

IAFP EUROPEAN SYMPOSIUM ON FOOD SAFETY



25–27 APRIL 2018
Stockholm, Sweden

PROGRAMME

HELD AT THE BREWERY CONFERENCE CENTRE STOCKHOLM

ORGANIZED BY



International Association for
Food Protection®

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

Room	Riddarsalen	Fogelstrom	Galleriet	Arkaden	Exhibit Hall
Wednesday, 25 April 2018					
Wednesday 8.30 - 10.30	Opening Session <i>Malarsalen</i>				
Wednesday 10.30 - 11.00	Coffee/Networking Break <i>Exhibit Hall - Masshallen</i>				Poster Session 1 – Beverages and Water, Communication Outreach and Education, General Microbiology, Meat and Poultry, Microbial Food Spoilage, Non-Microbial Food Safety, Risk Assessment, Sanitation, Seafood
Wednesday 11.00 - 12.30	RT1 - How Much of a Mystery Remains with Whole Genome Sequencing?	S1 - FSMA & FSVP – Impact on the Global Food Industry	S2 - Teaching Food Safety to Dietitians: Toward an International Network	Technical Session 1 – Response to Specific Environmental Conditions	
Wednesday 12.30 - 14.00	Lunch <i>Exhibit Hall - Masshallen</i>				
Wednesday 14.00 - 15.30	S3 - Drivers and Dynamics of Antimicrobial Resistance in Food: Beyond Antimicrobial Use in Animals	S4 - Microbiological Reference Methods, What's New, What's Coming?	S5 - Maximising Food Safety through Good Hygienic Design	Technical Session 2 – Intervention Strategies and Modeling	
Wednesday 15.30 - 16.00	Coffee/Networking Break <i>Exhibit Hall - Masshallen</i>				
Wednesday 16.00 - 17.30	S6 - How NGS Technologies Unravel Our Understanding of Food and Food Microbiology	S7 - Importance of Microbiological Criteria and Statistical Underpinning of Sampling and Testing for Food Safety Assurance	S8 - Biofilms and Environmental Monitoring	Technical Session 3 – Pathogens and Antimicrobials	
Wednesday 17.30 - 18.30	Exhibit Hall Reception				
Thursday, 26 April 2018					
Thursday 8.30 - 10.00	S9 - New Approaches for Safety and Quality of Fermented Foods	RT2 - Prediction of Spoilage and Safety with Models: How to be on the Safe Side in a World Full of Variability?	S10 - Validation and Verification – Successes, Pitfalls and Disasters	Technical Session 4 – Risk Assessment	
Thursday 10.00 - 10.30	Coffee/Networking Break <i>Exhibit Hall - Masshallen</i>				Poster Session 2 – Antimicrobials, Applied Laboratory Methods, Dairy and Other Food Commodities, Epidemiology, Food Toxicology, Novel Laboratory Methods, Pathogens, Produce
Thursday 10.30 - 12.00	S11 - Biological Variability in Thermal Processing: Impact for Process Control and Validation - What You Need to Know about Microbiological Variability for Food Quality and Safety Control	S12 - Integrating Microbial Adaptive Trait in Food Safety: Added Value of Biomarkers	S13 - Allergen Control - from Problem to Solution	Technical Session 5 – Molecular Characterization and Risk Assessment	
Thursday 12.00 - 13.30	Lunch <i>Exhibit Hall - Masshallen</i>				
Thursday 13.30 - 15.00	S14 - Risk Benefit Assessment of Food: Past, Present and Future Trends	S15 - Interventions to Reduce Antibiotic Resistance and Antibiotic Use in Animal Production	RT3 - Assessment of Microbial Risk for Fresh Produce	Technical Session 6 – Intervention Strategies and Management	
Thursday 15.00 - 15.30	Coffee/Networking Break <i>Exhibit Hall - Masshallen</i>				
Thursday 15.30 - 17.00	S16 - The Rise of Whole Genome Sequencing: How Do We Share and Interpret the Data Globally?	S17 - Global Occurrence of Mobile Colistin Resistance in Foodborne Pathogens	S18 - Control of Human Pathogens in Plant Production Systems	Technical Session 7 – Meat and Poultry, Seafood, Epidemiology	
Friday, 27 April 2018					
Friday 8.30 - 10.00	S19 - Turning Sequencing and Mass Spectrometry into Routine Testing Tools for Microbial Strain Characterization	S20 - Integrating Scientific Risk Assessment in the Prioritization and Management of Chemical Contaminants in Foods and Raw Materials	S21 - Natural Antimicrobial Preservatives in Foods: Where are We in Terms of Application and Commercialization?	Technical Session 8 – Detection and Typing Methods	
Friday 10.00 - 10.30	Coffee/Networking Break <i>Mastorget</i>				
Friday 10.30 - 12.45	Closing Session <i>Riddarsalen</i>				
Friday 12.45 - 14.00	Farewell Refreshments				

IAFP's 6th Latin American Symposium on Food Safety III Argentine Symposium on Food Safety

25–27 September 2018
Paseo La Plaza Complex – Av. Corrientes 1660
Buenos Aires, Argentina



CAIA
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Inocuidad Alimentaria
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IAFP's 6th Latin American Symposium on Food Safety

III Argentine Symposium on Food Safety

Conferences and round tables
with national and foreign experts

Presentation of papers

Deadline for submission of abstracts:
June 15, 2018

Questions:
caia@aam.org.ar
www.iafp-latino2018.com.ar

Scientific Program:

- Management of food safety in the food chain
- One Health
- New applications of genomics for safety
- Food fraud
- Non-conventional technologies and safety
- International food safety regulatory policies: impact on Latin America
- Antimicrobial resistance in food production and public health
- Physical pollutants, chemicals and allergens
- Biofilms
- Mycotoxins
- Food safety in Latin America: Problems and challenges





PROGRAMME

25-27 April 2018 – Stockholm, Sweden

Wednesday, 25 April – 8.30–10.00



WELCOME TO THE IAFP EUROPEAN SYMPOSIUM

7.30 – 17.00 Registration Open

7.30 – 8.30 Morning Coffee

Exhibit Hours 10.00 – 18.30

Opening Session

Malarsalen

Chair: Jeanne-Marie Membré

8.30 Introduction to IAFP

DAVID THARP, Executive Director, International Association for Food Protection, Des Moines, IA, USA

8.45 Introduction to IAFP's European Symposium

MICKEY PARISH, U.S. Food and Drug Administration, Washington, D.C., USA

8.55 Programme Notes and Recognition of Organising Committee

JEANNE-MARIE MEMBRÉ, INRA, UMR 1014 Secalim, Nantes, France

9.00 The Public Health Agency's Role in Foodborne Disease Outbreaks

KARIN TEGMARK WISELL, The Public Health Agency of Sweden, Solna, Sweden

9.30 Global Lessons of the Swedish Model

IVAR VÅGSHOLM, Swedish University of Agricultural Sciences (SLU), Uppsala, Sweden

10.00 Science and Food Safety Policy at the U.S. Food and Drug Administration

MICKEY PARISH, U.S. Food and Drug Administration, Washington, D.C., USA

10.30 Networking Coffee Break in the Exhibit Area

Poster Session 1 – Beverages and Water Communication Outreach and Education, General Microbiology, Meat and Poultry, Microbial Food Spoilage, Non-Microbial Food Safety, Risk Assessment, Sanitation, Seafood

Authors present during scheduled break times

RT1 How Much of a Mystery Remains with Whole Genome Sequencing?

Riddarsalen

Organizer and Convenor: Tobias Recker

Sponsored by ILSI Europe

11.00 Panelists:

LEEN BAERT, Nestlé, Vers-ches-les Blanc, Switzerland

KATHIE GRANT, Public Health England, Glasgow, United Kingdom

RENE HENDRIKSEN, National Food Institute, Denmark
Technical University, Lyngby, Denmark

PETER MCCLURE, Mondelez International, Birmingham,
United Kingdom

SARAH O'BRIEN, University of Liverpool, Liverpool,
United Kingdom

DANIEL PALM, ECDC, Stockholm, Sweden

12.30 Lunch Available in the Exhibit Area

S1 FSMA & FSVP – Impact on the Global Food Industry

Fogelstrom

Organizer: Richard Brouillette

Convenor: Roger Scheffler

11.00 Exporting to the United States – What Changed for Us?
PRIYA SUNDARAM, Danone, Amsterdam, The Netherlands

11.30 How to Prepare for a Possible FDA Visit
ANETT WINKLER, Cargill, Munich, Germany

12.00 Sanitation Programs under FSMA Regulation
ANDRES RODRIGUEZ LOZANO, Commercial Food Sanitation,
Amsterdam, The Netherlands

12.30 Lunch Available in the Exhibit Area

S2 Teaching Food Safety to Dietitians: Toward an International Network

Galleriet

Organizer: Ellen W. Evans

Convenors: Ellen W. Evans and Victoria J. Gould

11.00 Engaging Dietitians to Promote Safe Food-handling
Messages – Opportunities in the Curriculum
JENNIFER QUINLAN, Drexel University, Philadelphia, PA, USA

11.30 In the Classroom with Dietitians: Student Interest and
Engagement in Food Safety
SANJA ILIC, The Ohio State University, Columbus, OH, USA

12.00 The Importance of Teaching Food Safety to Dietitian
Students and Visions for an International Network of
Dietetic Food Safety Educators
INGELA MARKLINDER, Uppsala University, Uppsala, Sweden

12.30 Lunch Available in the Exhibit Area

T1 Technical Session 1 – Response to Specific Environmental Conditions

Arkaden

Convenor: Koen De Reu

T1-01 Evaluation of Two Surface Sampling Methods for
11.00 Microbiological and Chemical Analyses to Assess
the Presence of Biofilms in Food Companies
Sharon Maes, Son Nguyen Huu, Marc Heyndrickx,
Stephanie Van Weyenberg, Hans Steenackers, Alex
Verplaetse, Thijs Vackier, Imca Sampers, Katleen
Raes, KOEN DE REU, Flanders Research Institute for
Agriculture, Fisheries and Food (ILVO), Melle, Belgium

T1-02 Membrane Proteocomplexomic Approach for *Campylo-*
11.15 *bacter jejuni*
ALIZÉE GUÉRIN, Sheiam Sulaeman, Lucile Bugros,
Armelle Ménard, Emmanuelle Dé, Odile Tresse, SECALIM,
INRA, Oniris, Université Bretagne Loire, Nantes, France

T1-03 *Lactococcus lactis* subsp. *lactis* as a Natural Anti-
11.30 listerial Agent in the Mushroom Industry
Lionel Kenneth Dygico, Paula O'Connor, Maria Hayes,
Cormac Gahan, Helen Grogan, CATHERINE M. BURGESS,
Teagasc, Dublin, Ireland

T1-04 Identification, Spoilage and Biofilm-forming Properties
11.45 of Microorganisms from Surface Contamination
in Food Companies
SHARON MAES, Thijs Vackier, Hans Steenackers,
Alex Verplaetse, Marc Heyndrickx, Koen De Reu,
Flanders Research Institute for Agriculture, Fisheries
and Food (ILVO), Melle, Belgium

T1-05 Evaluation of *Bacillus thuringiensis* Strains Abts-351
12.00 and Abts-1857 within a Simulated Gut Environment
DANIEL ZOMMICK, Valent Biosciences LLC, Libertyville,
IL, USA

T1-06 Butyrate Effect on Extracellular GABA Production
12.15 in *Listeria monocytogenes* 10403s WT and Lactic
Acid Bacteria Isolated from Cheese and Gut
CAROLINA BRUSCHI, Kimon Andreas Karatzas, University
of Reading, Reading, United Kingdom

12.30 Lunch Available in the Exhibit Area

S3 Drivers and Dynamics of Antimicrobial Resistance in Food: Beyond Antimicrobial Use in Animals

Riddarsalen

Organizer: Jeffrey LeJeune

Convenor: Sarah Cahill

Sponsored by Food and Agricultural Organization of the United Nations, funders of this research

14.00 Does Biocide and Disinfectant Use in Food Production Drive Antimicrobial Resistance?

SANJA ILIC, The Ohio State University, Columbus, OH, USA

14.30 The Environment: Source or Sink for Antimicrobial Resistance in Food and Agriculture?

ELIZABETH PARKER, The Ohio State University, Wooster, OH, USA

15.00 Antimicrobial Use and Resistance in Plant-based Agriculture
JEFFREY LEJEUNE, Food and Agriculture Organization of the United Nations, Rome, Italy

15.30 Networking Coffee Break in the Exhibit Area

S4 Microbiological Reference Methods, What's New, What's Coming?

Fogelstrom

Organizer: David Tomás Fornés

Convenor: Bertrand Lombard

Sponsored by Merck

14.00 New Reference Standard Methods Developed, Validated and Standardized by CEN and ISO
ALEXANDRE LECLERCQ, Institut Pasteur, Paris, France

14.30 Culture Media and Reagents Development – Alignment with the New Standards
BARBARA GERTEN, Merck KGaA, Darmstadt, Germany

15.00 Implementation of Reference Methods in Food Industry – Impact on Alternative Methods
DAVID TOMÁS FORNÉS, Nestlé, Lausanne 26, Switzerland

15.30 Networking Coffee Break in the Exhibit Area

S5 Maximising Food Safety through Good Hygienic Design

Galleriet

Organizer and Convenor: Deb Smith

14.00 Hygienic Design – A Regulatory, Standards and Guidance Perspective
PATRICK WOUTERS, EHEDG & Cargill, Amsterdam, The Netherlands

14.30 Hygienic Design – A Food Manufacturer's Perspective
LAURENCE BLAYO, Nestlé, Lausanne, Switzerland

15.00 Hygienic Design – An Equipment Manufacturer's Perspective
DEB SMITH, UK:IE EHEDG & Vikan, Swindon, United Kingdom

15.30 Networking Coffee Break in the Exhibit Area

T2 Technical Session 2 – Intervention Strategies and Modeling

Arkaden

Convenor: Vasileios Valdramidis

T2-01 Quantifying the Heat Resistance of *Alicyclobacillus acidoterrestris* at Different pH of the Heating and Recovery Media
14.00

Ivan Leguerinel, Melina Maucotel, Noémie Desriac, Maria Gaspari, Christina Chatzitzika, Thibault Arnoux, VASILEIOS VALDRAMIDIS, University of Malta, Msida, Malta

T2-02 Growth/No Growth Boundaries of Heat-resistant Moulds as a Function of Temperature and °Brix
14.15
JULIANA LANE PAIXÃO DOS SANTOS, Simbarashe Samapundo, Enrique Algarra Alexandre, Anderson de Souza Sant'Ana, Jan Van Impe, Frank Devlieghere, Ghent University (UGent), Faculty of Bioscience Engineering, Department of Food Technology, Safety and Health, Research Unit Food Microbiology and Food Preservation (FMFP-UGent), Ghent, Belgium

T2-03 *Thermoanaerobacterium* Species are Emergent Spore-forming Bacteria Involved in Spoilage of Canned Food
14.30
Stella Planchon, STÉPHANE ANDRÉ, CTCPA, Avignon, France

T2-04 Quantifying the Food Safety Risk of Federally Registered Dairy Establishments in Canada Using the Canadian Food Inspection Agency's Establishment-based Risk Assessment Model (2016-2017)
14.45
Sylvain Quessy, MANON RACICOT, Alexandre Leroux, Raphael Plante, Sunny Ng, Romina Zanabria, Hargun Chandhok, Genevieve Comeau, Suzanne Savoie, Anna Mackay, University of Montreal, Saint-Hyacinthe, QC, Canada

T2-05 The Transfer Rate of *Salmonella Typhimurium* from Contaminated Parsley to Other Consecutively Chopped Batches Via Cutting Boards under Different Food-handling Scenarios
15.00
DIMA FAOUR-KLINGBEIL, DFK for Safe Food Environment, Hannover, Germany, Victor Kuri, Ewen Todd

T2-06 *Listeria monocytogenes* is Prevalent in Retail Grocery Produce Environments and is Influenced by Infrastructure, Sanitation, and Management Practices
15.15
HALEY OLIVER, Tongyu Wu, John Burnett, Susan Hammons, Deklin Veenhuizen, Jingjin Wang, Manpreet Singh, Purdue University, West Lafayette, IN, USA

15.30 Networking Coffee Break in the Exhibit Area

S6 How NGS Technologies Unravel Our Understanding of Food and Food Microbiology

Riddarsalen

Organizers: Noémie Desriac, Florence Postollec and Monique Zagorec

Convenors: Jerome Combrisson and Monique Zagorec

Sponsored by UMT ACTIA 14.01 SPORE RISK and IAFP Foundation

16.00 The Use of NGS to Ensure Food Authenticity and Avoid Food Fraud

VALENTINA PARACCHINI, European Commission Joint Research Centre, Ispra, Italy

16.30 The Use of NGS to Apprehend the Biodiversity of Microbial Communities in Food and Food-related Surfaces

BIRGITTE MOEN, Nofima, Oslo, Norway

17.00 Use of NGS to Investigate the Biodiversity and Activity of Cheese Microbial Communities

CHRISTOPHE MONNET, INRA, Thiverval-Grignon, France

17.30 Exhibit Hall Reception

S7 Importance of Microbiological Criteria and Statistical Underpinning of Sampling and Testing for Food Safety Assurance

Fogelstrom

Organizer and Convenor: Leon Gorris

16.00 International Perspective on the Role of Microbiological Sampling and Testing in Food Safety Assurance

LEON GORRIS, Unilever R&D Vlaardingen, Vlaardingen, The Netherlands

16.30 The Science and Statistics Underlying Sound Microbiological Sampling and Testing Approaches

MARCEL ZWIETERING, Wageningen University, Wageningen, The Netherlands

17.00 ICMSF Guidance on Microbiological Sampling and Testing for Key Commodities

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

17.30 Exhibit Hall Reception

S8 Biofilms and Environmental Monitoring

Galleriet

Organizer: Richard Brouillette

Convenor: Andres Rodriguez Lozano

16.00 Biofilms in the Food Industry

ANNETTE FAGERLUND, Nofima, Norwegian Institute of Food, Fisheries and Aquaculture Research, Ås, Norway

16.30 Biofilms, Pathogenic Bacteria and Environmental Monitoring – An Industry Perspective

PETER MCCLURE, Mondelez International, Birmingham, United Kingdom

17.00 Importance of Sanitation Programs and Hygienic Design to Control Biofilms

ROGER SCHEFFLER, Commercial Food Sanitation, Amsterdam, The Netherlands

17.30 Exhibit Hall Reception

T3 Technical Session 3 – Pathogens and Antimicrobials

Arkaden

Convenor: Panagiotis Skandamis

T3-01 Lessons from Growing *Listeria monocytogenes* in High Oxygen Environment – Detaching Exponential Phase from Anaerobicity

MARCIA BOURA, Carolina Bruschi, Kimon Andreas Karatzas, University of Reading, Reading, United Kingdom

T3-02 Characterization of the Virulence Potential of Environmental Shiga Toxin-producing *Escherichia coli* Reveals a Novel Plasmid Encoded Biomarker

BEATRIZ QUIÑONES, Bertram Lee, William Zaragoza, Jaszemyn Yambao, Clifton Fagerquist, U.S. Department of Agriculture-ARS-WRRC-PSM Unit, Albany, CA, USA

T3-03 Comparison of Regional Residue Assessments Evaluating the Safety of Antimicrobials and Cleaners in Food Contact Applications

Ludger Grunwald, Kathryn Sande, Eric Ditzel, CHRISTIAN BRIEDEN, Ecolab Deutschland GmbH, Monheimam Rhein, Germany

T3-04 The Absence of *N*-Acetylglucosamine in Wall Teichoic Acids of *Listeria monocytogenes* Modifies Biofilm Architecture and Tolerance to Cleaning and Disinfection Procedures

THOMAS BRAUGE, Christine Faille, Irina Sadovskaya, Alain Charbit, Thierry Benezech, Yang Shen, Martin Loessner, Jean Romain Bautista, Graziella Midelet-Bourdin, ANSES, Laboratory for Food Safety, Boulogne sur Mer, France

T3-05 Chitosan Coating as an Alternative Seed Disinfection Treatment for Alfalfa and Leek Seeds Intended for Sprouting

INGE VAN DER LINDEN, Marc Heyndrickx, Frank Devlieghere, Mieke Uyttendaele, Ghent University (UGent), Faculty of Bioscience Engineering, Department of Food Technology, Safety and Health, Research Unit Food Microbiology and Food Preservation (FMFP-UGent), Ghent, Belgium

T3-06 Development and Validation of Growth Models for *Listeria monocytogenes* in Mediterranean Fish Species from Aquaculture Production

ARACELI BOLÍVAR, Guiomar Denisse Posada-Izquierdo, J.C.C.P. Costa, Antonio Valero, Gonzalo Zurera, Fernando Pérez-Rodríguez, Department of Food Science and Technology, University of Cordoba, Cordoba, Spain

17.30 Exhibit Hall Reception

7.30 – 17.00 Registration Open

7.30 – 8.30 Morning Coffee

Exhibit Hours 10.00 – 16.00

Poster Session 2 – Antimicrobials, Applied Laboratory Methods, Dairy and Other Food Commodities, Epidemiology, Food Toxicology, Novel Laboratory Methods, Pathogens, Produce

Authors present during scheduled break times

S9 New Approaches for Safety and Quality of Fermented Foods

Riddarsalen

Organizers and Convenors: Luca Cocolin and Kalliopi Rantsiou

8.30 Application of Metataxonomics and Metagenomics in Fermented Sausages
LUCA COCOLIN, University of Torino-DISAFA, Grugliasco, Italy

9.00 Innovative Approaches to Reduce Fungal Spoilage in Dairy Foods
JEROME MOUNIER, University of Brest, Brest, France

9.30 Bioprotection in Vegetable Foods
ANTONIO GALVEZ, University of Jaen, Jaen, Spain

10.00 Networking Coffee Break in the Exhibit Area

RT2 Prediction of Spoilage and Safety with Models: How to be on the Safe Side in a World Full of Variability

Fogelstrom

Organizer and Convenor: Olav Sliekers

8.30 Panelists:
JEAN-CHRISTOPHE AUGUSTIN, National Veterinary School of Alfort, Maison-Alfort, France
KARIN BEEKMANN-METSELAAR, Corbion, Gorinchem, The Netherlands
MARIEM ELLOUZE, Nestlé, Lausanne, Switzerland
ANNEMARIE PIELAAT, Unilever R&D, Vlaardingen, The Netherlands

10.00 Networking Coffee Break in the Exhibit Area

S10 Validation and Verification – Successes, Pitfalls and Disasters

Galleriet

Organizers: Alvin Lee and Purnendu Vasavada

Convenors: Roy Betts and Alvin Lee

Sponsored by the IAFP Foundation

8.30 Validation and Verification – Concept and Practice
PURNENDU VASAVADA, University of Wisconsin-River Falls, River Falls, WI, USA

9.00 Non-Thermal and Thermal Process Validation
ALVIN LEE, Institute for Food Safety and Health, Illinois Institute of Technology, Bedford Park, IL, USA

9.30 Microbiological Test Methods: Validation and Verification, What Does It Mean?
ROY BETTS, Campden BRI, Gloucestershire, United Kingdom

10.00 Networking Coffee Break in the Exhibit Area

T4 Technical Session 4 – Risk Assessment

Arkaden

Convenor: Anett Winkler

T4-01 Integrating WGS Data into Quantitative Microbial Risk Assessment: Refinement of the *Listeria monocytogenes* in Cold Smoked Salmon Model
8.30 LENA FRITSCH, Laurent Guillier, Jean-Christophe Augustin, Anses, Maisons-Alfort, France

T4-02 The Canadian Food Inspection Agency Establishment-based Risk Assessment Model for Hatcheries: Principles and Algorithm
8.45 MANON RACICOT, Alexandre Leroux, Sunny Ng, Genevieve Comeau, Sylvain Quessy, Marie-Lou Gaucher, Canadian Food Inspection Agency, St-Hyacinthe, QC, Canada

T4-03 Development of a Software Tool for Risk Assessment of *Listeria monocytogenes* in Selected Ready-to-Eat Food Categories in the European Union
9.00 Fernando Pérez-Rodríguez, Elena Carrasco, ARACELI BOLÍVAR, Sara Bover, Anna Jofré, Antonio Valero, Department of Food Science and Technology, University of Cordoba, Cordoba, Spain

T4-04 Application Potentials of Network Science to Food Chain Safety Risk Analysis
9.15 TEKLA ENGELHARDT, Mátyás Milkovics, Zoltán Lakner, Akos Jozwiak, National Food Chain Safety, Budapest, Hungary

T4-05 Exposure Assessment of Process-related Contaminants in Food by Biomarker Monitoring
9.30 Pierre Dussort, Ivonne Rietjens, Helmut Günther, Paul Hanlon, Hiroshi Honda, Angela Mally, Sue O'Hagan, GABRIELE SCHOLZ, Albrecht Seidel, Justin Teeguarden, James Swenberg, Gerhard Eisenbrand, Nestlé Research Center, Lausanne, Switzerland

10.00 Networking Coffee Break in the Exhibit Area

S11 Biological Variability in Thermal Processing: Impact for Process Control and Validation – What You Need to Know about Microbiological Variability for Food Quality and Safety Control

Riddarsalen

Organizer: Marcel Zwietering

Convenor: Leon Gorris

10.30 Impact of Natural Diversity in Heat Resistance of Bacteria and Bacterial Spores on Food Safety and Quality
MARCEL ZWIETERING, Wageningen University, Wageningen, The Netherlands

11.00 Combining Challenge Tests and Predictive Microbiology in Thermal Process Validations of Low-moisture Food
MARIEM ELLOUZE, Nestlé, Lausanne, Switzerland

11.30 Impact of Variability in Regulation and Inspection
MICKEY PARISH, U.S. Food and Drug Administration, Washington, D.C., USA

12.00 Lunch Available in the Exhibit Area

S12 Integrating Microbial Adaptive Trait in Food Safety: Added Value of Biomarkers

