparison to conventional primary and commercially available single-step media. Stainless steel, cold-smoked salmon, pasteurized milk and raw-milk blue cheese samples were artificially contaminated with Listeria and then enriched with the broth at 30 to 35°C for 20 to 24 h followed by detection using real-time PCR and direct plating Results: In-vitro culture studies showed enhanced recovery and growth of Listeria grown in the broth, including fastidious and slowly growing L. ivanovii, while a complete inhibition of Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa and Bacillus licheniformis was observed. Overall, the results obtained from 300 environmental and food samples indicated a comparable recovery of Listeria after as soon as 20 h of incubation when compared to the USDA-FSIS MLG 8.10 and US-FDA BAM 10 reference methods. The samples enriched with the one-step broth showed reduced loads of microflora on selective and differential agars that allowed for easy and rapid reading of the plates.

Significance: The new one-step broth has a strong ability to provide an ideal growing environment for sublethally injured *Listeria* enriched at a wide range of incubation temperatures in particularly challenging matrices containing a high level of competing microflora. This enabled accelerated, reliable and faster detection of *Listeria*.

#### P1-50 Investigation of the Suitability of Neutralising Swabs for Use in an ISO 18593-based Programme

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Introduction: To ensure the safety of the consumer, it is essential to detect microbial contamination in the food supply chain. This requires suitable methods for sampling the immediate environment and food contact surfaces in manufacturing and processing facilities. The recently updated ISO 18593 provides guidance on the methods which can be used. Sampling devices and media should include neutralizers to prevent traces of disinfectant from interfering with the detection of microorganisms from recently cleaned surfaces. Performance testing for such media is specified in ISO 11133.

**Purpose:** NRS II Transwabs sampling swabs are formulated in accordance with ISO 18593, and this study investigated their performance in accordance with ISO 11133.

**Methods:** Suspensions of *Escherichia coli* or *Staphylococcus aureus* were prepared in phosphate buffered saline to a concentration of  $10^{\circ}$  CFU per ml. One hundred µl aliquots were dispensed into microplate wells. For each aliquot, the swab was used to absorb the liquid and then placed in its transport tube. All tubes were vortexed. For each tube,  $100 \,\mu$  was withdrawn immediately and plated onto tryptic soy agar. The tubes were closed again and retained at ambient temperature for 60 minutes, after which a further  $100 \,\mu$  was withdrawn and plated as before. All plates were incubated at  $37^{\circ}$ C for 24 hours and then counted. ISO 11133 requires that the counts for 60 minutes should not differ by more than 30% from the time zero counts **Results:** For *E. coli* the 60-minute counts were 10% higher, while for *S. aureus* the counts were 17% higher. Both these results are well within the specification of ISO 11133

Significance: The study demonstrates that the NRS II Transwab performs in accordance with ISO 11133 and can be used in an ISO 18593-based programme.

#### P1-51 From Lab Scale to Pilot Scale: Survival Dynamics of *Listeria monocytogenes* and Impact of Biocontrol Agents in Mushroom Production

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Introduction: Listeria monocytogenes is a growing concern for the mushroom industry, as studies have shown that this pathogen can be found in mushroom production facilities, which therefore pose a risk of product contamination. Despite the lack of listeriosis reports due to the consumption of fresh cultivated mushrooms (*Agaricus bisporus*), recalls of mushroom products have occurred in recent years which have resulted in an economic and reputational loss for the industry. Thus, it is important to take proactive steps to maintain this industry's reputation for food safety by exploring novel applications of biocontrol agents to provide enhanced assurance of product quality and safety. **Purpose:** The aim of this study is to evaluate the survival dynamics of artificially inoculated *L. monocytogenes* in mushroom growth substrate and the effect of anti-listerial biocontrol agents in the mushroom growth substrate and on the mushrooms themselves during the growing process.

**Methods:** A cocktail of five *Listeria monocytogenes* strains were inoculated on the mushroom growth substrate. The same level of *Lactococcus lactis* was also added to the casing to test for antagonistic activity. Casing and mushroom samples were taken at different time points and assessed for the presence of *L. monocytogenes*.

**Results:** *L. monocytogenes* was found to be present in mushroom growth substrate until the end of the crop production cycle, although levels decreased continuously. The use of *Lactococcus lactis* as a competitive exclusion organism had no effect on the levels of *L. monocytogenes*, having previously shown efficacy in the lab. **Significance:** This study provided valuable information on the survival of *L. monocytogenes* in mushroom growth substrates and its transfer potential onto crops if present. Additionally, this highlights the importance of upscale testing of potential biocontrol agents in industry-like conditions.

#### P1-52 Translation of Science into Guidance for Primary Processing of Onion and Basil: Management of Microbiological Food Safety during Air Drying and Steaming

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**Introduction:** Dried vegetables and herbs undergo only mild processing, such as air-drying and steaming. There are gaps to be filled in the industry to better understand the level of inactivation of pathogenic vegetative bacteria by these processes. **Purpose**: The objective of this study was to generate scientific data and translate them into guidance booklets for the training of Nestlé

and variable them into guidance booklets for the training of Nestle auditors and suppliers on the microbiological safety achieved during drying and steaming. **Methods:** Scientific data on bacterial inactivation achieved during

hot-air drying of fresh onion and low-temperature steaming of freezedried basil were produced. The data allowed the determination of safe process parameters and their inclusion in the guidance booklets developed. Air-drying was performed in a pilot plant dryer and steaming in a microbiology laboratory using a bench scale steamer. For air drying, the surrogates *Escherichia* coli P1 and *Enterococcus faecium* were tested. For steaming a cocktail of *Salmonella* spp. and the surrogates *E. coli* P1 and *E. faecium*, were tested. Samples of sliced onion and dry basil pieces were inoculated then processed for inactivation. Microorganisms that survived the process were recovered and counted using serial dilution methods.

**Results**: *E. coli* P1 and *E. faecium* were shown to be suitable *Salmonella* surrogates for drying and steaming, respectively. Airdrying of onions at 100°C delivered >4-log reduction of *Salmonella* after 90 minutes. Steaming of dry basil at 85°C delivered approximately 4-log reductions of *Salmonella* after 5 minutes. For both air-drying and steaming, dedicated booklets were produced reporting best practices for good manufacturing, process management, safe process conditions, validation, verification and equipment maintenance.

Significance: The drying and steaming booklets were distributed internally to auditors and externally to suppliers and product-specific organizations to create shared value and improve the food safety of mildly processed plant-based raw materials.

#### P1-53 Formal and Informal Spinach Safety from Farm to Fork: A South African Case Study

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**Introduction:** In South Africa, a complex supply chain exists with parts ranging from effectively implemented food safety management systems in the formal sector to no system in the informal sector. Spinach is a popular leafy green in South Africa, with 20% of the population consuming it raw.

**Purpose:** A comparison of the microbiological safety and quality of spinach grown and sold formally and informally was conducted in South Africa.

Methods: Samples of spinach (n=345) and water (n=195) were analyzed. Total coliforms, *Enterobacteriaceae*, and *Escherichia coli* were enumerated on violet red bile glucose agar, *E. coli*/coliform petrifilms and eosin methylene blue (EMB) agar. *Salmonella* spp., *Listeria monocytogenes*, *E. coli*, and presumptive ESBL-producing *Enterobacteriaceae* were detected using iQ Check (*L. monocytogenes* and *Salmonella* spp.) and enrichment followed by isolation on EMB, Rapid *L. mono*, XLD, *Salmonella* Brilliance and ChromID ESBL agar. All presumptive positive isolates were confirmed by MALDI-TOF MS.

**Results:** Spinach at the point of harvest in the formal and informal sectors had coliform counts ranging from 2.9 to 4.6 CFU/g and 1.15 to 5.13 CFU/g, respectively. *Enterobacteriaceae* counts ranged from 2.89 to 4.30 CFU/g and 2 to 6.04 CFU/g, respectively.

Enterobacteriaceae and coliform counts from spinach at the point of sale were similar between formal and informal supply chains, except for spinach being sold from one area in the informal markets, where counts were higher. No *L. monocytogenes* was detected from any spinach samples. Salmonella spp. were only isolated from spinach in the field at one informal farm, the irrigation water from this farm was also found to be contaminated. Presumptive ESBL-producing organisms were found to be present on all spinach from farms and at the point of sale.

Significance: Spinach in the informal sector is not less safe than spinach in the formal sector, despite robust food safety management systems.

#### P1-54\* Impact of Starter Culture and Cassava Variety on the Composition and Rheological Properties of Lafun

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**Introduction:** Lafun is a traditional fermented product of cassava; however consumer acceptability has declined due to its variable quality. This study is part of a wider project to standardise lafun production to overcome this.

Purpose: The objective of this study was to determine the impact of starter culture and three cassava varieties (bitter (IBA30572), sweet (TMEB117) and fortified (IBA011371) (IITA, Ibadan, Nigeria)) on the chemical and rheological properties of optimized lafun. Methods: The starter cultures were Weissella koreensis (two strains), Lactococcus lactis, and Leuconostoc mesenteroides, identified from spontaneous fermentation of cassava. They were used singly and in combination to produce 24 lafun samples. A traditionally produced lafun sample from Bodija market, Ibadan, Nigeria served as a control. Proximate analysis (moisture, ash, crude protein, crude lipid, crude fibre, and digestible carbohydrates) was used to partition both nutrients and non-nutrients found in the samples into classes based on chemical properties. The rheological evaluation of the samples was carried out by dynamic oscillatory tests and analysis was performed using a controlled stress-strain rheometer and a serrated parallel plate sensor. Frequency sweep tests were set up at frequencies between 0.1 and 10 Hz with a percent strain of 0.1 in the viscoelastic region. Storage modulus (G') and loss modulus (G") were then calculated.

**Results:** Generally, fortified cassava products had the highest ash (4.37%), protein (2.99%), lipids (2.20%), and fibre (7.43%) content. Interestingly, most of the optimised lafun samples had considerably higher elastic (G') and viscous (G'') moduli than the control. Products of fortified cassava roots had the best viscoelastic gel-like behaviour compared to the remaining two and did not have much significant difference from the standard.

Significance: Taken together, the findings of this work suggest that processing fortified cassava roots (IBA011371) into lafun will enhance its physicochemical properties and quality.

#### P1-55 Influence of Biological Soil Amendments and Soil Parameters on Pathogen Persistence and Transfer Dynamicsin a Pre-Harvest Environment

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**Introduction:** Biological soil amendments of animal origin (BSAAO) provide nutrients to soil but may contribute to contamination. BSAAO type and soil parameters may influence pathogen survival in soils and potential transfer to fruits and vegetables.

**Purpose:** Evaluate *Escherichia coli* population dynamics in soils amended with BSAAO and transfer to cucumbers along with soil extrinsic and intrinsic factors.

Methods: Twenty plots (three m<sup>2</sup>) were amended with: poultry litter (PL), heat-treated PL pellets (HTPP), composted PL (CPL), or unamended inorganic fertilizer (UN). Each plot was spray inoculated with one liter of E. coli TVS355 (six log CFU/ml). Cucumber plants (Supremo) were planted five days post inoculation (dpi). Composite soil samples (n=234) were collected every 10 days until 120 dpi, and cucumbers (n=160) upon maturation to determine E. coli levels, Salmonella spp. presence/absence, and extrinsic (temperature, moisture) and intrinsic (conductivity, soluble carbon) factors. Results: Significant (P<0.05) decreases in E. coli (4.7 to <0.4 log MPN/g), soil conductivity (1.5 to 0.1 mmhos/cm), and soluble carbon (1984.4 to 103.2 mg/kg) were observed between 0 and 120 dpi across all amendments. At 60 dpi, PL and HTPP plots had higher *E. coli* (2.4 to 5.3 log CFU/g) and soluble carbon (171.3 to 175.6 mg/kg) compared to CPL and UN (<0.4 log MPN/g, 76.8 to 95.3 mg/kg), suggesting soluble carbon could be critical for bacterial survival in soil. Although UN plots had lower bacterial populations than PL and HTPP, they supported significantly (P<0.05) greater *E*. *coli* transfer to cucumbers (1.7 log MPN/g) at 60 dpi. *E*. *coli* was detected on all cucumbers (*n*=120) from PL, CPL, and HTPP plots (0.7 to 1.0 log MPN) . Salmonella spp. were detected and confirmed in HTPP plots. Significance: BSAAO type affected *E. coli* survival duration in amended soils, but longer survival did not correlate with greater transfer to cucumbers. Results suggest soluble carbon could be a critical factor in understanding the potential for contamination.

#### P1-56 Evaluation of Microbial Contamination in Fresh Produce and Its Growing Environment

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**Introduction:** The incidence of foodborne outbreaks caused by consuming contaminated fresh produce has increased, however, the original sources of contamination cannot be determined in many cases.

Purpose: The purpose of this study was to identify the route of pathogen contamination in fresh produce by analyzing the microbial distribution in vegetables and environments at the cultivation stage. Methods: One hundred ninety-two samples of fresh produce (lettuce, young radish, cucumber and red pepper), soils, and water were collected from randomly selected fields and growing environments. The samples were analyzed for total aerobic bacteria, coliform/ *E. coli*, yeast and mold, norovirus I & II, and six pathogenic bacteria (*Escherichia. coli* 0157:H7, *Listeria monocytogenes, Staphylococcus aureus, Bacillus cereus, Salmonella* spp. and *Clostridium perfrin-*

**Results:** Total aerobic bacteria in lettuce, young radishes, cucumbers and red peppers were 4.79, 7.12, 4.36 and 4.61 log CFU/g, and coliforms were 1.75, 4.30, 1.07 and 2.54 log CFU/g, respectively. The average detection of norovirus group I was 0, 0, 9.09 and 5.88%, and group I was 6.25, 33.33, 9.09 and 11.76%. This showed the high level of microorganisms, as well as higher detection rate of norovirus, in vegetables grown in summer. *E. coli* was found in both cucumbers (0.3 log CFU/g) and irrigation water (0.2 log CFU/ ml). Norovirus was detected in vegetables, soil and water in same cultivation areas (Group I 5.26, zero, and 2.04%; Group I 15.79, 10.45 and 4.08%. This indicates that the contamination originated from the irrigation water collected from the surface, especially when irrigated using water sprayed over the produce. **Significance:** These results suggest that *E. coli* and norovirus in

Significance: These results suggest that *E*. *Coll* and norovirus in fresh produce would be transmitted by spray irrigation of surface water over the field, and thus, changes in water resources or irrigation methods should be considered.

#### P1-57 Examining the Efficacy of New and Currently Used Mushroom Industry Biocides on *Listeria monocytogenes* Biofilms

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Introduction: Many biocides in use in mushroom production are utilised for their efficacy against plant disease causing agents. Nonetheless, they may also have activity against foodborne pathogens such as *L. monocytogenes*. However, a number of studies have demonstrated that biocides are not always effective against *L. monocytogenes* in biofilms.

**Purpose:** The aim of this study is to test the efficacy of new and currently used biocides from the mushroom industry for inactivating *L. monocytogenes* biofilms.

**Methods:** A cocktail of five *L. monocytogenes* strains was used to grow three-day biofilms on stainless steel coupons. Eleven biocidal products were chosen and prepared according to the manufacturers' instructions. The anti-listerial efficacy of these biocides was then tested under clean and dirty conditions based on the EN 13697:2015 method.

**Results:** All the biocides tested resulted in a 3.85-log reduction of *L. monocytogenes* biofilm, on average. The dirty conditions generally reduced the efficacy of the biocides. Two biocides which contained a number of different active ingredients resulted in complete inactivation of *L. monocytogenes* biofilms, below detectable limits.

**Significance:** This study has a direct impact on the industry as it provides information on the efficacy of currently used biocides against *L. monocytogenes.* Additionally, new biocidal products with higher efficacy against *L. monocytogenes*, even under dirty conditions, had been identified for potential implementation.

#### P1-58 Quantitative Risk Assessment Model for Salmonellosis from Milk Chocolate Consumption in Brazil

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Introduction: Chocolate is a popular confectionery product consumed worldwide, and *Salmonella* is the main pathogen that has concerned the government and the cocoa-chocolate industry. Brazil stands out

as the fifth-largest chocolate consumer market and has all stages of the production chain, from cocoa farming to chocolate making. **Purpose:** This study estimates the risk of salmonellosis due to consumption of milk chocolate in Brazil using a quantitative microbial risk assessment (QMRA).

**Methods:** A QMRA model predicted *Salmonella* fate during milk chocolate production encompassing three modules: i) cocoa preprocessing, ii) milk chocolate processing, and iii) milk chocolate consumption. Sixteen scenarios were evaluated considering different levels of *Salmonella* contamination in cocoa seeds (lower and higher bacterial concentration), roasting conditions (110°C or 140°C, whole beans or nibs), and cross-contamination after roasting (occurring or not). Simulations (10,000 iterations per scenario) were carried out using the @Risk add-in for Excel.

**Results:** Consumption of milk chocolate contaminated with *Salmonella* would result in means of  $1.3 \times 10^5$  and  $3.2 \times 10^7$  salmonellosis cases per week for the best (low initial concentration, nibs roasted at  $140^\circ$ C) and the worst (high initial concentration, nibs roasted at  $110^\circ$ C) scenarios, respectively. The occurrence of cross-contamination after roasting at the level analyzed did not influence the risk. Further, the roasting of whole beans instead of cocoa nibs resulted in lower risk at  $110^\circ$ C. Sensitivity analysis revealed that the *Salmonella* inactivation rate during fermentation and percentages of cocoa ingredients added to chocolate formulation are the main risk influencers for the best and worst scenarios, respectively.

**Significance:** QMRA simulations predicted that consumption of contaminated milk chocolate can result in a relatively low risk of salmonellosis. Higher roasting temperatures effectively reduce the concentration of *Salmonella* in cocoa, which is a critical control point in the chocolate production process.

#### P1-59\* Application of Risk Assessment in Fungal Spoilage Control: Case Study of Aspergillus Niger on Bakery Products

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**Introduction:** Fungal spoilage is an issue of concern in the bakery industry. Some molds are resistant to thermal processing and also could exist in the industrial environment, leading to post-baking contamination which might reduce product shelf life. Quantitative microbial risk assessment has expanded to be used to estimate microbial risk when the population is exposed to microorganisms in the environment. It also has emerged as an effective tool in the determination and management of risk posed by foodborne pathogens. However, these approaches have not been exploited in the arena of fungal spoilage.

**Purpose:** This study aimed to build effective bakery quality management throughout the chain based on the use of risk assessment.

**Methods:** Risk assessment of bakery spoilage by *Aspergillus niger* is developed with a modeling approach for mycelium growth which includes the important sources of variability. These include extrinsic factors of time-temperature, relative humidity, and crosscontamination, and the intrinsic factor of water activity during the different stages of the chain. The input parameters are fitted to the appropriate distributions and the colony diameter of *Aspergillus niger* at each stage of the chain was estimated using Monte Carlo simulation.

**Results:** The preliminary results indicate the ability of *Aspergillus niger* to form visible mycelium (diameter of three mm) is detected at 1.83 days, which is in the retailer stage. Thus, quality management in the retailer stage would be critical. For future research, scenario analysis would result in the best condition of the variability, rather for the extrinsic or intrinsic factors. by which this factor conditions are expected could be provided to the bakery industry as references or based evidence for their risk-based quality management. **Significance:** By which this factor conditions are expected could be provided to the bakery industry as references or based evidence for their risk-based quality management.

#### P1-60\* A Growth Prediction and Time-Temperature Criteria Model of Vibrio parahaemolyticus on Korean Raw Sliced Konosirus Punctatus (jeoneo-hoe) at Different Storage Temperatures

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Introduction: *Vibrio parahaemolyticus* is one of the most common causes of seafood-borne illnesses in South Korea and is typically found on raw sliced *Konosirus punctatus (jeoneo-hoe)*. Time-temperature criteria (TTC) specify the storage period that can ensure microbiological safety from a certain growth level at several different temperatures. TTC models can be used to predict the growth of a particular foodborne pathogen for any time and temperature combination and set the maximum allowable organism levels. **Purpose:** In the present study, we developed a TTC model to predict

the growth kinetics of *V. parahaemolyticus* on *jeoneo-hoe* at different storage temperatures.

**Methods:** We investigated the growth of a cocktail of three *V. parahaemolyticus* strains (NCCP13712, NCCP13714, and NCCP14551) from *jeoneo-hoe* stored at different temperatures (5, 10, 15, 20, 28, and 35°C) for up to 36 h. The growth data were fitted to a curve to estimate primary kinetic parameters including lag time (LT), specific growth rate (SGR), and maximum population density (MPD) using DMFit.

**Results:** The model performance was evaluated by calculating bias ( $B_i$ ) (1.0002 to 1.0025) and accuracy ( $A_i$ ) (1.0241 to 1.0431). A secondary model of LT, SGR, and MPD was developed as the four-parameter polynomial model ( $R^2$ =0.8902 to 0.9756). Based on the developed predictive model, we established the time-temperature criteria (TTC) model for determining the time and temperature for ensuring microbiological food safety. According to the model, *V. parahaemolyticus* on *jeoneo-hoe* increases about one log growth ratio (LGR) from the initial cell number after storage for 27.8, 16.2, and 1.8 h at 6, 10, and 20°C, respectively. If one LGR is set as the food safety standard, these time and temperature values are recommended as the appropriate TTC values for *V. parahaemolyticus* on *jeoneo-hoe*. **Significance:** The predictive TTC model can provide a guideline for safe storage parameters that ensure microbiological food safety in the context of certain foodborne pathogens.

#### P1-61\* A Meta-Analysis of Major Foodborne Pathogens in Korean Food Commodities between 2010 and 2016

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Introduction: Food safety is a persistent and challenging issue in South Korea due to the diversity and complexity of foods and food production systems. Prevalence of pathogenic bacteria in food commodities in South Korea has been reported in numerous publications over time. Meta-analysis is a body of summarising statistical techniques whose objective is to synthesize, integrate and contrast the results from a large number of primary studies investigating the same research question.

**Purpose:** In order to prioritise research on microbial hazards, a metaanalysis study was conducted to synthesize frequency and severity information on the presence of foodborne pathogens in Korean food items in South Korea.

**Methods:** Data on the prevalence of bacteria in various food commodities were extracted and analyzed from food incident statistics of the Korea Ministry of Food and Drug Safety (KMFDS) between 2010 and 2016. Prevalence of the most frequently reported pathogens in food categories was used for a logit-transformed proportion as effect size parameterisation, and a number of multilevel random-effect meta-analysis models were fitted to estimate mean occurrence rates of pathogens.

**Results:** As a result of the meta-analysis, *Escherichia coli* O157:H7, *Staphylococcus aureus, Salmonella* Enteritidis, *Bacillus cereus, Vibrio parahaemolyticus, Campylobacter jejuni,* and *Clostridium perfringens* showed the highest level of hazard, and their highest risk foods (risk = frequency × severity) were Kimchi (0.044), raw sliced fish (*fish-hoe*) (0.175), rolled omelet (*gyeran-mari*) (0.201), fried rice (*bokkeum-bap*) (0.086), traditional Korean raw crab marinated in soy sauce (*ganjang-gejang*) (0.334), chicken (0.084), and Korean sausage (*sundae*) (0.123), respectively.