Fogelstrom

Organizers: Noémie Desriac and Sandrine Guillou

Convenors: Noémie Desriac and Ronald Lebofsky

Sponsored by UMT ACTIA 14.01 SPORE RISK and IAFP Foundation

10.30 RNA Used as Biomarkers: Requirements, Validation and Challenges in Clinical Applications
EDDY VAN COLLENBURG, Bio-Rad Laboratories, Eindhoven, The Netherlands

11.00 Use of Biomarkers to Refine Microbiological Exposure Assessment Associated with *Listeria monocytogenes*, *Bacillus cereus* and *Campylobacter jejuni* in the Dairy and Poultry Food Chains
BENJAMIN DUQUÉ, UMR1014 SECALIM, INRA, Oniris, Nantes, France

11.30 Reliable Identification of mRNA as Biomarkers: Workflow of the Quality Assurance Procedure
LAMIA BELKADI, LUBEM UBO University – UMT14.01 SPORE RISK, Quimper, France

12.00 Lunch Available in the Exhibit Area

S13 Allergen Control – From Problem to Solution

Galleriet

Organizer and Convenor: Deb Smith

Sponsored by Vikan and Holchem and IAFP Foundation

10.30 The Problem – Allergens as a Food Safety Hazard
LYNNE REGENT, Anaphylaxis Campaign, Farnborough, United Kingdom

11.00 The Solution I – Effective Strategies for Minimising Allergen Cross-contamination
DEB SMITH, UK:IE EHEDG & Vikan, Swindon, United Kingdom

11.30 The Solution II – Allergen Removal Validation, Monitoring and Verification
JOHN HOLAHA, UK:IE EHEDG & Holchem Laboratories Ltd., Bury, United Kingdom

12.00 Lunch Available in the Exhibit Area

T5 Technical Session 5 – Molecular Characterization and Risk Assessment

Arkaden

Convenor: Daniel Sohler

T5-01 16s Metagenomic Sequencing of a Facility to Determine Point Source Contamination of Products
10.30
EDAN HOSKING, Andrew Benson, Rohita Sinha, Joe Heinzlmann, Susanne Hinkley, Jaehyoung Kim, Emily Rose, Barry Simpson, Will Sawyer, Robert Donofrio, Neogen Corporation, Lansing, MI, USA

T5-02 Whole Genome Sequencing for Source Tracking: A Validation Approach of the End-to-End Workflow for *Listeria monocytogenes* and *Salmonella enterica*
10.45
Ann-Catherine Portmann, Coralie Fournier, Johan Gimonet, Catherine Ngom-Bru, Caroline Barretto, LEEN BAERT, Nestec Ltd. Nestlé Research Center, Lausanne, Switzerland

T5-03 Development of Abiotic Bacterial Surrogates for Various Sanitation Processes
11.00
LAURIE CLOTILDE, Antonios Zografos, Stilianos Arhondakis, Sharon Horgan, Carol Lauzon, SafeTraces, Pleasanton, CA, USA

T5-04 Surveillance of *Salmonella* Prevalence in Animal Food and Characterization of the *Salmonella* Isolates by Serotyping
11.15
XIN LI, Linda Benjamin, David Edwards, Daniel McChesney, Food and Drug Administration, Rockville, MD, USA

T5-05 The Effect of Phage Presence on PFGE and WGS in *Listeria monocytogenes*: Who is Right?
11.30
KATLEEN VRANCKX, Sanne Kiekens, Bernadette Hickey, Eoin O'Brien, Philip Curran, Brian Byrne, Applied Maths NV, Sint-Martens-Latem, Belgium

T5-06 A Bi-Phasic Model to Predict Heat Inactivation of *Salmonella* in Low-moisture Foods
11.45
Annemarie Pielaat, CHRISTINE VD SWALUW, Erik de Vries, Joerg Ueckert, Unilever R&D, Vlaardingen, The Netherlands

12.00 Lunch Available in the Exhibit Area

S14 Risk Benefit Assessment of Food: Past, Present and Future Trends

Riddarsalen

Organizers: Jeanne-Marie Membre and Sara Monteiro Pires

Convenor: Jeanne-Marie Membre

Primary Sponsor: IAFP Foundation

- 13.30** Introduction to Risk-benefit Assessment of Food
GÉRALDINE BOUÉ, UMR1014 SECALIM, INRA, Oniris, Nantes, France
- 14.00** Current Practices of Risk-benefit Assessment in a National Food Agency
HANNA ENEROTH, Livsmedelsverket, Uppsala, Sweden
- 14.30** Future Trend and Research Directions in Risk-benefit Assessment
SARA MONTEIRO PIRES, Technical University of Denmark, Lyngby, Denmark

15.00 Networking Coffee Break in the Exhibit Area

S15 Interventions to Reduce Antibiotic Resistance and Antibiotic Use in Animal Production

Fogelstrom

Organizers: Wael Abdelrahman, Lionel LeVen and Chris van Anne

Convenor: Lionel Le Ven

- 13.30** Antibiotic Resistance Transmission Pathways in the Food Chain
KATHARINA D.C. STÄRK, SAFOSO AG, CH-3097 Bern-Liebefeld, Switzerland
- 14.00** Review of Existing In-feed Solutions to Reduce Antibiotic Dependence on Farm
STEVEN RICKE, University of Arkansas, Fayetteville, AR, USA
- 14.30** Strategies for Controlling the Risk of Foodborne Pathogens and Antibiotic Resistance
J. ALLEN BYRD, Diamond V, Cedar Rapids, IA, USA

15.00 Networking Coffee Break in the Exhibit Area

RT3 Assessment of Microbial Risk for Fresh Produce

Galleriet

Organizer and Convenor: Tobis Recker

Sponsored by ILSI Europe

- 13.30** Panelists:
JOHN BASSETT, John Bassett Consulting Ltd., Bedford, United Kingdom
ROY BETTS, Campden BRI, Gloucestershire, United Kingdom
TIMOTHY JACKSON, Driscoll's, Watsonville, CA, USA
JIM MONAGHAN, Harper Adams University, Newport, Shropshire, United Kingdom

15.00 Networking Coffee Break in the Exhibit Area

T6 Technical Session 6 – Intervention Strategies and Management

Arkaden

Convenor: Annemarie Pielaat

- T6-01** Innovative Strategy to Improve the Food Safety Standards in the Emirates of Dubai – Happiness Inspection TEAM
13.30 SULTAN ALI AL TAHER, Dubai Municipality, Dubai, United Arab Emirates
- T6-02** FSMA Regulatory Audit: A European Experience, in Italian Manufacturers
13.45 CLAUDIO GALLOTTINI, Ferruccio Marelllo, Andrea Gentili, Franco Rapetti, Giovanni La Rosa, ITA Corporation, Miami, FL, USA
- T6-03** Development of Food Safety Interventions Using a Patient-centered Approach to Reduce the Risk of Foodborne Illness among Patients Receiving Chemotherapy Treatment
14.00 ELLEN W. EVANS, ZERO2FIVE Food Industry Centre, Cardiff Metropolitan University, Cardiff, United Kingdom
- T6-04** The (FAO/WHO) International Food Safety Authorities Network (INFOSAN): How Responsive Were Members during Food Safety Emergencies between 2010 and 2016?
14.15 CARMEN SAVELLI, Frances Widjaja, Adam Bradshaw, Peter Ben Embarek, World Health Organization, Geneva, Switzerland
- T6-05** One-hour NMR-based Metabolomics Approach to Study the Toxic Effects of Phoxim on Crucian Carp (*Carassius auratus gibelio*)
14.30 Hong Li, XIAOYU LIU, Si Li, Huazhong Agricultural University, Wuhan, China
- T6-06** Effects of Food Safety Training on Achieving Food Safety Knowledge and Practices in Restaurants in the Emirates of Dubai
14.45 ABDUL AZEEZ MULLATTU EBRAHIM, M R S International Food Consultants, Dubai, United Arab Emirates

15.00 Networking Coffee Break in the Exhibit Area

S16 The Rise of Whole Genome Sequencing: How Do We Share and Interpret the Data Globally?

Riddarsalen

Organizers: Maria Hoffmann, Jesse Miller and Eric Stevens

Convenor: Jesse Miller

15.30 How to Get Governments to Discuss the Benefits of Sharing Microbial WGS Data across Borders

JØERGEN SCHLUNDT, Nanyang Technological University, Singapore, Singapore

16.00 Different Implementation Options for Countries Looking to Utilize WGS

ERIC STEVENS, U.S. Food and Drug Administration–CFRAN-ORS-DM, College Park, MD, USA

16.30 Developing the Standards for Generating and Utilizing WGS Data by ISO Working Group 25

ARTHUR PIGHTLING, U.S. Food and Drug Administration, College Park, MD, USA

S17 Global Occurrence of Mobile Colistin Resistance in Foodborne Pathogens

Fogelstrom

Organizer: Xiaohua He

Convenors: Xiaohua He and Rick Meinersmann

15.30 Screening for MCR-carrying STEC Recovered from a Major Produce-production Region in California

XIAOHUA HE, USDA, ARS, WRRRC, Albany, CA, USA

16.00 Whole-plasmid Multilocus Sequence Typing for Incl2

RICK MEINERSMANN, USDA, ARS, Russell Research Center, Athens, GA, USA

16.30 Colistin Resistance and Link to Animals

JEAN-MARC ROLAIN, Aix-Marseille Université, Marseille, France

S18 Control of Human Pathogens in Plant Production Systems

Galleriet

Organizers: Gro Johannessen and Mieke Uyttendaele

Convenor: Mieke Uyttendaele

Sponsored by HUPLANTcontrol (COST Action 16110)

15.30 Occurrence of Human Pathogens in Plant Production Systems

LEO VAN OVERBEEK, WUR Plant Research, Wageningen, The Netherlands

16.00 How Microbiome Studies Can Help Us to Define Preharvest Measures for Increased Biosafety of Field Grown Crops

BEATRIX ALSANIUS, SLU (Swedish University of Agricultural Sciences), Alnarp, Sweden

16.30 To Sanitize or Not to Sanitize as Control Measure for Ensuring Safety of Seeds and Sprouted Seeds
INGE VAN DER LINDEN, Ghent University (UGent), Faculty of Bioscience Engineering, Department of Food Technology, Safety and Health, Research Unit Food Microbiology and Food Preservation (FMFP-UGent), Ghent, Belgium

T7 Technical Session 7 – Meat and Poultry, Seafood, Epidemiology

Arkaden

Convenor: George-John E. Nychas

T7-01 Does Sodium Reduction Compromise the Food Safety of Cooked Ham?

15.30 Cristina Serra-Castelló, Anna Jofré, Margarita Garriga, SARA BOVER-CID, IRTA. Food Safety Programme, Monells, Spain

T7-02 Inter- and Intra-generic Interaction between Meat Plant1 Environmental Bacteria and *Escherichia coli* O157:H7 in Co-Culture Biofilms

15.45 Jeyachandran Visvalingam, XIANQIN YANG, Agriculture and Agri-Food Canada, Lacombe, AB, Canada

T7-03 Two Studies Show Association between Consumption of Dry Raw Pork Sausages and Hepatitis E in The Netherlands

16.00 Anna D. Tulen, Harry Vennema, Sofie H. Mooij, Boris M. Hogema, Wilfrid van Pelt, Eelco Franz, Hans L. Zaaijer, Michel Molier, AGNETHA HOFHUIS, Centre for Infectious Disease Control, National Institute for Public Health and the Environment (RIVM), Bilthoven, The Netherlands

T7-04 Don't Wash My Chicken!? Identifying Barriers to Consumers Adopting the Practice of Not Washing Raw Poultry

16.15 Alaa Aljarboua, Gabriella Balla, Liyi Chang, JENNIFER QUINLAN, Drexel University, Philadelphia, PA, USA

T7-05 Deriving Personalized Recommendations for Fish Intake Using Mathematical Optimization Methods
MARIA PERSSON, Sisse Fagt, Sara Pires, Morten Poulsen, Maarten Nauta, National Food Institute, Kgs. Lyngby, Denmark

T7-06 Attributing STEC Infections to Food Sources to Inform International Food Standards: Results of an International Consultation and Evidence Synthesis

16.45 SARA M. PIRES, Brecht Devleeschauwer, Shannon Majowicz, Division of Diet, Disease Prevention and Toxicology, National Food Institute, Technical University of Denmark, Kgs. Lyngby, Denmark

7.30 – 12.00 Registration Open

7.30 – 8.30 Morning Coffee

S19 Turning Sequencing and Mass Spectrometry into Routine Testing Tools for Microbial Strain Characterization

Riddarsalen

Organizer: Patrice Arbault and Daniele Sohier

Convenor: Anne Brisabois

8.30 Regulation Recognition of Omics Technologies: The Pathway for Implementation in Routine Testing
PAUL IN'T VELD, VWA, Utrecht, The Netherlands

9.00 Harmonization of Omics-based Routine Methods and Database: Make It Easy and Fully under Control
JESSICA PRYOR, G2S at Centers for Disease Control and Prevention (CDC), Atlanta, GA, USA

9.30 Never Forget the Needs and the True Goal in Routine Testing
ERIN CROWLEY, Q Laboratories, Inc., Cincinnati, OH, USA

10.00 Networking Coffee Break in Marstarget

S20 Integrating Scientific Risk Assessment in the Prioritization and Management of Chemical Contaminants in Foods and Raw Materials

Fogelstrom

Organizer and Convenor: Gabriele Scholz

8.30 Application Example of a Global Scientific Tool for the Assessment and Prioritization of Chemical Hazards in Food Raw Materials
GABRIELE SCHOLZ, Thomas Stroheker, Paolo Mazzatorta, Nestlé Research Center, Lausanne, Switzerland

9.00 A Methodology for Risk Evaluation of Chemical Contamination in Food
TILEMACHOS GOUMPERIS, European Food Safety Authority, Parma, Italy

9.30 An Industry Approach to Integrate Scientific Risk Assessment to Prioritize Chemical Contaminants
PAUL HANLON, Abbott Nutrition, Columbus, OH, USA

10.00 Networking Coffee Break in Marstarget

S21 Natural Antimicrobial Preservatives in Foods: Where are We in Terms of Application and Commercialization?

Galleriet

Organizers: Armitra Jackson-Davis and Aubrey Mendonca

Convenor: Armitra Jackson-Davis

8.30 Overview of Natural Antibacterial Compounds with Potential for Use in Foods
ARMITRA JACKSON-DAVIS, Alabama A&M University, Madison, AL, USA

9.00 Food Safety Applications of Natural Antimicrobials Via Hurdle Technology: An Industry Perspective for Commercialization
MORTEN HYLDGAARD, DuPont Nutrition & Health, Braband, Denmark

9.30 Applications of Natural Antimicrobials in Foods: Strategies for Overcoming Current Challenges
BYRON BREHM-STECHER and Aubrey Mendonca, Iowa State University, Ames, IA, USA

10.00 Networking Coffee Break in Marstarget

T8 Technical Session 8 – Detection and Typing Methods

Arkaden

Convenor: Luca Cocolin

T8-01 8.30 Genome Diversity of *Salmonella enterica* Sub-species *enterica* Serotype Derby from Animal to Human: Sporadic Cases Source Attribution Study and Specific Host-association Identification
YANN SÉVELLEC, Simon Le Hello, Laurent Guillier, Laetitia Fabre, Sophie A. Granier, Carole Feurer, Renaud Lailier, Michel-Yves Mistou, Sabrina Cadet-Six, Anses, Maisons-Alfort, France

T8-02 8.45 Rapid *Salmonella* Serotyping Via Targeted Amplicon Sequencing
EDAN HOSKING, Rohita Sinha, Andrew Benson, Barry Simpson, Jaehyoung Kim, Jean Guard, Eric Tovar, Mark Mozola, Jennifer Rice, Robert Donofrio, Neogen Corporation, Lansing, MI, USA

T8-03 9.00 Comparison of Typing Reference Methods and Whole Genome Sequencing (WGS) Analyses for the Characterization of *S. aureus* Strains Isolated from Food Outbreaks
NOÉMIE VINGADASSALON, Déborah Merda, Alexandra Cauquil, Benjamin Félix, Thomas Méheut, Frédéric Auvray, Jacques-Antoine Hennekinne, Anses, Maisons-Alfort, France

T8-04 9.15 Bacteriophage Receptor Binding Proteins for the Isolation of *Yersinia enterocolitica* in Foods
CARLOS LEON-VELARDE, Shu Chen, Roger Johnson, Joseph Odumeru, AFL, University of Guelph, Guelph, ON, Canada

T8-05 9.30 Monitoring of Foodborne Viruses in Berries and Considerations on the Use of RT-PCR Methods in Surveillance
DAN LI, Sophie Butot, Sophie Zuber, Mieke Uyttendaele, Ghent University, Ghent, Belgium

T8-06 9.45 Method Validation for Staphylococcal Enterotoxins Detection in Food Matrices from Various Food Poisoning Outbreaks
YACINE NIA, Isabelle Mutel, Berivan Boran, Joan Ojenima, Jacques-Antoine Hennekinne, Université Paris-Est, ANSES, Maisons-Alfort, France

10.00 Networking Coffee Break in Marstarget

CS Closing Session

Riddarsalen

Convenor: Daniele Sohier

- 10.30** Challenges in Swedish Food Protection Control
ARJA HELENA KAUTTO, National Food Safety Agency, Uppsala, Sweden
- 11.00** Lowering the Use of Antibiotics in Swedish Farms
MY SAHLMAN, The Federation of Swedish Farmers, Stockholm, Sweden

- 11.30** Obstacles of Ready-to-Eat Vegetables and Fruits: The Swedish Approach
BEATRIX ALSANIUS, SLU (Swedish University of Agricultural Sciences), Alnarp, Sweden
- 12.00** Food Safety Management in a Changing World
TIMOTHY JACKSON, Driscoll's, Watsonville, CA, USA
- 12.30** Awards Presentation and Concluding Remarks
MICKEY PARISH, U.S. Food and Drug Administration, Washington, D.C., USA

12.45 – 14.00 Farewell Refreshments

Blue – 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*. Remember, when you support the IAFP Foundation everyone benefits, including you.



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INVITED SPEAKER BIOGRAPHIES

25-27 April 2018 – Stockholm, Sweden

INVITED SPEAKER BIOGRAPHIES



Beatrix Alsanius

SLU (Swedish University of Agricultural Sciences), Sweden

Beatrix Alsanius has a Ph.D. from Bonn University (1991), Germany. During 1992–1998, she was assistant professor and was habilitated in horticulture in 1999 and in plant protection ecology in 2006 at the Swedish University of Agricultural Sciences. In 2007, she was appointed professor in horticulture with on horticultural production systems and 2009 full professor at the SLU:Department of Horticulture, Alnarp, Sweden. During 2011–2014, she was adjunct professor in phytology at Université Laval, Québec, Canada. Dr. Alsanius leads the Microbial Horticulture unit. Her research program mainly emphasizes interactions between

microorganisms and the horticultural value network. Since 2013, she leads the multidisciplinary research program “Safe Salad,” funded by the Swedish research council Formas. Her approach includes laboratory studies and studies under controlled experimental conditions as well as trials at commercial facilities.



Wayne Anderson

Food Safety Authority of Ireland, Ireland

Dr. Wayne Anderson, Director of Food Science and Standards, Food Safety Authority of Ireland 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.



Jean-Christophe Augustin

National Veterinary School of Alfort, France

Prof. Jean-Christophe Augustin teaches food safety at the Veterinary School of Alfort and carries out his research activities in the Laboratory for Food Safety at Anses Maisons-Alfort. He has been working for 20 years in the field of quantitative microbiology including predictive microbiology, quantitative risk assessment, performance of analytical methods, and surveillance of food safety and quality. He currently chairs the scientific committee of the Sym'Previs consortium.



Leen Baert

Nestlé, Switzerland

Leen Baert obtained her Ph.D. in Applied Biological Sciences in 2009 at Ghent University, Belgium on the topic foodborne viruses. She continued as post-doctoral fellowship at Ghent University, Laboratory of Food Microbiology and Food Preservation until 2011. In 2011, she started to work at the Nestlé Research Center in Lausanne, Switzerland in the Microbial & Molecular Analytics group to develop and evaluate molecular detection methods for microbial targets. The last years she specialized on whole genome sequencing used for preventative food safety microbiology, establishing and validating a WGS methodology for source tracking

of *Salmonella*, *L. monocytogenes* and *Cronobacter*.



John Bassett

John Bassett Consulting Ltd., United Kingdom

John Bassett has over 20 years' experience of risk assessment and risk management, in both industry and government roles. A veterinarian by training, he brings a "farm to fork" perspective on food safety challenges. His consultancy, John Bassett Consulting Ltd. works with clients in the commercial food industry as well as governments and inter-governmental agencies.

John is particularly passionate about the practical use of 'risk-based' approaches in innovation and food safety management systems for industry, and in the application of optimal regulatory models – he is currently a member of FSA's expert advisory group supporting their 'Regulating our Future' activity.



Karin Beekmann-Metselaar

Corbion, The Netherlands

Karin Beekmann-Metselaar studied Food Safety at Wageningen University, The Netherlands. She obtained a Ph.D. in Food Microbiology from the same university. During her Ph.D. research, Karin isolated, quantified, and studied the ecological behavior of stress-resistant subpopulations of *Listeria monocytogenes* which are revealed upon stress exposure. Karin is currently working as Scientist of Microbiology & Modeling at Corbion in Gorinchem, the Netherlands. In her current position, she develops and validates predictive growth models for pathogens and spoilers that are used both internally and by customers within food industries.



Lamia Belkadi

LUBEM UBO University – UMT14.01SPORE RISK, France

Lamia Belkadi is a second year Ph.D. student in Food Microbiology at the University of Western Brittany in France. She is working at the UMT SPORE-RISK, collaboration between LUBEM University lab and Adria Food Technology Institute. Her research project aims at identifying molecular biomarkers to predict the growth and resistance potential of two microbial pathogens: *Bacillus cereus* and *Listeria monocytogenes*. Otherwise, Lamia has held a master's degree in Fundamental and Applied Microbiology from the University of Brest. Upon graduation, she strengthened her academic background by a second MSc in Microbiology at the University of Caen before starting her doctoral research in 2016.



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. Roy 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.



Laurence Blayo

Nestlé, Switzerland

Laurence Blayo started her career in 1994 as factory quality manager in the meat industry in France. In 1996, she joined the Nestlé group in Sweden as a microbiologist in their research and development center dedicated to frozen and chilled food. In 2000, she moved to Switzerland as a hygienist for 3 years, during which she was exposed to the challenges of hygienic engineering during trouble shooting and training in various Nestlé co-manufacturer's and supplier's factories all over the world. In 2003, she returned to France as quality manager for the chilled culinary division. In 2006, she became food safety manager for Nestlé France

until 2009, when she returned back to Switzerland to occupy her present position. She is currently leading the Food Safety Microbiology team at the International Nestlé Research Center based in Lausanne, Switzerland. Her team is dedicated to emerging microbial issues and microbiological risk assessment. She is leading as well the Nestlé R&D Microbiological Safety Expert Network aiming at developing the critical mass of food safety microbiologists

and leveraging the overall microbiology competences throughout the company. Laurence has built over the years considerable expertise in food safety, hygiene and hygienic engineering. Her passions include not only microbiology and food safety sciences, but also people management and development.



Géraldine Boué
UMR1014 Secalim, INRA, Oniris, France

Géraldine Boué is a Lecturer-Researcher in Food Safety at Secalim in Nantes, a research unit of INRA (French National Institute for Agricultural Research) and Oniris (Nantes Atlantic College of Veterinary Medicine, Food Science and Engineering). She has a Ph.D. in risk-benefit assessment in foods and a master's degree in food science and engineering. She is currently working on methodological development of risk-benefit and risk assessment of several applications.



Byron Brehm-Stecher
Iowa State University, USA

Byron Brehm-Stecher received his Ph.D. from the University of Wisconsin-Madison in 2002, where he studied rapid methods for the detection of foodborne pathogens. Dr. Brehm-Stecher continued his work in Madison as a postdoctoral research scholar in the laboratory of Professor Eric A. Johnson before taking a job in the biotechnology industry. He came to Iowa State University in 2004 from a position as a senior scientist and molecular biologist with Applied Biosystems, Inc., Bedford, MA. He now heads the Brehm-Stecher Rapid Microbial Detection and Control Laboratory in the Department of Food Science and Human Nutrition at Iowa State University. Dr. Brehm-Stecher's research is focused on two major areas: development of new approaches for rapid detection of foodborne and clinical pathogens and development of new antimicrobial treatments for control of pathogens and spoilage organisms. Naturally-sourced, value-added antimicrobials and hurdle-based strategies for enhancing antimicrobial efficacy are of particular interest in Dr. Brehm-Stecher's work.



J. Allen Byrd
Diamond V, USA

Dr. Allen Byrd is a native of Texas, receiving his B.S. in Animal Science (1984), M.S. in Nutrition (1987), Ph.D. in Poultry Science (1994) and D.V.M. (1996) from Texas A&M University. In January 1996, he joined the USDA-ARS Food and Feed Safety Research Unit in College Station as a Post-doctorate Animal Scientist. In 1997, Dr. Byrd accepted a position as a Research Microbiologist in the same unit and was the Project leader for pre-harvest food safety research in College Station, TX. In May 2017, he joined Diamond V. He is also an adjunct faculty member in the Departments of Poultry Science and Veterinary Pathobiology at Texas A&M University. His research interests have focused on investigating cost-efficient pre-harvest Food Safety intervention strategies in poultry. Dr. Byrd is a member of the Poultry Science Association, Southern Poultry Science Society, American Veterinarian Medical Association, American Association of Avian Pathologist, and World's Poultry Science Association. He has received the USDA-ARS, Southern Plains Area Early Career Research Scientist (2003) and was selected by the USDA-ARS Administrator as the Agency nominee for the Dr. Daniel E. Salmon Award for Exemplary Achievement in Federal Veterinary Medicine (2004). In 2006, he received the Frank Perdue Live Poultry Food Safety Award from the Poultry Science Association. He authored or co-authored over 150 manuscripts.



Luca Cocolin
University of Torino-DISAFA, Italy

Luca Cocolin is a full professor of food microbiology at the University of Torino, Italy. He is an executive board member of ICFMH, editor-in-chief of the *International Journal of Food Microbiology* and academic editor of *PLOS One*. Luca is 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. He is an 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.



Erin Crowley
Q Laboratories, Inc., USA

Erin Crowley is the Chief Scientific Officer at Q Laboratories, Inc. in Cincinnati, Ohio. Prior to this, she was the Microbiology Research and Development Supervisor at Q Laboratories, Inc. in Cincinnati, Ohio since 2006. For the past 11 years, Erin and her R&D team have served as an independent third-party laboratory with a primary focus on providing high quality method validation for microbiological rapid detection methods. These validations include independent laboratory evaluations for pathogen detection, qualitative methods and confirmatory biochemical assays for AOAC Official Methods of Analysis, AOAC Research Institute Performance Tested Methods Program, MicroVal and AFNOR NF Certification Programs. In addition to being an active member of the International Association for Food Protection (IAFP) and AOAC, Erin currently serves as Chair of the AOAC Official Methods Board, Chair of the International Stakeholder Panel on Alternative Methods (ISPAM) and a member of the MicroVal Technical Committee (MVTC). Erin earned a B.S. from the University of Cincinnati in Cincinnati, Ohio and an M.A. from Tufts University in Medford, MA.



Benjamin Duqué
UMR1014 Secalim, INRA, Oniris, France

Benjamin Duqué started his studies in the Engineering School of Microbiology and Quality of Brest, France (2013–2016, equivalent of M.Sc degree). During his studies, he conducted his Master's thesis in the Secalim unit from INRA specialised in food safety. Eager to learn more about food safety and to develop expertise in this area, he started a Ph.D. in 2017, still in the Secalim unit. He is working on the prediction of contamination on chicken carcasses using omic data.



Mariem Ellouze
Nestlé, Switzerland

Dr. Mariem Ellouze holds a Ph.D. in predictive microbiology and risk assessment conducted at the Veterinary School of Alfort, France. She is a now Food Safety Microbiology Specialist at Nestlé Research Center, Lausanne, Switzerland. She is involved in the evaluation of new and existing products/processes/concepts throughout different product categories. She provides technical assistance and develops internal tools and guidelines to help R&D and operations units to conduct microbiological risk, exposure or safety assessments. She animates different workshops and seminars, and published several articles on the topic.



Hanna Eneroth
Livsmedelsverket, Sweden

Hanna Eneroth has an MSc in human nutrition and a Ph.D. in medical science, international health from Uppsala University. In her current position as a risk-and benefit assessor at the National Food Agency, Sweden, she evaluates and communicates scientific evidence in nutrition to support decisions on dietary recommendations. One of her research interests is the methodology of assessments combining microbiological and toxicological risks and nutritional benefits of food and food components. She is a member of European and Nordic networks for risk-benefit methodology and has recently published an evaluation of risks and benefits of nut consumption.



Annette Fagerlund
Nofima, Norwegian Institute of Food, Fisheries and Aquaculture Research, Norway

Annette Fagerlund is a research scientist at the Department of Food Safety and Quality at Nofima, and has a Ph.D. in molecular microbiology (2008) from the Norwegian School of Veterinary Science (Norwegian University of Life Sciences). Her main research interests are foodborne pathogens such as *Listeria monocytogenes*, bacterial genomics and biofilm formation. Current research topics include investigation into how bacteria relevant in the food industry survive and adapt in food processing environments, and the use of DNA sequencing techniques to characterize and track the spread of bacteria in the food chain.



Antonio Galvez
University of Jaen, Spain

Antonio Galvez is a full Professor of Microbiology at University of Jaen since 2003. He has co-authored over 180 publications in peer-reviewed journals. His main research topics: Application of bacteriocins and other natural antimicrobials as part of hurdle technology in food preservation. High-pressure processing of vegetable foods. Impact of food preservation methods on the food microbiota. Antimicrobial resistance in the food chain. Academic activities: Supervisor of master and doctoral studies on food safety at University of Jaen.



Barbara Gerten
Merck KGaA, Germany

After her studies in microbiology and biochemistry, Barbara Gerten was employed in different companies responsible for culture media Quality Control and R&D and since 2008 at Merck in different positions. She is Chair of the German DIN NAL Working Group “Microbiology in the Food Chain” and member of the German delegation for ISO/CEN Food Microbiology.



Leon Gorris
Unilever R&D Vlaardingen, The Netherlands

Dr. Leon Gorris is Director for Regulatory Affairs at Unilever, with responsibility for food safety globally. He joined Unilever in The Netherlands in 1997. He was based in the UK from 2001–2010, in Shanghai from 2010–2014. Currently he is based again in The Netherlands. 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.



Tilemachos Goumperis
European Food Safety Authority, Italy

Tilemachos Goumperis is currently a scientific officer in EFSA’s Scientific Committee and Emerging Risks unit; the main task of the Scientific Committee is the preparation of scientific advice in the area of new and harmonised approaches for risk assessment of food and feed. Mr. Goumperis has been working for EFSA for over nine years, the first five of which were spent working in the team putting in place a process for the early identification of emerging risks in the European food and feed chain. He is working on crisis preparedness, insects as food and feed and currently being the scientific project manager of the EFSA working group on the development of a risk evaluation model for chemical contaminants in food. Mr. Goumperis has previously worked in the food industry in the area of food safety and quality assurance. He holds a degree in chemistry and a Master’s in food science and technology.



Kathie Grant
Public Health England, United Kingdom

Dr. Kathie Grant is an internationally recognised expert in the field of foodborne pathogens with 30 years experience in clinical and public health microbiology and a research interest in exploiting whole genome sequencing (WGS) of bacterial pathogens to improve the understanding and control of foodborne bacterial illness. Head of Public Health England’s Gastrointestinal Bacteria Reference Unit in United Kingdom which is the national reference laboratory for a range of foodborne pathogens including *Salmonella*, *E. coli* VTEC, *Campylobacter*, *Listeria monocytogenes* and *Clostridium botulinum*. Since 2001, she has championed the use of molecular methods leading to improvements in the detection and investigation of bacterial foodborne disease within the UK. Her laboratory is one of the first laboratories in the world to implement the use of WGS for routine bacterial reference service delivery.



Paul Hanlon
Abbott Nutrition, USA

Paul Hanlon earned his doctorate in molecular toxicology from the University of Wisconsin – Madison. After post-doctoral work at the National Institute of Environmental Health Sciences, he has worked as a toxicologist in both the pharmaceutical and food industries. He is a member of the Society of Toxicology and a Diplomate of the American Board of Toxicology. Paul is currently a Director of Regulatory Affairs at Abbott Nutrition where his primary roles are overseeing the regulatory approvals of novel food ingredients as well as providing guidance to food safety programs that govern the control of chemical contaminants. As part of his novel ingredient support, he has coordinated the submission of novel ingredient petitions to multiple regulatory agencies including the US FDA, Health Canada and the China Food Safety Authority. Paul also participates in a number of Codex Alimentarius committees, including the Committee on Contaminants in Foods (CCCF) and the Committee on Food Additives (CCFA). Paul currently chairs the food safety groups for two trade associations: the International Special Dietary Foods Industries (ISDI) Food Safety Working Group and the Grocery Manufacturers Association (GMA) Chemical Management Committee. He is also the current co-chair of the International Life Sciences Institute (ILSI) Food and Chemical Safety Committee, where he has been responsible for organizing workshops and publishing papers on risk-based evaluations of chemical contaminants in food.



Xiaohua He
USDA, ARS, WRRRC, USA

Dr. Xiaohua He is a Research Molecular Biologist in the Foodborne Toxin Detection and Prevention Research Unit at the Western Regional Research Center, USDA Agricultural Research Service, Albany, California. She received her Ph.D. in plant pathology from the University of California, Riverside, and had post-doctoral experience at Purdue and Cornell universities. Her research focuses on development of molecular tools and technologies for sensitive detection of zoonotic pathogens and toxins in food, environment and clinical samples; investigation of toxin synthesis and novel mechanisms of host cell injury by toxins; and development of animal models to study toxicokinetics.



Rene Hendriksen
National Food Institute and Denmark Technical University, Denmark

Dr. Rene Hendriksen is currently employed as Professor MSO at the Technical University of Denmark, National Food Institute and Acting Director and Deputy for the Reference Centres; WHO Collaborating Centre (WHO CC) for Foodborne Pathogens and Genomics and the European Union Reference Laboratory in antimicrobial resistance (EURL AR), respectively.

His main focus is research in global surveillance, epidemiology, antimicrobial resistance, and population structure of mainly foodborne and waterborne pathogens. In addition, his other main focus is advisory service to EC, EFSA, WHO, FAO in the area of AMR and WGS conducting yearly proficiency testing. He has conducted research with 403 scientists in 42

countries and authored 91 peer-reviewed published and accepted articles in international refereed journals. He represents the institute in the WHO GFN, advisor for the WHO AGISAR network, and institute focal point for INFOSAN and ECDC. Currently, he is involved in multiple projects such as the Global Sewage Surveillance project, GMI initiative, EU projects: COMPARE, ListAdapt, and IMPACT, and EFSA projects: ENGAGE, GENCAMP, and ASK.

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



Dr. John Holah is an Applied Microbiologist whose work has focussed on the prevention of microbial, chemical and foreign body contamination of food during its manufacture, distribution and retail. John has an extensive knowledge of the food industry, having worked within >500 food factories and catering establishments, in the UK, Europe, North and South America, Africa, Asia and Australia. John has a passion for food safety and food hygiene 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 undertaken specific investigations into microbial contamination incidents in factories from SMEs to multinational

companies and has recently worked at a corporate level to advise major international companies on developing HACCP, prerequisite and quality systems to provide an integrated food safety plan. He has specific expertise in the hygienic design of food factories and food processing equipment; factory services and water systems; maintenance; cleaning and disinfection; personal hygiene and environmental sampling. At an academic level, John has led several European and national research projects, has written over 100 publications, given over 200 external presentations, edited several books, has a wide range of teaching experience at all levels from industry to University MSc courses, and has been an external supervisor to more than 15 Ph.D. students. John has represented the UK on CEN/TC 216/ chemical disinfectants and antiseptics, chaired ISO/TC 199/WG2 on the Hygiene requirements for the design of machinery, is a member of the Executive Committee of the European Hygienic Design of Equipment Group and until

recently, was a member of the National Health Service Rapid Review Panel. John 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 was previously Head of the Food Hygiene Department at Campden BRI.



Morten Hyldgaard
Dupont Nutrition & Health, Braband, Denmark

See online programme for biography,



Sanja Ilic
The Ohio State University, USA

Dr. Ilic's research interests include microbial safety of foods with a special focus on fresh fruits and vegetables. She investigates transmission and dissemination routes, and interventions to prevent and reduce risks of contamination of fresh produce and other foods with human pathogens within the food systems approach. She uses formal knowledge translation to move research knowledge into practice. Dr. Ilic's research provide novel food safety interventions to control and eliminate foodborne pathogens and innovative approaches of knowledge transfer targeting food producers, handlers, and consumer.



Paul in't Veld
VWA, 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 continued working from 1987 to 1999 at the National Institute of Public Health and the Environment (RIVM). He is currently working at The Netherlands Food and Consumer Product Safety Authority (NVWA), the competent authority in The Netherlands. His 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), to coordinate method development activities and support the organisation with microbiological advice on methods. Mr. in't Veld is a Technical Assessor for various accrediting bodies. He was also a member of AOAC RI Board of Directors until 2014.



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 employment with Nestlé, he was a research scientist and assistant lecturer at Texas A&M University in College Park. An active IAFP member since 2001, Dr. Jackson currently serves on the IAFP Executive Board as President-Elect.

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.



Armitra Jackson-Davis
Alabama A&M University, USA

Dr. Armitra Jackson-Davis is an Assistant Professor in the Department of Food and Animal Sciences at Alabama Agricultural and Mechanical University (AAMU) in Normal, Alabama. She received her Bachelor of Science in Animal Science from the University of Arkansas at Pine Bluff and her Master of Science and Doctor of Philosophy (Meat Science with a research emphasis in meat microbiology) from Iowa State University. Before joining AAMU in 2013, she was a Post Doctoral Research Associate at Iowa State University. In her academic position, she teaches the following classes: Food Microbiology, Food Microbiological Techniques, Advanced Food Microbiology and the Regulation of Food Safety and Quality.

In addition to teaching, she oversees the Food Microbiology Laboratory at AAMU where she mentors high school and undergraduate students in experiential research experiences. Currently, she serves as the Chair of the AAMU Institutional Biosafety Committee. Her research interest focuses on the use of natural antimicrobials in different food systems and the microbiological safety of unpasteurized juices. She has received grant funding to conduct research in these areas and collaborations with researchers in both the academic and governmental sectors. She currently advises five Master of Science graduate students and one Post Doctoral Research Associate. She is a member of the International Association for Food Protection and the Institute of Food Technologists.



Arja Helena Kautto
National Food Safety Agency, Sweden

Arja Helena Kautto graduated as a biologist in University of Oulu, Finland, in 1986, a veterinarian in Swedish University of Agriculture, Uppsala, in 1993 and as a specialist in food hygiene in 2006. The M.Sci. Thesis, 1983–1986, concerning the ecological grazing issues in Finnish reindeer husbandry gave insight into the holistic ecosystem of this semi-domesticated management system. She has been working as a veterinarian and director of veterinary clinics, 1993–2001. Meat inspection, general food control and implementing the EU legislation has been a part of her curriculum in National Food Agency in many ways; as meat inspector 1993–2001, veterinarian inspector 2001–2006 and head of the unit 2006–2012. She has a broad leader education. Since 2012, she has been working with control analysis and especially modernization of meat inspection.

She has been an invited specialist concerning animal welfare for farmed game Ungulates in Wild Animals and Plant Preservation Department, Heilongjiang Province, Peoples Republic of China, August 2005. She was leading the education of food inspectors in food hygiene security and control 2004–2006 at the Institute of Biosciences, Environment and Geosciences at the University of Umeå. In 2012, she took place as a hearing expert for public health hazards to be covered by meat inspection of Farmed Game in EFSA, Parma. During 2013–17 she was a delegate on the Veterinary Program Board, Swedish University of Agricultural Sciences, Uppsala, as well as teaching the control of farmed and wild game for veterinary students. Currently, she is a resident for Diplomate in Veterinary Public Health, Population Medicine, European College of Veterinary Public Health.



Alexandre Leclercq
Institut Pasteur, France

Alexandre Leclercq is deputy director of the French Reference Centre and WHO Collaborating Centre for *Listeria* and biologist at the Laboratory for Urgent Responses to Biological Threats at Institut Pasteur, Paris. Beginning in 1996 as assistant professor in microbiology and chemistry (UCL, Belgium) where he studied molecular epidemiology of STEC. He became deputy director of the department of teaching and head of laboratory for Food Safety and Microbiology at Institut Pasteur Lille in 2000. In 2003, he joined Institut Pasteur at Paris and first studied molecular epidemiology of *Yersinia* and manages microbiological surveillance of *Listeria* since 2007. He is technical auditor in several

accreditation bodies and legal expert on microbiological methods. He has over 70 peer-reviewed publications. He was nominated in 2005 convenor of CEN TC275 “Food Analysis” Working Group 6 “Microbiology of the Food Chain” in Vienna agreement with ISO TC34/SC9 and scientifically manage the CEN mandate M381 of European Commission on validation of fifteen reference CEN ISO methods in microbiology of the food chain.



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, US FDA and various industry contracts. Dr. Lee is an instructor for food microbiology in the Illinois Institute of Technology’s Masters 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 and Agriculture Organization of the United Nations, Italy

Jeff LeJeune serves as Food Safety and Antimicrobial Resistance Specialist, in the Food Safety and Quality Unit of the Food and Agriculture Organization of the United Nations (FAO). His research has primarily focused on understanding the ecological mechanisms involved with the survival, dissemination, and prevention of pathogen contamination, notably antimicrobial resistant bacteria, between reservoir species and water, wildlife, and edible crops. His veterinary training was completed in Canada (UPEI), at the University of Prince Edward Island and his Ph.D. at Washington State University. He is concurrently appointed as Professor, The Ohio State University.



Ingela Marklinder
Uppsala University, Sweden

Ingela Marklinder, Associate Professor and Senior Lecturer in Food, Nutrition and Dietetics teaches dietitian and food science students in food microbiology, food science and health communication. Her research has a health perspective and focuses mainly on food safety, consumers, and health communication. Her thesis (defended 1996) was dealing with lactic acid bacteria and fermented oats and barley. One of her special competence is sourdough processes and wholemeal bread quality.



Peter McClure
Mondelez International, United Kingdom

Peter McClure gained his B.Sc. 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 Mondelez 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 Mondelez, 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.



Rick Meinersmann
USDA, ARS, Russell Research Center, USA

Richard (Rick) Meinersmann, VMD (University of Pennsylvania '85), Ph.D. (Temple University '82) began with the USDA Agricultural Research Service in 1987 on a project to design vaccine strategies to intervene with colonization of chickens with *Campylobacter jejuni*. He studied the diversity of *Campylobacter* and learned to apply powerful statistical analyses on DNA-sequence data to study the population genetics of the organism. These studies contributed to understanding the epidemiology and tracking of the organism. The technology was applicable to other problems and he was able to track *Listeria monocytogenes* in poultry processing plants and described the evolution of a multi-drug resistance plasmid, IncA/C. This led to studies on other antimicrobial resistance genes including mcr-1.



Jeanne-Marie Membré
INRA, UMR 1014 Secalim, France

Jeanne-Marie Membré, Ph.D., is a senior scientist in the quantitative microbiology domain. Her experience encompasses predictive microbiology, microbial risk assessment, applied statistics and food safety. She has been working in research for more than 20 years, particularly at the French National Institute for Agricultural Research (INRA). Currently, she is leading the group “Microbiological Risk Assessment in Food” of the unit research Secalim, at Nantes. In the 2003–2009, Dr. Membré worked at the Safety & Environmental Assurance Centre of Unilever, developing microbiological models in an industrial context. J.-M. Membré is a member of IAFP, of the scientific board of the *Journal of Food Protection* and *International Journal of Food Microbiology*.



Birgitte Moen
Nofima, Norway

Birgitte Moen has a Ph.D. in molecular microbiology from the Norwegian University of Life Sciences (2005), and has worked as a research scientist for the last 12 years at Nofima. She is author of 28 peer-reviewed publications. Her main research interests are microbiota analyses in food, environmental samples, biofilms and in the human and animal gut. Current research topics also include investigation of the mechanisms behind disinfectant resistance in *Serratia marcescens*.



Jim Monaghan
Harper Adams University, United Kingdom

Jim Monaghan has worked in crop science for over 25 years. Following a Biology degree at UCNW Bangor, Jim researched aspects of crop production at Harper Adams University College and John Innes Centre (Ph.D.), Newcastle University, HRI-Efford and HRI-Wellesbourne. Jim then had a look at the real world for three years at Marks and Spencer as Salads Technologist, where he had responsibility for food safety, pesticide residue minimisation, and compliance with codes of practice for all salad products and salad ingredients in minimally processed foods before heading back to Harper Adams to develop

teaching and research in the area of fresh produce production in 2005.

Jim leads the Fresh Produce Research Centre at HAU which is focused on fresh produce production, particularly leafy vegetables and covers three areas: (1) identifying genetic traits that may lead to more sustainable crop production; (2) agronomic manipulation of post-harvest quality and nutritional content in crops; and (3) developing and implementing food safety systems in fresh produce. Jim chaired the Technical Advisory Committee for Red Tractor Produce from 2010–2017.



Christophe Monnet
INRA, France

Christophe Monnet after his Ph.D. in 1994, was concerned with aroma compound production by lactic acid bacteria, Dr. Monnet joined the French Agricultural Research Institute INRA in Grignon, where he still works on the team “Microbial ecosystems of cheeses” at the Joint Research Unit for Food Process Engineering and Microbiology. His research aim is to better understand the interactions between cheese microorganisms, their adaptation to the cheese habitat and the generation of the desired functional properties. Dr. Monnet’s main fields of expertise are the metabolism of cheese bacteria, genomics of cheese bacteria, microbial ecology and the study of gene expression in dairy products.



Sara Monteiro Pires
Technical University of Denmark, Denmark

Sara Pires’ main research area is the health impact of foods and foodborne diseases. She has focused on burden of disease and risk-benefit assessments, and on developing methods for attributing the burden of foodborne diseases to the responsible sources, on surveillance of foodborne hazards, and on risk assessment. Currently, Sara coordinates the Danish initiative to estimate the burden of food-associated diseases in Denmark. She is a member of several international working groups focused on source attribution of foodborne pathogens, including WHO, FAO and EFSA initiatives.



Jérôme Mounier
University of Brest, France

Jérôme Mounier after obtaining a MSc in the French engineering school in Food Science, Agrosup Dijon, Dr. Mounier earned his Ph.D. in Microbiology in 2002 at the Moorepark Food Research Centre (Ireland) on the microbial diversity of smear-ripened cheeses. After a 2-year post-doctoral position in 2005 at INRA (France), he was hired in 2007 as an associate professor at the University of Brest (Laboratoire Universitaire de Biodiversité et Ecologie Microbienne, France). In 2013, he was hired as a full Professor in this university. He is currently the head of one research group within the LUBEM lab. His research interests focus on food

microbial ecology and mainly bacterial and fungal community structure and functions of fermented foods as well as the use of bioprotective cultures in food products.



Sarah O'Brien
University of Liverpool, United Kingdom

Dr. Sarah O'Brien qualified in Medicine in 1986 at Newcastle University. She undertook Higher Specialist Training in Public Health Medicine in Oxford and Newcastle-upon Tyne. She has held consultant positions in health protection in Birmingham, Glasgow and London before joining the University of Manchester in 2004 as Professor of Health Sciences and Epidemiology. Dr. O'Brien moved to the University of Liverpool in 2011 as Professor of Infection Epidemiology and Zoonoses.



Daniel Palm
ECDC, Sweden

Dr. Daniel Palm is working in the European Centre for Disease Prevention and Control (ECDC) as Group Leader for the Molecular Surveillance team. He received his Master of Science from Uppsala University, Sweden and in 2005 his Ph.D. from Karolinska Institutet, Sweden. His research topic for the thesis was the host-pathogen interaction during infections of the protozoan pathogen *Giardia lamblia*. Next, he held a position at the Swedish Institute for Infectious Disease Control where he focused on development of diagnostic tools and piloting novel laboratory technologies, including next generation sequencing. Since 2008, he has worked in ECDC. During the first years in the agency, he was involved in defining strategies and scope for the implementation of molecular typing into surveillance and outbreak investigation. Since 2015, he has led the operational team involved in the routine collection, analysis and interpretation of molecular typing data for EU-level public health purposes.



Valentina Paracchini
European Commission Joint Research Centre, Italy

Dr. Valentina Paracchini holds a BSc in Natural Sciences and a BSc in Biology, providing her with a solid background in applied biomedical sciences. Her Ph.D. degree and post-doctoral research employment dealt with genetic diseases, focusing on biotechnology, therapy and human health. Specifically, she has worked on the epidemiology of cancer and molecular biology of genetic diseases, in particular, cystic fibrosis. Currently, she works as a scientific and technical officer at the Joint Research Centre, Directorate for Health, Consumers and Reference Materials, in the Fraud Detection and Prevention Unit. During her JRC years, she obtained valuable insight in food quality and safety policies, as well as considerable experience in applying NGS technology in the context of food fraud detection and prevention, with a particular focus on fish species identification.



Mickey Parish
U.S. Food and Drug Administration, USA

Dr. Mickey Parish is the Senior Science Advisor for the FDA Center for Food Safety and Applied Nutrition where he had previously served as Senior Advisor for Microbiology in the Office of Food Safety. Prior to coming to FDA, Dr. Parish was Professor and Department Chair in the University of Maryland's Department of Nutrition and Food Science, and a Professor of Food Microbiology at the University of Florida. Dr. Parish is the President of the International Association for Food Protection.



Elizabeth Parker
The Ohio State University, USA

Ms. Elizabeth Parker graduated from the University of Queensland Veterinary School in 1993 and has since worked in mixed, companion and government practice. She was senior lecturer in the College of Public Health, Veterinary and Medical Sciences at James Cook University, (Townsville, Australia), from 2009 until 2015 and Associate Professor in the College of Food, Agriculture and Environmental Science at the Ohio State University, Columbus Ohio from 2015 until the present. Ms. Parker became a member, by examination, of the Australian and New Zealand College of veterinary epidemiology in 2007 and completed a Masters in Veterinary Public Health at the University of Sydney in 2008. She became a Diplomate of the American College of Veterinary Preventive Medicine. She is also currently enrolled in a Ph.D. in the Ohio State University's College of Veterinary Medicine.



Annemarie Pielaat
Unilever R&D, The Netherlands

Annemarie Pielaat has a Masters 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 setting-up biologically relevant experiments, sampling plans and subsequent statistical data analysis as input for microbiological risk assessment. The last few years she's working on methodology development for the implementation of molecular data in microbiological risk assessment of foodborne pathogens. As of December 2016, she started to work for Unilever R&D in the Microbiology&Analytics group as a Science Team Leader Microbiology.



Arthur Pightling
U.S. Food and Drug Administration, USA

As a member of the U.S. Food and Drug Administration's bioinformatics team, Arthur Pightling analyzes whole-genome sequence (WGS) data, develops tools for bioinformatic analysis, and provides interpretation of results that are used in outbreak investigations and compliance decisions. He was a post-doctoral fellow at the Listeriosis Reference Service for Canada where he developed the use of WGS data for characterizing *Listeria monocytogenes*. He is an active member of the Global Coalition for Regulatory Science Research and the ISO working group "Whole-genome sequencing for typing and genomic characterization."



Jessica Pryor
G2S at Centers for Disease Control and Prevention (CDC), USA

After graduating from SPSU, Jessica Pryor started at CDC in the genomics unit sequencing laboratory. She then moved to the MicrobeNet project where she worked capturing information about bacterial species. In 2017, Jessica started as the MicrobeNet Unit Lead coordinating the efforts of the MicrobeNet team and organizing training workshops both domestically and internationally.