Significance: This meta-analysis highlighted important gaps in knowledge, and may assist food safety authorities in the prioritisation of microbiological hazards and the implementation of essential food safety assurance systems.

#### P1-62 Open Food Safety Model Repository (openFSMR): An Update of the Database in the Service of Risk Assessment Authorities and Food Business Operators

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Introduction: Numerous publications, models and data sets describing the fate of microbial hazards in food already exist. However, this information is widely spread in the scientific literature and usually not available in a harmonized format. In 2015, a web-based, community-driven information portal on predictive food safety models, the "openFSMR", was released to the public. **Purpose:** The aim of this study is to provide an update on those

predictive models that were made available in software tools or in a standard format.

**Methods:** openFSMR is a user-friendly tabular web portal, containing detailed meta-information on each model that is available in a 3<sup>rd</sup> party software tool or as a publicly accessible file in the Predictive

Modelling in Food Markup Language (PMF-ML) format. PMF-ML is a software-independent information exchange format for models specifically designed for predictive microbial models. The openFSMR portal provides now also an opportunity to add new models into the open FSMR-DB through an online Google form template. Entries in open FSMR can be sorted/filtered and/or subgrouped according to metadata like microorganism, environment, type of the model, dependent and independent variables and lastly the software it comes from.

**Results:** Currently openFSMR (https://sites.google.com/site/ openfsmr/) contains information on predictive models for 144 microorganisms including spoilers and pathogens, 151 food matrices, 12 model-types (e.g., growth, inactivation, lethality, boundaries of growth/no growth), 45 independent variables (pH, a<sub>w</sub>, temp, acids, etc) covering 17 public or commercial software tools.

Significance: This updated food safety model repository openFSMR is now a useful tool for risk assessors and/or food business operators that want to identify existing ready-to-use predictive microbial models in a time-efficient way. It is also a dissemination channel for service providers or scientists that want to promote the use of their (new) models or tools.

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#### P1-63 Similarities and Differences between *D*-Values and Time Point Analysis: Critical Comparison and New Design Proposal

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Introduction: It is key for the food industry to understand the ability of a certain process to inactivate foodborne pathogens. D-values from the literature are often used to predict the inactivation of a target pathogen. Meanwhile, critical single time points specific to a certain process-food matrix combination are of great interest. The critical comparison of both data sets is important to design a safe process. Purpose: To compare D-values and inactivation results from single time points within one experimental setup and critically evaluate the results to propose a new design for validation studies, especially when a large group of target microorganisms needs to be evaluated. Methods: Results from heat inactivation in broth from 15 microorganisms (six Salmonella, three Escherichia coli O157, three Listeria monocytogenes and surrogates Enterococcus faecium, E. coli P1 and L. innocua) treated at 60°C for 1, 1.5, 2.5 and 5 min were used to compare D-values and inactivation results from single time points.

**Results:** The *D*-values of the 15 microorganisms were ranked, tested for significance and compared to the ones obtained within each single time point. Our analysis showed a lot of similarities between the two approaches. However, when an inactivation curve deviated from linearity, the point-by-point analysis reflected the true inactivation more accurately.

**Significance:** When a large group of bacterial strains needs to be taken into consideration, the following design is proposed: three fixed time points for all strains and two additional strain-specific time points. In this way both time point analysis and reliable estimation of *D*-values can be performed, allowing to distinguish strains which appear clustered and represents a useful approach when selecting surrogates for industrial validation.

# NOTES

#### THURSDAY, 25 April – 10.00 – 15.30 Poster Session 2 – Antimicrobials; Beverages and Water; Dairy and Other Food Commodities; Food Toxicology; Meat and Poultry; Pathogens; Sanitation; Seafood

#### The Use of Competition Models for Describing P2-01 the Fate of Listeria monocytogenes in Minas Fresh Cheese during Refrigerated Storage

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Introduction: Minas fresh cheese (MFC) is a soft refrigerated product very popular in Brazil, which can be produced with raw or pasteurized milk using either naturally present lactic acid bacteria (LAB) or added selected LAB

Purpose: This study compares the performance of four competition models to describe Listeria monocytogenes growth in MFC as affected by LAB during refrigerated storage.

Methods: Four treatments of MFC were prepared from raw or pasteurized milk with or without the addition of selected LAB with known anti-listerial activity. Cheeses were analyzed for LAB and L. monocytogenes counts throughout cold storage (7±1°C). Microbial competition models, including the simplified Lotka-Volterra and three Jameson-effect variants (simple; using the interaction parameter gamma, and considering the total maximum cell density  $N_{max tot}$ ), were used to describe the simultaneous growth of L. monocytogenes and LAB.

Results: The Lotka-Volterra model represented L. monocytogenes growth better in treatments using raw milk, although it presented the poorest fit among the four competition models in pasteurized cheeses. The three Jameson-effect models adequately described the simultaneous L. monocytogenes and LAB growth in each cheese treatment (P<.0001). The Jameson-effect models estimated that addition of anti-listerial LAB reduced L. monocytogenes growth rates (from 0.0184 to 0.0078 In CFU/h and from 0.0633 to 0.0350 In CFU/h for treatments with and without LAB addition, respectively). The Gamma-Jameson-effect model yielded a better fit for cheeses without selected LAB, resulting in lower goodness-of-fit measures for L. monocytogenes growth. On the other hand, parameters from the simple and  $N_{max tot}$ -Jameson-effect model were the easiest to optimize and provided a better representation of the treatments containing added LAB.

Significance: Competition models based on multi-species experiments demonstrate a more accurate representation of microbial community dynamics in cheese. The variants of the Jameson-effect model for MFC will prove valuable for future risk assessments to predict L. monocytogenes concentrations and assist in managing listeriosis risk.

#### P2-02 Fate of Salmonella enterica and Listeria monocytogenes in the Pulp of Eight Exotic **Tropical Fruits**

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Introduction: There is a growing trend of consumption of exotic tropical fruits due to their functional properties and distinct flavors. However, data on the incidence and/or behavior of foodborne pathogens such as Salmonella enterica and Listeria monocytogenes in these fruits are limited.

Purpose: This study aimed to assess the fate of S. enterica and L. monocytogenes in the pulp of eight exotic tropical fruits stored at different temperatures.

Methods: Pulps of jenipapo, umbu, maná, cajá-manga, physalis, feijoa and cupuaçu (pH<4.5) and abiu (pH>4.5) were evaluated for the fate of S. enterica and L. monocytogenes. Three S. enterica and two L. monocytogenes strains were used as pathogen pools. Low and high acid fruit pulps were inoculated (10<sup>3</sup> or 10<sup>6</sup> CFU/g, respectively) and stored at 10, 20, 30 and 37°C for up to 12 h (survival assessment) or 7 days (growth assessment), respectively. Pathogen

counts and growth potentials were determined at all temperatures for comparison of exotic tropical fruits.

Results: S. enterica and L. monocytogenes were able to grow in abiu (low acid fruit) at 20, 30 and 37°C, but presented reduced growth rates at 10°C. On the other hand, high acid fruits did not favor the S. enterica and L. monocytogenes development, since growth was reduced or even not detected at all temperatures. Only in physalis and feijoa pulps were S. enterica and L. monocytogenes able to keep the initial inoculum concentration after 12 h. Regarding growth potential for both pathogens, the only abiu, maná, cupuaçu and cajámanga presented significant differences between means for each temperature (P<0.05).

Significance: The assessment of S. enterica and L. monocytogenes growth potential in exotic tropical fruits is highly relevant in order to derive measures to protect public health, including the possibility of creating communication strategies aiming to educate consumers, as well as to allow the development of regulations for subsistence and large-scale commercial exploitation of these fruits.

#### P2-03 **Application of Plasma Activated Water for Decontamination of Alfalfa and Mung Bean** Seeds

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Introduction: Microbial contamination of fresh produce is a major public health concern, with the number of microbial disease outbreaks increasing in recent years. The consumption of sprouted beans and seeds is of particular concern, as these foodstuffs are generally consumed raw, and are produced in conditions favorable to the growth of pathogenic microorganisms if they are present in seeds prior to sprouting.

Purpose: This work aimed to evaluate the activity of plasma activated water (PAW) as a disinfecting agent in alfalfa (Medicago sativa) and mung bean (Vigna radiata) seeds during seed soaking.

Methods: Each seed type was inoculated with Escherichia coli O157, E. coli O104, Listeria monocytogenes or Salmonella Montevideo, and treated with PAW for one hour, three hours or overnight. Seeds treated for one and three hours were subsequently soaked in water to replicate commercial practices. Microbial counts for each pathogen were determined after treatment and soaking.

Results: Observed reductions in alfalfa seeds range from a 0.86-log reduction in Salmonella Montevideo concentration after overnight treatment, to a 1.13-log decrease in E. coli O104 levels after three hours of treatment and soaking. For mung bean seeds, observed results range from a 1.17-log decrease in the levels of Salmonella Montevideo after overnight treatment, to a 2.48-log reduction in the concentration of E. coli O104, after three hours of treatment followed by seed soaking

Significance: These results demonstrate the potential for PAW to be used in the inactivation of pathogenic microorganisms on sprouted seeds and beans, ensuring the safety of the products for consumers.

#### P2-04\* The Potential Use of Endophytic Bacteria from **Tropical Fruits to Control Foodborne Pathogens**

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Introduction: Listeria monocytogenes and Cronobacter sakazakii play an important role in food safety. They both can survive for a long period of time in low-moisture foods and can potentially grow in products such as reconstituted powdered infant formula and a wide variety of other RTE foods. Endophytic bacteria are those that reside inside surface-sterilized plants and have no visible harmful effects on the plants. Bacterial endophytes from fruits and vegetables can assist plant cells by providing beneficial antimicrobial compounds and protecting the host organism from plant pathogens. These biological compounds could be used in agriculture, medicine, as well as in the food industry.

Purpose: The purpose of this study is to isolate bacterial endophytes from tropical fruits and to examine their ability to reduce and/or inhibit the growth of L. monocytogenes and/or C. sakazakii.

Methods: Tropical fruits such as papayas, dragon fruits, sugar apples, lychees and guavas collected from retail stores in Canada were processed under sterile conditions. Subsequently, bacterial endophyte libraries were constructed from these tropical fruits.

In addition, isolated bacterial endophytes were screened for their inhibitory activity against L. monocytogenes and C. sakazakii by using a growth inhibition test.

**Results:** The results demonstrated that bacterial endophytes can be cultured In Vitro from tropical fruits. In addition, *Bacillus* spp. and *Pantoea* spp. demonstrated antagonistic activity against *L. monocytogenes* and *C. sakazakii*. Future work will include competitive challenge studies with these foodborne pathogens and our isolated bacterial endophytes in food products.

Significance: Bacterial endophytes isolated from tropical fruits have the potential to be used as biocontrol agents to control foodborne pathogens such as *L. monocytogenes* and *C. sakazakii.* 

#### P2-05\* A New Weapon against *Staphylococcus aureus*: Antimicrobial and Anti-Biofilm Activity of a Novel and Rapid Antimicrobial Peptide

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Introduction: Staphylococcal food poisoning – caused by certain enterotoxigenic staphylococci – is one of the most common foodborne diseases worldwide. In this context, bacterial biofilms on food processing plant surfaces can represent critical sources of contamination, being more resistant to cleaning and disinfection procedures. Therefore, it is important to prevent and control biofilms in food facilities. In the last few years, antimicrobial peptides (AMPs) have emerged as biofilm control tools. The recently developed AMP 1018-K6 (derived from the innate defense regulator peptide IDR-1018) showed significant bactericidal and anti-biofilm efficiency against *Listeria monocytogenes* and remarkable stability in different environmental conditions.

**Purpose:** The present study aimed at investigating the bactericidal efficiency of 1018-K6 against planktonic and biofilm-embedded cells of *Staphylococcus aureus* ATCC 35556 (a well-known strong biofilm producer) in comparison with IDR-1018.

**Methods:** The minimum inhibitory concentration (MIC) and the minimum bactericidal concentration (MBC) were determined by the standard broth microdilution method following recommendations of the Clinical & Laboratory Standards Institute. Inhibition of biofilm formation was assessed using crystal violet staining in 96-well microtiter plates, after incubation of *S. aureus* cells in the presence or in absence of the peptide for 24 h. The kinetics of action of 1018-K6 (80 µM) against 24-hour biofilms was performed using the Calgary Biofilm Device.

**Results:** The peptide showed an impressive rapidity of action, being able to cause the complete killing of established staphylococcal biofilms within the first 10 min. Furthermore, it was able to prevent biofilm formation and had potent bactericidal activity against planktonic cells.

Significance: The remarkable antimicrobial and anti-biofilm properties of 1018-K6, as well as the uncommon stability at different temperature and pH conditions, make it a promising candidate for applications in food safety and quality control, e.g., antimicrobial packaging systems and solutions for cleaning and disinfecting.

#### P2-06 Effect of Active Packaging in Extending the Shelf-life of Bread and Cheddar Cheese

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Introduction: The combination of natural antimicrobials with novel preservation techniques such as active packaging has been suggested as a means of extending shelf life and satisfying consumer demands for products without synthetic preservatives. **Purpose:** To investigate the effect of active packaging film containing the encapsulated natural antimicrobial carvacrol in halloysite nanotubes (HNTs) as an alternative strategy for extending the shelf life of cheese and bread.

**Methods:** Preservative-free bread loaves and shredded cheddar cheese were spray inoculated with a cocktail of three *Penicillium commune* strains at a 1:1:1 ratio to a final concentration of 100 conidia/g.

Each bread loaf was individually packaged using an active HNT film (containing one percent carvacrol) and a commercial control film (based on biaxially-oriented polypropylene, BOPP). Packed loaves were stored at 20°C and monitored for mold development. Cheese samples were individually packed and stored at 8°C for 60 days. Four different packaging types based on commercial BOPA-CPP film were used: i) air as headspace, ii) modified atmosphere (MA) as headspace (30% CO<sub>2</sub>/70% N<sub>2</sub>), iii) two percent carvacrol HNT film and air as headspace, iv) two percent carvacrol-HNT film and MA as headspace. Samples were visually inspected for the presence of fungal colonies with a diameter ≥1mm and the time to colony detection was noted. Each trial was repeated three times.

**Results:** The bread had 14 days of shelf life when packed in carvacrol-containing film and 7.8 days of shelf life when packed in the control film. Cheese with air as headspace had visible colonies within 14 days of storage and within 50 days of storage with MA as headspace. There was no significant difference in time to colony detection between cheese samples packed in carvacrol-containing film or commercial film.

**Significance:** The use of active packaging containing carvacrol can significantly extend the shelf life of bread. MA packaging can significantly extend the shelf-life of shredded cheddar.

#### P2-07 Selection of an *Escherichia coli* Gentamycinresistant Variant in Biofilms Exposed to Polyhexamethylene Biguanide

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**Introduction:** Antibiotic resistance is one of the most concerning issues facing modern medicine. Some biocides have demonstrated the potential to select resistance to antibiotics in bacteria, but data are still very scarce and it is important to better identify the molecules concerned and the underlying mechanisms.

**Purpose:** This study aimed to assess the potential of polyhexamethylene biguanide (PHMB), a widely used biocide in a variety of sectors, to select resistance toward clinically important antibiotics in *Escherichia coli* grown in biofilms.

**Methods:** Three *E. coli* strains isolated from pig industries and one collection strain were selected. Biofilms were grown on inox coupons and then exposed daily to sublethal concentrations of PHMB for 10 days. Antibiotic-resistant mutants were then selected and characterized phenotypically (resistance profiles against 14 antibiotics and three biocides, growth rate) and genotypically to identify mechanisms of resistance.

**Results:** Biocide exposure led to the selection of an *E. coli* variant with stable resistance to gentamycin (16-fold increase in MIC compared to wild type). This was also associated with a 40% decrease in the growth rate of the variant. Susceptibility to gentamycin was recovered in presence of the efflux pump inhibitor phenylalanine-arginine  $\beta$ -naphthylamide (PA $\beta$ N) in the variant. Moreover, *acrA*, a gene encoding an AcrAB-ToIC multidrug efflux pump module, was overexpressed fivefold in the variant. Sequencing and comparison of wild type and variant whole genomes are currently in progress to further decipher the underlying mechanism.

**Significance:** Data collected demonstrated the potential of PHMB to select antibiotic resistance in the model bacteria *E. coli*, and the involvement of the AcrAB-ToIC system in the cross-resistance observed. Such observations support the central role of multidrug efflux pumps in cross-resistance between biocides and antibiotics in gram-negative bacteria.

#### P2-08 Evolution of Antibiotic and Biocide Resistance of *Listeria monocytogenes* from Diverse Ecological Niches Following In Vitro Exposure to Biocides and Disinfectants

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**Introduction:** The LISTADAPT project belongs to the European Joint Program One Health. Its aim is to study the adaptive traits of *Listeria monocytogenes* to its diverse ecological niches. *L. monocytogenes* is a ubiquitous bacterium that is an important cause of bacterial foodborne infections in Europe with a significant increase in the prevalence of listeriosis cases for a decade. The capacity of some strains to adapt to the environmental conditions found in the food industry makes the production of high quality, safe food a major challenge.

**Purpose:** Part of this project concerns the determination of antimicrobial resistance profiles of a large panel of *L. monocytogenes* strains from various ecological niches and their ability to adapt after biocide exposure.

**Methods:** Antimicrobial resistance profiles of 200 *L. monocytogenes* strains were investigated and analyzed to determine their minimum inhibitory concentration (MIC) for a series of representative antibiotics (14) and biocides (eight) using a standard broth dilution method. Assessment of the ability to adapt to one or two biocides after repeated daily exposure to sublethal concentrations and to develop cross-resistance against antibiotics for some illustrative *L. mono-cytogenes* strains was also studied.

Results: Results obtained from the first 97 strains revealed slight differences in antimicrobial resistance profiles. Concerning the biocide resistance profiles of these *L. monocytogenes* strains, we can observe a greater variation in MIC values for quaternary ammonia as didecyl dimethyl ammonium chloride, for which MICs may vary by a factor of sixteen between these strains. Moreover, there is one strain resistant to meropenem and five resistant to tetracycline. Significance: Comparison of antimicrobial resistance profiles will enable discovery of the existence of resistance specificity according to the origin of the strain (food, human, animal and environment) and will be combined with genomic analyses (done in other work packages) to identify genetic determinants involved in the adaptation of *L. monocytogenes* to the different ecological niches.

#### P2-09 Chitosan and Its Phenolic Conjugates as Antibacterial and Antibiofilm Drugs against Pathogenic Bacteria

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**Introduction:** Emergence of antibiotic resistance in several pathogenic bacteria leads to a need for discovery of alternative strategies to treat infections.

Purpose: The aim of this study was to evaluate the antibacterial and antibiofilm potential of phenolic conjugated chitosan against Pseudomonas aeruginosa and Listeria monocytogenes. Methods: In the present study, we have evaluated the antibacterial and biofilm inhibiting properties of chitosan and its phenolic conjugates against pathogenic bacteria such as P. aeruginosa and L. monocytogenes. The phenolic compounds used for the conjugation with the chitosan were caffeic acid (CA), ferulic acid (FA) and sinapic acid (SA). The resulting conjugates were designated as CCA, CFA and CSA, respectively. The antibacterial and antibiofilm activity was evaluated by the assay of minimum inhibitory concentration (MIC), minimum bactericidal concentration (MBC), biofilm inhibitory concentration (BIC) and biofilm eradication concentration (BEC). Results: The MIC, MBC, BIC and BEC values of phenolic conjugated chitosan are lower than the unconjugated chitosan against the P. aeruginosa and L. monocytogenes. The values of phenolic conjugated chitosans are as follows: MIC, 0.05 to 0.33 mg/mL; MBC, 0.30 to 0.45 mg/mL; BIC, 0.42 to 0.83 mg/mL; and BEC, 1.71 to 3.70 mg/mL. The mechanism of biofilm inhibition by phenolic conjugated chitosan in *P. aeruginosa* and *L. monocytogenes* is a result of increasing the membrane permeability of the cells embedded in the biofilm matrix. Furthermore, we observed the subinhibitory concentration of phenolic conjugated chitosan significantly reduced the adhesion of the cells. The most effective material against *P*. aeruginosa and L. monocytogenes is the CCA, followed by CFA and CSA.

**Significance:** Based on the above study, it is concluded that the phenolic conjugated chitosan should be considered as potential antibacterial and antibiofilm drugs against foodborne pathogenic bacterial infections.

#### P2-10 A Natural Anti-Salmonella Solution to Preserve Poultry Fat, a Key Raw Material for the Pet Food Industry

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Introduction: Only half of the products generated from livestock animals are consumed by humans. The pet food industry enables most of the unconsumed part to be used as byproducts. For instance, poultry fat is a key raw material since it contributes to energy requirements and palatability. However, it is highly susceptible to pathogen contamination. Despite the cooking step, *Salmonella* could survive the process, in addition to the occurrence of post-cooking contamination. Additional preservatives are therefore needed to ensure microbial safety. They need to be natural in order to meet growing demand from the market.

**Purpose:** To show the performance of a blend of natural antimicrobial compounds to eliminate *Salmonella* from poultry fat without modifying pet food palatability.

Methods: One kg of poultry fat was inoculated with a cocktail of six Salmonella serovars (Typhimurium, Mbandaka, Senftenberg, Infantis, Montevideo, and Anatum) at 6 log CFU/g at 30°C. After adding 0.05% natural antimicrobials or 0.1% phosphoric acid as a reference, microbial counting on XLD agar and Salmonella detection on RAPID'Salmonella in 125 g was carried out for 14 days. Palatability of fat containing the antimicrobial solution was evaluated by pairwise comparison trials in cat and dog panels.

**Results:** A Salmonella cocktail inoculated into poultry fat is stable over time to around 6 log CFU/g while with the antimicrobial solution, Salmonella is uncountable after five hours (<1 log CFU/g) and undetectable in 125g after one day. The reference sample becomes uncountable only after nine days (detectable up to 14 days). In addition, while each component of the antimicrobial mix tested alone at 500 ppm results in a significant decrease in palatability (P<0.01), the natural blend at the same concentration doesn't significantly affect palatability.

**Significance:** Our mix is a natural solution that ensures the microbial safety of poultry fat without affecting the palatability of pet food.

#### P2-11 Role of Rhizobacteria in Modulating Plantpathogen Interactions for Improved Food Safety

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Introduction: Salmonella contamination of plants can involve leaf colonization and internalization, depending on flagellar and virulence genes. Preharvest control is difficult, making rhizobacteria that disrupt *Salmonella* persistence a potential biocontrol.

**Purpose:** Evaluate the role of specific Salmonella traits in lettuce leaf colonization and internalization and the subsequent effects of rhizobacteria as a biocontrol.

**Methods:** Roots of lettuce plants (*Latuca sativa*) were inoculated with a live culture of *Bacillus subtilis* UD1022 (10<sup>8</sup> CFU/ml) and maintained in a bio-growth chamber for 48 hr. Plant leaves were inoculated (nine log CFU/ml) with *Salmonella* Typhimurium (ATCC 14028) or its mutants:  $\Delta invA$  (*Salmonella* pathogenicity island-1; SPI-1),  $\Delta hilD$  (SPI-1 transcriptional regulator),  $\Delta sseB$  (SPI-2),  $\Delta fliC$  and  $\Delta fljB$  (flagellin subunits), and control S. Newport. Leaf samples were collected on days zero, one, three, five, and seven, and processed to enumerate populations, internalization, stomatal closure (width of apertures), and plant immune response (semi-quantitative PCR). Data were analyzed using one-way ANOVA (*P*<0.05).

**Results:** Salmonella populations were significantly (P<0.05) reduced in all UD1022 treated groups by day seven except  $\Delta fliB$  and  $\Delta invA$ . The  $\Delta fliB$  and  $\Delta invA$  showed similar survival (4.3 and 2.7 log CFU/g, respectively) to their untreated-control groups. Other mutants  $\Delta fliC$ ,  $\Delta hilD$ , and  $\Delta sseB$  were reduced to undetectable levels in UD1022 treated plants compared to controls, by day three. Salmonella Typhimurium and Salmonella Newport populations decreased (2.2 to 2.8 log CFU/g) significantly (P<0.05) in UD1022 treated compared to untreated plants. Salmonella internalization was not detected in plants after UD1022 treatment and these had significantly (P<0.05) higher

stomatal closure rate (aperture width=2.34 µm) by day one, compared to controls (8.5 µm). However,  $\Delta hilD$  and  $\Delta sseB$  mutants were not internalized in both treated and untreated controls (mean=1.49 and 1.26 µm), suggesting that these genes are vital for Salmonella internalization in lettuce.

Significance: Salmonella survival on plants is complex, involving flagella. Biocontrol UD1022 reduces Salmonella populations and internalization suggesting its utilization as a preharvest biocontrol.

#### P2-12 Antimicrobial and Cinnamoyl Esterase Activities of Lactic Acid Bacteria Isolated from Infant Feces

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Introduction: Lactic acid bacteria (LAB) having antimicrobial activities are very useful for enhancing food safety because they are natural inhibitors of pathogens. Cinnamoyl esterase (CE) is considered to be a prime constituent of cereal-based functional foods because it maximizes health benefits by improving the bioavailability of ferulic acid.

**Purpose:** The objective of this study was to screen and characterize LAB having antimicrobial and high CE activities.

Methods: To investigate CE activity, a total of 219 representative lactobacilli were obtained from fecal specimens of healthy Korean infants. The specimens were tested for the presence of lactobacilli with CE activity by plating on MRS agar containing ethyl ferulate. The strains that formed a clear zone were quantitatively analyzed using ultra-performance liquid chromatography. The selected isolates possessing high CE activity were evaluated for antimicrobial activity. **Results:** The screening results revealed that 12 out of the 219 strains assessed had high CE activities (>1mM, ferulic acid production). Six isolates (B1, G2, N2, E2, I2 and I3) were identified as *Lactobacillus gasseri* and the other 6 isolates (I1, I4, I5, J2, J5 and N1) were identified as *L. fermentum*. Among these, J2 showed the highest CE activity (>2.1 mM), which corresponded to an ethyl ferulate-to-ferulic acid conversion rate of nearly 70%. All the 12 isolates demonstrated strong antibacterial activities against *Escherichia coli* O157:H7. *L. gasseri* G2 and N2, and *L. fermentum* 1 exhibited broad inhibitory activities against three foodborne pathogens including *E. coli* O157:H7, *Salmonella* Enteritidis and *Bacillus creus*.

Significance: The probiotic LAB having antimicrobial activities could be considered as safe and natural preservative for food. In particular, CE activity can lead to synergistic effects, such as antioxidant effects, by increasing the bioavailability of ferulic acid.

#### P2-13 Antimicrobial Resistance Profiles of Lactic Acid Bacteria Isolated from Fermented Meat Products of European Origin

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Introduction: LAB are a diverse group of gram-positive anaerobic non-spore forming bacteria which play a critical role in food processing and are often added as starter cultures in a wide range of fermented products. Although it is recognised that the food chain plays a significant role in the complex pathway of antimicrobial resistance (AMR) transmission to humans, relatively little is known about the role of the food chain and how much food contributes to the burden of AMR.

**Purpose:** The aim of this study was to determine the AMR profiles of LAB isolates obtained from fermented food products of European origin.

**Methods:** LAB were isolated from chorizo, ham, salami and sausage and were subjected to 18 different antibiotics using the disk diffusion method. The diameter of inhibitory zones was measured after incubation at 37°C under anaerobic conditions.

**Results:** Antibiotic susceptibility profile of 40 LAB isolates revealed that 100% of isolates were resistant to nalidixic acid, 93% to vancomycin, 90% to cefoxitin, 88% to colistin and 83% to polymyxin B. Over 70% percent of isolates tested show sensitivity to streptomycin (82%), bacitracin (75%), erythromycin (73%), ampicillin (73%), imipenem (73%) and rifampicin (70%). In terms of multiple resistance to antibiotics, our results show that 38 of 40 (95%) of isolates were resistant to at least four antimicrobial agents and the highest AMR index recorded in our study was 0.89 while 0.11 was the lowest value recorded.

Significance: Determination of AMR profiles of LAB as the dominant microbial community in fermented products is highly important in terms of identifying and understanding mechanisms of persistence and spread of resistance genes within the food supply chain microbial community. The results obtained here highlights the need for continuous monitoring and identification of practices that could be reviewed in order to control and prevent further spread of AMR in the food supply chain.

#### P2-14 Evaluation of White Wines' Origin by Raman Spectroscopy Coupled with Chemometric Methods

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Introduction: France is the largest exporter of wine in the world. The export turnover is estimated at 8.7 billion euros in 2017 for 13 million hectoliters sold. This lucrative business pushes scammers to increase the value of some low-end wines, leading to losing 1.3 billion euros each year for the European Union's wine and spirits companies. The control of wine quality is performed by analytical methods such as infrared, NMR or HPLC. Nevertheless, the complexity of the wine matrix and the presence of water and ethanol interfere with the determination of the other wine molecules. Consequently, the development of sensitive, fast and automated procedures remains a real need for investors and stakeholders in this area. **Purpose:** Our study aims to evaluate the ability of Raman spectroscopy to determine the origin and the grape variety of wines based on its Raman spectral fingerprint.

Methods: Seven white wines have been studied: Cabernet Val de Loire (Anjou), Chardonnay (Bourgogne), Riesling (Alsace), Gewurztraminer (Roumanie), Muscadet (Sèvre-et-Maine sur Lie), Sauvignon blanc (Bordeaux) and Sauvignon blanc (Val de Loire). **Results:** The obtained results show that white wine has a rich spectral signature which reflects its molecular composition and the spectra presented very characteristic bands. The principal component analysis and discriminant analysis showed perfect discrimination between the different wines. The validation of the database showed very good discrimination between all samples. Nevertheless, confusion was observed between the two Sauvignon despite their different origins. Raman spectroscopy allows the rapid identification of the grape variety. Nevertheless, a large number of samples must be analyzed in order to evaluate the industrial viability of this technique (variability between years, batches). New chemometric models must also be developed to improve discrimination between wines. **Significance:** The final goal is to propose a portable device able to provide reliable and fast analysis for wine companies.

#### P2-15 Evaluation of Alternative DNA Extraction Protocols for Discriminating *Legionella* Viable Cells

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**Introduction:** Water quality is a real concern for food industries and can put food production plants at risk. Compared to the traditional culture method (ISO 11731), real-time PCR methods can significantly shorten the time-to-results for *Legionella* detection in water samples from three to 10 days to few hours. However, the drawback of molecular methods remains its discrepancy with traditional culture-based methods due to the detection of free DNA, viable but nonculturable cells, and dead cells.

**Purpose:** The GeneDisc *Legionella* method in clean water samples and two alternative DNA extraction protocols, including a smaller test portion (A) or an additional centrifugation step (B), were compared to the culture method (ISO TS 13171).

**Methods:** Six different clean water samples were analyzed. After homogenization, sample sizes of 200 ml were analyzed by the culture method and the three GeneDisc protocols.

**Results:** The results showed that the reference method gave the presence of *Legionella* in three out of six samples. The alternative protocol A, based on a reduced test portion, produced comparable results to the reference method for presence/absence, while the current GeneDisc method for *Legionella* detection in the clean water sample and the alternative protocol B detected presence in all six samples.

**Significance:** This new protocol A allows significant reduction of the number of false positives due to detection of dead cells and/or free DNA and can produce results comparable to the culture method in as short as two hours. This new GeneDisc *Legionella* method enables real-time monitoring of *Legionella* in clean water samples.

#### P2-16\* Culture-Dependent and Independent Analysis to Explore Microbiota of Robiola Di Roccaverano Production

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Introduction: Robiola di Roccaverano is a typical raw milk cheese from Piedmont (northwest Italy) which has received the PDO certification. Technical policies establish that during its manufacturing process, the addition of a commercial starter is not permitted. Thus, cheese-makers employ a natural milk starter (NMS) coming from acidification of raw milk. Quality and safety of cheese made with raw milk are mainly governed by microbiota inhabiting milk and, in this case, also from NMS.

Purpose: A comprehensive investigation of Robiola di Roccaverano's artisanal manufacturing process, monitoring each production step. Methods: Samples of raw milk, NMS, curd, five day ripened cheese and 15 day ripened cheese were collected from one artisanal cheese factory. Initially, the matrices were subjected to classical microbiology analysis using selective growth media to assess the presence of pathogenic bacteria and to isolate LAB. DNA from LAB strains were extracted and analyzed by RAPD-PCR, species-specific PCR and 16s rRNA sequencing. A metagenomic approach was also performed on DNA directly extracted from each sample. V3-V4 region of mitochondrial DNA was analyzed by Illumina MiSeq instrument following the manufacturer's guidelines.

**Results:** Each production step can be considered safe, because pathogens were not isolated. From the 175 LAB strains isolated and characterized by culture-dependent approaches, it was shown that *Lactococcus lactis* (54.86%) was the most abundant microorganism in Roccaverano's production, followed by *Leuconostoc mesenteroides* (26.86%). Less abundant genera of LAB were *Enterococcus* spp. and *Lactobacillus* spp. The metagenomic analysis confirmed whatever was reported by culture-dependent approaches and highlighted how milk samples show most variability compared to other matrices, with a high prevalence of non-LAB in which *Pseudomonas* spp. was predominant.

Significance: This is the first report in which culture-dependent analysis and metagenomic approaches were applied to monitor the microbial evolution during the manufacturing process of Robiola di Roccaverano.

#### P2-17 Assessment of the Microbial Quality of Membrane-processed Infant Formula

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Introduction: The thermal or membrane processing for infant formula (TOMI) project is concerned with the production of powdered infant formula by utilising a novel process, cascade membrane filtration, as an alternative to thermal processing. Thermal processing of infant formula is currently used to ensure a microbiologically safe product but there is potential for generation of undesirable Maillard reaction products during high intensity thermal processing. Purpose: To investigate the ability of a cascade membrane filtration system to produce an infant formula product with equivalent or improved microbial quality compared to standard formula. Methods: A pilot scale membrane filtration system was challenged with sterile media and non-pathogenic Lactobacillus delbrueckii subsp. bulgaricus that are naturally resistant to the antibiotic erythromycin. In line samples from the feed, permeate and retentate after filtration were selectively (with antibiotic) and non-selectively plated to determine exclusion efficacy of the filters. Results: The samples revealed a significant level of microbial contamination in the system and the passage of L. delbrueckii in low numbers through the membranes in the tested pilot scale configuration. In-line modifications to the pilot scale system improved the exclusion efficacy and the overall microbial quality of the permeate and retentate. However, these results highlight that microbes and potentially pathogens with similar characteristics may bypass the membrane if the system configuration is not optimised. Significance: TOMI has the potential to radically improve the quality of infant formula but further work on understanding the dynamics of microbial exclusion efficacy with the pilot scale membrane configuration is required.

#### P2-18 Microbiological Safety Indicators of Canastra Cheese in Brazil during and after the Ripening Period

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Introduction: Canastra Artisanal Minas Cheese is made from raw milk by small farmers in the Serra da Canastra region. The production process employs fermentation by an endogenous culture called "pingo", originating from the whey collected from the previous day's production. The use of raw milk is a risk factor for food safety and potential foodborne pathogens should be controlled during the 22-day ripening period required by legislation.

**Purpose:** This work analyzed the microbiological safety indicators of Canastra cheese from three rural properties during ripening and cheeses from 78 rural properties after ripening.

Methods: Total coliforms, Escherichia coli and coagulase-positive Staphylococcus counts were performed on Petrifilm plates (3M). The detection of Salmonella spp. and Listeria monocytogenes was performed using ISO6579:2002 and ISO11290-1:1996/(A)1:2004 methods. Enterobacteriaceae counts were determined by the APHA method 9.62:2015. The pH analyzes were performed according to the IAL 017-IV method and the water activity  $(a_w)$  in an AquaLab analyzer. **Results:** The study performed during the ripening period showed that the pH changed throughout time and all properties reached satisfactory microbiological limits before 22 days. However, the study performed with cheeses collected after the ripening period showed that only 38% of samples were satisfactory for all requirements of the legislation. In this study, pH and a<sub>w</sub> did not significantly influence whether the samples would be classified as satisfactory according to the legislation (P<0.05), suggesting no influence of these parameters on the microbiological indicators. No sample was contaminated with Salmonella, and L. monocytogenes was found in one sample. Significance: The high number of noncompliances and the high counts of E. coli, total coliforms and Staphylococcus in some samples after ripening indicates a need to implement or improve hygienic and sanitary conditions in the production, distribution or storage of Canastra cheese.

#### P2-19 Nisin Production by a Wild Strain of *Lactococcus lactis* Subsp. *Lactis* by Diacetylactis (SBR4) Obtained from a Dairy Environment

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Introduction: Lactococcus lactis subsp. lactis by diacetylactis has major importance in he dairy industry due to the production of aromatic compounds: diacetyl and acetoin. Some strains can present an additional beneficial feature: bacteriocin production. **Purpose:** This study aimed to characterize the bacteriocinogenic

potential of *L. lactics* subsp. *lactis* by biacterylactis strains obtained from dairy production systems.

**Methods:** A panel of 15 *L. lactis* subsp. *lactis* by biacterylactis was subjected to PCR to detect bacteriocin-related genes (nisin, lacticins 481 and 3147, lactococcins A and 972), and further investigated by PCR and sequencing for the nisin operon. Cell-free supernatants (CFS) of nisin-positive strains were tested by the spot-on-the-lawn assay against a panel of 16 targets (*Listeria monocytogenes*, four; *Listeria innocua*, one; *Staphylococcus aureus*. six; *Lactobacillus sakei*, one; *Lactococcus lactis*, four). Further, growth curves of six microbial targets (*Listeria monocytogenes*, two; *Listeria innocua*, one; *Staphylococcus aureus*, two; *Lactobacillus sakei*, one), alone and in the presence of the CFS of nisin producers, were obtained by optical density ( $\lambda$ =600 nm).

**Results:** Eight strains presented positive results only for *nisA*, and only one strain (SBR4) presented the full nisin operon, confirmed by sequencing as similar to nisin Z. Only SBR4 presented inhibitory activity by the spot-on-the lawn assay against the 16 targets. Growth curves of selected targets confirmed the inhibitory activity of the SBR4 strain, indicating its potential for nisin production.

**Significance:** The study has demonstrated the inhibitory potential of a *L. lactis* subsp. *lactis* by diacetylactis strain, SBR4, due to its ability in produce nisin Z. Considering the technological properties of this strain, this inhibitory potential indicates an additional beneficial feature of SBR4 to be considered in the dairy industry. Acknowledgments: CNPq, CAPES, FAPEMIG, FUNARBE.

#### P2-20\* Study of the Prevalence of Salmonella in Poultry Slaughterhouses and Cutting Plants

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Introduction: Salmonella is considered as the most common bacterial pathogen causing foodborne outbreaks. Most of the human salmonellosis cases are attributed to meat products and eggs. To prevent Salmonella in the poultry industry, the Salmonella status of broiler flocks is determined before slaughter in view of the logistics of slaughter in the EU. This means that flocks with a Salmonella-free status are slaughtered first each day followed by flocks with a positive status. However, this can only be efficient when the slaughter line and equipment before starting slaughter activities is Salmonella-free and thus cleaned and disinfected efficiently. This study was conducted to help the participating poultry slaughterhouses eliminate Salmonella contamination.

**Purpose:** This study aimed to investigate the possible contamination of the first slaughtered flock by sampling the equipment before starting slaughter activities.

Methods: A thorough sampling in the production environment of three broiler and two laying hen slaughterhouses was performed. Different swab and water samples were taken from the hanging area, the scalding tank, the plucking machine, the evisceration machines and the cutting line. Each slaughterhouse was visited twice. Salmonella detection on the samples was based on the ISO 6579 standard. The presumptive colonies were confirmed by Salmonella PCR. Results: From the 652 samples of the slaughter equipment collected after cleaning and disinfection in the five slaughterhouses 43 (6.6%) were positive for Salmonella. The prevalence of Salmonella in slaughterhouse A, B, C, D, E was 24 of 136, four of 134, eight of 130, three of 128, and four of 124, respectively. Among the five sampling stages, the results show that the plucking machine (26 of 150) was the most contaminated machine, especially the plucking fingers. Significance: This finding helps the elimination of Salmonella in the poultry slaughterhouse, especially preventing Salmonella contamination at the start of the slaughter activities.

#### P2-21 Hygienic Design: The Status of the Art in the Italian Cheese Plant Industries

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**Introduction:** Hygienic design (HD) is a common international voluntary standard manage by two main international association Ehedg in EU and 3A in the USA. Is this standard common, or is it used only by a few food industry sectors?

Purpose: We study the HD implementation in cheese food industries. Methods: We performed during 2018 food safety audits (n=60) linked to national and international food safety standards using our own developed checklist with a specific section made only for HD assessment. This section includes 15 requirements: five each for plant design, equipment design and construction materials design. Results: We observed cheese companies in Italy, the United Kingdom and France. Only 40 of the cheese companies observed are exporting their cheeses abroad (North America, China and Japan). About this 40, only 30 have planned a specific design for the plant. Of these 30 plants, only 12 have types of equipment in HD in some production areas and only two of these had a fully HD for pieces of equipment and construction materials. These two use glass containers for cream cheese with continuous production (24 hours). These two are the EU market leader brands. All the others do not know the Ehedg or 3A standard and what they have in place is random and unplanned, perhaps thanks to proposals from suppliers or consultants.

Significance: HD is not known for its complete implementation. Those who use it gain undoubted competitive advantages in the phases of maintenance and cleaning. In other food industries, for example in a meat plant, the situation is completely opposite. The answer to this discordance is linked to the standard requirements requested for export. HD implementation awareness is an opportunity to grow in a global market.

#### P2-22\* The (FAO/WHO) International Food Safety Authorities Network (INFOSAN): Looking inside This Global Community of Practice

#### CARMEN SAVELLI

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Introduction: Since 2004, INFOSAN has aimed to halt the international spread of contaminated food, prevent foodborne disease outbreaks, and strengthen food safety systems globally. However, INFOSAN has never been examined as a functional community of practice and its value, according to members, has not been determined in a systematic or rigorous way.

Purpose: A three-phase, mixed-method study is ongoing to explore the experiences of INFOSAN members with respect to their participation in network activities in order to improve global food safety and prevent foodborne illness. Details related to phase one are presented here and relate specifically to how the INFOSAN Community Website (ICW) is being used to support network activities. **Methods:** Registration data has been collected for each member on the ICW since 2012. Anonymized data was analyzed using descriptive summary statistics, and stratified by a number of variables including type of member, geographic region, and membership duration. Records of access to and participation on the ICW were also downloaded.

**Results:** As of January 2019, the ICW has 646 registered members from 188 WHO Member States; 298 (48%) of 622 are female. The majority of INFOSAN members (334 (52%) of 646) have been registered on the INFOSAN Community Website for three or more years and 327 (51%) of 646 have logged on to the ICW within six months. However, a relatively limited number of active members from a select group of member states contributed the majority of information shared on the ICW.

Significance: The data provide objective, foundational information about the engagement of all members and will be triangulated with data from phases two and three of the ongoing study to determine if members' reported attitudes and experiences reflect online behaviours. This information can be used by the INFOSAN Secretariat to increase active participation and improve international information exchange to mitigate the impact of food safety emergencies.

#### P2-23\* Utilisation of Tools to Facilitate Cross-Border Communication during International Food Safety Events, 1995–2019: A Realist Synthesis

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Introduction: Efficient communication and coordination between countries are needed for prevention, detection and response to international food safety emergencies. While communication tools/ networks/systems (e.g., RASFF, INFOSAN, etc.) exist, current evidence suggests that they may only be effective within certain contexts and cover certain geographic areas.

**Purpose**: This study explores the mechanisms of how and in what context such tools are effective at facilitating international communication and coordination to keep food safe and mitigate the burden of foodborne disease around the globe. Interconnections between such tools are also mapped.

**Methods**: Taking a realist approach, the focus of this review is explanatory and aims to develop and test theory regarding how contextual (C) factors trigger specific processes and mechanisms (M) to produce outcomes (O), termed a 'C-M-O configuration'. Preliminary work has involved a scoping review of literature and engagement of an international expert reference committee.

**Results**: An initial C-M-O configuration has been developed to suggest that when the context is such that a country: i) is an importer or exporter of food; ii) has the technical infrastructure to detect food safety events with international implications; and iii) is governed in accordance with regional and/or global laws and regulations relating to food control and global health security, then certain mechanisms including trust, experience, support, awareness, understanding, and a sense of community will facilitate the proximal outcome of using communication tools to relay information abroad and a potential range of distal outcomes to improve food safety. It is proposed that variations in the context will influence whether or not the proposed mechanisms will trigger the outcomes.

Significance: Preliminary results suggest that rigorous research is needed to understand how the various tools used to facilitate communication during international food safety events are actually working and in what contexts.

#### P2-24 Relationship between Heavy Metals and Alpha Particles as a Marker of Environmental Pollution in Rice Consumed in Najaf, Iraq

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**Introduction:** The rice examined in the present work is the primary food in Iraq. Recently, concerns have been raised about possible pollution of the crop by heavy metals. Many industrialized processes cause contamination by heavy metals (cadmium (Cd), Iron (Fe) and lead (Pb)) in the soil, food, water, and air

**Purpose:** This new study shows the pollution in the environment by alpha particles and heavy metals and the relationship between them. **Methods:** This study focuses on the emission of alpha particles rate (EAPR) and heavy metals concentrations (HMC) in rice from Najaf markets using nuclear track detectors (CR-39, UK) and a flame atomic absorption spectrophotometer (6300 AA, Shimadzu, Japan), respectively. Rice samples were collected from different sites in Najaf city and Najaf markets in Iraq. The EAPR in rice is determined using an  $\alpha$ -sensitive plastic track detector (PADC-TASTRACK CR-39, Track Analysis Systems Ltd, Bristol, UK).