Jennifer Quinlan
Drexel University, USA

Jennifer Quinlan is an Associate Professor in the Dept. of Nutrition Sciences at Drexel University. Her areas of expertise include food safety risks for low income and minority populations and consumer food safety education. She currently serves on the Editorial Boards of *Food Protection Trends* and the *Journal of Food Protection*. She has served on USDA's National Advisory Committee for Microbiological Criteria for Foods and is a former Fulbright Scholar to Corvinus University in Budapest, Hungary. She earned her B.S. and M.S. from Rutgers University and her Ph.D. in food science from North Carolina State University.



Lynne Regent
Anaphylaxis Campaign, United Kingdom

Lynne Regent joined the Anaphylaxis Campaign as CEO in October 2008. Her role is to lead the charity and ensure that it achieves its objective of helping people with severe allergies live their lives. Prior to taking up this role, she spent 30 years working in the National Health Service. The Anaphylaxis Campaign is the only UK-wide charity to exclusively meet the needs of people at risk from severe allergic reactions and provides help and information for everyone affected by food allergy. Guided by some of the UK's top allergy experts, the Campaign has become a leading adviser to the food industry, government and health professionals.

The Campaign provides services for individual and clinical professional members and has a corporate membership programme.



Steven Ricke
University of Arkansas, USA

Dr. Steven C. Ricke received his B.S. and M.S. from the University of Illinois and Ph.D. from the University of Wisconsin. Dr. Ricke was a USDA-ARS post-doctorate in the Microbiology Department at North Carolina State University then joined Texas A&M University as a professor in the Poultry Science Dept. In 2005, he became the first holder of the new Donald "Buddy" Wray Endowed Chair in Food Safety and Director of the Center for Food Safety at the University of Arkansas (UA) and is a faculty member of the Dept. of Food Science and Cellular and Molecular Graduate program. He received the Poultry Sci. Assoc. (PSA) Research Award, American Egg Board Award and honored as a Texas Agricultural Experiment Station Faculty Fellow, and the Division of Agriculture –UA John White Outstanding Research Award. He served as co-founder and former president of the Arkansas Association of Food Protection (AAFP) and named as an AAFP and a PSA Fellow. Dr. Ricke's *Salmonella* research projects have emphasized studies on the growth, survival and pathogenesis of the organism under conditions encountered during food animal production and processing.



Andrés Rodríguez Lozano
Commercial Food Sanitation, The Netherlands

Andrés Rodríguez Lozano joined Commercial Food Sanitation in March 2017, with over 9 years of experience in the food industry. His career started at Campden BRI (Chipping Campden, UK) where he worked in the Microbiology Department. He then joined the Mondelez International Food Safety team where he supported the business across different categories (confectionary, coffee and biscuits) by providing food safety support to R&D, internal and external plants and suppliers across Europe. Andrés has experience working on cross-functional and multicultural teams. At Commercial Food Sanitation, Andrés continues his passion for supporting and training the industry on their food safety and sanitation programs. Andrés holds a veterinary degree (University of Murcia, Spain) and both a MSc (University of Tennessee, USA) and a Ph.D. (University of Massachusetts, USA) in food microbiology.



Jean-Marc Rolain
Aix-Marseille Université, France

Professor Jean-Marc Rolain, Pharm.D., Ph.D. at Institut Hospitalo-Universitaire (IHU) Méditerranée Infection, Aix-Marseille University in Marseille, France. He is the head of the teaching Microbiology laboratory at the Pharmacy School of Marseille, France since 2005. In the IHU he is the leader of a research team dedicated to the study of resistance to antimicrobial agents since 2005. The main objective of the team is to decipher the molecular mechanisms of antibiotic resistance and genomic evolution in terms of resistance in new and/or emerging pathogens. He has strong experience in those fields and in emerging infectious diseases with translational knowledge in other fields, including new technologies, virology and tropical diseases (parasitology and mycology). He is currently author/coauthor of more than 450 international indexed publications in Pubmed and ISI web of knowledge (H factor = 48; 10000 citations), had more than 150 international communications, 10 patents and is co-founder of two startups. Since 2015 he is the Editor in Chief of *International Journal of Antimicrobial Agents* (IF = 4.307).



My Sahlman
The Federation of Swedish Farmers, Sweden

My Sahlman has worked as a senior advisor in animal health and food safety at the Federation of Swedish Farmers for the last eight years; an interest and business organisation for the green industry in Sweden, with approximately 150,000 individual members representing some 90,000 enterprises. After graduating from the Veterinary College in Uppsala, My has been working with bacteriological and chemical investigations for food and water as assisting laboratory manager with a focus on pathogens and chemical hazards. She has also been dealing with animal diseases, nationally and internationally at the Swedish Board of Agriculture.



Roger Scheffler

Commercial Food Sanitation, The Netherlands

Roger Scheffler has a background in design engineering. He first became involved in food protection and processing while working as an account manager for Intralox. In this role, he advised meat-, poultry-, and seafood-processing customers on selecting conveyance solutions and services in order to ensure product quality and safety. In his current role at Commercial Food Sanitation, Roger combines a thorough knowledge of the machinery and equipment used in food processing plants with field-tested experience in food safety practices to assist customers in enhancing sanitation efficiency while meeting regulations.

Roger supports customers in Europe in South Africa in a variety of areas. These areas include sanitation down-time reduction, cleaning sequencing, sanitation program analysis for continuous improvement, equipment design reviews, development of SSOPs and associated documentation, and sanitation and hygiene training.



Jøergen Schlundt

Nanyang Technological University, Singapore

Jøergen Schlundt, Professor, Nanyang Technological University (NTU), Singapore JS, has a DVM and Ph.D. from the Royal Veterinary University in Denmark. He has worked nationally and internationally on food safety and security, including 11 years as Director Department for Food Safety and Zoonoses at the World Health Organization, and 4 years as Director National Food Institute in Denmark. Dr. Schlundt has participated in international development of food safety Risk analysis principles, as well as international evaluations of the importance of antibiotic use in agriculture. He chairs the Global Microbial Identifier, an international initiative suggesting a global database of DNA-sequences of all microorganisms.



Gabriele Scholz

Nestlé Research Center, Switzerland

Gabriele Scholz is a food safety scientist at the Nestlé Research Center in Lausanne, Switzerland. She is a molecular biologist by training and obtained her Ph.D. at the Freie Universität Berlin. She then moved into the field of the validation of alternatives to animal experimentation and in vitro toxicological assays at the Federal Institute for Risk Assessment (BfR) in Berlin. Gabriele changed to work in the pharmaceutical industry where she applied in vitro safety testing using organ cell culture models in early drug development. Since more than 13 years at Nestlé, she has built a strong expertise in the health risk assessment of chemical contaminants in foods, particularly those that arise through heat processing. Being active in working groups (e.g., ILSI Europe) with various stakeholders to evaluate the safety relevance of such compounds for human dietary consumption, she has a good record of publications in the area. She is a member of the European Society for Alternatives to Animal Testing (EUSAAT) and the International Maillard Reaction Society (imars).



Deb Smith

UK:IE EHEDG & Vikan, United Kingdom

Deb Smith has >30 years food safety/research training and experience. She has previously worked in the food industry; the food safety division of DEFRA; and the Food Hygiene Department at Campden BRI. She is a qualified microbiologist, food scientist and FSSC 22000 auditor; an active Committee Member of the EHEDG and IFST; and is currently Chair of Campden BRI's Microbiology Members Interest Group. Deb has authored/co-authored numerous food safety/hygiene publications, including peer reviewed papers, book chapters and guidelines. She has also presented her work at many national and international food safety events. At Vikan, Deb is responsible for providing expert hygiene advice, training and support to the food industry.



Katharina D.C. Stärk
SAFOSO AG, Switzerland

Katharina Stärk graduated as a veterinarian from Zurich Veterinary School and obtained her Ph.D. from Massey University (New Zealand). Over the last two decades, she has conducted applied research on infectious diseases and zoonosis as well as risk analysis, surveillance and animal health decision making, particularly in relation to the safety of animal-derived food. She has worked in diverse environments including government, private industry and academia in several countries. Katharina has repeatedly served on international working groups and expert panels. Currently, she is director for Science and Quality SAFOSO AG,

Bern, and honorary Professor of Veterinary Public Health Policy at the Royal Veterinary College, London, UK.



Eric Stevens
U.S. Food and Drug Administration—CFSSAN-ORS-DM, College Park, MD, USA

Eric L. Stevens, Ph.D., is a Staff Fellow in the Office of Regulatory Science at the Center for Food Safety and Applied Nutrition. He received his Ph.D. in Human Genetics and Molecular Biology from The Johns Hopkins School of Medicine with an emphasis on human population genetics and estimating genetic relatedness. From 2013–2014 he completed a Postdoctoral Fellowship analyzing RNA sequencing data to find differentially expressed genes and isoforms related to schizophrenia.

In 2014, he became an FDA Commissioner's Fellow. Dr. Stevens' current work continues to focus on engaging other federal and international stakeholders in applying WGS methodologies to track foodborne outbreaks through CFSAN's GenomeTrakr. He travels extensively to both domestic and international locations to represent the Office of Regulatory Science and CFSAN in how FDA is using WGS for its food program. Dr. Stevens works closely with international organizations.



Priya Sundaram
Danone, The Netherlands

Priya Sundaram graduated with a Ph.D. in microbiology from Iowa State University where she began her career in food safety teaching HACCP, food safety principles and microbiology as part of the university extension education program. Her thesis on shelf-life extension of meat products led to a job in the meat processing industry in the Chicago area and onto food safety roles in quick service restaurants, dairy, acidified and low-moisture foods, etc. with Kraft Foods and baked goods manufacture with Aryzta. As part of the Danone Food Safety Board, Priya supports all supply chain food safety for all Danone business units worldwide. She is passionate about methodical science-based decision

making and robust training and communication as hallmarks of quality culture.



Karin Tegmark Wisell
The Public Health Agency of Sweden, Sweden

Dr. Karin Tegmark Wisell is the Director of Microbiology and the Head of the Department of Microbiology at The Public Health Agency of Sweden and is as such responsible for the national microbiological surveillance programme including food and waterborne diseases, antibiotic resistance and health-care associated infections, vaccine-preventable diseases, parasitology and tuberculosis. She graduated in Medicine from the Karolinska Institutets and trained to become a specialist in Clinical Microbiology at The Karolinska University Hospital. Her Ph.D. thesis on Regulation of Virulence Gene Expression in *Staphylococcus aureus*

explored the intricate agr regulatory system through, at that time advanced genetic analyses. Her main research during the past years has focused on various microbiological and epidemiological aspects of antibiotic resistance and communicable disease control. She has served as a member of The Nordic Society of Clinical Microbiology and Infectious Diseases, The Swedish Reference Group for Antibiotic Resistance, the Karolinska Institutets Biosafety Committee, the Swedish Society for Clinical Microbiology Educational Committee, The Programme committee of the Diploma programme in Infection Control and Hospital Hygiene at the Nordic School of Public Health and was the Scientific Secretary for the Annual Conference of Scandinavian Society for Antimicrobial Chemotherapy in Stockholm 2009, and is currently the Chairperson of the Swedish Reference Laboratory Network in Microbiology.



David Tomás Fornés
Nestlé, Switzerland

David Tomás Fornés is senior scientist in the Microbial and Molecular Analytics group at Nestlé Research Center in Lausanne, Switzerland. He works on the evaluation, development and validation of microbial methods for pathogen detection and sample preparation. He participates in ISO and CEN technical committees developing reference methods for microbiological food analysis. He has been expert in international cooperation projects supported by the European Committee for Standardisation (CEN) and the Food and Agricultural Organization (FAO).



Ivar Vågsholm
Swedish University of Agricultural Sciences (SLU), Sweden

Ivar Vågsholm graduated as a veterinarian in 1984 and received a Ph.D. from University of California. He has worked with European Union questions relating to animal health and food safety the last 30 years, working in the EFTA Surveillance Authority 1992–1995, thereafter as a scientific expert. Professor Vågsholm is a member of the EFSA scientific panel on Animal Health and Welfare from July 2012 and has been member of the Biological Hazards panel 2003–2012, The Scientific Committee of Veterinary Measures relating to Public Health 1997–2003, member ECVPH education Committee since 2002, and member editorial board – Preventive Veterinary Medicine, member advisory group Swedish medical products agency. He has participated in 2 WHO consultations during 2012 on *Campylobacter* control and environmental health. In particular the revision of meat inspection and food safety legislation, and control of TSE have been a major topic during the last 6 years.



Eddy Van Collenburg
Bio-Rad Laboratories, The Netherlands

Eddy Van Collenburg has been working for Bio-Rad Laboratories for over 25 years, the last 6 years as a Droplet Digital PCR Specialist for Europe. After working for >10 years in sales, he started to work in 2000 as a qPCR application specialist. where he developed and supported multiple applications. During this time, new applications in several fields (gene expression, environmental analysis and diagnostic) were developed. As a ddPCR specialist, he is involved in development and supporting new and existing applications. His main focus in this area involves liquid biopsy in cancer, environmental DNA detection, gene expression, residual disease monitoring, copy number alterations and targeted genome editing applications. He has experience in optimizing and designing specific assays for ddPCR applications and sample preparation for the mentioned applications.



Inge Van der Linden
Ghent University (UGent), Faculty of Bioscience Engineering, Department of Food Technology, Safety and Health, Research Unit Food Microbiology and Food Preservation (FMFP-UGent), Belgium

Dr. Inge Van der Linden graduated as Master of Science in Biology at Ghent University in 2005. During her Ph.D. (2009–2013), she studied the behaviour of *Escherichia coli* O157 and *Salmonella enterica* during greenhouse butterhead lettuce production at the Flemish Research Institute for Agriculture, Fisheries and Food (ILVO). Her academic promotor was Prof. Dr. Ir. Mieke Uyttendaele from the Research Unit Food Microbiology and Food Preservation (FMFP) of Ghent University. She further pursued research at FMFP-UGent working as a postdoc on the internalisation of foodborne pathogens in minimally processed lettuce (within the framework of the EU P7 Veg-i-Trade project). Since 2014 she has been working on the safety of sprouted seeds, a collaborative project of FMFP-UGent and ILVO, which is – as was her Ph.D. project – funded by the Belgian Ministry of Public Health.



Leo Van Overbeek
WUR Plant Research, The Netherlands

Leo Van Overbeek is senior scientist at Wageningen UR. His main scientific interest is on the roles that microorganisms play in plant production systems. These interactions can be positive from a human perception such as plant growth stimulation, but also negative such as contamination with human pathogens. He received his Masters degree in molecular microbiology at Utrecht University in 1992 and his Ph.D. at Leiden University in 1997. He is principle investigator in endophyte and plant microbiome research at Wageningen University and Research. He recently was elected as chair of Cost Action 16110 on 'control of human pathogenic microorganisms in plant production systems (HUPLANTcontrol)'.



Purnendu Vasavada

University of Wisconsin-River Falls, USA

Dr. Purnendu C. Vasavada is a Professor Emeritus of Food Science, University of Wisconsin-River Falls and principal and managing member of the PCV & Associates, LLC. Previously, Dr. Vasavada served as a FDA-ORISE Fellow (2011–2013) and the coordinator of the Food Safety Preventive Controls Alliance (FSPCA). He is currently serving on several FSPCA committees, including the FSPCA outreach and technical assistance program. Dr. Vasavada is the author or co-author of over 100 publications. He has received numerous awards and professional recognition including a Fellow of the American Academy of Microbiology (1990), the IFT (2009), the IAFP (2010) and IAFST (2017).



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 & 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.



Patrick Wouters

EHEDG & Cargill, The Netherlands

Patrick Wouters leads the Global Hygienic Design Centre of Expertise and community of practice, and is responsible for increasing collaboration and visibility of food safety engineering principles, standards and technologies throughout the Cargill organisation. Core responsibilities include the development and promotion of hygienic process, equipment, and facility design and other technologies that contribute to the production of safe products in existing plants and in new projects. Next to his position in Cargill, Patrick is the Vice President of the EHEDG – European Hygienic Engineering & Design Group – organisation. Prior to Cargill, Patrick worked in Unilever and had various roles in R&D and quality management.



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).



SYMPOSIUM ABSTRACTS

25-27 April 2018 – Stockholm, Sweden

SYMPOSIUM ABSTRACTS

OPENING SESSION

The Public Health Agency's Role in Foodborne Disease Outbreaks

KARIN TEGMARK WISELL, The Public Health Agency of Sweden, Solna, Sweden

The Public Health Agency of Sweden has a national responsibility for public health issues and works to ensure good public health. The agency works with protection of the population against communicable diseases and other health threats and is responsible for national coordination of communicable disease control within the human sector. The work involves drawing up strategies for developing and improving communicable disease prevention and control, recommendations based on the latest scientific results in the field, monitoring, and analysis of the epidemiological status of communicable diseases in the population, with particular emphasis on diseases covered by the Swedish Communicable Diseases Act. The agency moreover participates in extensive international cooperation with international bodies such as the EU and WHO.

Within the area of foodborne disease outbreaks the agency has extensive collaboration with the National Food Agency, the National Veterinary Institute, the County Medical Officers and the Clinical Microbiological Laboratories throughout the country. Specifically, the agency closely monitors the reporting of salmonellosis, shigellosis, campylobacteriosis, yersiniosis, cryptosporidiosis and EHEC-infections through notifications according to the Swedish Communicable Diseases Act. The agency is moreover conducting national microbiological surveillance where specimens from the Clinical Microbiological Laboratories are collected for characterization below species-level by epidemiological typing. During the past years whole genome sequencing has replaced most of the traditional typing methods and by doing so also demonstrated the superiority of this method to priorly established methods. This presentation will focus on the benefits and opportunities whole genome sequencing has enabled in the work with foodborne disease outbreaks.

Global Lessons of the Swedish Model

IVAR VÅGSHOLM, Swedish University of Agricultural Sciences (SLU), Uppsala, Sweden

History never repeats itself but it rhymes said Mark Twain. The same applies to lessons in Food Safety — the insights gained from success stories and not so successful stories can be useful. The global food chains are complex and opaque and operate at high volumes and with low margins. The consumers have to trust the official controls and the warranties implicit in the food business operators' trademarks. The Swedish consumers have high trust in food safety authorities (Eurobarometer 2010).

The Swedish approach to food safety has been guided by the tenets of (a) radical solutions in primary production; (b) proactive approaches – taking action when risks are identified; (c) close collaboration between public health, food safety and animal health authorities; (d) close collaboration and dialogue between food industry and official control agencies; and (e) focus on specific hazards such as *Salmonella*.

The drivers for Swedish food control are and have been (1) the Swedish membership of the European Union (EU) – all food safety legislation is harmonized, (2) the One Health – One Medicine paradigm – public, animal and environmental health is linked with food safety, (3) adapting to climate

change novel foods – e.g., insects, vegetarian lifestyles, (4) globalization and added complexity of the food chains, and linked to the previous point (5) food frauds e.g., fish sold of unclear origin, (6) the increased use of quality assurance standards, industry guidelines to ensure safety and quality of the foodstuffs.

Using microbiological risks as a case study the success of *Salmonella* control is showing the success of proactive approaches in primary production. The use of a hazard-based approach focusing on *Salmonella* misses other important foodborne hazards, such as enterohaemorrhagic *Escherichia coli* (EHEC) and *Campylobacter* that should have been included in a risk-based approach.

Science and Food Safety Policy at the U.S. Food and Drug Administration

MICKEY PARISH, U.S. Food and Drug Administration, Washington, D.C., USA

The Food and Drug Administration regulates the safety of most foods (except meats, poultry, catfish and certain egg products) within interstate commerce in the USA by writing, implementing and enforcing rules that regulate production and transport of foodstuffs. These rules, also known as regulations, are written to support laws passed by Congress and signed by the President. Regulations written by federal agencies have the force of law and firms engaging in the production of foods must comply with the regulations. Production of new regulations is a science-driven and science-informed process involving review and analysis of existing information prior to publishing a "draft" regulation that is open for public comment. Those comments are then reviewed and changes deemed appropriate are made to the rule before issuing a "final" regulation that requires compliance by industry. This presentation will describe this process and the utilization of science for decision-making purposes at FDA.

S1 FSMA & FSVP – Impact on the Global Food Industry

Many sub-rules have been put in place since the United States Food Safety Modernization Act (FSMA) was signed in 2011. One of these, in particular, has a big, long-term effect on the Global Food Industry: the Foreign Supplier Verification Program (FSVP). Under the Foreign Supplier Verification Program, any company exporting feed, food or food ingredients to the United States has to comply with FSMA regulation. That means the exact same regulation any food processing company inside the United States of America has to adhere to.

Under FSVP, the importer is responsible for proving compliance of its foreign suppliers. The responsibility of complying is with the food industry. The U.S. Food and Drug Administration (FDA) is planning to increase their audit intensity (FDA visits to food processors outside the U.S. are planned for 2018) to ensure FSMA requirements are met and products can safely be allowed into the U.S.

What is the impact on a food processor outside of the U.S.? What does it mean for the industry? What changes have to be implemented to comply and how do we prepare for a possible FDA visit? All of these questions and more will be covered in this session. After attending you will have a good understanding of the compliance needs when exporting to the United States.

Exporting to the United States – What Changed for Us?

PRIYA SUNDARAM, Danone, Amsterdam, The Netherlands

The Cycles and Procurement Business Unit of Danone procures materials and services for the various business units of Danone. This 'buy and sell' model poses unique challenges in that we are both a supplier (and sometimes a Foreign Supplier) of Danone factories as well as a customer (and sometimes importer). Additionally, the recent inclusion of Whitewave into the Danone family brought with it added complexity of products that were being regulated very differently than in the past.

What changed was not so much the way we manufacture products or our standards and processes for quality and Food Safety. It was the way we thought about, talked about and managed regulatory compliance and supplier food safety.

This presentation covers the complexity and multiple challenges faced in FSMA compliance and developing a Roadmap for achieving it. It starts with building awareness that FSMA was not just another set of regional regulations and bridging both the language and cultural divide between HACCP and Risk Based Preventive controls. It also highlights the development of a FSMA platform in collaboration with all stakeholders-business units, regions, internal functions and suppliers. This includes a common language and metrics for the elements of the food safety plan: a science based risk assessment, definitions and rationale for monitoring, verification and validation actions.

How to Prepare for a Possible FDA Visit

ANETT WINKLER, Cargill, Munich, Germany

The final Foreign Supplier Verification Programs (FSVP) rule requires that importers perform certain risk-based activities to verify that food imported into the United States has been produced in a manner that meets applicable U.S. safety standards, and to ensure that the supplier's food is not adulterated and is not misbranded with respect to allergen labeling.

There are many different types of verification activities that can be used to meet the requirements: a review of the supplier's relevant food safety records, sampling and testing, and onsite auditing as examples of verification activities that may be appropriate, either individually or in combination. Going further, suppliers may be subject to FDA inspector visits to review compliance with FSVP requirements, where applicable. Preferably, the preparation for such visits starts when manufacturers/processors know that they would need to comply with FSVP requirements. It is helpful to get familiar with the language used by FDA in order to avoid misunderstandings and/or lengthy discussions. That requires a throughout understanding of the current U.S. Food Safety standards, starting with a "Food Safety Plan," its verification activities and how is that integrated into the business to ensure smooth operations and actions at appropriate times.

Part of the preparation should also include defining clear responsibilities and contacts during an FDA visit to ensure that valuable information is not lost. There a catalogue of respective contacts could be a helpful tool, especially for businesses which are not only managed on a local basis.

Further on, manufacturers/processors should be aware of which materials/production components would be part of an FDA visit – and which ones would not fall into that, since that would set the overall scope of the visit.

Sanitation Programs under FSMA Regulation

ANDRES RODRIGUEZ LOZANO, Commercial Food Sanitation, Amsterdam, The Netherlands

Improper or suboptimal sanitations programs can lead to food safety incidents. FSMA related activities are looking to tighten this gap via risk-based and preventative controls under sanitation controls and hygiene zoning. Whilst these chapters have not been yet published, this presentation will give an overview on how FDA's new guidance and publications under FSMA are gearing towards higher control,

monitoring, verification and documented corrective actions of the manufacturing facilities sanitation and hygiene zoning programs.

S2 Teaching Food Safety to Dietitians: Toward an International Network

The symposium focuses on translating collaborative research findings into action, developing efficient methods to work with dietitians to facilitate the delivery of food-safety information and explore the need for developing an international network of food-safety educators for dietitians. The first presentation will review how dietitians are trusted health professionals for food and nutrition messages and how dietitians are exposed to food-safety science in their curriculum under the 2017 Standards of the Academy of Nutrition and Dietetics (USA) and the British Dietetic Association Curriculum Framework for the Education and Training of Dietitians (UK).

Perspectives on the food safety education that dietitians receive will be provided by the second presenter. Identified barriers to classroom teaching, students' attitudes and opportunities for alternative approaches to teaching food-safety to future dietitians will be presented. Collaborative USA and UK research will emphasize the need for adjustments in current curricula.

Finally, a first-hand account of engaging Swedish dietetic students in food-safety will be shared. The growing interest in the food-safety training and education of dietitians will be discussed and the need for the development of an international network of food safety educators for dietitians explored. Visions for the network will be communicated, and the need for a training toolkit to facilitate appropriate training and education will be explored.

Cumulatively, the session will give insight on how a group of like-minded academics aspires to integrate dietetics students in effective food safety education to empower dietitians to inform and enable patients to implement food safety practices and reduce the risk of foodborne illness.

Engaging Dietitians to Promote Safe Food-handling Messages – Opportunities in the Curriculum

JENNIFER QUINLAN, Drexel University, Philadelphia, PA, USA

This presentation will review the role and importance of dietitians as trusted health professionals for food and nutrition messages in the health care system. While most consumers will not directly interact with a food microbiologist, individual and group consultations with dietitians are not uncommon with the increase in obesity and accompanying chronic diseases. The dietitian, therefore, may serve as a partner to food microbiologists in delivering safe food-handling messages directly to consumers. To do so, however, food microbiologist must ensure that their messages complement and accommodate the intense education training dietitians already receive. Dietetic programs are required to meet strict accreditation requirements in their curriculum under the 2017 Standards of the Academy of Nutrition and Dietetics (USA) and the British Dietetic Association Curriculum Framework for the Education and Training of Dietitians (UK). Required student learning outcomes in the curriculum that offer the opportunity to teach safe food handling messages to dietitians will be reviewed. Current food safety education efforts by the profession will also be highlighted. This presentation will identify where in the current curriculum and continuing education requirements food microbiologists can potentially partner with dietetic educators and dietitians to most efficiently and effectively promote safe food handling messages.

In the Classroom with Dietitians: Student Interest and Engagement in Food Safety

SANJA ILIC, The Ohio State University, Columbus, OH, USA

For optimal health, food needs to be nutritious and safe for consumption. Dietitians are the recognized professional group primarily engaged with delivery of nutritional and

therapeutic dietary/food counsel. Moreover, they are trusted health professionals for food related messages. The studies with immune compromised patients demonstrate that they are commonly not aware of the increased food safety risks due to their condition. This disconnect can have a serious impact on this country's ability to improve the public health of the consumer. Targeting dietetics program trainees who are about to begin their professional career is the ideal focus for this study, a focus that will most likely have a long-term impact. The purpose of the study is to describe the food safety risk awareness, knowledge and attitude of dietetic program students as well as their attitudes and self-efficacy delivering food safety messages to patients. A total of 80 (92.5% between age 19–22) students, future dietitians participated in the study. Of the students that received food safety education (n=73/80), one half (46%) thought that they have not received sufficient food safety training to deliver education to the clients. Only 33 (41%) of students have ever heard about listeriosis, and only one understood the association with high risk foods. Only 15% have ever heard about *Campylobacter*, and none of the students were able to relate the pathogen to high risk foods. Further, 80% of students did not identify pregnant women as population at risk and only 13% knew that people at diabetes have increased susceptibility to foodborne diseases. The students in general did not like food safety classes and did not consider food safety important to their practice. This collaborative USA and UK research emphasized the need for adjustments in current curricula. The findings of this study will be used to develop novel approaches to teaching food safety to dietetics students.

The Importance of Teaching Food Safety to Dietitian Students and Visions for an International Network of Dietetic Food Safety Educators

INGELA MARKLINDER, Uppsala University, Uppsala, Sweden

Dietitians in Sweden work mainly in hospitals, primary healthcare and communities as well as in the food industry. They are responsible for dietary treatment and counseling to individual patients. They will meet vulnerably groups susceptible to food poisoning. Thus, dietitian students need to know how to handle food in a safe way.

The dietitian program at Uppsala University provides the students to develop their knowledge regarding pathogenic microorganisms and to relate them to food handling and critical health risks. The students also learn how to apply food safety control programs based on the European food legislation. Further, to describe hygienic principles for serving different diets in hospitals. My experience as a food safety lecturer is that the dietitian students very soon become interested in the complex food safety area; however, this is just a small part of the current curriculum.

In 2011, a National network in food safety teaching was established at seven universities in Sweden. It has been fruitful to meet other senior lecturers teaching food safety. This session initiates the start of an international network in order to increase the possibilities to discuss food safety literature, exchanging tools for teaching in food microbiology laboratories, and, last but not least, to highlight the importance of the food safety topic for dietitian students.

Dietitians are the recognised professional group primarily engaged in the delivery of nutritional and therapeutic dietary/food advice. The symposium consists of international speakers with a shared belief that food should be nutritious and safe for optimal health. To enable this, they believe dietetic-curriculum could be enhanced to incorporate stronger food-safety messaging.

diversity and variety of food production systems provide innumerable niches for the growth of microbial populations and the selection antimicrobial-resistant bacteria. For example, the types and kinds of resistant bacteria and the pressures exerted in the production of meat and poultry differ greatly than those found in the aquaculture and the cultivation of plants. This problem is exacerbated by the use of antimicrobials in food production systems. The use of sanitizers and biocides through the food chain may also further contribute to AMR. This multi-sectoral dimension of AMR highlights another challenge – the need for a cross-cutting and integrated approach to address this serious food safety hazard. Using a One Health perspective, this symposium will explore the dynamics, drivers, and possible control mechanisms of antimicrobial resistant bacteria in the food chain—other than during the primary production of animal protein—including environmental, processing, and horticultural contributions to food contamination with antimicrobial-resistant microorganisms.

Does Biocide and Disinfectant Use in Food Production Drive Antimicrobial Resistance?

SANJA ILIC, The Ohio State University, Columbus, OH, USA

Biocides are chemicals with antimicrobial properties that play an essential role in limiting the spread of infectious diseases. The food industry is dependent on these chemicals, and their increasing use is a matter for concern due to their potential to promote emergence of resistant microbial strains. Specifically, the emergence of bacteria demonstrating increased tolerance to biocides, coupled with the potential for the development of a phenotype of cross-resistance to clinically important antimicrobial compounds is of public health relevance.

Biocide tolerance has become an important food safety issue because it may increase persistence of human pathogens in the food chain. Bacterial resistance may emerge following the inappropriate use or inadequate storage of biocides, resulting in a decrease in the effective biocide concentration. Moreover, the use of antimicrobial compounds at inappropriate doses in the food industry may lead to enhanced ability of bacteria to produce biofilms, which are common source of food contamination with biocide bacteria during processing. In addition, bacteria may elicit common cellular responses to counteract the effects of biocides that confer cross-resistance to antibiotics, a phenomenon that has been known as biocide-antibiotic cross-resistance.

The role of biocides as selectors of resistant strains, or as inducers of mechanisms involved in antimicrobial resistance has been described in numerous genetic and bacteriological experiments. Studies reporting cross-resistance in clinically important antibiotics are conflicting. However, data establishing risks in food processing environment as well as data linking the cross-resistance with antibiotics in clinical and food isolates is lacking. Upon the synthesis of the available evidence, prudent use of biocides might be recommended in order to maintain their effectiveness and to limit release in the environment. However, additional research including epidemiologic investigation and analysis of the data from commercial settings is required to better assess the implications of biocide resistance in possible monitoring recommendations for food industry.

The Environment: Source or Sink for Antimicrobial Resistance in Food and Agriculture?

ELIZABETH PARKER, The Ohio State University, Wooster, OH, USA

Antimicrobials are used in large quantities (1000's of tonnes per year), in both human and animal medicine. Active and partially metabolized antibiotics are found in human and animal waste after excretion in either urine or feces. The effectiveness of waste treatment in removing antibiotic residues is variable and in many developing countries human and animal wastes may not be treated at all. Treated and

S3 Drivers and Dynamics of Antimicrobial Resistance in Food: Beyond Antimicrobial Use in Animals

When it comes to the transmission of antibiotic-resistant bacteria, our food supply is a critical link between animals, plants, the environment, and humans. The

untreated waste products may be used as crop and pasture fertilizer or simply disposed of in terrestrial and aquatic environments. Antibiotic residues have been detected in most environmental media including soil and surface, underground and coastal water and sediment. In addition to the direct effects of antimicrobial contamination on the environmental and human and animal health there is also a concern that selective pressure will cause the proliferation of antimicrobial resistant pathogens. Globalization of trade and human movement results in spread of these resistant organisms and their genetic material to populations of humans and animals at locations distant from the source of contamination. Antimicrobial resistance (AMR) is considered by the World Health Organization to be a serious threat to global public health and that action should be taken by all government and societies to address this growing concern.

Whilst scientific research has confirmed that environmental contamination with antibiotic residues is widespread and that exposure to antibiotics supports the development of antibiotic resistance the impact on human and animal health is still largely unknown and unquantified.

Antimicrobial Use and Resistance in Plant-based Agriculture

JEFFREY LEJEUNE, Food and Agriculture

Organization of the United Nations, Rome, Italy

Bactericides, fungicides and other plant protection products play an important role in the management of plant diseases. However, their use can result in residues on plants and in the environment with detrimental consequences. Use of streptomycin and oxytetracycline is correlated with increased resistance among these plant pathogens to these agents. Resistance to copper compounds and other fungicides is also frequently observed. Importantly genes can be exchanged among a variety of bacteria in the plant production environment, including phytopathogens, soil bacteria, and zoonotic bacteria that are occasionally present in the plant production environment and in the food chain. Through co-resistance, cross-resistance and gene up-regulation, resistance to one compound may confer resistance and multi-drug resistance to other similar or even very dissimilar compounds. Given the alarming and global rise in antimicrobial resistant organisms worldwide and their effects on plant, animal and human health, the prudent use of plant protection products is required to maintain their effectiveness and limit the emergence and transmission of AMR microorganisms from horticultural sources.

and at the European level by CEN TC275/WG6. Between 2011 and 2017, European Commission financially supported the European and international validation of the 15 main reference methods of the microbiology of the food chain concerning bacteria, virus, toxins and histamine. This validation and the subsequent standardization of these methods have been managed by CEN TC275/WG6. These validations were conducted according to the content of the EN ISO 17468, a standard which defines the steps to develop and validate a reference method. This standard completes the series of EN ISO 16140 standards that cover now validation and verification of methods used in microbiology of the food chain.

New insights in the microbiology of the food chain, such as ddPCR and isothermal amplification but also whole genome sequencing, are in development at standardization level, replacing step by step the classical microbiology used in reference methods. New fields of standardization are opened such as the standard on challenge tests. New models of standard, such as modular standards, are established to follow new analytical technologies or emergent virus/bacteria.

Standardization is often considered as a slow process, rigid and using old methods but it is moving rapidly to propose to the users to work with update analytical technologies. Knowing its analysis target and need of its customers, the user of standards will now have all the tools to use the appropriate and accurate methods and to show its performance and competence.

Microbiological Reference Methods, What's New, What's Coming

BARBARA GERTEN, Merck KGaA, Darmstadt, Germany

Recently new published ISO-CEN standardized methods are mostly using culture media and reagents.

During development of new or revision of existing standards it is of high importance to consider different aspects for the used culture media and reagents. This includes a standardized description of the composition, preparation and performance testing of all culture media and reagents. It enables the user to prepare these from the single components but also the usage of prepared dehydrated complete media/reagents or ready-to-use media/reagents provided by manufacturers. During the development and validation of the new EN ISO standards, it was checked that the method works using media/reagents made up in the laboratory from individual ingredients as well as from several commercial media.

Performance testing requirements (performance criteria, control method and targets) are included now in each EN ISO standard, following a standardized layout based on the methods and principles given by EN ISO 11133. Test strains strains included are only from the WDCM (World Data Centre for Microorganisms) catalogue. It enables the usage of strains from National Culture Collections providing the strains included in the WDCM catalogue.

During the process of revision or development of EN ISO standards and their subsequent validation a close inclusion of commercial available ingredients, culture media and reagents ensuring a worldwide possibility for implementation in the user's laboratories.

It is the objective to present also the aspects for a manufacturer to be considered, especially for the release of these media/reagents aligned with the new standards under ISO 17025 accreditation.

Implementation of Reference Methods in Food Industry – Impact on Alternative Methods

DAVID TOMÁS FORNÉS, Nestlé, Lausanne, Switzerland

According to European Regulation EC 2073/2005 on microbiological criteria for foodstuffs, food business operators shall perform testing against the microbiological criteria when validating or verifying the correct functioning of the procedures based on HACCP and Good Hygiene Practice.

S4 Microbiological Reference Methods, What's New, What's Coming?

Reference analytical methods are a key tool to guarantee food safety and quality in the food chain. Standard methods are widely used by official, industrial and third party laboratories as part of official and quality controls. These standards are also required to validate alternative methods in comparison to them, allowing to obtain equivalent results between food industry operators and between official controls.

Impact of the recent publication of several ISO-CEN standardized methods, including validation studies, should be considered at different levels of the food chain, including regulators, small and big size industry companies, diagnostic companies and third party laboratories.

The objective of this session is to present the new ISO-CEN Standards recently published and give examples about the changes, improvement, applications and impact in the food sector.

New Reference Standard Methods Developed, Validated and Standardized by CEN and ISO

ALEXANDRE LECLERCQ, Institut Pasteur, Paris, France

Reference methods in food microbiology, for the analysis of food, animal feeding stuff, samples from primary production and food processing and handling, are standardized at the international level by ISO TC34/SC9

The EN/ISO analytical methods included in the regulation shall be applied as the reference methods. However, food business operators should have the possibility to use alternative analytical methods other than the reference methods, if the alternative methods are validated against the reference method in accordance with the protocol set out in EN ISO 16140.

The new ISO/EN reference methods, developed and validated by a multiple stakeholder collaboration and consensus, have been optimized and, in some cases simplified allowing a reduction of workload, costs and time to results compared with previous versions, without compromising the validity of the results.

Performance criteria included in the new standards bring also valuable information. Reference values (e.g., LOD, repeatability, reproducibility) are now available to compare against when the method is implemented in a quality control laboratory.

Last but not least, new methods have a direct impact on alternative methods validated against these standards. When changes in the standard are considered major with a significant effect on method performance, re-validation and new verification should be conducted to guarantee results from the samples analysed are still valid.

and promoting the importance of hygienic design through the provision of training, guidance, certification, networks and expert advice. These activities will continue to increase knowledge and awareness with food manufacturers, suppliers and other organizations active in the whole food chain.

Hygienic Design: A Food Manufacturer's Perspective

LAURENCE BLAYO, Nestlé, Lausanne, Switzerland

One-hundred eighty deaths in South Africa due to *Listeria monocytogenes*, more than 7000 tons of products from a French manufacturer recalled due to *Salmonella*. Recent outbreaks and the accompanying media headlines remind us that, despite great strides forward in the area of microbiological food safety, the burden of foodborne disease persists!

To prevent food contamination, food industries need to have in place strong food safety management systems that must include hygienic design of facilities and equipment. Although hygienic design principles are not "rocket science" and sound more like "common sense concepts", reality shows that hygienic design remains a major weakness in food production and is too often underestimated by food manufacturers and equipment suppliers.

This talk will address challenges of food manufacturers with regards to hygienic design; requirements of food manufacturers to equipment suppliers and how "common sense concepts" of hygienic design combined with state of the art technologies (e.g., WGS, Biofilm monitoring technology) contribute to keep food safe and improve public health.

Hygienic Design – An Equipment Manufacturer's Perspective

DEB SMITH, UK:IE EHEDG & Vikan, Swindon, United Kingdom

The use of equipment that is easy to clean and maintain, i.e., hygienically designed, and made of food contact compliant materials, is fundamental to ensuring food safety. It is also a requirement in law and of global food safety standards. Surprising then that hygienic design is often unknown, or overlooked, by those that purchase and manufacture food industry equipment. This presentation provides valuable information on the requirements for food industry equipment hygienic design, as stated by regulators and food safety management auditing systems, and the guidance provided by bodies such as EHEDG and 3A, with the aim of raising awareness of such and assisting the food industry in procurement of appropriate equipment.

S5 Maximising Food Safety through Good Hygienic Design

The use of equipment that is easy to clean and maintain, i.e., hygienically designed, is fundamental to ensuring food safety. It is also a requirement in law and of Global food safety standards. Surprising then that hygienic design is often unknown, or overlooked, by those that purchase and manufacture food industry equipment. This seminar brings together valuable information from the food industry, regulators, food safety management auditing systems, guidance providers, and equipment manufacturers, with the aim of raising awareness of hygienic design and assisting the food industry in its application.

Hygienic Design – A Regulatory, Standards and Guidance Perspective

PATRICK WOUTERS, EHEDG & Cargill, Amsterdam, The Netherlands

In the food industry, the relationship of food manufacturers with their consumers is built on trust. This trust is established over time but can be lost in a moment. Each food safety incident erodes consumer confidence in the food supply and, thus, our complete industry. Therefore, we must work together to learn from incidents and share these learnings to advance our programs and our designs.

One area that plays a critical role in safe food production is the hygienic or sanitary design of the food manufacturing facility, the building, the installed processes, equipment and utilities. The hygienic design is foundational to HACCP as a pre-requisite program and supports the success of the overall food safety management program. The discussion regarding food safety programs often focuses on hazard management and this is critical if a hazard is unable to be removed. However, an even more successful approach is to eliminate the hazard by successful design.

This session will provide an overview of regulatory requirements, existing standards, guidelines and organizations active in the area of hygienic design. There are specific legal requirements with regard to food contact materials and hygienic design of the equipment. However, awareness and especially the application of these regulations is often poor or difficult.

Hygienic auditing will be addressed and the role of hygienic design requirements in Global Food Safety Initiative (GFSI) benchmarked certification programs. GFSI recognized food safety management schemes, require the use of hygienically designed equipment and premises, but are these requirements clear enough and is the food industry aware of these requirements and of what hygienic design is? The European Hygienic Engineering and Design Group (EHEDG) has for many years been active in defining

S6 How NGS Technologies Unravel Our Understanding of Food and Food Microbiology

Now almost a decade old, the term next-generation sequencing (NGS) remains the popular way to describe very-high-throughput sequencing technologies that read millions of small fragments of DNA in parallel, during a single instrument run. In recent years, sequencing technology has made dramatic steps forward with an explosion of new methodologies, instruments and advanced bioinformatic tools to ensure and cope with data storage, analyses, and management solutions. But what about the use of NGS in food? Which applications for food producers nowadays?

Rather than exploring these fascinating cutting-edge technologies, this session aims at illustrating several applications of NGS in order to improve outreach and demonstrate its huge potential in food.

The first talk will deal with food authenticity using DNA barcoding, which offers an alternative and feasible taxonomic toolbox for rapid and robust species identification. If implemented it is a fast and cheap way to monitor biodiversity within of complex mixture. It may be used to detect high valuable spices such as saffron or elucidate the composition of spice blends... or to detect mislabelling or food cross-contamination as recently highlighted by the European horse meat scandal.

The other two talks will deal with taxonomic and functional microbial diversity. Indeed NGS is revolutionizing our understanding of food microbiology. It is resolving previously intractable questions concerning the stability, resilience, and function of a given microbial community in food or within the industrial plant. While patterns of diversity and abundance are well characterized for microbial hazards, this knowledge lacks for technological, spoilage or residential microflora where subtle shift may have a huge impact on the food quality. The implementation of NGS in the food factory will surely allow us to move from monitoring indicator species to take into account the whole biodiversity. Are we ready to sequence the food factory?

The Use of NGS to Ensure Food Authenticity and Avoid Food Fraud

VALENTINA PARACCHINI, European Commission
Joint Research Centre, Ispra, Italy

The development of an efficient food traceability framework is crucial for the monitoring of potential substitution fraud across the food chain, and therefore for consumer protection.

Recent scientific advances, particularly in the fields of genetics and genomics, have led to the development of novel and improved technologies, and efforts are under way to harness their potential for the species identification. DNA-based approaches, like DNA barcoding (using either nuclear or mitochondrial DNA), are most commonly used in this case, due to their high specificity and to the high resilience of the target molecules to food processing techniques.

The main caveat of DNA barcoding is the traditional use of Sanger sequencing, which does not produce a useful output when the sequencing reaction contains a mixture of different DNAs. A possible solution is the use of Next Generation Sequencing technologies, that can sequence hundreds of thousands DNA strands in parallel, allowing characterisation of multiple species in mixed and processed samples.

In this context, the JRC recently published the identification of sets of DNA barcodes candidates in the nuclear genome sequences of fishes, using available whole genome sequences. Specific primers pairs were shown to efficiently identify fish species of different families, like flatfishes or gadoids, a choice driven by an analysis on the frequency of documented fraud cases. The method proposed could complement existing fish identification strategies in establishing an efficient framework to detect and prevent frauds along the food chain.

The Use of NGS to Apprehend the Biodiversity of Microbial Communities in Food and Food-related Surfaces

BIRGITTE MOEN, Nofima, Oslo, Norway

Food undergoes many processing steps before reaching the consumer and knowledge of the microbiota throughout this process is important to control the safety and quality of the final product. The majority of the bacteria present in the products and in the food processing environments are non-pathogenic. Most food processors do not know the identity of the non-pathogenic, residential bacteria found in their production plant. Increased knowledge is important to understanding how these bacteria can affect food quality and food safety.

In the last years, the development of NGS techniques has enabled researchers with a broader and deeper understanding of the biodiversity of microorganisms in food and food environments. 16S rDNA sequencing, metagenomics and metatranscriptomics are basic sequencing strategies used in the taxonomic identification and characterization of food-related microbiomes. To date, most food-related investigations have used a 16S rDNA approach, but recently metagenomic approaches have resulted in improved understanding of a microbiome by providing a species-level/strain-level characterization.

In this talk, I will touch upon some of the different sequencing approaches available and present work we have done at Nofima using NGS in shelf-life studies and food-related surface studies.

Use of NGS to Investigate the Biodiversity and Activity of Cheese Microbial Communities

CHRISTOPHE MONNET, INRA, Thiverval-Grignon, France

Microbial communities from the surface and interior of cheeses include a large variety of bacteria, yeasts and moulds, whose activity contribute to the development of the typical organoleptic properties (flavour, texture and colour) and also limit the growth of spoilage microorganisms or of pathogens. For a long time, investigation of cheese microbial communities was hampered by a lack of methods that can be applied *in situ* during cheese manufacturing. However, during the last years, the development of Next Generation Sequencing (NGS) technologies provided new perspectives. One important application of NGS for cheese is targeted metagenomics, a technique based on sequencing of a locus that provides phylogenetic information, which makes it possible to get a good picture of the phylogenetic composition of the community. This composition can also be assessed by shotgun metagenomics, in which is the whole DNA of the community is sequenced. Shotgun metagenomics is also useful for the study of the functional potential of the microbial communities. Another interesting application of NGS for cheese is the sequencing of the metatranscriptome, which provides information about the activity of cheese microorganisms. Examples concerning metagenomics and transcriptomics applications for cheesemaking processes will be presented.

S7 Importance of Microbiological Criteria and Statistical Underpinning of Sampling and Testing for Food Safety Assurance

Assuring food safety poses ever increasing challenges to various public and private stakeholders along the food supply chain from primary production to consumption. Sampling and testing of food products as well as food production, processing and handling environments plays a key role in food safety assurance.

Erroneously, the importance is often considered to be so big that it is believed that sampling and testing alone would be sufficient to assure food safety. However, this symposium will provide important insights in the role of sampling and testing among the many other control measures that, in an orchestrated way, are needed to assure food safety day by day in the many different food operations and outlets that exist in the world.

The experts contributing to the symposium are members of the International Commission on Microbiological Specifications for Foods (ICMSF). They will share their expertise concerning the regulatory views on microbiological criteria, the statistical basis of sound sampling and testing approaches, as well as sampling and testing recommended by ICMSF for different food commodities.

International Perspective on the Role of Microbiological Sampling and Testing in Food Safety Assurance

LEON GORRIS, Unilever R&D Vlaardingen, Vlaardingen, The Netherlands

Assuring food safety poses ever increasing challenges to various public and private stakeholders along the food supply chain from primary production to consumption. Sampling and testing of food products as well as food production, processing and handling environments plays a key role in food safety assurance.

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The session will start with an international perspective on the role of microbiological sampling and testing in food safety assurance. It will discuss the role of Codex Alimentarius in designing useful standards and guidelines for food safety management within the risk-based framework referred to as Risk Analysis and how local and regional governments consider (or not) Codex output when establishing food safety regulations for their jurisdiction.

The Science and Statistics Underlying Sound Microbiological Sampling and Testing Approaches

MARCEL ZWIETERING, Wageningen University, Wageningen, The Netherlands

The importance of food safety is at an all-time high and it remains an important priority for many stakeholders around the world, including food enterprises, regulatory agencies and consumers. To manage and assess food safety risks, a variety of methods and tools are available, including microbiological testing of foods.

Microbiological testing can be applied at all stages of food production from the farm to manufacturing facilities to the retail level. When using microbiological testing to assess the safety of a food, it is important to select the appropriate test method and sampling plan, based on good understanding of the underlying statistics as well as knowledge of the limitations of such testing.

In the session, the statistics underlying useful testing will be elaborated on, providing examples of how microbiological criteria are developed and sampling plan performance is assessed.

Sampling by nature is a stochastic process. However, uncertainty regarding results is made even greater by the uneven distribution of microorganisms in a batch of food. Different batch contamination scenarios are illustrated: a homogeneous batch and a heterogeneous batch with high- or low-level contamination.

ICMSF Guidance on Microbiological Sampling and Testing for Key Commodities

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

The International Commission on Microbiological Specifications for Foods (ICMSF) was formed in 1962 through the action of the International Committee on Food Microbiology and Hygiene, a committee of the International Union of Microbiological Societies (IUMS). Through the IUMS, the ICMSF is linked to the International Union of Biological Societies (IUBS) and to the World Health Organization (WHO) of the United Nations.

The ICMSF provides basic scientific information through extensive study and makes recommendations without prejudice based on that information. Results of the studies are published as books, discussion documents or refereed papers (see Publications at www.icmsf.org). ICMSF recommendations have no official status – official promulgation of such recommendations are the province of governments and international agencies.

At an early stage, the Commission recognized that no sampling plan can ensure the absence of a pathogen in food. Testing foods at ports of entry, or elsewhere in the food chain, cannot guarantee food safety. However, testing can play important roles within the wider suite of measures and systems underlying food safety assurance.

Over the years, ICMSF has compiled practical guidance on appropriate testing of food processing environments, processing lines, shelf life and finished product to enhance the safety and microbiological quality of the food supply. Examples will be given in this session.

S8 Biofilms and Environmental Monitoring

Food environments provide ideal conditions for bacterial growth. Once bacteria colonize the factory environment or equipment, they can grow and form a biofilm, allowing them to survive for long periods of time. Bacterial biofilms in food processing environments are normally made up of multiple bacterial species. Pathogenic organisms like *Salmonella* or *Listeria monocytogenes* can be part of the biofilm community. The biofilm provides them with increased resistance to desiccation and common sanitizers used in the food industry, which makes them a lot harder to remove during routine cleaning. Having a strong environmental monitoring and sanitation programs in place to identify and control biofilms and pathogens in the processing environment is key to food safety. These programs will seek to find and eliminate the presence of pathogenic organisms in the environment before they contaminate the food product.