**Results:** The highest EAPR was found to be 0.0249 mBq cm<sup>2</sup> in basmati rice (Southern Iraqi Company), whereas the lowest EAPR (0.0092 mBq cm<sup>2</sup>) is found in Indian basmati rice. The highest Fe was found to be 2.7237 ppm in basmati rice (Southern Iraqi company), and the lowest (0.3997 ppm) was found in USA basmati rice. The highest Cd was 0.0468 ppm in Iraqi Alnasryah rice, and lowest HMC (0.0034 ppm) was found in Indian basmati rice. The highest Pb was 0.2431 ppm in Babil Anbar Iraqi rice, and the lowest (0.0695 ppm) was in Indian basmati rice. Pb and Cd were lower than the FAO/WHO recommended limits (Pb: 0.50 ppm, Cd: 0.50 ppm) and the European Union acceptable dietary limits.

Significance: With recent rice consumption data, estimated weekly intakes of toxic elements are calculated for the Iraq population. No statistically significant correlation between EAPR and HMC was found in studying rice.

#### P2-25 Deoxynivalenol Screening in Wheat-derived Products in Paraguay

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Introduction: Fusarium graminearum is one of the most important pathogens affecting wheat production in Paraguay. This fungus not only decreases yield and grain quality; the most important consequence is the production of mycotoxins from the trichothecene group, secondary metabolites toxic to humans and other animals. The toxin reported in Paraguay is deoxynivalenol, which has effects on the gastrointestinal tract and immune system.

**Purpose:** In order to determine the presence of deoxynivalenol in flour and baked goods, samples of wholemeal flour and white flour, white and wholemeal bread, and bran crackers were taken in the Gran Asunción Area.

Methods: The presence of deoxynivalenol was determined by monoclonal antibodies through the use of quantitative test strips with the Vertu Lateral Flow Reader, Vicam Technologies. Results: In this study, the presence of deoxynivalenol in samples of wholemeal bread, white bread, local and wholemeal flour, and white flour was confirmed; the results were variable, and below those established by international standards. The imported whole-grain crackers had the highest contamination levels with a maximum of 11.86 ppm on average, above international standards that establish a maximum of one ppm of deoxynivalenol for products derived from cereals intended for human consumption.

**Significance:** This is the first time that this type of study has been carried out in Paraguay in products destined for human consumption. At present, Paraguay lacks any regulations regarding acceptable levels of deoxynivalenol in flour and baked goods. It is important to consider the need for national legislation that establishes acceptable limits of deoxynivalenol in products intended for human consumption, and the implementation of management systems at the field level to prevent contamination with *F. graminearum* and deoxynivalenol.

#### P2-26 Detection of Chicken Vaccine Strain Salmonella enteritidis 441/014 (ade-/his-) and Differentiation between Salmonella Field Strains and the Vaccine Strain

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Introduction: Salmonella can be present in poultry meat, on eggs, or on the feathers of the animals. The detection of Salmonella in primary poultry production is relevant for its control on poultry farms. **Purpose:** For poultry production sites using live vaccines, differentiation between the live vaccine strain and Salmonella field strains is of importance for newly vaccinated poultry. **Methods:** Enrichment of chicken samples was performed for 18±2 h in buffered peptone water (BPW). DNA preparation was performed with a new four-channel real-time PCR assay.

Results: We detected the live vaccine strain Salmonella Enteritidis 441/014 (ade-/his-) in 66 different samples (channel 1), and we could differentiate between this strain and Salmonella field strains (channel 2). Samples containing the vaccine strain were positive in the vaccine channel and in the SE/STM channel (channel 3). In 59 samples with Salmonella Typhimurium we could differentiate between Salmonella Enteritidis and Salmonella Typhimurium serotypes in a one-tubereaction by melting curve analysis in the SE/STM channel (channel 3). In addition, we use an internal positive amplification control in one channel (channel 4). We positively tested different cyclers. Significance: BIOTECON Diagnostics developed a new multiplex real-time PCR assay, the vetproof SE Vaccine Detection 1 Kit, which enables specific detection of the live vaccine strain Salmonella Enteritidis 441/014 (ade-/his-) present in Salmovac SE, Salmovac 440 and Gallivac SE). It can differentiate between this strain and Salmonella field strains, and additionally allows differentiation between Salmonella Enteritidis and Salmonella Typhimurium serotypes in a one-tube-reaction. The vetproof SE Vaccine Detection 1 Kit is prefilled and lyophilized within individual strip tubes for direct loading into real-time PCR instruments

#### P2-27 Study of the Transfer of *Listeria monocytogenes* during Cattle Slaughter by Molecular Typing

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**Introduction:** As *Listeria monocytogenes* is a ubiquitous organism, bacterial contamination may occur at every process step in cattle slaughter, making it hard to prevent pathogen transfer to carcasses during the slaughter process.

**Purpose:** The objectives of this study were to assess the occurrence of *L. monocytogenes* and identify the most important contamination sources by genetic characterisation.

**Methods:** In total, 110 carcasses were tracked through the slaughter process in four different slaughterhouses. Before evisceration, swab samples (400 cm<sup>2</sup>) were taken from the hide at both hind legs, the brisket centre line and the inside foreleg. Immediately after slaughter, and also before chilling, these locations were sampled on the carcasses of the tracked animals. Also, environmental samples such

as knives, air, and evisceration robots were taken during slaughter. *L. monocytogenes* was detected and enumerated according to ISO11290-1 and 2. Confirmed *Listeria* isolates were submitted to molecular typing by serotype identification and pulsed-field gel electrophoresis (PFGE).

**Results:** In 96% of the animals involved, the skin appeared to be positive for this pathogen for at least one of the sampled locations. Forty-eight percent (95% CI 39% to 58%) of the carcasses were found to be *L. monocytogenes*-contaminated at the end of the slaughter line. The first identified serotypes belong to the groups 1/2a (23%), 1/2b (25%), 1/2c (35%) and 4b (17%). Serotypes 1/2a, 1/2b and 4b are commonly found on the hide of the animals, while 1/2c is predominantly found on the carcasses at the end of the slaughter line. Twenty-seven pulsotypes have been identified on hides of the different animals.

**Significance:** At this moment, the results show that in addition to the hides, other sources of *L. monocytogenes* are possible within the production process. Furthermore, it seems that in some slaughterhouses certain pulsotypes are quite persistent.

#### P2-28 Microval Validation of the Thermo Scientific Brilliance CampyCount Enumeration Method in Comparison to EN ISO 10272-2:2017 in Accordance with ISO 16140-2:2016

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Introduction: The Thermo Scientific *Brilliance* CampyCount Agar (BCCA) (alternative method) has been previously validated in comparison to the EN 10272-2:2006 reference method in accordance with EN ISO 16140-2:2016 for the selective enumeration of thermotolerant *Campylobacter* species in raw poultry products. **Purpose:** To renew the MicroVal validation in line with EN ISO 16140-2:2016, and assess the alternative method performance in comparison to the updated EN ISO 10272-2:2017 reference method. **Methods:** Ten grams of raw poultry sample were added to 30 ml of diluent and then serially diluted. In duplicate, *Brilliance* CampyCount agar plates were inoculated with 100 µl aliquots and incubated at 41.5±1°C for 48±1 hours in microaerophilic conditions. The Thermo Scientific O.B.I.S. Campy Test was used to confirm presumptive colonies. The alternative method was compared to the EN ISO 10272-2:2017 reference method.

**Results:** The relative trueness and accuracy profile study results satisfied requirements of EN ISO 16140-2:2016. The results from 14 laboratories in the interlaboratory study results showed no statistical bias between the reference and alternative methods, the average repeatability across all inoculum levels was 0.21 for the reference method and 0.18 for the alternative method. The average reproducibility across all inoculum levels was 0.35 for the reference method and 0.23 for the alternative method.

Significance: The *Brilliance* CampyCount Enumeration method is equivalent to the EN ISO 10272-2:2006 reference method for the enumeration of *Campylobacter* species from raw and ready to cook poultry samples.

#### P2-29 Effect of Ascophyllum nodosum as a Control Strategy against Campylobacter in Broilers

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Introduction: *Campylobacter* is responsible for the first bacterial zoonosis in Europe with more than 246,000 cases in 2017. Risk assessment studies demonstrated that reducing *Campylobacter* loads during primary production would greatly decrease the risk for humans. Therefore, the implementation of effective control strategies at the farm level is needed. Seaweeds are a valuable source of bioactive compounds. In particular, polyphenols demonstrate interesting antibacterial activities.

**Purpose:** This work aimed to evaluate the effect a polyphenolenriched extract of the brown seaweed *Ascophyllum nodosum* on *Campylobacter* contamination in broilers.

**Methods:** A polyphenol-rich extract of *A. nodosum* was produced and studied In vitro to determine the bactericidal effect on different *Campylobacter* spp. strains and evaluate its cytotoxicity on eukaryotic cells. An In Vivo trial was performed: one group of Ross PM3 broilers was fed with the extract added at 0.01% from day 14 to day 35 (treated) and another one was not treated (control). Broilers were orally challenged with *Campylobacter jejuni* at day 18 and caecal *Campylobacter* spp. loads were assessed following the decimal dilution method at day 22 and 35. The statistical analysis was performed using an unpaired *t*-test. **Results:** The seaweed extracts contained 61.9% phloroglucinol equivalents/100 g dry weight. The minimal bactericidal concentration was 0.12 to 0.47 mg/ml depending on *Campylobacter* strains. The extract at 0.9 mg/ml presented cytotoxicity against Caco-2 cells; after contact of 24 h, cell viability was only 20% compared to the control. In Vivo, the addition of the extract in feed did not reduce (*P*=0.053) *Campylobacter* spp. in broilers at day 14. However, *Campylobacter* spp. loads significantly increased (*P*=0.028) by one log CFU/g compared to the control group at day 35.

**Significance:** The product was not able to reduce *Campylobacter* spp. in broilers at the tested concentration, however, additional work using higher concentrations is in progress.

#### P2-30 Detection of Minced Beef Adulteration by Means of Multispectral Vision Technology

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Introduction: Food fraud is an issue of increasing concern among regulatory authorities and consumers. The type of food fraud referred to as "economically motivated adulteration" is rather common in certain food commodities such as minced meat. Hence, the rapid and reliable detection of minced meat adulteration is of apparent value in terms of food protection throughout the food supply chain. **Purpose:** This study was conducted in order to assess the potential of multispectral imaging in detecting minced beef adulteration with offal, both in fresh and frozen-thawed samples.

Methods: Beef and offal (bovine hearts), purchased from four different butcher shops, were minced and appropriate portions of the two tissue types were mixed in order for three adulteration levels (25, 50 and 75%) to be attained, and two levels of pure meat or offal (100% beef, 100% offal) also were studied. Six different samples were prepared for each one of the levels. In total, 120 multispectral images (five levels×six samples×four batches) were acquired. The meat/offal samples were then stored (-20°C for three months), thawed (six to eight hour incubation at 4°C), and multispectral images were acquired once again. The collected data corresponding to fresh and frozen-thawed samples was subjected to partial least-squares discriminant analysis (PLSDA) for the samples' classification in three groups (pure beef, pure offal and adulterated beef). Three of the tested meat/offal batches were used for model calibration and one batch for external validation.

**Results:** Overall correct classification for calibration and validation of the fresh samples was 83 and 77%, respectively, while the corresponding values for the frozen-thawed samples were 100 and 97%.

Significance: Multispectral vision technology was evaluated as propitious for the rapid detection of minced beef adulteration, both for fresh and frozen-thawed samples.

This work has been supported by the project "PhasmaFOOD".

#### P2-31 Spoilage Potential of *Hafnia alvei* B295 and Serratia liquefaciens B293 in Sterile Beef Meat

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**Introduction:** Metabolome formation in food and especially in meat during storage is a complex phenomenon depending on various factors. The contribution of microbes in meat spoilage is still a challenge that needs further elucidation.

**Purpose:** The aim of this study was to apply a metabolomics approach through HPLC and GC/MS in order to monitor metabolite formation of two representative strains belonging to the *Enterobacteriaceae* family inoculated on sterile beef fillets. **Methods:** Sterile beef fillets were inoculated with monocultures of *Hafnia alvei* B295 and *Serratia liquefaciens* B293 in order to obtain an initial inoculum of two to three log<sub>10</sub> CFU/g on the meat. Inoculated samples were stored aerobically at four and 10°C and analyzed microbiologically for *Enterobacteriaceae*, while sterile non-inoculated meat samples were used as a control. Selected meat samples were analyzed for pH and metabolic changes by SPME-GC/MS and HPLC-PDA-RI. In addition, growth kinetic parameters of microorganisms were calculated.

**Results:** Both strains were able to produce acetic and hexanoic acid, dimethyl disulphide, methanethiol, 2,3-butanediol and esters of acetic acid. *H. alvei* B295 also produced isoamyl formate, while *S. liquefaciens* byproducts were 2,3-hexanedione, butyl and isobutyl acetate. In addition, formic and succinic acids, one unknown compound (rt: 29.53 min) and glucose were found to decrease during storage for both strains. Discrimination of the samples was not easy due to the fact that significant changes in the metabolome appeared when the microorganisms reached high numbers (>6.5 log CFU/g). **Significance:** Metabolomic analysis can provide useful information about the dynamic changes of metabolites in the meat from specific spoilage microorganisms and further on identify microbial biomarkers.

#### P2-32 Shelf-life Estimation of Horse Fillets during Aerobic Storage

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**Introduction:** It is well-established that the growth of microorganisms is dependent on various intrinsic and extrinsic factors, with the temperature being the most important.

**Purpose:** The aim of this study was to (i) explore the microbial association of horse fillets stored under different isothermal conditions, (ii) estimate the shelf life of horse fillets during aerobic storage and (iii) monitor the evolution of metabolites.

**Methods:** Horse fillets were derived from a block of deboned horse meat and stored aerobically at zero, five, 10 and 15°C. At appropriate time intervals, they were analyzed microbiologically for total viable counts (TVC), *Pseudomonasspp., Brochothrix thermosphacta*, lactic acid bacteria, yeasts and enterobacteria. In parallel, pH and HPLC analysis for glucose and organic acids were performed. A two-step modeling approach was employed based on primary models to calculate the growth kinetic parameters of microorganisms followed by secondary models to explore the effect of temperature on the growth rate. Shelf-life estimation took place using the Arrhenius equation.

**Results:** At all temperatures tested, the dominant microorganisms were *Pseudomonas* spp. followed by *B. thermosphacta* and *Enterobacteriaceae*, while lactic acid bacteria and yeasts had the lowest growth rate. Finally, it was observed that the higher the storage temperature, the higher the growth rate ( $\mu_{max}$ ) and activation energy (Ea) obtained. On the other hand, pH changes were not significant. Considering HPLC results, the concentration of glucose, lactic and propionic acids decreased, while citric, acetic, formic and butyric acids increased.

Significance: This study demonstrated for the first time the association of microorganisms and metabolites in raw horse fillets during aerobic storage.

#### P2-33 Rapid Assessment of Fish Microbiological Quality with Spectroscopy-based Sensors

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**Introduction:** Due to the time-consuming and sample-destructive nature of microbiological analyses, the real-time monitoring of food spoilage using rapid and non-invasive methods has been the objective of several recent studies.

**Purpose:** The aim of the present study was the investigation of the efficacy of Fourier transform infrared spectroscopy (FTIR) and multispectral imaging (MSI) in tandem with chemometrics in estimating the microbiological quality of fish.

Methods: Whole gilthead seabream (Sparus aurata) were stored under modified atmosphere packaging (30%  $O_2$ , 40%  $CO_2$ , 30%  $N_2$ ,) at zero, four, and 8°C for a maximum time period of 19 days. At regular time intervals, duplicate samples of different fish were subjected to microbiological analysis (determination of total mesophiles), and FTIR spectra and multispectral image acquisition. Two independent experimental replicates were conducted (n=4). Partial least-squares regression (PLSR) models were developed, with the collected spectral data (FTIR, MSI) and microbiological data constituting the X-variables and the Y-variable, respectively. Model calibration was performed using the data corresponding to samples stored at 0 and 8°C, whereas model external validation (prediction) was carried out using the data collected during storage at 4°C. Results: The performance of the PLSR models was evaluated using the correlation coefficient (r) and the root mean square error (RMSE) of prediction. The model based on the FTIR data exhibited better performance than the model based on the MSI data. The estimated values of r and RMSE for the FTIR model were 0.83 and 0.71 respectively, while the corresponding values for the MSI model were 0.68 and 0.98

Significance: Spectroscopy-based sensors appear to be promising for the rapid assessment of the microbiological quality of gilthead seabream, with FTIR spectroscopy appearing to better describe fish spoilage than MSI. This work has been supported by the project "PhasmaFOOD".

#### P2-34 Exploring Yeast Biodiversity from Dry-Salted Naturally Black Olives with Culture Dependent and Independent Molecular Methods

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Introduction: Dry-salted olives are cultivated mainly on the island of Thassos and they are traditionally prepared by placing the fully mature olives in layers with coarse salt. Despite the low water activity of the final product due to high NaCl concentration, the olives are susceptible to spoilage microorganisms such as yeasts and fungi. **Purpose:** To explore the yeast biodiversity of dry-salted table olives from Greek retail outlets with culture-dependent and independent molecular methods.

**Methods:** Nine samples of dry-salted olives were analyzed. One hundred eighty isolates were clustered after PCR amplification of repetitive DNA with the oligo-nucleotide primer (GTG)<sub>5</sub> and profiles with identity percentage higher than 90% were considered to belong to the same group. The typical isolates of each group were further clustered after the amplification of TS region using ITS, and ITS<sub>4</sub> as primers and the implementation of RFLP analysis with the restriction endonucleases Hinf I, Hae III and Cfo I. A total of 20 isolates with discriminant differences were subjected to sequencing of the ITS region of rRNA gene and the results obtained were aligned with BLAST to determine the closest known relatives. Next generation sequencing was performed after the amplification of 26S rRNA gene using LS2 f and NL4MS as primers.

**Results:** Pichia membranifaciens, Candida sorbosivorans, Citeromyces nyonsensis, Candida etchelsii, Wickerhamomyces subpelliculosus, Candida apicola, Wickerhamomyces anomalus, Torulaspora delbrueckii, and Wickerhamiella versatilis were the dominant species among the samples according to ITS sequencing (culture-dependent method), while the dominant species showed by NGS (culture-independent method) were Candida etchelsii, Pichia triangularis, Pichia membranifaciens and Candida versatilis. **Significance:** Dry-salted olives are an economically important product for the Greek table olive industry and the biodiversity of yeasts in the final product is revealed for the first time by means of molecular techniques.

#### P2-35 Control of Undesirable Microbial Growth in Table Olive Fermentation Using Selected Yeast Strains with Multifunctional Potential

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**Introduction:** The implementation of microbial starters related to table olives with multifunctional features during the fermentation process can reduce the risk of spoilage and the growth of undesirable microorganisms.

**Purpose:** To assess the survival of five autochthonous yeast strains belonging to *Candida boidinii* and *Saccharomyces cerevisiae* inoculated in the brines of Kalamata natural black table olives during fermentation and monitor the changes in the microbiological and physicochemical profile.

**Methods:** Olives were fermented in seven percent w/v NaCl and inoculated in monoculture with four yeast strains belonging to *C. boidinii* (Y27, Y28, Y30, and Y31) species and one strain of *S. cerevisiae* (Y34) during the first day of the fermentation to a final concentration of  $3 \times 10^6$  CFU/ml. Changes in microbiological counts and pH were analyzed for a period of 150 days. The survival of the inoculated strains has been estimated by gel electrophoresis from PCR amplification of repetitive DNA elements with the oligonucleotide primer (GTG)<sub>s</sub> at different fermentation points.

**Results:** At the end of the fermentation, yeasts' populations ranged from 3.8 to 5.3 log CFU/g in olives, while they reached a population between 3.8 and 6.4 log CFU/mL in brines. Moreover, pH values ranged from 3.7 to 4.9 among the treatments for both the olives and the brines. The results obtained from rep-PCR for the first day of fermentation showed 100% survival of the starters. At the end of the process three out of five starters namely, Y27, Y30, and Y31 (seven isolates, one isolate and two isolates, respectively) showed survival on the olive flesh.

**Significance:** The survival of the strains used as starters during fermentation is very important as table olives are considered to be a good matrix for several microorganisms with probiotic potential.

#### P2-36 Prevalence and Relevance of Salmonella spp. Testing in Camellia Sinensis Tea

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<sup>1</sup>R&D Unilever, Sharnbrook, United Kingdom, <sup>2</sup>Unilever, Sharnbrook, United Kingdom Introduction: Tea is the second most commonly consumed beverage in the world. Its forecasted growing consumption is driven by consumer demand for new flavours and recognition of the health benefits, which results in the emergence of innovative herbal tea and tea blends with the addition of herbs and spices. Currently, no legislation specifies microbiological criteria for tea in Europe. However, available guidelines require the absence of pathogenic *Salmonella* spp. in finished products.

**Purpose:** This study evaluates the prevalence of *Salmonella* spp. in leaf and herbal teas to mitigate the risk associated with this pathogen. Findings are used to assess the relevance of finished product *Salmonella* testing for these commodities.

**Methods:** Information relating to the prevalence of *Salmonella* spp. in *Camellia sinensis* was collected following searches of electronic databases, scientific journals, books, technical reports and company internal data. The search strategy included keywords "microbiology", "tea", "prevalence, "quality", "*Salmonella*".

**Results:** An exhaustive search of the literature (over 2,100 publications screened) and internal data yielded 17 publications. The overall analysis indicated low prevalence (48 positive samples of total 2,338 identified) of *Salmonella* at the level of 2%, with no study reporting pathogens in black and green tea. All positive samples were in herbal teas (e.g., jasmine, lemongrass, rooibos, peppermint or lemon verbena tea).

**Significance:** Findings of this review align with a history of safe use of tea over thousands of years and equally confirms that control of *Salmonella* spp. within factory facilities manufacturing black and green tea should rely on preventative approaches, e.g., ingredient quality programmes, Good Manufacturing Practices, Hazard Analysis Critical Control Point and environmental monitoring programmes, rather than finished product testing. Considering that the pathogen has been sporadically isolated from herbal teas, robust sampling plans aligned to finished product testing based on statistical methods are suitable to control the risk of *Salmonella* spp. associated with tea blends containing herbs, dried flowers, fruit pieces, and spices.

#### P2-37 Exploring the Virulence Gene Expression of Arcobacter butzleri during Simulated Infection of Human Gut Models

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Introduction: Arcobacter butzleri is an emerging foodborne pathogen often isolated from pork, chicken and beef meat, which causes different gastrointestinal diseases in human due to its invasive behaviour. However, the pathogenicity of *A. butzleri* is still underestimated due to a substantial lack of information on its virulence mechanisms, metabolic and genomic features. **Purpose:** This study aims to explore, in simulated host-pathogen

interactions, the expression of nine genes that are currently correlated with virulence traits of *A. butzleri*.