This symposium combines academic knowledge with industry examples on environmental monitoring and sanitation strategies to better address the challenge of biofilms in processing equipment and factory environments.

Biofilms in the Food Industry

ANNETTE FAGERLUND, Nofima, Norwegian Institute of Food, Fisheries and Aquaculture Research, Ås, Norway

The microbiota found in the food processing environment after cleaning and disinfection may include food spoilage bacteria and foodborne pathogens. These represent a potential major source of bacteria which can compromise food quality and food safety. The resident background microflora is recognized to play an important role with respect to protecting and sheltering pathogenic strains within food processing environments. One of the pathogens regularly encountered in food processing environments is *Listeria monocytogenes*. The transfer of *L. monocytogenes* from food contact surfaces such as conveyor belts to processed food products has been documented and, in some cases, shown to result in outbreaks of listeriosis. The use of chemical disinfectants in food processing environments is usually based on their efficacy in tests performed with planktonic bacteria. However, in both natural and industrial environments, bacteria often grow as biofilms, which are complex and structured microbial communities encased in a self-produced protective extracellular matrix composed of polysaccharides, proteins, and/or extracellular DNA. The formation of biofilms is important for microbial survival in the food industry, and cells in biofilms are typically more tolerant towards biocides and other antimicrobial agents compared with that of their planktonic counterparts. In recent work, we show that *L. monocytogenes* survives in complex biofilms after exposure to cleaning and disinfection. Biofilms established preferentially in surface irregularities of conveyor belts, potentially constituting harborage sites for persistent contamination.

Biofilms, Pathogenic Bacteria and Environmental Monitoring – An Industry Perspective

PETER MCCLURE, Mondelez International, Birmingham, United Kingdom

Foodborne diseases remain a serious concern for international and national public health agencies around the world, affecting both developed and developing countries. These continue to drive development of more robust food safety programs, reinforcing the importance of effective control procedures including pre-requisite programs that are a key cornerstone underpinning HACCP. Collectively, these are fundamental to successful implementation of effective controls and prevention of build-up of contamination sources in manufacturing environments. Several foodborne outbreaks associated with commercially manufactured foods reveal that weaknesses in Good Hygienic Practice implementation can lead to post-process contamination with pathogens. One of the key targets for good hygienic and manufacturing practices are biofilms, since these can harbour pathogenic microorganisms and protect them from

cleaning and disinfection regimes used to eliminate them in manufacturing environments. Biofilms can cause serious hygiene problems on processing equipment and are also a concern for environmental surfaces such as floors and walls, where they can provide indirect sources of contamination by vectors such as air, people, equipment and cleaning systems. In this presentation, some of the aspects related to biofilm control are described, together with the important role of environmental monitoring in food manufacturing facilities. The presentation aims to provide a summary of the key considerations where environmental contamination can result in unsafe foods, including establishment of a routine sampling program to assess control of the environment for verification of processing environment controls, use of appropriate cleaning and disinfecting procedures that are targeted toward the pathogens of concern for the particular food and process, and effective follow up investigations when pathogens are detected.

Importance of Sanitation Programs and Hygienic Design to Control Biofilms

ROGER SCHEFFLER, Commercial Food Sanitation, Amsterdam, The Netherlands

Cleaning of food processing facilities is key for safe food manufacturing and should be seen as the first step in the food supply chain. This presentation emphasizes on the importance of sanitation as well as on hygienic design for food processing equipment. Hygienic Design is a key tool to prevent food residues to be staying and harboring in niches of the equipment. These programs work hand in hand with programs like PIC and PEC, Periodic Infrastructure Cleaning and Periodic Equipment Cleaning. Practice oriented activities will be outlined on how to execute a proper PIC and PEC program in order to prevent biofilms and to contribute to safe food processing.

Application of Metataxonomics and Metagenomics in Fermented Sausages

LUCA COCOLIN, University of Torino-DISAFA, Grugliasco, Italy

Fermented sausages are the result of a complex microbial transformation in which different bacteria, yeasts and filamentous fungi metabolize meat proteins and fats and other ingredients included in the recipe. Several microbial activities are key for the correct manufacturing such as acidification, proteolysis and lipolysis. Microorganisms also help the dehydration process by creating micropores on the casing, allowing for a more efficient water loss. Through the microbial activity, foodborne pathogens eventually coming from the raw materials and the processing environment are killed and the product at the end of the manufacturing is safe. Lastly, the microbiota of fermented sausages helps the development of a product with specific quality characteristics.

From the end of the '90s, the study of microbial ecology in complex ecosystems has taken a new direction in which the potential of molecular biology methods are fully exploited. New approaches based on culture-independent methods have been developed and now to study microbial ecology and interactions is not anymore necessary to cultivate, isolate and identify the microorganisms. Pattern-based techniques, such as DGGE, have recently been replaced by more efficient sequence-based methods with which is not only possible to understand who is there (metataxonomics), but also what they are doing (metagenomics and metatranscriptomics).

In this talk, we will describe the application of next generation sequencing techniques to investigate the microbial ecology of fermented sausages, looking at both the ecology and the functions. Specifically, a more detailed picture on how the microorganisms contribute to the development of the organoleptic was obtained comparing fermented sausages inoculated or not with starter cultures.

Innovative Approaches to Reduce Fungal Spoilage in Dairy Foods

JEROME MOUNIER, University of Brest, Brest, France

Fungi are commonly involved in the spoilage of dairy products which may provide a favorable niche for their growth. They are responsible for visible or non-visible defects, such as off-odor and -flavor and lead to significant food waste and food losses. In the dairy industry, traditional hurdle technologies are implemented and combined to prevent and control fungal spoilage. Among emerging control methods, biopreservation is gaining more and more attention. Biopreservation is not a new concept as it has been used for thousands of years in fermented foods. Also called biocontrol, it refers to the extension of food shelflife and increases in food safety using natural or added microbiota and/or their antimicrobial compounds. Food bioprotective cultures can thus be defined as food-grade bacterial or fungal strains that have been specifically selected for their antimicrobial properties. This presentation will focus on different aspects concerning the development of antifungal cultures in dairy foods from in vitro screening to pilot scale applications as well as the understanding of their mechanism of action.

Bioprotection in Vegetable Foods

ANTONIO GALVEZ, University of Jaen, Jaen, Spain

Lactic acid bacteria (LAB) may produce different antimicrobial substances, including metabolites and antimicrobial peptides (also known as bacteriocins). The antimicrobial activity of LAB can be exploited for biopreservation of different types of foods by either using bioprotective cultures or partially-purified bacteriocin preparations. Vegetable foods are prone to contamination with spoilage and human toxinogenic or pathogenic bacteria. LAB and their bacteriocins have been exploited in vegetable foods with several purposes such as decreasing the risk for proliferation of spoilage bacteria, displacing biogenic amine producers, or inhibiting foodborne pathogens. In addition to

S9 New Approaches for Safety and Quality of Fermented Foods

Fermentation is one of the most ancient technique man has used to extend the shelf life of highly perishable raw materials (e.g., meat and milk) and to improve their microbiological safety. Through the metabolic activity of beneficial microorganisms, the spoilage and pathogenic microbiota are inhibited allowing for the final product to be microbiologically stable and safe. While in the past, it was believed that the stabilization process was mainly associated with the acidification ability of some bacteria (namely Lactic Acid Bacteria, LAB), nowadays it is recognized that other mechanisms are involved in the competition process, such as production of antimicrobial compounds (bacteriocins), competition towards nutrients and others.

In the last decades, food fermentations have attracted a lot of attention from researchers due to the availability of powerful tools able to culture-independently profile the microbial ecology of these complex ecosystems, opening up new possibilities to impact the safety and the quality of the final products.

The symposium aims at showcasing advancements in the field of fermented foods taking into consideration three product categories, namely meat, dairy and vegetables. The application of metataxonomics and metagenomics in fermented sausages will be introduced, giving emphasis on the opportunities to improve and advance the knowledge on microbial interactions in this complex ecosystem. Then, the use of bioprotective agents to reduce fungal spoilage in dairy foods will be presented with a special regard on the integrated approaches required for their development, i.e., from in vitro screening to in situ applications and the understanding of their action mechanisms. Finally, the impact of bacteriocins and LAB as part of hurdle technology on the microbiota of vegetable foods will be discussed.

The symposium represents an interesting opportunity for students, industries and professionals for an update on the possible approaches to improve the safety and quality of fermented foods.

the above-mentioned effects, LAB and their bacteriocins may also influence the whole bacterial communities of vegetable foods as well as the dynamics of bacterial populations during food storage. Application of high-throughput sequencing technologies can provide novel information on the main bacterial groups found in food systems (including vegetable foods) and how they are affected by food processing and preservation methods. This may be important to better understand the key factors to improve food preservation and product shelf life, but at the same time may also reveal other points of interest related with food safety such as the behavior of emerging human pathogens and reservoirs of antimicrobial resistance in bio-preserved foods.

S10 Validation and Verification – Successes, Pitfalls and Disasters

In the modern age in food safety, validation and verification processes are critical components of a good food safety plan and are complementary to each other. However, food manufacturers often confuse the meaning and function between the two terms. Most food products undergo a kill step of some form at the point of production and yet, most of these control points lack scientific validation. Similarly, some are unclear of details required in validation reports that could result in regulatory agencies not accepting validation reports. Once validation is completed, a food manufacturer needs to establish verification procedures to ensure that the implemented processes are effectively and consistently carried out and a confirmation that the food manufacturer is doing what is intended and that it is effective and activities are properly documented.

Likewise in testing laboratories, a fundamental requirement is to ensure that test methods actually work, and will detect/enumerate the organisms of concern. To achieve this we rely on method manufacturers to “validate” methods. But does this give the laboratory enough confidence to use the method? Should they undertake their own verification that the method works in their hands? What should they do? Does it depend on the extent of the validation process, or the range of food that they test? All good questions that will be reviewed within this session.

The symposium will highlight the concepts of validation and verification, their differences and how various food processing technologies and microbiological analytical methods are validated and verified to ensure they serve their intent, the parameters used in validation studies and record keeping.

Validation and Verification – Concept and Practice

PURNENDU VASAVADA, University of Wisconsin-River Falls, River Falls, WI, USA

Validation and verification are essential activities for an effective food safety system. The purpose of validation is to provide objective evidence that food safety plan and preventive controls employed to control the hazards associated with a specific product and process do have a scientific basis and can, in fact, control the significant hazards that need to be controlled to assure food safety. Verification, on the other hand, is an ongoing process designed to provide evidence that the Food Safety Plan is being properly implemented and operating as intended. Validation demonstrates that following the plan will actually control the identified hazards and is usually done before implementation of the Food Safety Plan. While using scientific principles and data from the literature or expert opinion may have been adequate in the past for validation strategies, conducting in-plant validation may be necessary for developing validation to adequately address food safety. Verification is an important component of supply-chain, sanitation, allergen and process preventive controls. Verification activities and procedures vary depending on the food, the facility, hazards requiring preventive controls, the processes used and the nature of the preventive control, etc. and may include periodic in-process or end product testing, internal audits, and third-party audits. This presentation is designed to address validation and verification, in the context of contemporary food safety management systems.

Non-Thermal and Thermal Process Validation

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

Food manufacturers use either a combination of thermal-based and non-thermal-based processing technologies or both to manufacture food and to ensure the safety and quality of their products. These processes will need, unless there is a demonstrated safe harbor, to prove efficacy and reduce/eliminate the identified risk. There have been discussions on validation design, who and where to conduct validation studies, what constitutes a scientifically valid study and what detail is required in a validation report. Validation of processes can take on different forms and can be conducted on-site or off-site using either pilot scale or actual manufacturing equipment. Most thermal-based technologies have a long history of safe use but “modern” food matrices as a result of consumer desires may require re-validation. Similarly, validating non-thermal-based processes can pose challenges in validation study design because many food regulations are currently based of thermal processes e.g., processes related to canning and juice products. The presentation will provide a perspective on how process validation studies are conducted and how processing conditions, bacterial strain selection, risk identification and potential research gaps are addressed.

Microbiological Test Methods: Validation and Verification, What Does It Mean?

ROY BETTS, Campden BRI, Gloucestershire, United Kingdom

Microbiologists have seen over many years, the development of a range of new test methods. Designed to replace more traditional methods, manufacturers of these methods will claim all sorts of advantages over agar/broth based traditional methods which reside in national and international standards. Terms such as faster results, more accurate, sensitive, more automated, higher throughput will be encountered. However, where food quality and safety is concerned, what the microbiologist needs to know is—will they give me the right results. Now one may ask for a definition of “right”, but to many, it means if the organism is there, will I find it.

For years we have spoken about method evaluation, validation, verification. We have looked at third-party validation and in-house validation. Definitions of what we mean have become confused and misused.

This presentation will consider the current thinking in method validation, what is it and what does it do, and method verification within user laboratories. It will look into existing and forthcoming standards and explain what to look for in methods that get it right.

S11 Biological Variability in Thermal Processing: Impact for Process Control and Validation – What You Need to Know about Microbiological Variability for Food Quality and Safety Control

Microbial limits are used to evaluate and validate processes that aim to control food safety and stability. However, biology is highly variable; microorganisms, raw materials, and humans are variable and diverse. And this variability is challenging the ‘line in the sand’ of our microbial limits. When we would control safety and spoilage for the average case, half of the times it would go wrong. When we aim to control at the 99 percentile all our process parameters, we might over-process. Understanding, quantifying and determining the impact of variability is needed to control spoilage and food safety. In this symposium, we will present experimental modelling and risk assessment aspects, including the points of view of academia, government and the industry.

Impact of Natural Diversity in Heat Resistance of Bacteria and Bacterial Spores on Food Safety and Quality

MARCEL ZWIETERING, Wageningen University, Wageningen, The Netherlands

Heat treatments are widely used in food processing often with the aim of reducing or eliminating spoilage microorganisms and pathogens in food products. The efficacy of applying heat to control microorganisms is challenged by the natural diversity of microorganisms with respect to their heat robustness. This presentation gives an overview of the variations in heat resistances of various species and strains, and describes modeling approaches to quantify heat robustness. It particularly addresses the relevance and impact of the natural diversity of microorganisms when assessing heat inactivation. This comparison of heat resistances of microorganisms facilitates the evaluation of which (groups of) organisms might be troublesome in a production process in which heat treatment is critical to reducing the microbial contaminants, and also allows one-tuning of the process parameters. Various sources of microbiological variability are discussed and compared for a range of species, including spore-forming and non-spore-forming pathogens and spoilage organisms. This benchmarking of variability factors gives crucial information about the most important factors that should be included in risk assessments to realistically predict heat inactivation of bacteria and spores as part of the measures for controlling shelf life and safety of food products. Furthermore an approach is presented to handle the variation to be included in the targeted reduction.

Combining Challenge Tests and Predictive Microbiology in Thermal Process Validations of Low-moisture Food

MARIEM ELLOUZE, Nestlé, Lausanne, Switzerland

Validating pathogen inactivation processes is a regulatory requirement which is even more challenging in low moisture foods, where the fat content, the heterogeneity and several interactions between the product water activity, temperature, and process humidity play a major role. In fact, sparse data and tools are available to enable robust validation of such processes.

This presentation discusses the use of challenge testing, surrogates and predictive microbiology to perform such validations for *Salmonella* in low moisture foods at the lab scale and at the industrial scale. The effects of the variability in microbial behavior and in the product and process characteristics are discussed as well as practical ways to assess the impact of such variabilities with a focus on the opportunities, limitations and complementarities offered by the challenge testing and the modelling approaches.

Impact of Variability in Regulation and Inspection

Mickey Parish, U.S. Food and Drug Administration, Washington, D.C., USA

Process validations are commonly reviewed by regulators to ascertain compliance by a firm with a specific regulation, or to determine the usefulness of a firm's proposal to recondition contaminated food lots. Process variability may be impacted by a variety of factors such as pH, buffering capacity, water activity, food composition (e.g., fat content), seasonality, and geography, as well as temperature, relative humidity, and time, among others. Validation studies need to account for variability that can impact thermal process efficacy, and thermal processes should not be extended outside the boundaries of the parameters validated. When reviewing process validations, the regulator must evaluate how the uncertainty that exists in observed results impacts public health and a firm's compliance with regulatory requirements. This presentation will provide a broad overview of how variability impacts a regulatory process review, and will provide an example to illustrate regulatory concerns.

S12 Integrating Microbial Adaptive Trait in Food Safety: Added Value of Biomarkers

Recent scientific and technological advances open new perspectives for an in-depth understanding of microbial cellular mechanisms. In the field of food safety, the implementation of the bacterial adaptive traits through the identification and quantification of the expression of microbial gene markers or biomarkers would enable to refine microbiological exposure assessment. Nevertheless, up to now, the integration of such data remains a scientific challenge. Gene transcription allows cells to adapt rapidly to environmental changes. Thus gene expression profiling, by delivering genetic and regulatory information, is a powerful means of identifying bacterial biomarkers. In this context, the symposium will focus on the use of mRNA as biomarkers to account for bacterial fitness.

First, biomarker discovery, quantification or validation, which it is routinely used for improved diagnostics and therapeutics in the medical domain, will be described. Some examples which aims at integrating gene expression measurements and clinical parameters will be used to illustrate these aspects. A special attention will be paid to the critical parameters such as accuracy, sensitivity, specificity for the successful use of RNA as biomarkers.

Then, results of a French collaborative project which aims at evaluating and predicting the capacity of bacteria to survive stress or to grow after a preliminary stress encountered in processing steps in the dairy and poultry food chains, by using biomarkers will be presented. More precisely, work on *Listeria monocytogenes*, *Campylobacter jejuni* and *Bacillus cereus* will be presented. Special attention to the methodological techniques to obtain reliable and biological-diversity-representative phenotypic and transcriptomic data, and to interpret them afterwards, will be issued.

RNA Used as Biomarkers: Requirements, Validation and Challenges in Clinical Applications

Eddy Van Collenburg, Bio-Rad Laboratories, Eindhoven, The Netherlands

Biomarkers are of increasingly high importance in medicine, particularly in the frame of 'personalized medicine'. In food safety and quality issues, biomarkers open the avenue to take strain diversity or physiological variability into consideration in microbiological risk assessment (MRA). Biomarkers are characteristics that are objectively measured and evaluated as indicators of biological processes and include nucleic acid-based biomarkers, proteins, lipids and other small molecules. Compared with DNA biomarkers, RNA biomarkers have the advantage of providing dynamic insights into cellular states. Compared with protein biomarkers, RNA biomarkers have more sensitivity and specificity since PCR enables traces of RNA sequences to be amplified.

While quantitative PCR (qPCR) has been the gold standard for nucleic acid quantification, some applications are more demanding in sensitivity or precision. Digital PCR (dPCR) is a recent technology that has become commercially available since 2011 and which allows sensitive nucleic acid detection and quantification. Thus rare event detection (higher sensitivity) and gene copy number determination (higher precision) could benefit from dPCR. The great advantage of dPCR is that no specific calibrator samples are required to determine the actual copy number. Indeed, the absolute quantification relies on two elements: positive and negative fluorescence data from the sample droplets and data fitting to a Poisson distribution. Nevertheless, such as for qPCR, the ability of dPCR to perform robust meaningful experiments requires careful design and adequate controls. All these aspects will be presented and discussed in this presentation.

Use of Biomarkers to Refine Microbiological Exposure Assessment Associated with *Listeria monocytogenes*, *Bacillus cereus* and *Campylobacter jejuni* in the Dairy and Poultry Food Chains

BENJAMIN DUQUÉ, UMR1014 SECALIM, INRA, Oniris, Nantes, France

During their entire shelf life, food products have to be safe and do not have to represent a risk to human health. Quantitative Microbiological Exposure Assessment (QMEA) describes the dynamics of pathogens propagation until the consumer's plate by estimating the pathogen contamination level at each processing step through inactivation or growth predictive modelling. So far, these models have been mainly built considering that pathogen behaviour is not dependent on the conditions previously encountered during the food process. However, minimizing the impact of bacterial adaptation may lead to an under- or over-estimation of growth or survival capabilities.

In a French collaborative project, predicting the ability of bacteria to survive or grow after a preliminary stress, based upon gene expression biomarkers, was challenged. This was applied in parallel to the dairy and the poultry food chains. *Bacillus cereus* and *Listeria monocytogenes* were selected as relevant pathogens in the dairy food chain, whereas *Campylobacter jejuni* was chosen for its high prevalence in the poultry food chain.

In the present talk, the different steps associated with the identification of gene expression biomarkers to refine exposure assessment model, will be exposed. For each bacterial species, the common methodology of selection of representative bacterial strains and study of their phenotypic behaviour following stress will be presented. The comparison of gene expression with phenotypic behaviour of pathogens preliminary stressed will lead ultimately to construct a quantitative model based on biomarkers.

Reliable Identification of mRNA as Biomarkers: Workflow of the Quality Assurance Procedure

LAMIA BELKADI, LUBEM UBO University - UMT14.01SPORE RISK, Quimper, France

The recent development of "omics" technologies has largely influenced the way to study biological responses of microorganisms. Transcriptomics data from foodborne pathogens under different environmental stresses have been used to identify biological markers or biomarkers related to specific resistance characteristics of the pathogens.

Real-time PCR (qPCR) and reverse transcription qPCR (RT-qPCR) are recognized as the methods of choice to analyze specific genes and their expression patterns. They offer many practical and sensitivity advantages. However, lacks in the reporting of RT-qPCR quality controls may have an impact in result interpretation, and it is more difficult for reviewers to evaluate the work or for investigators to reproduce the experiment. Therefore, appropriate quality procedures are needed to guarantee the reliability of RT-qPCR data acquisition and confirm the robustness of the quantitative results. Thus, the data generated will be integrated to build robust mathematical models to predict microbial behavior in food industry process. Furthermore, this approach will allow the use of data in decision support related to food safety and quality issues.

Throughout an example of bacterial behavior biomarker of *Listeria monocytogenes*, *Bacillus cereus*, and *Campylobacter jejuni* study, the strategy to select relevant biomarkers, as well as the step-by-step RT- qPCR workflow including numerous necessary quality controls, will be presented and discussed.

S13 Allergen Control – From Problem to Solution

The control of allergens within food production facilities is essential to ensure food safety. It is also a requirement in law and of global food safety standards. This seminar brings together valuable information from the Anaphylaxis Campaign, regulators, audit bodies, guidance providers, and food industry equipment manufacturers, with the aim of raising awareness of the impact allergens, can have on susceptible consumers, and advice on how to minimise this impact through the application of good hygienic design and appropriate cross-contamination control strategies.

The Problem – Allergens as a Food Safety Hazard

LYNNE REGENT, Anaphylaxis Campaign, Farnborough, United Kingdom

Food allergies and intolerances are life changing and their prevalence is increasing. This presentation provides prevalence data and shows what the physical impact of severe allergy can be. It will also describe how severe allergies directly affect the everyday lives of individuals and their families and carers. It will give details of the regulatory and standards requirements related to allergen control and discuss the significant implications of not meeting these requirements.

The Solution I – Effective Strategies for Minimising Allergen Cross-contamination

DEB SMITH, UK:IE EHEDG & Vikan, Swindon, United Kingdom

The control of allergens within food production facilities is essential to ensure food safety. It is also a requirement in law and of global food safety standards. This presentation provides study data on the spread of contamination and advice on practical measures to assist in the reduction of allergen cross-contamination through the application of good hygienic design, appropriate use of area and equipment segregation; and good cleaning practices.

The Solution II – Allergen Removal Validation, Monitoring and Verification

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

Cleaning programmes that have the objective of removing allergens from food contact surfaces are effectively controlling a major food hazard. The cleaning programme can thus be described as an Operational Prerequisite and as such, GMPs and GFSI based audits require such programmes to be validated.

Information relating to cleaning validation in the food industry is scarce and this presentation reflects the views of a new EHEDG Working Group on Cleaning Validation and the practical experiences of Holchem. Cleaning validation can be retrospective, using existing data, or prospective, using a planned validation programme, and the pros and cons of these approaches are noted.

Prospective validation is preferred and its basic principle is that the cleaning programme is validated under worst case scenarios, which can include:-

- The hardest area of the line to clean
- The longest production run
- The most difficult soil to remove
- The production run containing the highest percentage of the allergen in the product
- The acceptable minimal cleaning parameters – chemical solution and rinse temperatures, chemical solution concentrations

The primary objective of the validation is to ensure that the first product down the line of the subsequent production run following cleaning for open surfaces, and the first product, mid product and final product in the production run for enclosed surfaces, is free of allergen. This is traditionally assessed using the most sensitive allergen detection technique, with ELISA as the minimum acceptable sensitivity

method. The secondary objective is to ensure that food processing surfaces are free of allergen following cleaning, using LFDs or, if possible, rapid ATP or Protein detection methods.

S14 Risk Benefit Assessment of Food: Past, Present and Future Trends

Risk-benefit assessment (RBA) of foods is a relatively new discipline that integrates scientific knowledge on nutrition, toxicology, microbiology and human epidemiology to estimate the beneficial and negative health impacts of foods, food ingredients or diets. RBA is useful to inform food safety policies or to provide dietary advice based on the available scientific knowledge, to prevent food-associated diseases and promote well-being in populations. Significant progress has been made in the development of methods in RBA and in the recognition of the utility of RBA as a public health decision support tool. However, several challenges remain.

Methodological development of RBA has come along with studies performed at the European Food Safety Authority (EFSA) or within collaborative programs (e.g., BRAFO and QALIBRA). The first RBA studies have concerned the assessment of consumption of fish, well-known for its health benefits (omega-3 fatty acids, vitamin D, iodine) and risks due to environmental pollutants (dioxins, PCBs and methylmercury). Beside RBA on fish, in the last decade, many other case studies have emerged: nitrates and nitrites in fruits and vegetables, acrylamide created during the manufacturing process, water treatment, replacement of sugar by intense sweeteners, meat cooking practices, infant milk diet, etc.

In their day-to-day work, agencies face issues of RBA methodology (data gaps, weighing different types of evidence, difficulty in the quantification of effects, etc.) as well as with risk-and benefit communication to national authorities and/or to consumers.

The aim of this symposium is to introduce RBA using previously developed case-studies, and then, to move to RBA current practices in national food agencies. The symposium will end by future trend and research directions. The latter talk will be based upon the conclusions of a workshop supported by EFSA that took place in Copenhagen in May 2017 and included participants of 11 countries.

Introduction to Risk-benefit Assessment of Food

GÉRALDINE BOUÉ, UMR1014 SECALIM, INRA, Oniris, Nantes, France

Quantifying the overall impact of food consumed on human health appears as a key issue to improve population's public health. Up to the last decade, risks and benefits associated with food consumption were assessed separately in each area of research (microbiology, chemistry and nutrition) leading to rather incomplete or unsatisfying recommendations.

Risk-benefit Assessment (RBA) of foods has emerged in Europe to evaluate at the same time all potential adverse and beneficial health effects resulting from human exposure to specific agents in foods. This comprehensive evaluation was initiated at the beginning of the 21st century by European scientists with EFSA in 2006 and 2010 and European projects (BRAFO, QALIBRA and BEPRARIBEAN). They have settled a general approach to conduct RBA.

So far, about 100 RBA were carried out. While the first studies focused on fish consumption, more recently a wider range of food categories (e.g., nuts, milk and meat) and practices (e.g., fortification and effect of cooking) have been covered. These case studies have highlighted different RBA methods with for instance different ways to compare risks and benefits.

RBA is now recognized as a valuable tool to increase the comprehensiveness of the scientific evidence underlying public health recommendations on food but there is still a need for a harmonized, internationally recognized step-by-step method.

Current Practices of Risk-benefit Assessment in a National Food Agency

HANNA ENEROTH, Livsmedelsverket, Uppsala, Sweden

Since 2010 the National Food Agency has had a department of risk and benefit assessment (RBA), including microbiologists, toxicologists and nutritionists. Full RBAs are time consuming and demand input from several areas of expertise. Furthermore, data to make a full RBA evaluation is often incomplete. Thus, comprehensive assessments of all of the potential risks and nutritional benefits associated with a food or food component are rarely done. However, even in cases when not performing a complete RBA, the National Food Agency is benefitting from considering both risks and benefits to human health to support decisions on management options. Examples include recommendations to limit consumption of red and processed meat, recommendations to limit rice-based foods due to high arsenic content or legislation about trading of non-pasteurized milk. At the department, we have had several activities to overcome barriers to working in multidisciplinary teams. For the future we see opportunities for RBA as an efficient tool to evaluate complex issues in relation to public health, in order to support updates of food policy, control and dietary advice. To realize the potential of applying RBAs to public health, countries and regions could benefit from more collaboration regarding for example data Exchange.

Future Trend and Research Directions in Risk-benefit Assessment

SARA MONTEIRO PIRES, Technical University of Denmark, Lyngby, Denmark

Risk-benefit assessment (RBA) of foods is a relatively new discipline that incorporates knowledge of nutrition, toxicology, microbiology, chemistry and epidemiology for integrated health assessments. The overall aim of RBA is to assess the beneficial and negative health impacts of foods, food ingredients or diets, and compare them using common health metrics. RBA is useful to inform food safety policies or to provide dietary advice, with the ultimate aim of preventing food-associated diseases and promoting the well-being of consumers.

While several national-level and international projects have in the last decade led to substantial developments in RBA methodologies, several challenges remain. These include data and knowledge gaps, methodological limitations, lack of harmonization of concepts and new research questions and agendas, for example, linked to sustainability and economic issues.

Recent progress in RBA has been equally evident in terms of method development and data collection and analysis. As examples, while the first RBA studies focused on one single food (e.g., fish) or one single food compound (e.g., folic acid) and investigated risks and benefits in the population as a whole, recent work accounts for the health effects of substitution of foods in overall dietary patterns, or for the variation in the population in terms of susceptibility or dietary preferences.

By sharing ongoing research at DTU Foods' Risk-Benefit Research Group, as well as the views of a recently established Risk-Benefit Assessment International Network, we present current progress with RBA of foods, discuss how to further develop and optimize RBA methodology, and present our arguments to increase collaboration within RBA internationally.

S15 Interventions to Reduce Antibiotic Resistance and Antibiotic Use in Animal Production

Antibiotic usage in animal production is recognized to be an important driver of antibiotic resistance.

Initiatives to curb indiscriminate antibiotic use in animal production are therefore promoted by many global and regional organizations, including the FAO, OIE and WHO.

In this session, we will focus on the need for new interventions for preventing and controlling antibiotic resistance.

There will be a review and assessment of existing alternatives to antibiotics, including in-feed interventions such as nutritional additives and feed materials.

We will focus in particular on an all-natural fermentation solution proven to have a direct effect on the prevalence, virulence and antibiotic susceptibility of foodborne pathogens in all systems of production.

By reducing pathogen load in animals prior to entering the processing facility, such technology could be used as a tool to help improve compliance with safety regulations and mitigate the risk of recalls. Additionally, it could enhance antibiotic stewardship on the farm, supporting the public health goal of preserving the efficacy of antibiotics.

Antibiotic Resistance Transmission Pathways in the Food Chain

KATHARINA D.C. STÄRK, SAFOSO AG, CH-3097 Bern-Liebefeld, Switzerland

Antimicrobial-resistance genes are present in diverse ecosystems and can be transmitted across various pathways, including food. The relative importance of the public health of each pathway is currently uncertain. Yet undisputedly, the cumulative exposure of consumers to bacteria and genes via food is substantial. Therefore, the relative contribution of this pathway deserves priority in terms of research efforts. Several projects are currently aiming at the quantification of resistance exposure of consumers via retail meat as well as other pathways. Initial results from Europe-wide sampling efforts are being used to derive a first quantification (www.effort-against-amr.eu). The analysis is conducted based on PCR results for selected resistance genes in five retail meat categories. However, many information gaps are likely to be present regarding the impact of such exposure on public health outcomes. This presentation will provide an overview of the current knowledge, the approach used for exposure assessment and data gaps that need to be addressed before health risk estimates can be provided.

Review of Existing In-feed Solutions to Reduce Antibiotic Dependence on Farm

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

As on-farm antibiotic use declines, there is an increasing emphasis on developing alternative approaches to provide feed additives that can replicate at least some of the benefits associated with antibiotics. Certainly, reduction of foodborne human pathogens and animal disease-causing microorganisms in the gastrointestinal tracts of farm animals continues to be of high interest. Feed additives that accomplish this can be broadly categorized as either preventative of future pathogen colonization and invasion or remove pathogens that have already colonized the gastrointestinal tract of the respective farm animal. Preventive agents that potentially limit pathogen colonization include vaccination, prebiotics and probiotics. While prebiotics selects for indigenous beneficial bacteria already present in the gastrointestinal tract, probiotic cultures are externally administered with the intention of becoming established and/or modulating host responses for the benefit of the animal. Feed additives that directly remove pathogens already colonized in the gastrointestinal tract include a variety of botanical agents such as essential oils, bacteriophage, and bacteriocins. Some of these are more selective than others and the range of selectivity can limit efficacy in certain cases. There is more recent commercial interest in using these feed additives to improve gastrointestinal health, immune response, and digestibility of feeds but less is known about mechanisms associated with these host responses. As advances are made in molecular tools for assessing the gastrointestinal tract responses to these feed additives, it is anticipated that optimization of delivery administration and improving outcome predictability will enhance management for routine use.

Strategies for Controlling the Risk of Foodborne Pathogens and Antibiotic Resistance

J. ALLEN BYRD, Diamond V, Cedar Rapids, IA, USA

New challenges are constantly emerging that impact animal health and nutrition. These challenges can affect animal productivity, food safety, and profitability. On farm, conventional approaches like increased biosecurity, better hygiene, and improvements in management, husbandry and nutrition play critical roles. To address these new challenges, innovative tools are needed to reduce the risk of foodborne pathogens to help assure improved food safety. Finding solutions to these new challenges involves significant investment in research and innovation. One such solution is provided by the all-natural functional metabolites contained within Diamond V technologies, which help to support and strengthen the immune system. These technologies are designed to address specific real-world challenges in the poultry, ruminant, swine and aquaculture industries. For example, reducing stress is an important goal in animal production and research has demonstrated that feeding Diamond V Original XPC™ (XPC) can reduce the stress response in poultry, which in turn improves the animal's ability to respond to pathogen challenges. Feeding XPC improves animal production parameters, reduces food borne pathogens such as *Salmonella*, *Campylobacter*, and *Clostridium*; as well as reducing antibiotic resistance to clinically relevant antibiotics and invasiveness in health and foodborne pathogens. The strengthened immune response to pathogenic bacteria results in a healthier and more productive animal. In addition, by utilizing an in feed pre-harvest intervention like XPC to reduce resistance to antibiotics currently used in human medicine, we can provide a safer wholesome product to the consumer.

S16 The Rise of Whole Genome Sequencing: How Do We Share and Interpret the Data Globally?

Whole Genome Sequencing (WGS) technologies are revolutionizing how we respond to foodborne disease surveillance. The increased adoption and implementation of WGS for pathogen detection, diagnostics, and traceback within a few countries have highlighted the importance and necessity of having access to both sequence data and the corresponding metadata in order to respond to outbreak events in real time. This need to be able to share and analyze the data across continents has only become more apparent as the food supply chain continues to become more global in nature. The use of genomic data during the recent Ebola and Zika virus outbreaks has only underscored the importance of a shared international commitment and shared values and standards regarding the collection and use of sequence data. This session addresses some of the issues of data sharing across governments and industry, such as data ownership and trade implications. Additionally, we will highlight recent guidance documents on WGS implementation strategies so that all countries might benefit equally from this game-changing technology. Finally, we will also provide insight into data interpretation and harmonization given the multiple sequencing platforms and analysis techniques that exist in order to provide insight on the efforts of ISO WG 25 to create a standard for WGS and how this standard may affect communication of sequencing data globally.

How to Get Governments to Discuss the Benefits of Sharing Microbial WGS Data across Borders

JØERGEN SCHLUNDT, Nanyang Technological University, Singapore

As microbial Whole Genome Sequencing (WGS) spreads fast, the need to enable the use of microbiological genomic data for identification and epidemiology is increasingly recognized – also at the global level. In the not-so-distant future, WGS data collections will be used as diagnostic tools as well as for surveillance purposes, but if

we want to take full advantage of these new opportunities we need to be able to share our data internationally. Global databases will provide the basis for a platform for genomic investigations of all microorganisms, human and animal pathogens and microorganisms used in food production (probiotics and industrial strains), etc. This presentation will describe the first attempt to suggest a global database of all microbial DNA sequences to characterize (and monitor) microorganisms from animals, food and humans: the Global Microbial Identifier initiative.

Different Implementation Options for Countries Looking to Utilize WGS **ERIC STEVENS, U.S. Food and Drug Administration–CFSAN-ORS-DM, College Park, MD, USA, Amy Cawthorne, World Health Organization, Geneva, Switzerland**

Globally, there is increasing recognition that foodborne diseases are a public health priority. From a public health perspective, foodborne diseases are largely preventable and can be controlled through an effective food safety system. An integrated food chain surveillance system can identify and monitor trends of foodborne bacteria across the food chain. Whole genome sequencing (WGS) has the potential to change the way we detect and monitor microbial hazards in the food chain and how we assess, investigate and manage these food safety risks.

With WGS, the genomes of foodborne pathogens can be compared at the nucleotide level, and this offers greater insight into whether the isolates are closely genetically related or not when compared with traditional subtyping methods. WGS also provides extensive information about an isolate which can be entered into a surveillance system, such as subtyping and the presence or absence of antimicrobial resistance genes and virulence factors. As this technology is being introduced globally, WGS is becoming the typing method of choice for foodborne pathogens. However, there are still some uncertainties around the use of WGS for surveillance and outbreak response. This presentation will focus on an upcoming WHO Guidance Document on implementing WGS for foodborne disease surveillance, covering the generic principles of sequencing and surveillance and highlighting the different implementation options available to countries. Some of the items to discuss include: a standardized approach to WGS analysis for microbial subtyping; training requirements for laboratory personnel, epidemiologists and surveillance officers in public health institutions; and data sharing.

The purpose of this presentation and the forthcoming manual will provide guidance on: the capacities that need to be in place before WGS can be useful for foodborne diseases surveillance and response purposes; options for implementing WGS; how to implement WGS within the existing surveillance and response system.

Developing the Standards for Generating and Utilizing WGS Data by ISO Working Group 25 **ARTHUR PIGHTLING, U.S. Food and Drug Administration, College Park, MD, USA**

The rapid and economical whole-genome sequencing (WGS) of microbes has led to the application of WGS to an expanding number of problems in food microbiology. In public health, WGS-based analyses are used to detect outbreaks, to contextualize isolates from facility investigations, and to identify mutations, genes and other genetic features which characterize the virulence and survival potential of foodborne bacteria. WGS analyses are used to make decisions and inform legal, business, and public health activities, so standards are important for providing a basis for general confidence in WGS data and its implications in food safety. The International Organization for Standardization (ISO) Working Group 25, which includes subject matter experts from around the world, is producing guidance on the generation and analysis of WGS data from foodborne bacterial pathogens. This international

standard (“Microbiology of the food chain — whole genome sequencing, typing and genomic characterization of foodborne bacteria”) specifies requirements for the activities involved in the generation and analysis of WGS data, including i) laboratory procedures (bacterial culture handling, sequencing, raw DNA and sequence quality assessment, and data storage); ii) bioinformatic analyses (including *hq*SNP and *wgMLST*) and bioinformatic pipeline validation; and iii) metadata reporting and maintenance of sequence repositories. This guidance provides a framework for implementing and using but does not specify, chemistries, analytical methods, and software. These standards ensure that WGS applied to food microbiology provide well-understood data and analyses whose conclusions are widely accepted by all those involved with food safety.

S17 Global Occurrence of Mobile Colistin Resistance in Foodborne Pathogens

Development of bacterial resistance to antibiotics has been a global problem. The discovery of the plasmid-borne colistin resistance gene *mcr-1* in *E. coli* isolates from China in 2016 drew even more concerns among researchers because colistin is used as a last-resort drug to treat patients with multi-drug resistant infections. Following the initial report, the horizontally transmissible *mcr* gene has been found in different bacterial species from patients, livestock, pet foods and wild animals. Recent phylogenetic analyses suggest close relationships between *E. coli* from humans and livestock. Therefore, it is essential that we quantify the contribution of foodborne pathogens to antibiotic-resistant human infections.

This symposium intends to provide an overview of the occurrence of mobile colistin-resistance bacteria in different countries, the potential dissemination of the *mcr* genes, and the relationship between colistin use and anti-multidrug resistance in foodborne pathogens.

Screening for *mcr*-Carrying STEC Recovered from a Major Produce-production Region in California

XIAOHUA HE, USDA, ARS, WRRRC, Albany, CA, USA

The rapid spreading of polymyxin E (colistin) resistance among bacterial strains through the horizontally transmissible *mcr-1* and *mcr-2* plasmids has become a serious concern. The emergence of these genes in Shiga toxin-producing *Escherichia coli* (STEC), a group of human pathogenic bacteria was even more worrisome, urging us to investigate the prevalence of *mcr* genes among STEC isolates. A total of 1000 STEC isolates, recovered from livestock, wildlife, produce and other environmental sources in a major production region for leafy vegetables in California during 2006–2014, were screened by PCR for the presence of plasmid-borne *mcr-1* and *mcr-2*. All isolates tested yielded negative results, indicating if any, the occurrence rate of *mcr-1/mcr-2* among STEC was very low in this agricultural region. This study provides valuable information such as sample size needed and methodologies for future surveillance programs of antimicrobial resistance.

Whole-plasmid Multilocus Sequence Typing for *Incl2*

RICK MEINERSMANN, USDA, ARS, Russell Research Center, Athens, GA, USA

Incl2 type plasmids are medium-sized (~55 – 80 kb) conjugative plasmids that have been found carrying important antimicrobial resistance genes but have also been frequently found as cryptic plasmids. A whole-plasmid multilocus sequence typing (*wpMLST*) scheme was developed and the DNA sequences for 147 fully sequenced *Incl2* plasmids were studied. A total of 165 loci were identified of which 52 were considered core (carried by greater than 95% of the plasmids). There were 121 haplotypes found. Any single plasmid had between 74 and 101 genes (average = 84.4). Locus diversity varied from 0.067 up to 1.0 with toxin/

antitoxin genes showing remarkably low diversity scores. Most of the plasmids carrying the antimicrobial gene *mcr-1* were in a distinct clade while most of the antimicrobial gene-free plasmids were fairly unrelated. Geographic segregation of the plasmids was apparent, as was carriage of *mcr-1* and *blaCTX-M*. However, the evolutionary profile of the plasmids was not monophyletic, which was especially evident for ISAp1, the transposon associated with *mcr-1*. The wpMLST scheme is useful for tracing the evolution of plasmid-borne antimicrobial resistance genes and this will be enhanced by the addition of more world-wide data from IncI2 plasmids that do not carry those genes

Colistin Resistance and Link to Animals **JEAN-MARC ROLAIN**, Aix-Marseille Université, Marseille, France

Colistin is an old antibiotic that is currently used for the treatment of critical infections caused by multidrug-resistant Gram-negative pathogens. However, the recent emergence of colistin resistance in patients and the massive use of colistin in animals (poultry and pigs) worldwide have questioned the risk of zoonotic transmission of colistin-resistant bacteria from animals to humans. Colistin use and colistin-resistant bacteria in animals have been reported worldwide, suggesting that animals could be a source of transmission of colistin-resistant bacteria to humans. Indeed, there are reports of colistin resistance in humans that had never received the drug previously or without nosocomial transmission. The objective of this presentation is to provide an overview of resistance to colistin in animals and link to animals based on the current review of the literature on this topic.

S18 Control of Human Pathogens in Plant Production Systems

Contamination of plants meant for human consumption is of increased concern for food safety and human health. Many different disease outbreaks resulting from consumption of fresh fruits and vegetables demonstrated the possibility of occurrence of human pathogenic microorganisms (HPMO) in plant-derived products. Basic resources for agro-production, such as soils, water and fertilizers can play a role in contamination of plants, but microorganisms taxonomically closely related with HPMO are also present in plant microbiomes. HPMO must be considered as integral components of the plant microbiome. The new EU COST Action HUPLANTcontrol (COST Action 16110) will investigate the potential negative aspects of plant microbiomes on human health and aims to integrate novel scientific insight into sanitary measures and agricultural management practices. The COST action is unique in the creation of a network overarching classical disciplines in food and plant microbiology. The current IAFP Europe session wants to highlight the objectives of the recently started HUPLANTcontrol COST action (2017-2021) with a focus on two of its working packages

1. 'to improve current understanding of the role of the plant microbiome on ecological behaviour, colonization and physiological and genetic adaptation of human pathogenic micro-organisms in agriculture production systems'
2. 'to evaluate, improve and design new sanitary and agronomic practices, based on the plant microbiome concept and aimed at controlling human pathogenic microorganisms in agricultural production systems, e.g., in the open field or an industrial scale, for example in the production of sprouts.'

This session will also serve as a platform to inform on progress achieved in the interaction of human pathogenic microorganisms in plants and different environments relevant for agricultural production and to discuss within the scientific community and involve stakeholders on how new scientific concepts on the plant microbiome can be translated into practical applications and recommendations.

Occurrence of Human Pathogens in Plant Production Systems

LEO VAN OVERBEEK, WUR Plant Research,
Wageningen, The Netherlands

Foodborne disease outbreaks resulting from consumption of plant-derived fresh produce have been reported worldwide such as from spinach in the USA, from mung bean sprouts in Japan and most recently also in Europe from fenugreek sprouts (Hamburg, 2011). It is clear that particular groups of human pathogenic microorganisms (HPMO) can find their ecological niches in plant production systems. Contamination routes of HPMO to plants are poorly understood. Basic resources for agro-production, such as soils, water and fertilizers can play a role in contamination of plants, but microorganisms taxonomically closely related to HPMO are also present plant microbiomes. HPMO must be considered as integral components of the plant microbiome and it is the intention of HUPLANTcontrol to investigate the potential negative aspects of plant microbiomes on human health and to integrate novel scientific insight into sanitary measures and agricultural management practices. The HUPLANTcontrol network consists of five working groups: (1) on the ecology of HPMO in plants, (2) on taxonomical identification of HPMO from plants, (3) on characterization of the potential human-threatening nature of HPMOs, (4) on sanitary and agricultural management procedures to control HPMO in plant production facilities and (5) on dissemination of achieved knowledge via connections between science groups and relevant stakeholders from agriculture, industry and public health authorities. The proposed program integrates molecular biology, bioinformatics, microbiology, ecology, agronomy, veterinary and clinical sciences and places a strong focus on primary plant production, in principle covering all microorganisms posing potential threats to humans.

How Microbiome Studies Can Help Us to Define Preharvest Measures for Increased Biosafety of Field Grown Crops

BEATRIX ALSANIUS, SLU (Swedish University of Agricultural Sciences), Alnarp, Sweden

During the last decade, culture-independent methods to describe the microbiome have been developed. Might microbiome studies help to define preharvest methods for increased biosafety during primary production? We address this question in a series of greenhouse experiments with leafy green vegetables and *E. coli* O157:H7 *gfp*⁺ as a model organism, and fertilization as a factor. We studied the impact of nitrogen fertilization on the microbial community structure and prevalence of *E. coli* O157:H7 *gfp*⁺ in three leafy green species, i.e., spinach, rocket and Swiss chard as well as the associations with phenotypic and environmental parameters. Metagenomic analysis of the microbial community structure involved Illumina MiSeq.

Irrespective of crop and treatment, Proteobacteria dominated the phyllosphere microbiome. Actinobacteria, Firmicutes and Chlorobi were significantly more abundant in the rocket phyllosphere than in spinach and Swiss chard. Log CFU *E. coli* O157:H7 *gfp*⁺ was higher in spinach and Swiss chard. No consistent associations were found between the nitrogen supply as a single factor nor leaf nitrogen content and viable count of *E. coli* O157:H7 *gfp*⁺ when considering the crops individually. Based on all data, some families were correlated to plant nutrient content. Among these, *Enterobacteriaceae* and *Solimonadaceae* were positively correlated to the leaf nitrogen content. *Enterobacteriaceae*, *Pseudomonadaceae* and *Xanthomonadaceae* displayed similar interaction patterns with the leaf nutrient contents. Our study indicates significant associations between the log CFU *E. coli* O157:H7 *gfp*⁺ and leaf dry weight, leaf nutrient content as well as the relative abundance of the genera *Buttiauxella*, *Leminorella* and *Pantoea*.

To Sanitize or Not to Sanitize as Control Measure for Ensuring Safety of Seeds and Sprouted Seeds

INGE VAN DER LINDEN, Ghent University (UGent), Faculty of Bioscience Engineering, Department of Food Technology, Safety and Health, Research Unit Food Microbiology and Food Preservation (FMFP-UGent), Ghent, Belgium

In this presentation, a general overview is given on the status of the food safety of sprouted seeds seen from a European perspective. What does the European sprouted seed market look like? Which interventions are currently applicable in order to make the production process safer? What are the pros and cons of sanitizing? Are there already promising decontamination treatments as an alternative for the intensive chlorine wash which can pose environmental and worker safety risks? What are the current research needs?

These questions are tried to be answered and wherever possible supplemented with examples of the experience gained during a 3-year project on the food safety risk of sprouted seeds throughout the production process. For this project, alfalfa and leek seeds were used and sprouts were produced under semi-industrial conditions, mimicking a natural contamination event with *Salmonella* or *E. coli* O157. Therefore, low contamination levels of stressed pathogens or naturally contaminated seed batches (generic *E. coli*) were used.

(FBO) in routine testing. The basis for most of the currently recognised (reference) methodologies is still very traditionally based on culturing the organisms. This is a consequence of the fact that these newer technologies are mostly proprietary and therefore cannot be regarded as a (ISO and/or CEN) reference method.

The first step in opening the possibility of using proprietary methods was the publication of EN-ISO 16140 in 2003. This standard describes the comparison between the reference method and the (proprietary) alternative method. These validation studies are performed by independent organisations that conduct these studies and evaluate the results according to the criteria included in EN-ISO 16140. Based on these findings and the check of the manufacturer production quality system, a certificate is issued. Over 150 alternative methods have been validated according to this standard.

The status of EN-ISO 16140 (currently EN-ISO 16140-2) is recognised in the European directive 2073/2005 in article 5. So it is officially allowed to use these alternative methods by FBOs (and the official labs). Recent developments is that also a standard is drafted (EN-ISO 16140-6) for the validation of alternative confirmation methods. This could be a well-established biochemical confirmation galley but also omics technologies based on molecular (DNA/RNA) test or e.g., proteins. The recognition for the omics techniques is now simplified by the fact that a foreseen revision of the EU directive 2073/2005 will also allow for the EN-ISO 16140 to be used.

S19 Turning Sequencing and Mass Spectrometry into Routine Testing Tools for Microbial Strain Characterization

Omic-based-technologies have shown their capacity for generating robust data, offering new approaches to the understanding of food safety issues and new tools for routine testing. They are perceived as new ways to track and characterize microbial isolates. The power of these methods needs to be controlled in order most of the food industry users can benefit from them, and get a clearer understanding of their applications. Regulators are investigating the use of these technologies in order to define risk profile associated with food-borne pathogens, conduct outbreak investigations and strengthen regulation.

A transition is ongoing to turn sequencing and mass spectrometry into routine testing tools. What is the roadmap to ensure a global recognition by regulators and FBOs? How can these technologies fit in the day to day food testing applications? The upcoming ISO 16140-part 6 standards to acknowledge these technologies in European regulation will be introduced.