**Methods:** After the *ex novo* design of primers for the nine genes (*cadF, ciaB, cj1349, irgA, hecA, hecB, mviN, pldA, tlyA*) on an *A. butzleri* reference genome (type strain LMG 10828<sup>T</sup>), their relative expression was quantified by quantitative RT-PCR under simulated host-pathogen interaction conditions. Briefly, In Vitro gut models of mucus-producing (HT29–MTX) and non-producing (HT29, Caco-2) human cells were co-incubated with the pathogen and total bacterial RNA was recovered at different time points. At the same time, bacterial counts were performed to describe the colonization and translocation capabilities of *A. butzleri*, by using two dimensional and three dimensional gut models, respectively.

**Results:** As the first outcome, an RT-qPCR protocol, suitable to quantify the relative expression of the nine virulence genes of *A. butzleri* in the presence of human cells, was optimized. Applying this protocol, an upregulation of part of those genes along the coincubation time was observed. In addition, a favourable role of the mucus for the pathogen colonization was observed whereas the data from the three-dimensional models suggested an intracellular passage of *A. butzleri* through the epithelial barrier. **Significance:** The results of this study represent a first step in the understanding of *A. butzleri* pathogenicity and are important to explain its role in gastrointestinal diseases.

#### P2-38 Survival of *Listeria monocytogenes* in Food Residues on Packaging Materials for Dairy Products

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**Introduction:** *Listeria monocytogenes* is known for causing foodborne infections often associated with a high mortality rate. Survival under adverse conditions of *L. monocytogenes* for extended periods of time has been reported. *L. monocytogenes* contamination

in dairy products may be transferred on packaging materials which could then potentially serve as a source of cross-contamination in the home.

**Purpose:** The aim was to quantify the survival of *L. monocytogenes* on packaging materials soiled with cheese purge and stored at different temperatures.

Methods: Three poly-coated materials used for packaging of dairy products were selected, with different physicochemical properties: polythene/parchment paper (A); polythene/polyamide (B), and parchment paper (C). Each material (5×5cm) was inoculated with a five-strain mixture of L. monocytogenes (2.5 log CFU/cm<sup>2</sup>) of dairy origin suspended in a non-sterile-homogenate, simulating cheese purge. Samples were stored for up to 56 days at four, 12 and 37°C, and periodically analyzed for L. monocytogenes and total bacterial populations. Three samples were analyzed at each sampling point. **Results:** Survival of *L. monocytogenes* varied among packaging materials as well as at different storage temperatures. Counts decreased on all materials stored at 37°C and reached nondetectable levels on B by day four and on A and C by day seven. Initial levels (2.5 log CFU/cm<sup>2</sup>) of Lm increased to three log CFU/cm<sup>2</sup> within four days of storage at 12°C on A and C. After 56 days at 4 and 12°C, Lm was recovered from all the tested materials, with counts ranging from 1.64 log CFU/cm<sup>2</sup> (B) to 3.24 log CFU/cm<sup>2</sup> (C) and from 0.4 log CFU/cm<sup>2</sup> (B) to 3.68 log CFU/cm<sup>2</sup> (C), respectively. Significance: Survival of Lm on packaging materials raises concern because consumers may not expect pathogen contamination on the package and could consequently do nothing to prevent crosscontamination in the home environment.

#### P2-39 Performance Assessment of a New One-Broth-One-Plate Detection Method for Salmonella spp. in Food and Feed Samples

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**Introduction:** Neogen has developed a new alternative method to detect *Salmonella* spp.: OBOP-S.

Purpose: This method has been validated according to ISO 16140-2:2016

**Methods:** The method requires a supplemented buffered peptone enrichment followed by CASE, which is a selective chromogenic agar for the detection of *Salmonella*. It utilises a dual chromogenic system to differentiate between *Salmonella* and non-target organisms that grow on the agar. Utilisation of the first chromogen results in blue/ green *Salmonella* colonies. Competitive flora is inhibited or appears a different color.

Results: During the ISO 16140-2 validation study scheme, 468 samples in total were analyzed by AdGene to determine the sensitivity of the OBOP-S method versus the ISO 6579. The results showed a similar sensitivity between OBOP-S and the ISO 6579, even if only one broth and one agar plate have been used, versus three broths and four agar plates in the ISO standard. The sensitivity of the alternative method was 99% and the sensitivity of the reference was 99.5%. During the study, no false positives have been seen on CASE agar. These results have been confirmed by the inclusivity study. The 111 target strains gave typical colonies on the CASE agar. The 30 non-target strains that have been tested during the exclusivity study gave no false positive result. The relative detection limits of the alternative method and the ISO standard are similar for the six tested food and feed matrices. The relative levels of detection are all below the acceptability Limits (fixed at 2.5 whatever the matrix/strain pairs). Significance: According to this study on six categories, the OBOP-Salmonella method shows satisfactory relative accuracy, specificity and sensitivity.

#### P2-40 A Metabarcoding Approach to Study Norovirus Diversity in Contaminated Shellfish

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Introduction: Noroviruses (NoV) are small positive-strand RNA viruses and the leading cause of acute gastroenteritis worldwide. They are present in sewage-contaminated seawater and bioaccumulated by filter-feeding shellfish. NoVepidemics linked to shellfish consumption are among the most recognized causes of foodborne gastroenteritis in Europe. Twenty-five different NoV genotypes belonging to genogroups (G) I, II or IV are known to infect humans, with the frequent emergence of new strains that can be associated with increased epidemic burden. Thus, public health protection and management requires monitoring NoV emergence and diversity. NoV being prone to genome recombination, the identification of strain requires sequencing of its polymerase and capsid genes. Environmentally contaminated foods such as shellfish pose specific challenges such as low levels of contamination, involving preferentially certain NoV genotypes and often with multiple viral strains.

Purpose: To gain better insight into the diversity of NoV genotypes in contaminated shellfish associated with gastroenteritis outbreaks. Methods: We analyzed shellfish samples linked to confirmed NoV outbreaks from France, 2016. We compared the classical sequencing approach using cloned PCR products to a metabarcoding approach using MiSeq Illumina deep sequencing of amplicons, targeting both the polymerase and capsid genes. This approach was validated on different mixes of NoV GII strains of known composition Results: Through amplicon cloning, we identified the GII.17 genotype in all samples and NoV genotype diversity in three samples. Deep sequencing revealed minor NoV strains in all samples but one, with the identification of eight GII capsid genotypes, one GIV capsid, and five GII polymerases. In artificial mixes of NoV strains, most genotypes were identified, but with a high impact of sample dilution. Significance: Metabarcoding allowed the study of NoV diversity in samples with very low viral contamination such as shellfish and could be applied to NoV monitoring in other foods.

#### P2-41 Next Day Detection of *Cronobacter* Species in Powdered Infant Nutritionals, Milk Powders and Environmental Samples Using the Assurance GDS *Cronobacter* Tq II Assay

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Introduction: Cronobacter is a pathogen of concern in infant nutritionals. A MicroVal certification (Q Laboratories, Expert Lab) for the Assurance GDS Cronobacter Tq II (GDS) was performed for detection of Cronobacter in the food category of infant formula and infant cereals following ISO 16140-2 guidelines. Milk powder and environmental samples were also tested. In total, this data included 434 samples for the sensitivity study, and a relative limit of detection (RLOD) study on infant formula without probiotics, infant cereal containing probiotics, milk powder, and process water. Finally, an interlaboratory study was performed by 11 collaborators with one food type, powdered infant formula containing probiotics. **Purpose:** To validate the next-day detection of Cronobacter by Assurance GDS Cronobacter Tq II method in infant nutritionals, milk powder and environmental samples compared against the ISO reference method.

**Methods:** Lyophilized cultures of *Cronobacter* were inoculated into foods and dust sweepings and stabilized at room temperature for a minimum of two weeks. Environmental sponges and process water were inoculated as recommended by ISO 16140-2. *Cronobacter* were inoculated at less than three CFU/sample for the sensitivity study. *Cronobacter* were diluted in bulk uninoculated foods to achieve partial recovery for RLOD studies. Samples were enriched 1:10 in media for 20 h (24 h for 375 g samples) and analyzed by GDS. Environmental samples were enriched for 18 h and tested by GDS.

**Results:** For the sensitivity study, the observed values for positive and negative deviations were successfully below the acceptability limit (AL) required per category. The average combined RLODs were 0.858, all individually below the AL of 1.5 to 2.5. Statistics support that there is no difference between the alternative method and the reference method

Significance: Assurance<sup>®</sup> GDS *Cronobacter* Tq II provides a reliable next-day method for detection of *Cronobacter* in infant formula, infant cereals, non-probiotic ingredients, milk powders and environmental samples.

#### P2-42\* Evaluation of Oxygen Availability and Substrate Structure on Growth and Inter-Strain Interactions of *L. monocytogenes*

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Introduction: The growth and interactions of different *Listeria* monocytogenes strains present simultaneously in the same food product are affected by the structural properties of the food matrix. **Purpose:** To evaluate the effect of oxygen availability in different media on growth and inter-strain interactions of *L. monocytogenes*. **Methods:** Antibiotic-resistant *L. monocytogenes* strains of serotypes 4b(C5,ScottA), 1/2a(6179) and 1/2b(PL25), were inoculated in liquid media containing tryptic soy broth supplemented with yeast extract (TSB-YE) and on solid media (TSB-YE with 0.6% and 1.2% agar) at 2.5 log CFU/ml in single or two-strain cultures (1:1 ratio). Aerobic conditions (AC) were achieved with constant shaking (liquid) or surface inoculation (solid), while static incubation (liquid) or pour plated media (solid) were used for a hypoxic environment (HC). Anoxic conditions (AnC) were attained by flushing nitrogen (liquid) or adding 0.1% w/v sodium thioglycolate and a paraffin overlay (solid).

Bacterial growth was assessed during storage at 7°C (n=3×2). Results: Generally, inter-strain interactions, as indicated by the difference in the final population of single and co-cultured strains, seemed to be potentially serotype-dependent. C5 and ScottA(4b) suppressed 6179(1/2a) and PL25(1/2b). The extent of suppression increased with decreasing agar concentration, while the impact of oxygen availability on the inter-strain interactions was strain combination dependent. During co-cultivation in liquid and solid media, regardless oxygen availability, 6179 was suppressed by C5 by 4.6 (AC,TSB-YE) to 1.9 log CFU/ml (HC,TSB-YE+1.2), compared to the corresponding single culture, which attained a population of ca. 9.4 log CFU/ml (P<0.05). In broth, the growth of 6179 was also inhibited by ScottA by 2.2 and 2.6 log CFU/ml under HC and AnC, respectively. In broth under AC and AnC, ScottA was suppressed by C5 by 3.4 and 2.2 log CFU/ml, while in solid media, inhibition was 1.8 and 0.8 log CFU/ml (P<0.05).

**Significance:** Investigating growth interactions in different environments could explain the dominance of certain serotypes in foods of safety concern for *L. monocytogenes*.

## P2-43 Survival Study of Salmonella enterica in Inoculated Pistachios

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**Introduction:** Pistachios have been associated with multiple outbreaks and product recalls due to contamination with *Salmonella enterica*. Two serovars, *Salmonella* Montevideo and *Salmonella* Senftenberg, have been responsible for multiple contamination events since 2008.

**Purpose:** The objective of the study is to evaluate how long *Salmonella enterica* can survive in pistachios at different relative humidities and to look for differences between serovars. **Methods:** Pistachios were inoculated with either the *Salmonella* cocktail containing serovars *Salmonella* Montevideo, *Salmonella* Senftenberg, *Salmonella* Anatum, *Salmonella* Oranienberg, and *Salmonella* Newport or the individual strain by the soak method. The pistachios dried overnight in a hood at room temperature and then were stored at different humidities (30 or 60% RH) at 25°C over a nine-month period. At different time points, triplicate 10 g samples were enumerated by first macerating in a 1:9 dilution of buffered peptone water and then directly plating on mTSAYE to assess the total *Salmonella* counts. DNA was extracted to perform metagenomics and molecular serotyping.

**Results:** The direct plating showed the cocktail inoculation has a reduction of 3.75 (30% RH) and 4.53 (60% RH) log CFU/g after nine months of storage with a reduction of 3.00 log CFU/g after desiccation. Differences in the log CFU/g between the samples stored at 30 and 60% RH were observed after six months of storage. The single strain inoculation showed that serovar *Salmonella* Oranienberg has less potential for survival with a 1.4 (30% RH) and 1.8 (60% RH) log CFU/g reduction. This finding was also observed in the cocktail study based on metagenomics and serotyping. The *Salmonella* Montevideo, *Salmonella* Senfenberg, and *Salmonella* Anatum strains showed the lowest log CFU/g reduction.

**Significance:** This study clearly shows that different serovars of *Salmonella enterica* are able to survive in pistachios over an extended period of time. These findings indicate a potential risk if the product becomes contaminated during harvest or processing.

#### P2-44 Virulence Gene Characteristics of Enteropathogenic *Vibrio* spp. Isolated from Korean Fishery Products

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**Introduction:** As a result of high demand in the production and consumption of fisheries products in Korea, there is an urgent need to control marine pathogenic bacteria such as *Vibrio* spp. in seafood products. In the culture-based approach, *Vibrio* spp. can be detected on thiosulfate citrate bile salts sucrose (TCBS) agar, and the molecular approaches include PCR amplification. In the present study, both culture-based and molecular approaches were employed for the detection and characterization of *Vibrio* spp. from Korean seafood products.

**Purpose:** The present work was aimed at the detection and characterization of *Vibrio vulnificus* and *Vibrio cholerae* using a culture-based and molecular approach in Korean seafood products.

**Methods:** The serially diluted samples (1%) were inoculated in a test tube containing 10 mL alkaline peptone water and incubated at 30°C for 24 h. The tube which showed turbidity was selected, streaked on TCBS agar, and incubated overnight at 30°C. The bacterial colonies with yellow and green appearance on the agar plate were considered presumptive *V. cholerae* and *Vibrio vulnificus*, respectively. These colonies were further amplified by PCR using primers based on the species, serotype and virulence genes.

**Results:** We have isolated a total of 10 species of *V. cholerae*, two strains of *V. cholerae*-O139 and eight strains of *V. cholerae* non-O1. The *V. cholerae*-O139 were confirmed to possess virulence *tcpl* and *zot* genes. Among seven isolated *Vibrio vulnificus* strains, three possess virulence *vvh*, *viuB* and *vcgC* genes.

Significance: Although the culture-based approach is commonly used for the detection of *Vibrio* spp., there is a chance of getting false positive results. Hence, the combined application of both culture-based and molecular approaches would provide the most accurate and reliable results for the detection and characterization of *Vibrio* spp.

#### P2-45 The Effect of pH and Salinity on the Heat Resistance of *Listeria monocytogenes* in Sauce

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Introduction: The thermal resistance of pathogens in acid or salinity conditions of various foods have been evaluated, but there are few studies about the fate of pathogens under acidic and salinity conditions of sauces.

**Purpose:** The aim of this study was to evaluate the effects of pH and salt on heat resistance of *Listeria monocytogenes* in sauces. **Methods:** The pH (three, four, five, and seven) and salinity (three, seven, and 11%) of the sauce was adjusted, and zero percent salinity solutions were prepared by altering the pH of buffer. A cocktail of three *L. monocytogenes* strains was inoculated into samples resulting in 7 log CFU/mI and samples were treated at 55, 60 and 65°C in a water bath. One mI of serially diluted solution was spread onto Oxford agar and incubated at 37°C for 48 hr. *D*-values for each condition were calculated.

**Results:** At pH 3, all bacteria were killed after five minutes at 25°C regardless of salt concentration, while *D*-values were decreased from 5.54 to 2.53 min at 55°C as pH decreased and temperature increased at zero salinity. The variation in heat resistance was shown to be largely unrelated to the three factors; *D*-values at 55°C and pH 7 compared to zero salinity were shown to increase at three percent, decrease at seven percent and increase again at 11% (5.55, 23.04, 17.89 and 18.38 min), and similar variation was observed at 60°C. The value was highest at seven percent, pH 5, and 60°C or less, but at three percent, pH 4, 60°C or more. The variable effects of pH and salinity on heat resistance were diminished when heated at 65°C. **Significance:** These results suggest that the effects of pH and salinity became greater at a low temperature such as 55°C, and thus heating at a temperature above 65°C is recommended for microbial safety to minimize the effects of pH and salinity on heat resistance.

#### P2-46 Microbial Food Safety in Canteen Foods in Hanoi, Vietnam

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Introduction: Food safety is a major concern for Vietnamese society with about 250-500 reported outbreaks of foodborne toxi-infections each year, affecting 7,000 to 10,000 individuals and causing 100 to 200 deaths, and probably many more unreported cases. Among different safety issues, microbial safety is very prominent, and it is known that meals prepared in large kitchens are one of the major causes of foodborne toxi-infections.

**Purpose:** This contribution documents the microbiological safety of 20 dishes, mostly cooked and served at ambient temperature, in each of 20 canteens in Hanoi, Vietnam, based on different microbiological parameters including total aerobic count, coliforms, *E. coli, Salmonella, Staphylococcus aureus, Bacillus cereus,* and *Listeria monocytogenes.* 

**Methods:** The analyses were performed by traditional microbial plating techniques using different selective media and biochemical confirmation tests.

**Results:** Results show that average total plate count (TPC) in individual canteens ranges from three to five log CFU/g, including a number of samples with TPC as high as eight log CFU/g, while the acceptable level of TPC in RTE food is less than 10<sup>5</sup> CFU/g. *E. coli* and S. *aureus* were absent in most of the canteen meals, although some foods were contaminated with up to 10<sup>3</sup> to 10<sup>4</sup> CFU/g. *Salmonella* was occasionally found, mostly in poultry and pork meat. *Bacillus cereus* and *Listeria monocytogenes* were rarely present but in some cases reached numbers up to 10<sup>3</sup> CFU/g.

Significance: The results indicate that canteen foods are usually safe, but also that additional measures are in some cases needed to further improve the safety. The results of this work will help to raise consumers' and food producers' awareness and serve as guidance for policymakers to develop and implement better control measures to ensure the safety of canteen food in Vietnam.

#### P2-47\* Survival of *Listeria monocytogenes* on Pistachios, Corn Flakes and Chocolate Liquor at 4 and 23°C

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Introduction: Low-moisture foods (LMFs) such as dried fruits, cereals and confections, are characterized by water activity (a<sub>w</sub>) below 0.85 and have been recognized as being vehicles for foodborne illness. Although the growth of bacterial pathogens is inhibited by low a<sub>w</sub>, they have been shown to survive/persist for long periods of time in some LMFs. This presents a public health concern, especially when LMFs are consumed without undergoing any microbial inactivation steps.

**Purpose:** The main purpose of this study is to assess the survival of *Listeria monocytogenes* on artificially-inoculated LMFs (dry-roasted shelled pistachios, chocolate liquor and corn flakes).

**Methods:** Foods were inoculated with a four-strain cocktail of *L. monocytogenes* at an initial concentration of 8 log CFU/g by wet inoculation (pistachios) or misting (chocolate liquor, corn flakes). LMFs were then dried at 30°C, equilibrated, and stored at either 23°C, 30–35% relative humidity (RH) or 4°C, 55–81% RH for up to one year. Bacterial enumerations were done on tryptic soy agar with 0.6% (w/v) yeast extract, with the exception of pistachios, for which Oxford agar was used due to interfering background microbiota. Analysis of significant populations was determined by using a two-way repeated measures ANOVA.

**Results:** During the initial drying/equilibrium period, populations of *L. monocytogenes* declined by 1.2 to 1.9 log CFU/g on LMFs. Populations declined significantly ( $P \le 0.05$ ) by 2.7, 5.3, and 6.4 log CFU/g on dry-roasted pistachios, corn flakes, and chocolate liquor stored at 23°C, respectively. At 4°C, *L. monocytogenes* populations remained stable on LMFs throughout the storage period. **Significance:** As the presence of any *L. monocytogenes* on ready-to-eat foods can lead to food recalls, research regarding foodborne pathogens on LMFs is very important to understand the environmental mechanisms underlying pathogens survival and also has great relevance for predictive modeling used in microbial health risk assessments.

#### P2-48\* An Investigation of the Shedding Dynamics of Shiga-toxingenic *Escherichia coli* Including Super Shedding of O157 and O26 Serogroups in Irish Sheep

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Introduction: Ruminant animals are considered the primary reservoir of Shiga-toxigenic *Escherichia coli* (STEC). A wide range of STEC serogroups have been isolated from ruminant animal feces, with serogroups O157 and O26 the most frequently reported. STEC predominantly colonise the recto-anal junction (RAJ) in ovine hosts. The number of STEC shed in the feces may vary. Some animals, termed 'super-shedders', shed high volumes of STEC (>4 log CFU/g feces) and are considered high-risk carriers of the pathogen. **Purpose:** The purpose of this study was to assess the shedding of STEC and super-sheddeng of two serogroups, O157 and O26, in Irish sheep. The risk factors underpinning shedding dynamics were assessed, including the relationship between shedding and whole genome sequence profile.

Methods: RAJ swab samples (*n*=410) were collected over a ninemonth period from an ovine slaughtering facility. Metadata for each sample was recorded. Swabs were enriched in 30 ml of modified tryptone soya broth with novobiocin at 41.5°C for five hours and subjected to a quantitative real-time PCR assay to detect and enumerate serogroups O157 and O26 in super-shedding animals. Incubation was allowed to continue for 24 hours and Shiga-toxin prevalence was assessed using a targeted qualitative real-time PCR assay. Positive isolates were sequenced using the Illumina MiSeq platform and analysed In Silico for serogroup, phylogroup, Shiga-toxin subtype and sequence type. **Results:** Eight O157 strains were isolated, of which six were from super-shedding sheep. The incidence of culturally isolated *E. coli* with *stx* and STEC O157 and O26 positive sheep was 54.4, 1.95 and 0.24%, respectively. The prevalence of *stx1*, *stx2* and *stx1/stx2* in isolated strains was 17.1, 12.4 and 24.9%. The occurrence of *stx1/ stx2* in combination with *eaeA* is significant according to Pearson's correlation and a paired *t*-test.

Significance: These results underline the risk Irish sheep pose as a potential source of STEC infection.

#### P2-49\* Microbiological Safety of Portuguese Dry Fermented *Chouriço* Sausages as Affected by Processing and Physicochemical Factors

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**Introduction:** In Portugal, *chouriço* is mainly produced by small-scale artisanal manufacturers, employing their own traditional techniques. Accidental variations of the production process, even within the same processing unit, can occur, since process variables may not be fully controlled, thus resulting in final products of variable microbiological quality, stability and safety.

**Purpose:** This study aims to: (i) elucidate the critical process variables responsible for the varying levels of *Enterobacteriaceae*, *Staphylococcus aureus* and *Listeria monocytogenes* in *chourico*; and (ii) assess relationships between physicochemical properties of the product and microbial counts along processing.

**Methods:** Microbiological and physicochemical surveys at five stages of production were conducted in 12 batches sampled from two factories of *chouriço* (one using curing salts in their formulation). Longitudinal or stepwise multiple regression models ( $\alpha$ =0.20) were fitted to the counts of each microbial group, according to the objective of the analysis.

Results: Maceration was found to be a critical point of the process since Enterobacteriaceae (P=0.017) and S. aureus (P=0.087) could significantly increase until the end of this stage. Furthermore, the contamination of chouriço correlated positively with the maceration room temperature (S. aureus: P=0.034; L. monocytogenes: P=0.010) and the final pH of the macerated meat (L. monocytogenes P=0.048). Batches with lower salt concentration in smoked meat (P=0.021), higher maceration temperature (P=0.034) and shorter ripening time (P=0.171) presented higher S. aureus counts in the end product, whereas higher levels of contamination in the mixture (P<0.0001) and casings (P=0.044) and shorter production time (P=0.090) led to higher L. monocytogenes counts in the end product. Although nitrite showed a strong effect on reducing Enterobacteriaceae during smoking (P<0.0001) and controlling *L. monocytogenes* during ripening (P=0.036), it did not hinder (P=0.007) *S. aureus* growth. Significance: These results reveal the urgent need to standardise and optimise the production process of chouriço in order to assure consumers' safety.

#### P2-50 Thermal Inactivation of Salmonella spp. in Camel Meat Burger

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**Introduction:** Salmonella is one of the significant foodborne pathogens present mostly in raw meat products. Camel meat products are rich in proteins and provide a reliable source of healthy meat.

**Purpose:** The thermal inactivation of two *Salmonella* strains (*Salmonella* Typhimurium 02-8423, *Salmonella* Copenhagen PT99) was investigated in camel meat burger.

**Methods:** Camel lean meat (CLM) and camel lean meat with fat (CLMF) burgers were prepared and inoculated with *Salmonella* strains. Inoculated burger samples, packed in sterile bags, were completely submerged in a circulating water bath and each heated to one of four internal temperatures (55, 57.5, 60 and 62.5°C) for different time intervals. Viable *Salmonella* cells were enumerated by surface plating on xylose lysine deoxycholate agar overlaid with tryptic soy agar. *D*-values and *z*-values were calculated using SPSS software.

**Results:** There are significant differences (*P*<0.05) between the *D*-values of *Salmonella* spp. in CLM and CLMF burgers; *Salmonella* spp. in CLMF had higher thermal resistance than CLM. The average *D*-values of *Salmonella* Typhimurium 02 in CLM and CLMF burgers at temperatures 55 to 62.5°C ranged from 5.82 min to 6.3 sec and 8.69 min to 8.4 sec, respectively. Similarly, average *D*-values of *Salmonella* 

Copenhagen PT99 in CLM and CLMF burgers ranged from 5.22 min to 6.3 sec and 9.49 min to 9.2 sec respectively. The z-values of *Salmonella* Typhimurium 02 in CLM and CLMF were 4.4 and 4.2°C, respectively, while the z-values of S. Copenhagen PT99 in CLM and CLMF were 4.4 and 4.1°C, respectively. There were no significant differences between the z-values of *Salmonella* spp. **Significance:** These data would be useful for the processors of camel meat products to validate their process and reduce the risk of *Salmonella* in these products.

#### P2-51 Evaluation of Seasonal Variation of Enteric Viruses and Protozoa Contamination in Shellfish Samples

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Introduction: Coastal areas present a high density of human population and related activities such as animal farming. All these activities contribute to faecal contamination of the coastal marine environment. A large diversity of pathogens from a human or animal origins can be dispersed directly, or reach the coastal waters via runoff from urban, suburban, and agricultural lands or insufficiently treated wastewaters. Filter-feeding bivalve molluscan shellfish have the unique ability to actively accumulate and retain some pathogens. Purpose: The aim of the study was to evaluate E. coli, enteric viruses (human norovirus genogroup I and II, bovine norovirus, rotavirus) and protozoa (Cryptosporidium parvum) in oyster and mussel samples in a shellfish farming area. The study area is exposed to faecal contamination of human (important human population centers) and animal origin (intensive cattle and sheep farming). Methods: NF V 08-106 standard method for E. coli, real-time RT-PCR (enteric viruses) and PCR (protozoa) methods were used. Results: During one year, 74 samples were collected: 35 oysters and 39 mussels. The results for E. coli testing as an indicator of faecal contamination of the shellfish production zone were satisfactory for 67 samples (90%). Six unsatisfactory results, with a concentration varying from 4,800 to 20,000 *E. coli*/100 g of flesh and intra-valvular liquid were detected during the winter season. No *C. parvum* and no rotavirus were found. The presence of human noroviruses was confirmed in 12 samples. Only genogroup II was detected and at low concentrations (<600 RNA copies/g of DT). Bovine noroviruses were found in ten samples. All norovirus-positive samples were collected during late autumn and winter months (November to March). Significance: The results showed that despite faecal contamination of the shellfish growing area selected for study, the shellfish sampled had low levels of contamination, mainly during autumn and winter season. No relationship was observed between the presence of E. coli, enteric viruses and protozoa.

#### P2-52 Staphylococcus aureus and Risk Factors along the Dairy Food Chain in the Western, Southern and Lusaka Provinces of Zambia

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**Introduction:** Food safety remains a serious challenge in African countries such as Zambia. Milk has been considered as an ideal food for humans, especially children. However, milk in Zambia is usually marketed and consumed unpasteurized (raw). Raw milk can be a source of many pathogens including *Staphylococcus aureus* and methicillin-resistant *S. aureus* (MRSA).

**Purpose:** The SAD-Zambia project aims to characterize *S. aureus/* MRSA in the dairy-chains of different Zambian provinces and to reduce health risks associated with *S. aureus/*MRSA to consumers and producers.

Methods: A total of 1,938 samples (bulk milk, milk from mastitis cows, nasal and hand swabs of milkers, bucket swabs, water, and processed milk/milk products) were collected from around 320 facilities in three Zambian provinces and analyzed to detect *S. aureus* and MRSA. In parallel, 414 participants were interviewed to determine the milk handling and hygienic practices along the dairy-chain. **Results:** Two hundred ninety-five *S. aureus* isolates (confirmed by MALDI-TOF and real-time PCR) were extracted from samples from 183 facilities. Isolates were further characterized by *spa* type, antimicrobial resistance, and genes encoding virulence factors and enterotoxins. Differences and similarities in hygienic practices were found between facilities in the three Zambian regions with Southern and Western Provinces showing poorer hygiene and milk-handling

practices than Lusaka Province. However, in Lusaka Province, swabs from milkers more often tested *S. aureus*-positive. **Significance:** Results show wide contamination of raw milk with *S. aureus* in all three Zambian provinces; no *S. aureus* was detected in commercially processed milk. Characterization of *S. aureus* isolates provides information on the distribution pattern and genetic diversity among the different regions. Potential risk factors for *S. aureus* maintenance and transmission along the Zambian dairy chain could be identified. Training in hygienic practices and continued surveillance is expected to improve milk safety in Zambia.

#### P2-53\* High Pressure Processing Inactivation of Salmonella in Raw Pet Food Formulated with and without Lactic Acid

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**Introduction:** Interest in raw pet food has been growing consistently over the past few years. The safety concerns related to the occurrence of *Salmonella* pushes manufacturers to explore risk minimisation strategies such as high pressure (HP) processing, the efficacy of which has to be validated.

**Purpose:** The aim of the present study was to evaluate the effect of HP on the inactivation of *Salmonella* spp. in raw pet food, formulated without and with lactic acid.

**Methods:** Meat-based raw pet food was manufactured without (control) and with eight g/kg of a lactic acid-based ingredient. Samples were inoculated with one of three different *Salmonella enterica* strains (CTC1022, GN0082 and GN0085) at 10<sup>8</sup> CFU/g and pressurized at 600 MPa for up to 10 min. *Salmonella* spp. was enumerated on chromogenic medium immediately after HP and after holding for 24 h at 4°C. Inactivation kinetics were estimated with the log-linear with a tail model.

**Results:** Salmonella HP inactivation kinetics depended on the product formulation, the strain and the time when the enumeration was performed after HP. For all strains, the addition of lactic acid caused a higher inactivation rate ( $k_{max}$ =2.2 to 3.7 min<sup>-1</sup>), earlier appearance of resistance tails ( $t_{shift}$ =3.4 to 5.7 min), though at a lower concentration (log  $N_{max}$ =0.1 to 0.9 log CFU/g) than in the control ( $k_{max}$ =1.7 to 2.4 min<sup>-1</sup>;  $t_{shift}$ =5.5 to 6.4 min; log  $N_{res}$ =0.9 to 2.5 log CFU/g). Surprisingly, Salmonella spp. counts in products stored at 4°C for 24 h after HP were considerably higher than those observed immediately after HP, suggesting a quick recovery of sublethally injured cells, thus resulting in lower inactivation rates and resistance tails appearing at higher concentrations.

Significance: The validation of an HP treatment requires a tailormade approach to take into account the specific product formulation. The selection of the most appropriate strain(s) and the most appropriate time for product testing after HP is critical to avoid overestimation of HP efficacy.

#### P2-54 Prevalence of Salmonella spp. and Listeria monocytogenes in Irrigation Water Sources in the Mid-Atlantic United States: A Conserve Project

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Introduction: Outbreaks of salmonellosis have been associated with produce grown in the Mid-Atlantic region of the United States. Irrigation water may provide a reservoir of bacterial pathogens that may contaminate produce intended for human consumption. **Purpose:** To evaluate six surface and recycled water sources in the Mid-Atlantic United States for the prevalence of *Salmonella enterica* and *Listeria moncytogenes*.

**Methods:** Water was collected from six sites: one non-tidal freshwater (NF), one tidal brackish (TB), two pond water (PW), one recycled water (RW), and one processing water (PP), and filtered through modified Moore swabs (MMS). Sampling occurred from September 2016 to October 2018 (*n*=168 to 171 samples). MMS were analyzed for *S. enterica* and *L. monocytogenes* populations through a modified MPN procedure using 10 L, 1L and 0.1 L volumes. One-way ANOVA on recovered pathogen populations in JMP was performed.

**Results:** A large percentage of *S. enterica* and *L. monocytogenes* MPN/L values - 50% (*n*=168) and 69% (*n*=170), respectively – were at or below the limit of detection (0.03 MPN/L). The percentage of *S. enterica* and *L. monocytogenes* MPN/L values which reached the maximum detection limit (11 MPN/L) were 4.7% and 8.2%, respectively. *S. enterica* ±5.27 MPN/L) at the PP site were significantly (*P*<0.05) greater than at the NF, TB, RW and PW sites (0.06 to 1.44 MPN/L). *Se* ±2.70 MPN/L) were significantly greater at the TB site compared to PW sites (0.06 to 0.21 MPN/L). *L. monocytogenes* ±5.03 MPN/L) were significantly greater at the TB site compared to all RW, PW, and PP sites (0.04 to 0.54 MPN/L). Precipitation events after 24 h and 7 d were associated with increased recovery of *Se*.

Significance: Populations of *S. enterica* and *L. monocytogenes* were frequently low in these water sources but did differ significantly based on location and weather factors, indicating their potential as a reservoir of enteric pathogens.

#### P2-55 How to Establish Food Integrity Using Molecular Testing and the Maxwell RSC Purefood GMO and Authentication Kit

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Introduction: Consumers around the world are increasingly demanding information on and reassurance of the origin and content of their food. Protecting consumer rights and preventing fraudulent or deceptive practices such as food adulteration are important and challenging issues facing the food industry. **Purpose:** We report on the Maxwell RSC PureFood GMO and

Purpose: We report on the Maxwell RSC PureFood GMO and Authentication kit for efficient extraction of amplifiable DNA used for GMO sequences and PCR-based food testing using the Maxwell RSC and Maxwell RSC 48 instruments. The instruments are capable of processing one to 16 or one to 48 samples per run, respectively. **Methods:** DNA from ground beef, pork sausage, canola seeds, corn meal, soy flour, and crispy rice cereal was extracted using Maxwell RSC PureFood GMO and Authentication Kit, and analyzed for yield, purity, and amplifiability. For GMO testing, GMO-positive ground maize was spiked into GMO-free maize and DNA was extracted using the Maxwell RSC PureFood GMO and Authentication Kits. DNA from the eluates were amplified using the TaqMan GMO Maize 35S detection kit to identify the percent GMO in the samples. For authentication testing, DNA from ground pork was extracted using the Maxwell RSC PureFood GMO and Authentication Kits and the two competitor kits. DNA eluates were amplified using the RapidFinder Pork ID kit to identify swine DNA.

**Results:** For GMO testing, both isolation methods extracted DNA from the two g maize samples. Isolation methods used extracted DNA from the meat samples. DNA was amplifiable with the swine specific kit from all samples except the 100% beef sample. Based on the C, cut-off from the positive control, swine DNA was identified down to 0.01% pork content.

Significance: This shows the utility of the Maxwell RSC (Catalog # AS4500) and the Maxwell RSC 48 (Catalog # AS8500) for automated DNA extraction from food samples suitable for PCR amplification-based GMO and authentication testing.

#### P2-56 A Study of Cross-contamination Events of *Campylobacter* spp. in Domestic Kitchens Associated with Consumer Handling Practices of Raw Poultry

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Introduction: Campylobacteriosis is the most reported zoonosis in the European Union since 2005 and the most common cause of bacterial foodborne diarrhoeal disease worldwide. Contaminated chicken has been recognized as the major vehicle for consumers' exposure to *Campylobacter*. Handling, preparation and consumption of this product accounts for 20% to 30% of human infections. **Purpose:** The overall objective of the research presented here was to evaluate possible cross-contamination events that can contribute to the spread of *Campylobacter* spp. in domestic kitchen environments during food preparation.

**Methods:** Eighteen households were visited in October 2017 and between February and April 2018 to observe consumers preparing a recipe that included poultry and a raw vegetable salad. Poultry samples and swabs from several domestic kitchen surfaces and utensils were collected before and after food preparation. Detection

and/or enumeration of *Campylobacter* were done according to the methods recommended by the International Organization for Standardization and species confirmation was by a multiplex PCR assay. Genotypic characterization of 72 *Campylobacter* spp. isolates were carried out through antimicrobial susceptibility, pulse-field gel electrophoresis and *flaA*- short variable region sequencing. **Results:** Of the 18 chicken samples analysed, 14 were *Campylobacter*-positive at least by one of the methods applied (77.8%). Molecular typing of 31 *C. jejuni* and 41 *C. coli* isolates showed a high genetic diversity among isolates and demonstrated cross-contamination events from poultry to a kitchen cloth in one household, and from poultry to the cutting-board in other two households.

**Significance:** These results highlight the potential for crosscontamination and survival of this foodborne pathogen in the kitchen environment and the need to educate the consumer on appropriate handling of raw chicken meat products.

## P2-57 Internalization of *Listeria monocytogenes* in Mango (*Mangifera indica*) Variety Tommy Atkins

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**Introduction:** In recent years, significant cases of listeriosis caused by contaminated fruits were reported. Cross-contamination of fruit surfaces and postharvest internalization of pathogens may result in increased risks to consumers.

**Purpose:** This study investigated the internalization capacity of *Listeria monocytogenes* in the mango variety Tommy Atkins, under different experimental conditions.

**Methods:** Mature mangoes were preevaluated for the absence of *L. monocytogenes* on the surface using the swab technique. *Listeria*-negative fruits were challenged with *L. monocytogenes* ATCC 7644 and Scott A. Their internalization was evaluated after spot contamination of the peduncle region (six log CFU/mL) and after immersion in contaminated cooling water (six log CFU/mL, 21.1°C for 10 min) of fruits submitted to the classical "hot water treatment" (46.1°C for 90 min). Mangoes were kept up to 10 days under refrigeration (8°C) and at controlled room temperature (25°C). Counts of *L. monocytogenes* were performed after 24 h, five days and 10 days, in three regions of the challenged fruits: stem scar "SS", middle side "MS" and blossom end "BE").

Results: Internalization of *L. monocytogenes* was detected after 24 h, especially on the stem scar area. After 10 days at 8°C, the counts (log CFU/g  $\pm$  SD) in surface inoculated fruits were SS=4.2 $\pm$ 0.2; MS=3.7 $\pm$ 0.2; and BE=3.0 $\pm$ 0.1. For fruits at 25°C, the counts were SS=5.6 $\pm$ 0.1; MS=5.2 $\pm$ 0.2; BE=3.6 $\pm$ 0.1. After 10 days fruits immersed in contaminated water at 8°C had counts of SS=4.5 $\pm$ 0.2; MS=2.9 $\pm$ 0.1; and BE=<1. At 25°C, counts were SS=4.6 $\pm$ 0.4; MS=2.9 $\pm$ 0.3; and BE=2.2 $\pm$ 0.3.

Significance: The findings indicate that *L. monocytogenes* is able to internalize and spread through the pulp of mangoes. These data may help producers and health agencies to develop quantitative risk assessments and establish proper measures to prevent outbreaks.

#### P2-58 Effect of Low-temperature Steaming on the Survival of Foodborne Pathogens and Their Surrogates on Whole Black Peppercorns

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Introduction: Microbial safety of dried spices has been questioned following numerous reported foodborne outbreaks and safety alerts. Steaming is used in the production of black peppercorns to eliminate foodborne pathogens. However, intensive processing conditions of high temperatures and long times jeopardizes the quality of black peppercorns. Mild steaming conditions at lower temperatures are thus proposed, but limited information is available on the effectiveness and cortical processing parameters of these alternative conditions against foodborne pathogens.

**Purpose:** To evaluate the effectiveness of a low temperature steaming process (<80°C) on selected foodborne pathogens and identify suitable surrogates for this inactivation process. **Methods:** Whole black peppercorns at different water activities (a<sub>w</sub>) were inoculated with one percent of a single bacterial strain (four *Salmonella*, three *Listeria monocytogenes*, three *Escherichia coli* O157 and four non-pathogenic surrogates) to reach a final concentration of 10<sup>7</sup> CFU/g. Inoculated samples were equilibrated at 22°C for four days to final a<sub>w</sub> of 0.35, 0.57 or 0.69, and subjected

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to steam treatment for five min at 70 and 75°C. Bacterial cells were enumerated on selective media to quantify the inactivation effect. The product color and  $a_w$  were measured before and after steam treatment.

**Results:** All the tested pathogens at three  $a_w$  levels were substantially inactivated (5-log reductions) by steaming at 75°C. The resistance of tested *Salmonella* strains was the highest, followed by *Listeria* and *E. coli*, and increased significantly at reduced  $a_w$  of 0.35. No change in color was observed and  $a_w$  remained below 0.85 after steam treatment. *Enterococcus faecium* was the most suitable surrogate to validate the inactivation effect of steaming processes at low temperatures.

Significance: This work demonstrated that low temperature steaming could be applied in the production of black peppercorns to lower the risk of food safety issues while maintaining the product quality.

#### P2-59 Does Hot-air Drying Deliver Satisfactory Microbial Reductions during the Production of Dried Basil?

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Introduction: Drying technologies are often evaluated for their efficiency in removing moisture and their impact on the sensory quality of products. Decontamination effects against foodborne pathogens, however, are rarely investigated. With the increasing use of dried herbs in minimally processed foods and the need for alternative milder processing conditions, inactivation during drying should be verified to ensure the microbial safety of these products. Purpose: Investigate the inactivation of bacterial strains on basil under isothermal heating and dynamic drying to better understand the decontamination efficiency of mild hot-air drying. Methods: Inactivation kinetics of Salmonella Senftenberg and surrogates (Enterococcus faecium and E. coli P1) were first determined on basil (3 g/sample) at fixed a, (0.99, 0.95) in a water bath at 60°C. Afterward, fresh basil (40 g/sample) inoculated with the same strains was dried (decreasing  $a_{\!_W}$  from 0.99) at 60°C and 100°C in a lab-scale convective dryer for different durations. Lastly, to evaluate the overall process lethality by simulating industrial drying, fresh basil (300 g/batch) inoculated with two surrogate strains was dried in a pilot-scale dryer at 60°C and 100°C until  $a_w$  <0.3. **Results:** A treatment of 20 min at 60°C under fixed  $\ddot{a_w}$  inactivated Salmonella Senftenberg by 4.7 and 3.7 log at  $a_w 0.99$  and 0.95, respectively. Drying in the lab-scale dryer delivered a reduction of 1.8 and 6 log for Salmonella Senftenberg after 30 min at 60°C and 100°C respectively. A pronounced increase in thermal resistance was observed once the product  $a_w$  was ≤0.95 during hot-air drying. Inactivation was comparable between lab-scale and pilot-scale dryers, which resulted in 3-log reductions of *E. coli* P1 and ≤1-log reduction of E. faecium at 60°C after 90 min.

**Significance:** Results will contribute to the improvement of the food safety of dried herbs. *E. coli* P1 was a more suitable surrogate than *E. faecium* for the validation study of leafy herbs during hot-air drying.

#### P2-60 BRC Issue 8 Requirements for the Environmental Monitoring of Key Pathogens in Open and Ready-to-Eat Food Production Areas: *Listeria* Control

#### **STINE LOENNERUP BISLEV** *Vikan A/S, Skive, Denmark*

**Introduction:** BRC released issue 8 of its global standard for food safety in August 2018 with the new section 4.11.8 Environmental Monitoring that states: 'Risk-based environmental monitoring programmes shall be in place for pathogens or spoilage organisms. At a minimum, these shall include all production areas with open and ready-to-eat products.' *Listeria monocytogenes* is one of these key pathogens and is especially important to monitor in RTE food production environments.

**Purpose:** As part of a robust environmental monitoring programme, to emphasise the most important locations to look and sample for *L. monocytogenes* in RTE food production environments, and provide guidance on how to control *Listeria* in these environments. **Methods:** BRC section 4.11.8 Environmental Monitoring was reviewed and related to existing knowledge on *Listeria* hideouts and control and current EU legislation on *L. monocytogenes* in the environment and food<sup>1</sup>.

**Results:** As a result of this review *Listeria* problem areas were identified as: floors, floor mats, walls, drains, processing equipment, cleaning equipment, chillers, freezers, doors, cart-wheels, and air-handling systems<sup>2,3,4,5,6,7</sup>.

Consequently, these areas should be included in an environmental monitoring plan for *L. monocytogenes* in RTE food production. In order to provide guidance to the industry, publications on *Listeria* hideouts and controls have been published<sup>8</sup>.

**Significance:** It is crucial to food safety to control the presence of *L. monocytogenes* in RTE food production environments by using the appropriate cleaning and disinfection techniques and by confirming the efficacy of the controls through sampling in the right places at an appropriate frequency. By sharing knowledge on where *Listeria* most frequently hides, and on how it can be controlled, food producers will be able to develop an effective environmental monitoring plan that will keep their products safe and aid in compliance with regulatory and standard requirements, including those in BRC issue 8.

#### P2-61 Inactivation of Stress-resistant Ascospores of *Eurotiales* by Industrial Sanitizers

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Introduction: Different fungi, including the genera Aspergillus (Neosartorya), Paecilomyces (Byssochlamys) and Talaromyces, produce ascospores that survive pasteurization treatments and are regarded as the most stress-resistant eukaryotic cells. The sensitivity of the ascospores to treatments with industrial sanitizers containing chlorine dioxide and iodine (iodophors) has never been assessed before.

**Purpose:** In this study, we report the inactivation of dormant and activated ascospores by solutions of acidified sodium chlorite (ASC), chlorine dioxide and iodine (iodophors).

**Methods:** The activity of the sanitizers was tested in droplet tests in which growth of fungus was evaluated after inoculation of a fixed number of ascospores. In addition, colony count experiments were done. Stereo microscopy was used to evaluate if spores formed germ tubes or not.

**Results:** Ascospores of four species of *Eurotiales* were tested and showed clear variations in sensitivity. The most resilient species, *Talaromyces macrosporus* and *Paecilomyes variotii* (=*B. spectabilis*) survive 75 but not 200 ppm chlorine dioxide solution treatments. These species were able to survive 75 ppm iodine solution treatments, but low amounts of ascospores (100 to 1000 spores) could be inactivated after 16 h of treatment. Inactivated spores did not show any sign of germination after seven days following chlorine dioxide solution treatments as judged by microscopy, but iodine inactivation resulted in visibly distorted ascospores. Activation of the spores is important to evaluate if a spore is inactivated or (still) dormant.

**Significance:** Our data suggest that dormant ascospores can be eradicated by the used sanitizers.

#### P2-62 The Preservative Propionic Acid Damages Germ Tubes of Feed Spoilage Fungi

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**Introduction:** The weak organic acid propionate is an important preservative in food and feed and inhibits the growth of various spoilage bacteria, yeasts and fungi, including mycotoxigenic fungi. The mode of action of this compound on fungal survival structures (conidia) and germ tubes of xerophilic feed-spoiling fungi is scarcely studied.

**Purpose:** In this study, we have isolated xerophilic *Aspergillus* with very high predominance from spoiled poultry feed. We assessed the sensitivity of a panel of isolated fungi for propionic acid and evaluated the viability of treated conidia and germ tubes.

Methods: In this study, we have isolated and identified fungal strains from nine samples of poultry feed originating from different countries using a shelf-life test and molecular methods. MIC values were measured by means of a microtiter plate. Survival of conidia was tested after a 24-hour exposure to 31 and 62 mM propionic acid. To evaluate if propionic acid damaged germ tubes, a novel method was developed in which young biofilms of the fungi were tested in Erlenmeyer flasks using the live-dead fluorescent dye TOTO-1. Results: The MIC values of 4.6 to 32.1 mM of these poultry-feed-specific fungi were well in the range as described in the literature. Propionic acid has a fungistatic action on conidia (spores) that were still able to germinate for minimally 60% after a 24-hour treatment. Germ tubes) already after a 30 min treatment with 31 mM propionic acid.

**Significance:** Propionic acid can exhibit both fungistatic and fungicidic effects with the same fungi.

#### P2-63 Response of *Listeria monocytogenes* Biofilms to Sanitisers Used in Ready-to-Eat Processing Environments

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Introduction: Biofilms are omnipresent in the food processing environment and grow and proliferate under various conditions Dissimilar growth conditions result in diverse biofilm structures and communities that respond differently to sanitation efforts. Purpose: In this study, the response of L. monocytogenes monoculture biofilms to sanitation chemicals was evaluated using a CO, evolution measurement system (CEMS). The CEMS is an effective method with which to study biofilms under flow conditions since it accurately simulates conditions in a water drain environment. Methods: Protocol development was conducted since L. monocytogenes biofilms have not been studied using this system. Listeria monocytogenes isolates 51 and 135 were selected since they represent the most prominent ribotypes isolated from the RTE factory environment and they were also isolated from various drains. Four sanitisers representing quaternary ammonium compounds (QAC) peracetic acid, and alternative chemicals were evaluated using their manufacturer prescribed minimum concentration and contact time. Responses were classified as the biofilm displaying the development of resistance over time or being eradicated.

**Results:** Peracetic acid sanitiser and the proprietary QAC chemical showed no bactericidal effect. A proprietary QAC-free chemical yielded satisfactory results. This has laid the foundation on which to study the effect that flushing sanitisers down drains without detergent washing has on the persistence of *L. monocytogenes* biofilms. **Significance:** The possibility of QAC resistance among the isolates is further supported by the response of *L. monocytogenes* biofilms to the QAC-free (QFS) version. No resuscitation of the biofilm was observed after only two treatments. This is evidence of its effectiveness in the eradication of *L. monocytogenes* biofilms found within the drains of the RTE food factory environment.

#### P2-64 Validation of Abiotic Surrogates for Rapid Verification of Equipment Sanitation

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Introduction: Effective sanitation of equipment is a critical step in an integrated food safety system. Existing sanitation verification methods such as microbial sampling or ATP have significant shortcomings: they are not always specific and may require days to produce results. **Purpose:** This laboratory study is intended to validate abiotic bacterial surrogates (SaniTracers) for the rapid verification of solid-surface sanitation processes.

**Methods:** Abiotic bacterial surrogates were manufactured by encapsulating short, naturally occurring DNA sequences within food-grade material particles and were engineered to attach to stainless steel surfaces and degrade under the action of sanitizers in a manner comparable to non-pathogenic *E. coli*. Large (30 cm×30 cm) stainless steel coupons were inoculated with either the abiotic surrogates (*n*=55) or non-pathogenic *E. coli* (*n*=18) and treated with a standard protocol employing a 150 ppm sodium hypochlorite solution. Bacterial counts were measured using traditional microbiological methods, the surrogates were quantified by PCR, and the reductions of both were calculated.

**Results:** The reductions of both the *E. coli* and the surrogate formulation used in these experiments were normally distributed with W values of 0.97 for both. The mean surrogate reduction, expressed in Cq units, was 8.3 with sd=2.56. The mean log reduction for *E. coli* was 4.22 with sd=0.909. Studies with three pathogenic bacteria (*E. coli* O157, *Salmonella* Enteritidis, and *Listeria monocytogenes*) and a range of sodium hypochlorite concentrations, are in progress and the results will be reported.

**Significance:** The behavior of these abiotic bacterial surrogates under sanitation of stainless-steel surfaces can be measured with a simple 15-minute test and it can predict with high confidence the lethality of the sanitation process for non-pathogenic *E. coli*. These abiotic surrogates can become the basis for a rapid sanitation verification method.

#### P2-65 Effects of Land Pollutant Sources on the Shellfish Growing Area at Yangyang-Gun and Gangneung-Si in East Sea, Korea

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**Introduction:** The production, consumption and exportation of Korean marine products have increased every year since 1990. For this reason, shellfish farming is increasing in Korea. The assurance of safety is very important on shellfish farms because they are often affected by land-based pollutant sources, domestic water, stream water and sewage water.

**Purpose:** This study was carried out to evaluate the effect of land pollutant sources on shellfish farms at rainfall time in Yangyang-gun and Gangneung-si, Korea.

**Methods:** A total of 100 samples (35 of land pollutants, 55 of seawater, 10 of shellfish) were collected from Ganghyeon-myeon, Yangyang-gun to Okgye-myeon, Gangneung-si in East Sea, Korea. Quantitative analysis of the coliform group, fecal coliforms and *Escherichia coli* were carried out using the most probable number (MPN) method by the Recommended Procedures for the Examination of Seawater and Shellfish method of the American Public Health Association (APHA).

**Results:** The fecal coliform levels were affected by stream water and sewage water near the beach. The range of fecal coliforms was 6.8 to 17,000 MPN/100 ml before rainfall. The range of fecal coliforms sharply increased to 39 to 54,000 MPN/100 ml by the first day after rainfall, slowly decreased to 22 to 22,000 MPN/100 ml by the third day after rainfall, and recovered to 7.8 to 21,000 MPN/100 ml on the fifth day after a rainfall.

**Significance:** These results could contribute to assurance of the safety of shellfish from land pollutant sources at rainfall time and during the rainy season.

#### P2-66 The Effect of Combined Treatment with Sodium Hypochlorite and Citric Acid for Reducing *Bacillus cereus* in Dairy Processing Plants

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**Introduction:** In a dairy processing plant, there is a clean-in-place (CIP) system for sanitizing the circulating pipeline. Alkali and acid solutions are used for removing residual protein and milk stone and eliminating harmful bacteria. However, it is not easy to perfectly eliminate residual cells or milk dross. Especially since *Bacillus cereus* has heat resistance, high-temperature pasteurization may not be enough to eliminate the bacteria.

**Purpose:** The results show that the combination of sodium hypochlorite and a citric acid solution can effectively reduce *B. cereus* in the CIP system.

**Methods:** There were two types of experiments. In the first, sanitizing was performed of biofilm-coated stainless steel (SS) chips with sodium hypochlorite (30, 50, 100, and 200 ppm) and citric acid (one, three, five, and seven percent). Chips with sodium hypochlorite were shaken at 1,150 rpm for 10 min to detach sessile cells. After cleaning, the samples were washed with PBS to enumerate cells. The second involved applying combined sodium hypochlorite and citric acid treatment to the plant for removing the biofilm coating the pipeline. The pipeline was exposed to the combination with sodium hypochlorite (200 ppm, two liters) and citric acid (seven percent, two liters) for 20 min. The residual cells were captured after a final rinse (10 min).

**Results:** After sanitizing, we enumerated the residual biofilm. Compared to existing sanitizer, the combined treatment significantly (*P*<0.05) reduced it by 1.17 log CFU/ml. The final reduction of *B. cereus* was 0.93 log CFU/ml. Also, the residual chlorine after combined treatment with sodium hypochlorite and citric acid is reduced by about one-third less than after treatment with existing sanitizer.

**Significance:** As sodium hypochlorite and citric acid are designated as food additives in Korea, this condition can be used as an ecofriendly option for the circulating system in the plant. Although dairy plants already sterilized during manufacturing, this method can possibly be a preventive way to eliminate heat-resistant bacteria.

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#### P2-67 British Retail Consortium (BRC) Global Standard for Food Safety Issue 8 – Requirements for Housekeeping and Hygiene

#### DEB SMITH

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Introduction: Requirement 4.11: Housekeeping and Hygiene is a fundamental requirement of BRCs Global Standard for Food Safety. BRC defines a fundamental requirement as something that "relates to systems that are crucial to the establishment and operation of an effective food quality and safety operation." However, in 2015 BRC reported that, across all food product categories audited against Issue 6 in 2014, the most frequent and consistent pattern of non-conformance emerged in relation to Requirement 4.11<sup>1</sup>. Issue 8<sup>2</sup> of the BRC standard (BRCv8) was released in August 2018. **Purpose:** To raise awareness of the requirements of 4.11 and support the food industry in relation to improved food safety and BRCv8

compliance. Methods: With regard to Requirement 4.11, to

i. review the BRCs non-conformance data in relation to BRCv6 and BRCv7 audits;

ii. review the BRCv8 Standard;

iii. summarise and share the relevant key points from all  $\!\!^3$ 

#### Results:

i. In 2015 BRC reported that the documenting of cleaning procedures was the most frequent non-conformance globally, with over 18% prevalence across sites audited in 2014. In 2016 and 2017 non-conformance against Requirement 4.11 was >16%, with almost 10% related directly to cleaning procedures. The BRC's conclusion was that improvement in the maintenance of housekeeping and hygiene systems was needed.

ii. BRCv8 newly includes clauses specifically related to the design of an environmental monitoring programme (Clauses 4.11.8.1 & 4.11.8.2).

iii. A summary of the key points in BRCv8 relating to 4.11 has been published  $^{\scriptscriptstyle 3}$ 

**Significance:** Given the high rate of non-conformance observed by BRC to the requirements of 4.11, and the importance of these regarding food safety, it is essential to raise awareness and drive improvement in this area.

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#### P2-68\* Use of Predictive Modelling for Fish Quality Evaluation under Temperature Fluctuations

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Introduction: Fish is a highly perishable food due to its biological composition. It is easy to spoil in a relatively short time unless appropriately stored at a required temperature level. Unexpected temperature fluctuations that occur during distribution or storage could accelerate the deterioration of quality which could result in differences between the actual and labeled shelf life. Purpose: This study aims to develop a mathematical model for the effect of fish characteristics on the remaining shelf life under different storage temperature conditions, and to evaluate the impact of temperature fluctuations on the remaining shelf life of tilapia fish. Methods: Fresh tilapia was stored at controlled temperature (5, 10, 15, 20 and 25°C). The evaluation of fish characteristics and shelf life modeling were based on the changes of quality parameters (pH, TVBN, and sensory). Temperature dependence of quality loss rates was modeled by the Arrhenius relation and then validated under dynamic conditions (five hours at 20°C, one hour at 37°C, and five hours at 15°C).

**Results:** Quality of fish gradually deteriorated over storage time. The initial value of TVBN was found to be  $5.07\pm1.5$  mg/100mg. The remaining shelf life of tilapia fish in this study was estimated based on TVBN values. The remaining shelf life of tilapia fish at 5, 10, 15, 20 and 25°C was estimated to be 349, 145, 65, 25, and 15 h, respectively. The TVBN value was estimated to increase up to 14.7 mg/100mg after simulation under dynamic temperature conditions and it would have a remaining shelf life of up to 12.6 months if it is stored at -18°C.

**Significance:** The findings in this study offer a novel view of predicting the safe storage for fisheries products.

#### P2-69 Norovirus and *E. coli* Contamination in French Marketed Oysters

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Introduction: Shellfish are filter feeders that can accumulate human enteric pathogens (bacteria, viruses or protozoa). Consumption of raw or undercooked shellfish can lead to transmission of human pathogens, noroviruses (NoV) being the most frequently agents implicated in shellfish-borne disease. Microbiological monitoring of production area classifications for bivalve molluscs is carried out in European Union (EU) with *Escherichia coli* used as a faecal indicator. **Purpose:** The present study was conducted to evaluate the prevalence of NoV in French marketed oysters, as well as *Escherichia coli* concentrations

**Methods:** Sampling was performed by the veterinary service for one year. In total 608 samples were collected (152 between January and April, 233 between May and October and 223 between November and December). Nov was quantified according to the standard ISO 15216-1 (RT-qPCR) and *E. coli* according to the standard ISO 16649-3 (MPN method).

Results: Noroviruses were detected in 6.6% of samples with a seasonal impact (January through April: 11.8% May through October: 3.0% and November through December 6.7%). For the 16 samples above the limit of quantification, concentrations between 50 and 700 RNA copies/g of digestive tissue (DT) were obtained. All other samples were contaminated at levels below the limit of quantification (50 RNA copies/g TD). Only four samples were non-compliant with EU regulations, with an Escherichia coli count above the regulatory threshold of 230 MPN/100 g of flesh and intra-valvular liquid. Contamination levels ranged from 330 to 490 E. coli /100g of flesh and intra-valvular liquid. No correlation between Escherichia coli contamination and norovirus contamination was observed. Significance: Prevalence and levels of norovirus contamination detected in these oyster samples are low. However, this level detected may be sufficient to induce gastroenteritis symptoms in consumers.

#### P2-70 Marine Biotoxin Contamination of Raw Bivalve Molluscs Commercially Available in Poland from 2009 to 2018

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Introduction: Most plankton produce marine biotoxins which are harmful to humans. Molluscs, as filter-feeder organisms, accumulate different contaminants such as biotoxins. They include paralytic shellfish poisoning (PSP), diarrhoeic shellfish poisoning (DSP) and amnesic shellfish poisoning (ASP), caused by domoic acid. Serious health problems can appear for humans after consumption of contaminated shellfish. There are several symptoms caused by marine biotoxins such as nausea, cramps, vomiting, weakness, dysphasia, dysphonia, respiratory paralysis, diarrhea, abdominal pain, ataxia and memory loss, headaches, disorientation, coma, and dizziness. Severity depends on the individual sensitivity of the human, the type and the quantity of consumed toxins. In extreme cases of contamination, death can result. The legal limit for PSP content in the meat of mussels is 800 µg/kg, for ASP 20 mg/kg, and for DSP 160 µg/kg.

Purpose: The aim of this study was the determination of biotoxin levels in molluscs available at retail in Poland.

Methods: The following ELISA tests were used to detect biotoxins: PSP - Ridascreen Fast PSP S.C. R-biopharm, Germany; DSP -Okatest, Zeu-Immunotec, Spain; ASP Elisa kit for the quantitative determination of domoic acid, Biosense, Norway. Determination of the content of individual marine biotoxins was carried out on eight species of live bivalve mussels: blue mussel, cockle, dog cockle, great scallop, hard clam, Japanese carpet shell, oyster and razor clam. **Results:** In 465 samples of mussels marine biotoxins were detected below the permitted levels in 42.37% of the samples. The maximum amounts of PSP and ASP biotoxins were found in great scallops (532.6 µg/kg and 1.0 mg/kg respectively) and the peak for DSP was in blue mussels (107 µg/kg) but all were below the legal limits. **Significance:** The study showed that the raw shellfish available at Polish markets are safe for the consumers.

#### P2-71 Development of Most Probable Number-Polymerase Chain Reaction (MPN-PCR) as a Sensitive Method for the Detection and Enumeration of *Vibrio vulnificus*

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**Introduction:** *Vibrio vulnificus* is an opportunistic pathogen for humans and infection as well as mortality, can result from consuming contaminated seafood. The most frequent and common method employed for the detection of *V. vulnificus* is the plate-count method using thiosulfate citrate bile salts sucrose (TCBS) agar. However, the TCBS agar plate count method is not suitable to detect damaged *V. vulnificus* due to the difficulty in recovering damaged cells. Hence, a new quantitative counting method needs to be developed for the accurate and robust detection of *V. vulnificus*.

**Purpose:** The objective of the present study is to establish a most probable number polymerase chain reaction (MPN-PCR) method for the quantitative counting of *V. vulnificus* cells.

**Methods:** For the counting and detection of *V. vulnificus* we have developed and used an MPN-PCR method. The results obtained from the MPN-PCR method were also compared to the results of a viable cell count spread plate method (Luria-Bertani (LB) agar with 2% NaCI, or TCBS. The MPN-PCR method includes three steps: inoculation of serially diluted cell culture into peptone water for pre-incubation, selection of the tube which has turbidity, and PCR on the selected tube.

**Results:** The MPN-PCR method showed 2-log higher cell numbers compared to the TCBS and LB-agar dmethod.

**Significance:** The MPN-PCR method can be used as a robust and sensitive method for the quantitative detection of viable cell counts of *V. vulnificus* in fish as well as shellfish samples.

#### P2-72 Foodborne Outbreak of Gastroenteritis Caused by Norovirus in a Restaurant of Northern Italy

Guido Finazzi<sup>1</sup>, Barbara Bertasi<sup>1</sup>, **ENRICO PAVONI**<sup>1</sup>, Elisabetta Suffredini<sup>2</sup>, Camillo Gandolfi<sup>3</sup> and Marina Nadia Losio<sup>1</sup>

<sup>1</sup>IZSLER, Brescia, Italy, <sup>2</sup>Istituto Superiore di Sanità, Rome, Italy, <sup>3</sup>ATS of Bergamo, Bergamo, Italy

**Introduction:** In case of foodborne illness, in addition to diagnostic confirmation, the biggest challenge is to identify the offending food. Disease diagnosis and the patient's care are of primary importance, but it is equally fundamental to identify the food responsible in order to prevent the spread of infection.

**Purpose:** Object of this study was to describe a case of foodborne illness that occurred in Northern Italy at the end of 2017.

**Methods:** Two cases of gastroenteritis were reported to the local hospital of Bergamo as part of a larger outbreak involving subjects from two different groups (27 and 57 individuals) who participated in social dinners in the same restaurant. Gastroenteritis symptoms (abdominal pain, vomiting and diarrhea) were developed by 39 subjects (19 from the first group and 20 from the second) within 18 to 36 hours from dinner. The menus for both dinners included raw oysters and 39 persons out of 48 who had consumed them, were

symptomatic. The batch of oysters still available in the restaurant was collected and screened for *Salmonella* spp., pathogenic vibrios, hepatitis A virus (HAV), Norovirus (NoV) and liphophlic marine biotoxins.

Results: Oysters were positive for NoV genogroup I and II, and negative for all the other investigated hazards. Due to poor cooperation from involved subjects, clinical specimens could not be collected to perform a molecular comparison with the NoV detected in oysters. Based on the epidemiological investigation, oysters were considered as the main suspected source of infection. Significance: NoV contamination in bivalve shellfish remains a matter of public health concern. Several studies underline the importance of post-harvest depuration and good manipulation practices to guarantee the safety of these products. However, given the risk of contamination in primary production, it would be desirable to include virological parameters among Food Safety Criteria (EC Reg. 2073/05) for bivalve shellfish intended to be consumed raw.

#### P2-73 Investigation of Microbial Contamination Levels in Minimally Processed Fishery Products in Korea: Crustaceans, Echinodermata, Tumicata and Roe

HYEYOUNG SHIN, Hyeonjeong Kim and Ilshik Shin Gangneung-Wonju National University, Gangneung, South Korea

Introduction: Minimally processed fishery products (MPFs), seafood manufactured through primary processing such as washing, heading, gutting and shucking etc. or secondary processing including salting, drying, refrigeration andothers, account for the majority of processed fishery products (PFs) in Korea. However, MPFs have not been legally classified as PFs until now. For that reason, these commodities have not been managed for safety.

**Purpose:** The purpose of this study was to contribute to the safety of fishery products by investigating microbial contamination levels in Crustacean, Echinodermata, Tunicata and Roe frequently consumed by Koreans.

**Methods:** The samples, 14 frozen pre-cut swimming crabs, 17 frozen shrimp meats, 14 frozen crab meats, 25 dried shrimps, 10 dried sea cucumbers, 15 live sea cucumbers, four frozen *Styela clava*, five frozen *Styela plicata*, 15 shelled sea squirts, 15 frozen Alaska pollack roes, seven frozen salmon roes and nine frozen flying fish roes were purchased from retail outlets and online markets. Aerobic Plate Count (APC), *Escherichia coli, Staphylococcus aureus*and *Vibrio parahaemolyticus* were evaluated in samples, respectively, according to the method described in the Korean Food Code.

**Results:** APC was detected below  $5.0 \times 10^5$  colony forming unit (CFU)/g except in dried shrimp (range of CFU/g:  $1.5 \times 10^4$  to  $3.0 \times 10^6$ ). *E. coli* and *V. parahaemolyticus* were detected below 18 most probable number (MPN)/100 g and 15 CFU/g respectively. *S. aureus* in eight of 150 samples was detected above  $1.0 \times 10^2$  CFU/g (regulation limit in Food Code, Korea). Therefore, sanitation management is needed for insurance of safety against *S. aureus*. **Significance:** The findings of this study could contribute to reduce microbial hazards and to construct safety management systems of MPFs.

## STUDENT TRAVEL SCHOLARSHIP AWARD RECIPIENTS



#### Maria Gkerekou Agricultural University of Athens Athens, Greece

Maria Gkerekou is a Ph.D. candidate in the Department of Food Science and Human Nutrition at the Agricultural University of Athens in Athens, Greece.

Her research is focused on the evaluation of the impact of interactions

between *Listeria monocytogenes* strains mimicking their co-existence in dairy food matrices (different structure and/ or oxygen availability) and their subsequent passage from the gastrointestinal tract and how this co-existence is reflected on their phenotypic responses and gene expression of key virulence genes. Ms. Gkerekou's research team received the student competition for Best Poster presentation at IAFP's European Symposium in 2016, and she was one of five finalists in the student poster competition at IAFP's European Symposium in 2018.



#### Yifan Zhang ETH Zurich Zurich, Switzerland

Yifan Zhang is a Ph.D. candidate in the Sustainable Food Processing Group at ETH Zurich in Zurich, Switzerland. Her research includes investigating milder, non-thermal food processing approaches to control bacterial spores. The processing technologies currently under investi-

gation are high hydrostatic pressure (for high water activity foods) and low energy electron beam (for low water activity foods). Ms. Yang received second place for her research in the Developing Scientist Competition at IAFP 2018.

## PAST EUROPEAN STUDENT TRAVEL SCHOLARSHIP RECIPIENTS

2014 - Erika Georget

- 2015 Emily Jackson
- 2016 Amanda Demeter

2017 - Christian Hertwig

2018 – Katrien Begyn and Giannis Koukkidis

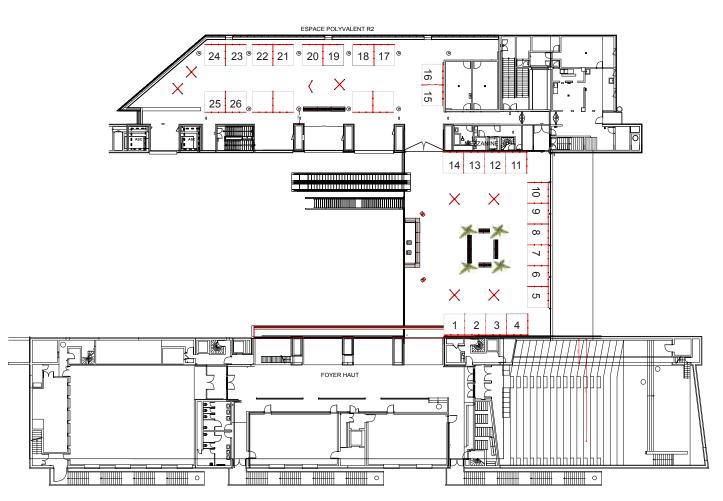


# **EXHIBITORS**

24–26 April 2019 – Nantes, France

## **EXHIBITORS**

## **EXHIBITION HALL**



#### Stand Number

#### Exhibitor

1	Bruker Daltonics
2	Medical Wire & Equipment Co. Ltd.
3	Tasmanian Institute of Agriculture
4	ILSI Europe
5	Check-Points B.V.
6	Novolyze
7	Chihon Biotechnology
8	R-Biopharm AG
9	bioMérieux
10	Pall Corporation
11	Eurofins NDSC Alimentaire France
12	ELISA Systems
13–14	3M
15–16	Hygiena International Ltd.
17	Bio-Rad Laboratories
18	Oxford Nanopore Technologies
19	Thermo Fisher Diagnostics
20	GFSI–The Consumer Goods Forum
21	Nemis Technologies
22	ICFMH (International Committee
	on Food Microbiology and Hygiene)
23	Decon7 Systems, LLC.
24	
25–26	Promega

#### **EXHIBIT HOURS**

Wednesday, 24 April 2019 – 10.00 – 18.00 Thursday, 25 April 2019 – 10.00 – 16.00

#### **EXHIBIT EVENTS**

#### Wednesday, 24 April 2019

10.00 Networking Coffee Break 12.00 Lunch 15.00 Networking Coffee Break 17.00 Reception

#### Thursday, 25 April 2019

10.00 Networking Coffee Break 12.30 Lunch 15.00 Networking Coffee Break

Be certain to visit our Exhibit Area to explore the latest techniques, applications, products and services in food safety demonstrated by our valuable exhibitors.

## **EXHIBITORS**

#### 3M Boulevard de l'Oise 95006 Cergy Pontoise Cedex France Phone: +33 130 31 85 70 www.3m.com

www.3m.com The Food Safety business is part of the global science-based technology company, 3M. With \$32.8 billion in sales, 93,000 employees and over 55,000 products, 3M connects with and serves

customers around the world. 3M invests about six per cent of its sales revenue in research and development and is recognised as a global innovator, producing more than 3,000 patents each year.

3M offers a comprehensive range of food safety and environmental monitoring solutions: hygiene indicator tests, rapid hygiene management and allergen control, advanced pathogen detection, sample handling, and supply chain product temperature monitoring.

bioMérieux 100 Allee Louis Pasteur Campusde L'Etoile Marcy L'Etoile, 69290, France www.biomerieux.com



BIO-RAD

Stand 17

Stand 13-14

bioMérieux develops and produces in vitro diagnostic solutions (instruments, reagents and software) for private and hospital laboratories, mainly for the diagnosis of infectious diseases. The results obtained from a patient sample (blood, urine, stool, cerebrospinal fluid, saliva, etc.) provide doctors with information to support their decisions.

For 25 years, bioMérieux has also applied the expertise acquired in the clinical sector to meeting industrial microbiology needs, making it possible to manage contamination risks in agrifood, pharmaceutical and cosmetic products, at each step of the production chain.

Bio-Rad Laboratories 3 Boulevard Raymond Poincare Marnes la Coquette, 94230, France Phone: +1.47.95.60.00 www.bio-rad.com/foodscience

As a powerful, accurate alternative method, iQ-Check real-time PCR kits provide fast, qualitative detection of major pathogens in a variety of complex food matrices and environmental samples. The 96-well thermal cycler has a small footprint and is well-suited for both low- and high-volume users. Our iQ-Check Prep Automation Solution, designed for maximum flexibility and traceability, is optimized for higher throughput labs providing true reliable walk-away automation, sample verification, liquid level monitoring and bi-directional LIMS integration. As a complete solution provider, we also offer enrichment media and RAPID'Chromogenic media for easy bacterial detection, identification and quality indicator enumeration.

Bruker Daltonics Fahrenheitstrasse 4 28359 Bremen, Germany Phone: +49.0.421.2205.0 www.bruker.com/microbiology



Bruker's leading MALDI-TOF system, the MALDI Biotyper<sup>®</sup>, has in no time revolutionized the way microbial identification and confirmation are performed, providing specific and reliable results within minutes.

The MALDI Biotyper's validation and certification according to the new ISO 16140-part 6 standard by MicroVal and according to the Official Method of Analysis program (OMA) of the AOAC International, offers now a reliable "one system – one workflow" solution for microbiology of the food chain, in terms of safety and quality. Implementing the MALDI Biotyper in the microbial workflow can directly translate to significant cost savings by accelerated testing along the entire process chain.

Check-Points B.V. Binnenhaven 5 Wageningen, 6709PD, The Netherlands Phone: +31.317.453.908 www.checkandtrace.com



Stand 5

Check-Points' innovative Check&Trace Salmonella method can discriminate over 300 Salmonella serotypes, including the most relevant ones like S. Typhimurium and S. Enteritidis, due to the differences in their DNA sequences. This allows the Check&Trace Salmonella test to significantly decrease serotyping lead times and enable quick tracing. The Check&Trace Salmonella confirms Salmonella presence and the serotype with a single test in one day. http://checkandtrace.com/ info@checkandtrace.com.

#### Chihon Biotechnology P.O. Box 9514 Naperville, IL 60564, USA Phone: +1 630.670.5701 www.chihonbio.com

CHIHONBIO Stand 7

Founded in 2003, Chihon Biotechnology is a leading manufacturer of natural preservatives of Nisin and Natamycin, as well as other preservatives, such as Lauryl arginate ethyl ester. We offer technical support to the research and development. We also welcome the opportunities of contract manufacturing of other preservatives and food additives.

Decon7 Systems, LLC. 8541 E. Anderson Drive, Suite 106 Scottsdale, AZ 85255 Phone: +1 480.339.2858 www.d7food.com



Stand 23

The D7 Difference: Decon7 manufactures and markets antimicrobial, sanitizing and degreasing solutions that are scientifically formulated to perform in the most challenging environments. In university studies, D7's patented technology consistently outperformed the competition in the presence of soil load conditions. D7 is registered and approved for use as a Food Contact Surface Sanitizer and to penetrate, kill and remove Biofilms. D7, the power of foam.

ELISA Systems 8 Cox Road Windsor, 4030, Australia Phone: +1 61.7.3625.9000 www.elisasystems.com



Stand 12

ELISA Systems offers an extensive range of high quality Food Allergen test kits to the food industry and our Food Allergen test kits are used extensively all over the world. We have a wide range of both traditional ELISA-based kits, which are developed and produced in Australia, as well as our new rapid detection devices. Our team are technical experts in the field of Food Allergen testing, as well as ensuring our ongoing participation in International technical special interest groups on Food Allergens. We look forward to working with you for all of your Food Allergen testing needs. 🔅 eurofins Stand 11

Stand 20

#### Eurofins 9 rue Pierre-Adolphe Bobierre 44300 Nantes, France Phone: +33.2.51.83.21.00 www.eurofins.fr

Eurofins is one of the leading service providers for microbiological testing in Europe. The microbiological testing portfolio includes analytical services to detect pathogenic microbes and their toxins (such as *Salmonella*, *Listeria*, *Legionella*, *Campylobacter*) or organisms causing decay (such as yeasts, fungi or moulds), to provide hygiene checks (e.g., site inspections but also sterility tests in packaging materials, measurement of airborne microbes) with the aim of making foods safer for our clients, meeting statutory requirements and protecting the consumer. Eurofins offers both classical methods and modern methods (e.g., PCR methods, MALDI TOF testing, NGS analysis) for rapid microbiological results.

During IAFP's European Symposium, please discover our poster 24/04 on "A New Validation Method for Untargeted NGS Analysis," assist to the presentation of Thomas Charrier 25/04 at 9h30 on "Fast MALDI Typing to Drive Decision Making and Source Tracking." And do not hesitate to visit us at our stand.

GFSI – The Consumer Goods Forum 22/24 rue du Gouverneur Eboue Issy-les Moulineaux, 92130, France Phone: +33.1.82.00.95.88 www.mygfsi.com

The Global Food Safety Initiative (GFSI) brings together key actors of the food ecosystem to collaboratively drive continuous improvement in food safety management systems around the world. With a vision of safe food for consumers everywhere, food industry leaders created GFSI in 2000 to reduce food safety risks and inefficiencies while building trust throughout the supply chain. The GFSI community is composed of experts from the full-stake-holder spectrum, across industry and international organisations to governments and academia. GFSI is powered by The Consumer Goods Forum (CGF), a global industry network working to support Better Lives Through Better Business.

#### Hygiena International Ltd. Whygiena Stand 15–16 8 Woodshots Meadows, Croxley Park Watford, Hertfordshire WD18 8YU, United Kingdom Phone: +01923.601.988 www.hygiena.com

Hygiena delivers rapid microbial and allergen detection, monitoring, and identification solutions to a wide range of industries, including food and beverage, health care, hospitality, pharmaceuticals, and personal care. Utilizing advanced technologies and patented designs, Hygiena provides industry-leading ATP monitoring systems, PCR-based foodborne pathogen detection, DNA fingerprint molecular characterization systems, allergen tests, environmental collection devices, etc. Hygiena is committed to providing customers with high-quality innovative technologies that are easy-to-use and reliable, backed by excellent customer service and support. Its headquarters are in California and it has offices in 5 countries and over 80 distributors in more than 100 countries worldwide. on Food Microbiology and Hygiene) Ghent University, Faculty of Bioscience Engineering Dept. of Food Technology, Food Safety and Health, Coupure links 653 Ghent, 9000, Belgium Phone: +34.972.630.052 www.icfmh.org

**ICFMH (IVZW International Committee** 

Since 1953, the ICFMH represents the IUMS in all issues related to food microbiology. Its major aim is to contribute to food safety internationally with activities such as the "FoodMicro" Conference, workshops, publications (*International Journal of Food Microbiology*), mobility grants and awards for young scientists, and by supporting and initiating education and training in food microbiology. The ICFMH particularly focuses on developing countries.

The 27th International ICFMH Conference, FoodMicro 2020, will take place in Athens (Greece), 7–10 September 2020, with the theme "Next Generation Challenges in Food Microbiology" (http:// foodmicro2020.com/). We shall be pleased to welcome you there!

#### ILSI Europe Avenue E Mounier 83 Brussels, 1200, Belgium Phone: +32.0.277.59.121 www.ilsi.eu

Founded in 1986, ILSI Europe fosters collaboration among the best scientists from industry, academia and the public sector to provide evidence-based scientific solutions and to pave the way forward in nutrition, food safety, consumer trust and sustainability. To deliver science of the highest quality and integrity, scientists collaborate and share their unique expertise in expert groups, workshops, symposia and resulting publications.

Medical Wire & Equipment Co. Ltd. Leafield Industrial Estate Corsham, SN13 9RT, United Kingdom Phone: +44.1225.810361 www.mwe.co.uk

Manufacturers of effective surveillance products for Food & Pharma since 1952. Medical Wire produces reliable microbiological sampling solutions of critical surfaces from swabs to sponges:

- NRSII Transwab<sup>®</sup> effective sampling of food contact surfaces after cleaning
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- Isolation Transwab<sup>®</sup> active surveillance for *Listeria*, *Salmon-ella* & Coliforms

ISO 18593 Compliance: NRS II Transwab<sup>®</sup> and Polywipe<sup>™</sup> products can already be used in surface monitoring programmes based on the new ISO 18593:2018. Diluents and neutralisers are fully compliant.

Nemis Technologies AG Ueberlandstrasse 109 Duebendorf, 8600, Switzerland Phone: +41 44 820 71 52 www.nemistech.com

Founded in 2018, Nemis Technologies AG is a Swiss diagnostics company in the field of rapid, precise, easy-to-use and low-cost screening and detection of pathogenic bacteria. Its AquaSpark<sup>™</sup> technology has proven to deliver a significant reduction in timeto-results for various bacteria over current market standard, thus providing a very powerful technology to prevent proliferation and dissemination of dangerous microorganisms at a large scale.





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Stand 2

Stand 21

Stand 4

#### NOVOLYZE Stand 6

#### Novolyze 50 rue de Dijon Daix, 21121, France Phone: +33.983.694.213 www.novolyze.com

Novolyze is a Food Safety company. Our mission is to develop and commercialize innovative technologies to help the food industry manufacture safer food, while ensuring strong compliance with international Food Safety & Quality Standards. Our innovative approach to Food Safety relies on the utilization of cutting-edge microbiology solutions, combined with the latest developments in digital, IoT and machine learning. To learn more about our company and solutions, visit www.novolyze.com.

#### Oxford Nanopore Technologies Stand 18 Gosling Building, Edmund Halley Road Oxford, OX4 4DQ, United Kingdom Phone: +44.845.034.7900 www.nanoporetech.com

Oxford Nanopore Technologies has developed the world's first nanopore DNA and RNA sequencing devices. The MinION is a portable, real-time, long-read, low-cost device designed to bring easy biological analyses to anyone, whether in scientific research, education or real-world applications such as disease/pathogen surveillance, environmental monitoring, food chain surveillance, self-quantification or microgravity biology.

Nanopore sequencing is used by a thriving community of thousands in more than 70 countries, enabling a myriad of applications within the laboratory environment and in the field. The GridION and PromethION devices serve users with larger projects or more samples. All devices are for research purposes only.

#### Pall Corporation Planiger Str. 137 Bad Kreuznach, 55543, Germany Phone: +49.671.8822.201 www.pall.com/foodandbev

Pall GeneDisc Technologies, part of Pall Corporation, is the provider of a unique qPCR-based platform. GeneDisc<sup>®</sup> Systems offers an easy-to-use and cost-effective multi-parametric molecular diagnostic solution, allowing the user to obtain up to twelve different results from a single-sample drop, in an hour.

Pall GeneDisc Technologies aims to provide you with proven, accurate and validated tests for real-time detection of microorganisms in food and beverage. GeneDisc products include a unique, high troughput and flexible solution for pathogenic *E. coli* O157 and Shiga Toxic *E. coli* monitoring as well as for *Listeria* and *Salmonella*. Easy, Fast and Reliable: no need to compromise.

#### Promega

#### Stand 25–26

Food and Beverage Stand 10

24, chemin de Verrieres Charbonnieres-les-Bains, 69260, France Phone: 33.4.37.22.50.00 www.promega.com

As a world leader in applying genomics and cellular biology expertise to develop high value products for the Life Sciences, Promega Corporation understands that today's food quality, GMO and authenticity testing challenges require creative solutions. We have developed systems that simplify plant and food DNA extraction and seamlessly integrate into food testing workflows. Stop by our booth to learn more about successful approaches and tools for enabling GMO and food pathogen testing.

#### R-Biopharm AG An der Neuen Bergstrasse 17 Darmstadt, 64297, Germany Phone: +49.6151.81020 https://food.r-biopharm.com

R-Biopharm has developed innovative products in the field of food and animal feed analysis and clinical diagnostics since 1988. Food producers, laboratories and hospitals throughout the world appreciate our high quality and customer-oriented solutions. R-Biopharm is family-owned in second generation and combines development, production and sales under a single roof. As a competent partner of the food industry, we offer test systems for a wide range of requirements:

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- Tests for the identification of substances, prohibited residues and adulteration of products
- Microbiological tests for pathogen detection and hygiene management

#### Tasmanian Institute of Agriculture College Road Tasmania Hobart, 7001, Australia Phone: +813.510.0277 http://www.utas.edu.au/tia

The Tasmanian Institute of Agriculture (TIA), located in Tasmania, Australia, is dedicated to research and development of sustainable agricultural industries. Founded in 1996, it is a collaborative effort of the University of Tasmania (UTAS) and the Tasmanian Government.

ComBase (www.combase.cc), a partnership between TIA and the U.S. Department of Agriculture, is a free public database that describes microbial responses to food environments, and is accessed by more than 56,000 registered users. CB Premium (www.cbpremium.org), founded by TIA, uniquely focuses on foodbased, peer-reviewed predictive models that help the food industry develop Food Safety Plans and comply with regulatory guidelines.

 Thermo Fisher Diagnostics
 Thermo Fisher
 Stand 19

 6 rte. Paisy
 Dardilly, 69570, France
 Phone: +33.4.72.52.3370

 www.thermofisher.com/microbiology
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Thermo Fisher Scientific supplies innovative solutions for the world's food manufacturing and testing industry. With applications for food safety and authenticity that span the food production process – from environmental monitoring, testing of raw materials and ingredients through to finished products and beyond – we provide a broad range of products and services.

At Thermo Fisher Scientific, we believe we are uniquely positioned to help the food industry effectively protect its customers, brand and reputation by delivering simpler, faster and smarter solutions.

To discover more, stop by our booth at the 2019 IAFP European Symposium or visit us at thermofisher.com/foodandbeverage.



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24–26 April 2019 – Nantes, France

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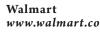
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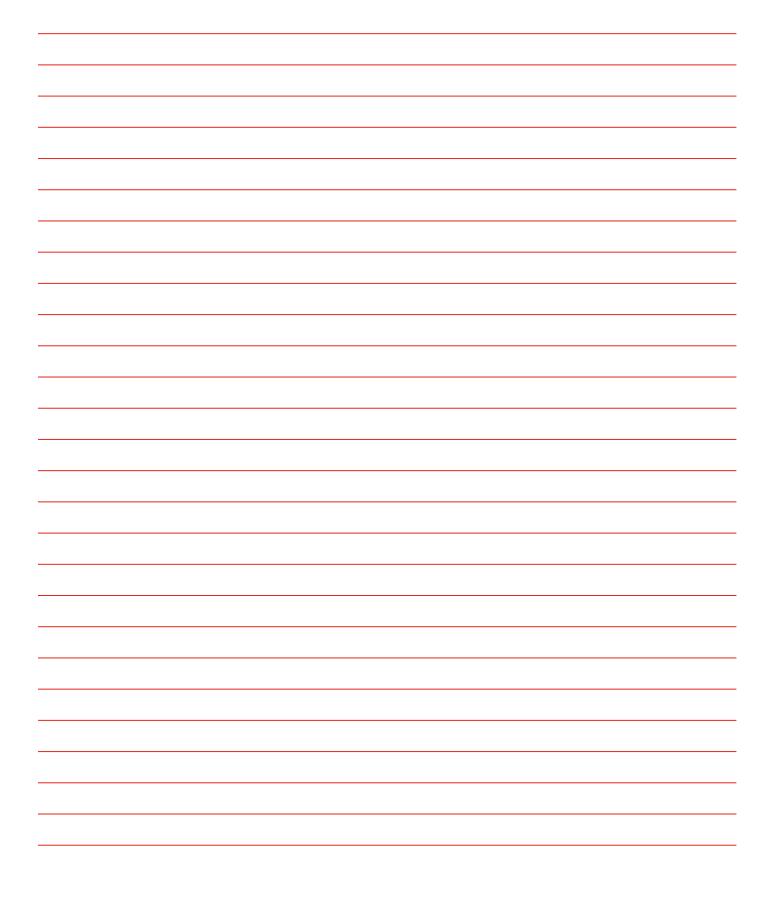
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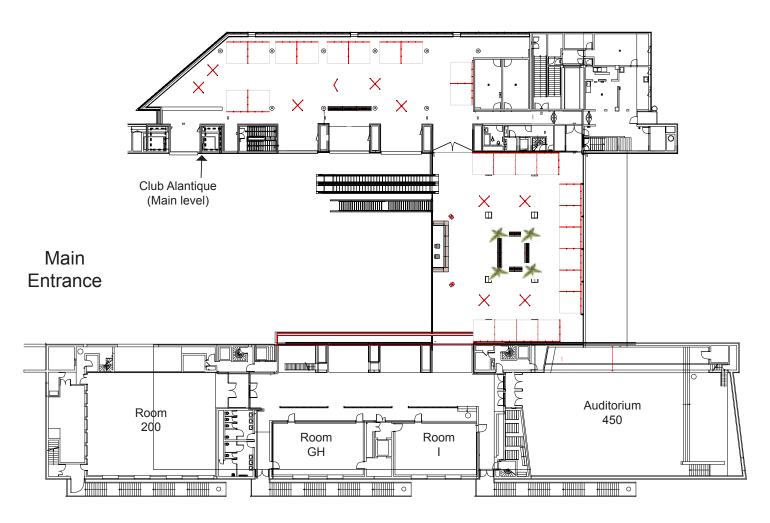
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