Omic-analyses also require harmonization and standardization in order to strengthen the use of bio-informatics platforms, to be implemented in food testing laboratories, to share and compare data sets. The ongoing set up of an international and available database for genomic sequences and for mass spectrometry profiles will be introduced. The need for quality assurance and method robustness to run a reliable comparison and enable information exchanges will be presented.

After one decade of overflow in communicating about Foodomics-data and technologies, implementing high-throughput technologies in routine testing laboratories is nowadays a hot topic. But there is definitely a need for global acceptance and education to facilitate the understanding of the results: What are the keys to success when selecting and implementing such technologies in routine testing? What are the wish-list and expectations of routine testing laboratories? Key studies will be presented.

Regulation Recognition of Omics Technologies: The Pathway for Implementation in Routine Testing

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

The Omics technologies pose a challenge for regulatory bodies whether these techniques are allowed to be used by their official laboratories and by the Food Business Operators

Harmonization of Omics-based Routine Methods and Database: Make It Easy and Fully under Control

JESSICA PRYOR, G2S at Centers for Disease Control and Prevention (CDC), Atlanta, GA, USA

As the era of omics-based technologies increase dramatically, the need for harmonization and standardization of the relative databases becomes ever more important. Curation of these databases is essential to ensure quality results and provide information which, in a data-driven diagnostic world, directly impacts patient care. MALDI-TOF, DNA sequence and whole genome analysis databases are expanding exponentially, many times without pausing to ensure the taxonomic basis for these data is correct. CDC has been constructing the MicrobeNet database system in recent years, as a subject matter expert-curated, virtual reference laboratory which is accessible from scientific platforms such as DNA sequencing and the Bruker MALDI-TOF instruments. This system allows for a user of these instruments or those performing traditional testing, to access CDC curated databases and information on over 2,400 bacterial and fungal species. The centralization of this information provides a cost-effective and resource-saving method for diagnostic lab to identify more pathogens than they would normally be able to with in-lab databases. This also decreases the likelihood of multiple independent strain databases which may not be representative of the species. With these databases being curated, the 1,600 current MicrobeNet users from all over the world can have 100% trust in the results that they are reporting.

Never Forget the Needs and the True Goal in Routine Testing

ERIN CROWLEY, Q Laboratories, Inc., Cincinnati, OH, USA

The concept of Omic-based technologies has emerged as the latest and greatest in our microbiological toolbox. We see them and their power to produce critical data supporting recalls in foodborne illness outbreaks thus allowing regulators to more accurately trace the epidemiology of an implicated strain. Contract laboratories offer a unique perspective in the discussion of Omics and their role in routine testing, identification and confirmation. This session will present the perspective of the contract laboratory on the use of Omic-based technology as one of many offerings for clients to meet their needs. These tools may often be just the answer a client is looking for but one can never forget the

role of the biochemical and molecular options that may be enough in the right situation. The spectrum of options within the Contract lab setting are provided to support the fit for purposes tools needed to come to the reliable conclusion. It is up to us to make the right choice! Discussions of this spectrum and scenarios from the routine testing world will be presented.

S20 Integrating Scientific Risk Assessment in the Prioritization and Management of Chemical Contaminants in Foods and Raw Materials

Food commodities may contain undesirable compounds from a multitude of possible sources, intrinsic or extrinsic, natural or man-made, introduced via raw materials or by processing. The entry of many contaminants into foods and raw materials is efficiently prevented or managed by the application of good agricultural or manufacturing practices, and local or international regulations. However, not all possible risks can be regulated, and non-safety based factors such as trade, food availability and risk-benefit considerations may impact regulation. The starting point for the risk assessment of any compound is the establishment of a health-based guidance value (HBGV), defined as the dose that does not pose a health risk to consumers at daily lifetime exposure. Food manufacturers are obliged to properly manage the ingredients and manufacturing processes to ensure that consumer exposure will not exceed HBGVs. This must take into account all other possible sources of exposure, including non-dietary sources or non-oral routes of exposure. Recently, technological improvements of analytical method sensitivities allow the detection of trace amounts of a plethora of chemicals, a fact that is generating the tendency to associate the mere presence of a compound with a risk for consumers regardless if, from a (quantitative) risk assessment perspective, it would not present a concern for the population. Therefore, the perceived of risks may not reflect the 'real' relative health significance. Prioritization and management actions, taking into account various sources of occurrence and total exposure, relative to existing HBGVs and regulatory limits, is a challenging exercise. In the present symposium, 3 recent approaches have presented that aim at prioritizing and managing undesirable contaminants integrating risk-based approaches. This is exemplified by two industry approaches and one approach being developed by the European Food Safety Authority (EFSA). Overall approaches and specific case studies will be presented.

Application Example of a Global Scientific Tool for the Assessment and Prioritization of Chemical Hazards in Food Raw Materials **GABRIELE SCHOLZ**, Thomas Stroheker Paolo Mazzatorta: Nestlé Research Center, Lausanne, Switzerland

We have recently developed a tool that integrates scientific risk-based approaches into the management and prioritization of chemical contaminants in raw materials. This includes the use of Health-Based Guidance Values (HBGVs), the severity of the toxic effect, occurrence data in raw materials and global dietary consumption information. Using the approach, we can not only define and prioritize ingredients at risk of leading to human overexposure in the context of the overall diet, but we can also derive target levels in the raw materials at risk that would not represent a concern, taking into account other known sources of exposure. This is one of the key scientific inputs in, for instance, setting limits on raw materials for use in HACCP studies, monitoring or for developing raw material specifications.

To illustrate the usefulness of the approach, the example of 3-MCPD esters is presented, a group of contaminants found in refined oils, particularly palm-based oils. HBGVs have recently been refined by EFSA and JECFA. Sources of exposure are relatively well defined and both free 3-MCPD (from ingredients like hydrolyzed vegetable proteins or soy sauces) and its esters (from refined vegetable oils) must be combined in the exposure assessment. Ample occurrence and analytical data have accumulated allowing to derive limits on fats and oils that

would protect consumers in the absence of regulatory limits. Overall, the tool provides flexibility to anticipate consequences of changes in HBGVs and dietary scenarios.

A Methodology for Risk Evaluation of Chemical Contamination in Food **TILEMACHOS GOUMPERIS**, European Food Safety Authority, Parma, Italy

The European Food Safety Authority (EFSA) was asked by the European Commission to propose a simplified approach for evaluation of risks to public health from chemical contaminants in food for the purpose of RASFF (Rapid Alert System for Food and Feed) notifications i.e., when to notify. The tool should be able to translate analytical findings (quantities found in a given food matrix) into a quantifiable risk level.

The working group, EFSA staff and external experts, developed a generic approach that can be applied to the large majority of situations when a non-compliance regarding exceedance of a legal limit or an analytical result showing potential concern is detected in food or food contact materials. The approach is a practical guide for hazard characterization based on toxicological information available for chemical contaminants in food that for the purpose of this work were classified in contaminants arising from food contact materials, pharmacologically active substances, and other food contaminants. Consecutive steps include the development of the approach to estimate dietary exposure and the IT tool development.

EFSA guidance documents, other EFSA scientific outputs, and scientific literature were used as the scientific basis for developing the tool. The proposed methodology does not replace safety evaluations carried out by European and/or National Food Safety Authorities, for instance prior to authorization of regulated products or a full-risk assessment to be performed by experts, but provide a tool to evaluate risks consistently and in a transparent manner and to harmonize the notification criteria.

An Industry Approach to Integrate Scientific Risk Assessment to Prioritize Chemical Contaminants **PAUL HANLON**, Abbott Nutrition, Columbus, OH, USA

Food manufacturers face many food safety challenges, including ensuring the foods they produce are safe in terms of chemical hazards, microbiological contamination, and allergens. One of the fundamental principles for food safety programs is the identification of risks, which includes information associated with both hazard (how toxic a substance is) and exposure (how much of that substance an individual consumes). Scientific, risk-based assessment programs allow companies to focus resources on the control of substances that pose a concern for consumers, rather than attempting to address all chemicals. Creating programs to prioritize chemical contaminant control for different ingredients is already quite complex, and in addition global companies need to ensure these programs take into consideration differences in regulatory frameworks that are often not harmonized. These risk-based programs also allow companies to address challenges associated with the continued advancement of analytical methodologies in food, which results in the detection of more and more chemicals which may have a hazard associated with them but because of insignificant exposure are not true food safety risks. These considerations are also being incorporated into new approaches to food safety, such as current activities at the Codex Committee on Contaminants in Foods (CCCF) that are exploring the integration of concepts such as the Threshold of Toxicological Concern (TTC) into guidance to control chemicals inadvertently found in food. This presentation will discuss how food manufacturers incorporate risk-based decision making into food safety programs, as well as where risk-based decision making may move to in the future.

S21 Natural Antimicrobial Preservatives in Foods: Where are We in Terms of Application and Commercialization?

Increasing health consciousness of consumers and their preference for natural food additives are major contributory factors to food manufacturers' decisions to replace synthetic food additives with those from natural sources. This global trend to ensure microbial food safety while satisfying consumer demand for natural food additives has fueled increased interest in the use of naturally derived antimicrobial agents from plant, animal and microbial sources. Currently the major considerations for food applications of natural antimicrobials include (i) effects on sensory and quality characteristics, (ii) efficacy against target organisms in specific food matrices, (iii) economic (cost) feasibility and (iv) regulatory and labeling issues. While there is a rapidly growing body of research-based knowledge on the antimicrobial efficacy of natural preservatives, information on practical applications, challenges in applications, and amounts and types of foods currently formulated with natural antimicrobials and commercially available need to be widely shared. This symposium aims to provide such information via presentations that will include: (i) introductory overview of natural antimicrobials and potential uses of natural antimicrobials in foods, (ii) strategies for overcoming current challenges in food applications, (iii) food safety applications via hurdle technology concept, (iv) commercially available foods with added natural antimicrobials, and (v) commercialization (cost) considerations. Additionally, information on future prospects for further research and novel applications will be discussed. Presentations in this symposium will be delivered by scientists from academia and the food industry and will benefit those with interest in the use of natural preservative ingredients. Those persons include food technologists, microbiologists, food processors, product development specialists, food toxicologists, and food regulatory personnel. This topic on applications of natural antimicrobials with specific emphasis on challenges in applications and considerations for commercialization has not previously been covered by the European IAFP Symposium in 2013, 2014, 2015, 2016 or 2017.

Overview of Natural Antibacterial Compounds with Potential for Use in Foods

ARMITRA JACKSON-DAVIS, Alabama A&M University, Madison, AL, USA

In an effort to control the growth and survival of undesirable microorganisms in food systems and to extend the shelf life of food products, antimicrobials are added as ingredients. These antimicrobials have been traditionally added in the form of synthetic antimicrobials. Because consumers are more health conscious and more aware of ingredients in foods, the use of synthetic antimicrobials is not as desirable. In addition, consumers are now focused on the long-term effects and interactions of synthetic chemicals in the body. Further, consumers are now more concerned with their quality of life versus how long they will ultimately live. Due to consumer concerns regarding the use of chemicals in the production of food, the use of natural antimicrobials has increased. To meet this demand, research that focuses on the use of antimicrobials derived from natural sources has also increased. The food industry has responded to this demand by creating "clean labels" on food products. Although the ultimate goal is to use natural antimicrobials that offer protection similar to its synthetic counterpart, there are benefits and challenges associated with their use.

Food Safety Applications of Natural Antimicrobials Via Hurdle Technology: An Industry Perspective for Commercialization

MORTEN HYLDGAARD, Dupont, Copenhagen, Denmark

See online programme for abstract.

Applications of Natural Antimicrobials in Foods: Challenges and Potential Opportunities

BYRON BREHM-STECHER and Aubrey F. Mendonça, Department of Food Science and Human Nutrition, Iowa State University, Ames, IA, USA

There is a growing consumer trend for foods that are fresh-like, minimally processed, nutritious, devoid of synthetic chemical additives, and safe for consumption. Although traditional processes such as heat pasteurization and canning are effective in enhancing food safety and shelf-life extension, those processes can alter sensory and nutritional characteristics of food products. Additionally, negative consumer perception of synthetic food additives is forcing food processors to seek alternatives from natural sources. Natural antimicrobials (NAs) including plant essential oils have good potential to improve the microbial safety and shelf-life of foods and are linked to the popular concept of food sustainability. While such benefits are attractive, the actual applications of NAs in foods are challenging. Unlike the traditional antimicrobials such as sorbates, sodium benzoate, sodium nitrite, and sodium sulfite, among which one antimicrobial may find suitable application in a variety of foods, the applications of NAs for microbial control in foods face several impediments. These include: (i) changes in food sensory characteristics, (ii) poor water solubility, (iii) binding to food matrices, (iv) natural variations in NA composition, and (v) alteration of NA activity by certain food processes. This presentation will highlight major challenges to the effective food applications of NAs and describe some potential strategies for overcoming those challenges.

CLOSING SESSION

Challenges in Swedish Food Protection Control

ARJA HELENA KAUTTO, National Food Safety Agency, Uppsala, Sweden

The effect and efficiency of food control has to be evaluated and improved continuously in order to be able to meet the goal with safe food and good animal welfare.

Control in food chain has mainly two central competent authorities: Swedish Board of Agriculture (SBA; animal welfare, epizootics) and National Food Agency (NFA; food safety and control in steps after primary production). Operational control is performed by NFA, County Administrative Boards (CBAs; primary production) and Local Control Units (LCUs). Performance of the LCUs is audited by CBAs and performance of CBAs and NFA-operational control is audited by NFAs special Evaluation Unit.

Recognized needs for improvement are many. Better co-operation between and sometimes within authorities in all levels is needed. Digitalized harmonized systems for control together with the national register over food enterprises in the whole food chain with open access for the authorities should be in place. Better capability to deal with black market and food fraud together with stronger legislation for consequences for illegal activities are urgently needed. New technical innovations for meat control in slaughterhouses and game handling is a must already. Advantage of risk management of different hazards in primary production should be considered before the hazards are entering the food chain. New legislation has to be flexible in order to meet challenges of novel foods and new emerging hazards.

Well-performed food control is directly beneficial for food enterprises, consumers and food producing animals. Control is not worth doing if it is not done well.

Lowering the Use of Antibiotics in Swedish Farms

MY SAHLMAN, The Federation of Swedish Farmers, Stockholm, Sweden

Due to lobbying activities from farmers, Sweden as the first country in the world banned antibiotics as a growth promoter in 1986. Unfortunately, we were unaware that the antibiotic did so much more than just promote growth—it had managing problems.

Luckily, changes in how to keep animals healthy had already started before the ban and as we learned more, we made huge progress. Good animal health depends on competent management, appropriate animal welfare, eradicating diseases, sectioning the stables with the “all in, all out” policy etc. As a result, we have good growth, a low disease incidence and the lowest use of antibiotics within the EU.

Reducing use of antibiotics in animal production is crucial. The way to do this is having healthy animals and a prudent use. Individual treatments are far more expensive but make a huge difference in the amount being used. Group treatments account for 10% of the antibiotics used in Sweden, the average in the EU is more than 90%. To lower the number of group treatments, actions must be taken to reduce the need for them.

Healthy animals are not just an interest for the farmer; it is also a national interest. A close cooperation between farmers, veterinarians and the authorities has been fundamental for the successful eradication of many diseases in Sweden. In order to maintain this freedom, we must have a secure trade in live animals. This is questioned by other countries who put trade before animal health.

Healthy animals don't need antibiotics!

Obstacles of Ready-to-Eat Vegetables and Fruits: The Swedish Approach

BEATRIX ALSANIUS, SLU (Swedish University of Agricultural Sciences), Alnarp, Sweden

See online programme for abstract.

Food Safety Management in a Changing World

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

Change can come from many directions and can impact the effectiveness in food safety management systems. Awareness of new hazards, new information on existing hazards, changes in the environment, consumer behavior and exposure are only a few examples of changes that would prompt a re-evaluation of a food safety system. Often needed modifications in the food safety system are not readily accepted by personnel or corporate leadership, particularly where they run counter to historic practices, affect production or result in increases in capital or management cost. This presentation will discuss approaches to change, both in assessing the impact of change, and in enacting needed modifications to the food safety system that impact the organization.



ROUNDTABLE ABSTRACTS

25-27 April 2018 – Stockholm, Sweden

ROUNDTABLE ABSTRACTS

RT1 How Much of a Mystery Remains with Whole Genome Sequencing?

Whole genome sequencing (WGS) is transforming the field of food safety microbiology. The technology has already cemented its place as a tool for the investigation of food-borne illness outbreaks and as a prospective surveillance tool for the public health authorities and regulators. In the private sector, there is also a growing appreciation of the benefits of using WGS in source tracking of microorganisms and its wider potential to improve food safety. Advances in the sequencing technologies and the bioinformatics analytical tools are happening at a breakneck pace, leading to significant changes even within a short span of time.

When a new, complex and rapidly changing technology such as WGS is being implemented, there are always concerns about the reliability of the technology. It is important to understand how reliable and reproducible results can be generated using a technology which is constantly evolving. It is also important to understand what is changing and how that affects the interpretation and potential use of the technology. Clarification on these aspects will facilitate widespread use of WGS in industry.

This roundtable panel, comprised of leading scientists from the government, academia, and industry will discuss how the industry can cope with the rapid technological developments to apply WGS as routine. The panel will tackle questions that need to be answered to transition WGS from research to routine application. Cornerstones to obtain reproducible analytical results such as benchmarking, validation, harmonization, standardization and verification of WGS – both current status and future needs will be addressed by the panel. The panel will discuss how these concepts work when applied to WGS compared to traditional microbial analytics. Finally, identification and implementation of fit-for-purpose tools that meet industry needs will be discussed.

Panelists:

LEEN BAERT, *Nestle, Vers-Ches-Les Blanc, Switzerland*

KATHIE GRANT, *Public Health England, Glasgow, United Kingdom*

RENE HENDRIKSEN, *National Food Institute, Denmark Technical University, Lyngby, Denmark*

PETER MCCLURE, *Mondelez International, Birmingham, United Kingdom*

SARAH O'BRIEN, *University of Liverpool, Liverpool, United Kingdom*

DANIEL PALM, *ECDC, Stockholm, Sweden*

RT2 Prediction of Spoilage and Safety with Models: How to be on the Safe Side in a World Full of Variability

Food processing companies benefit from predictive models when formulating their products and designing food processes. Preventing or slowing down pathogens and spoilers can be done with e.g., salt, heating and antimicrobials, and predictive models help to find the right conditions and concentrations. However, there is a trend towards milder processing, salt reduction and minimizing or replacing antimicrobials. On the other hand, it has become clear that microorganisms have a large variability in response to salt, antimicrobials and heat, and that there are strains that show a high tolerance to these hurdles. Not all models that are online or published are based on a large enough dataset to include the worst case scenarios and/or resistant strains. The models that are only based on a few data points or sets could benefit from including more variability.

On the other hand, it can happen that when worst case scenarios or exceptional resistant strains are taken into account, that this will rather lead to higher temperatures, higher amounts of salt and antimicrobials, instead of lowering them, leading to cost increase, unhealthy food or organoleptic challenges.

This roundtable session will aim at a discussion about the challenges and possible strategies to deal with worst case scenarios. When do you know that you are on the safe side or what does the safe side look like? What exactly defines a good model that takes variability into account without being too conservative? What are the challenges for food industry when taking worst case scenarios into account and at the same time producing tasty, healthy and safe food with a reasonable shelf life? How to deal with these challenges? These and related questions will be discussed by the audience and a panel consisting of scientists from the food industry as well as research institutes.

Panelists:

JEAN-CHRISTOPHE AUGUSTIN, *National Veterinary School of Alfort, Maison-Alfort, France*

KARIN BEEKMANN-METSELAAR, *Corbion, Gorinchem, The Netherlands*

MARIEM ELLOUZE, *Nestlé, Lausanne, Switzerland*

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

RT3 Assessment of Microbial Risk for Fresh Produce

The purpose of this roundtable session is to provide a platform to discuss the relevance of a formal risk assessment in the fresh produce chain. An easy 'Grower's Risk Assessment' tool could potentially provide a practical solution to mitigate risks for fresh produce. A webinar and previous sessions have been organized to introduce the issue; this roundtable session will provide the platform needed to further support all those involved in the fresh produce supply chain, to help food safety managers and growers to develop a practical and informed assessment of the risk that ultimately improves the safety of fresh produce.

Panelists:

JOHN BASSETT, *John Bassett Consulting Ltd., Bedford, United Kingdom*

ROY BETTS, *Campden Bri, Gloucestershire, United Kingdom*

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

JIM MONAGHAN, *Harper Adams University, Newport, Shropshire, United Kingdom*



TECHNICAL ABSTRACTS

25-27 April 2018 – Stockholm, Sweden

TECHNICAL ABSTRACTS

* Student Award Competitor

WEDNESDAY, 25 APRIL — 11.00 – 12.30

T1 Technical Session 1 – Response to Specific Environmental Conditions

T1-01 Evaluation of Two Surface Sampling Methods for Microbiological and Chemical Analyses to Assess the Presence of Biofilms in Food Companies

Sharon Maes¹, Son Nguyen Huu², Marc Heyndrickx¹, Stephanie Van Weyenberg¹, Hans Steenackers³, Alex Verplaetse⁴, Thijs Vackier⁴, Imca Sampers⁵, Katleen Raes² and **KOEN DE REU**¹

¹Flanders Research Institute for Agriculture, Fisheries and Food (ILVO), Melle, Belgium, ²UGent, Kortrijk, Belgium, ³KU Leuven, Leuven, Belgium, ⁴KU Leuven, Gent, Belgium, ⁵Ghent University, Gent, Belgium

Introduction: Biofilms are an important source of contamination in food companies, yet the composition of biofilms in practice is still mostly unknown. The chemical and microbiological characterization of surface samples taken after cleaning and disinfection is very important to distinguish free-living bacteria from the attached bacteria in biofilms.

Purpose: In this study, sampling methods that are potentially useful for both chemical and microbiological analyses of surface samples were evaluated and used in practise.

Methods: In eight food companies, food contact surfaces were sampled after cleaning and disinfection using two sampling methods: the scraper-flocked swab method and the sponge stick method. Microbiological and chemical analyses were performed on these samples to evaluate the suitability of the sampling methods for the quantification of extracellular polymeric substance components and microorganisms originating from biofilms in these facilities.

Results: The scraper-flocked swab method was most suitable for chemical analyses of the samples because the material in these swabs did not interfere with the determination of the chemical components. For microbiological enumerations, the sponge stick method was slightly but not significantly more effective than the scraper-flocked swab method. The amount of microbiological contaminated surfaces varied from 0 to 64% across the different food companies, with values varying from <1.00 to 7.23 log CFU/100 cm². Proteins were found in 20% of the chemically analyzed surface samples, and carbohydrates and uronic acids were found in 15 and 8% of the samples, respectively. For 0 to 33% of the sampled surfaces in the different food companies (with a global average of 17%), microorganisms (more than 10² CFU/100 cm²) were found in combination with biofilm matrix components; thus, these surfaces were characterized as carrying biofilm.

Significance: Depending on the desired output, the most appropriate sampling method can now be used to evaluate surface contamination and the presence of biofilm in food companies.

T1-02 Membrane Proteocomplexomic Approach for *Campylobacter jejuni*

ALIZÉE GUÉRIN¹, Sheiam Sulaeman¹, Lucile Bugros¹, Armelle Ménard², Emmanuelle Dé³ and Odile Tresse¹

¹SECALIM, INRA, Oniris, Université Bretagne Loire, NANTES, France, ²Université de Bordeaux, Laboratoire de Bactériologie, Centre National de Référence des *Helicobacters* et *Campylobacters*, Bordeaux, France, ³Université de Rouen, Laboratoire Polymères Biopolymères Surfaces, UMR 6270 and FR 3038 CNRS, IFRMP23, Mont-Saint-Aignan, France

Introduction: *Campylobacter* is a gram negative spiral-shaped bacterium and has emerged as the leading cause of bacterial foodborne infections in developed countries, with a significant increase in the prevalence of campylobacteriosis cases for a decade. *Campylobacter jejuni* infections occurred mainly after consumption of poultry. This pathogen requires fastidious growth conditions, as it is strictly microaerophile, capnophile, and thermophile. The cues from biotic or abiotic environments perceived by the bacteria are often related to bacterial surface and membrane proteins. These proteins mediate the cellular response for the adaptation of *C. jejuni* to the environment.

Purpose: *Campylobacter* rarely function as a unique entity and are often organized in functional complexes. In *C. jejuni*, these complexes are not fully identified and some of them remain unknown.

Methods: To identify functional multisubunit entities at the membrane subproteome level in *C. jejuni*, a holistic non-a priori method was addressed using two-dimensional blue native/sodium dodecyl sulfate (SDS) polyacrylamide gel electrophoresis (PAGE). Membrane protein complexes (MPCs) were subsequently denatured using SDS-PAGE, and each spot from each MPC was identified by mass spectrometry (nano-LC-MS/MS).

Results: Altogether, 20 MPCs could be identified including multihomooligomeric and multiheterooligomeric complexes. These MPCs are distributed in both inner and outer membranes. Function and conservation of MPCs across *C. jejuni* strains were inspected by functional and genomic comparison analyses. The MPCs identified in *C. jejuni* membranes are involved in protein folding, molecule trafficking, oxidative phosphorylation, membrane structuration, peptidoglycan biosynthesis, motility and chemotaxis, stress signaling, efflux pumps, and virulence.

Significance: This is the first time that a global method has been applied to *C. jejuni* to detect MPCs.

T1-03* *Lactococcus lactis* subsp. *lactis* as a Natural Anti-listerial Agent in the Mushroom Industry

Lionel Kenneth Dygico¹, Paula O'Connor¹, Maria Hayes¹, Cormac Gahan², Helen Grogan¹ and **CATHERINE M. BURGESS**¹

¹Teagasc, Dublin, Ireland, ²University College Cork, Cork, Ireland

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 biocontrol agents to provide enhanced assurance of product quality and safety.

Purpose: The aim of this study was to isolate and evaluate bacteriocins or bacteriocin-producing bacteria from mushroom growth substrate, which can prevent or eliminate *L. monocytogenes* and related biofilm formation.

Methods: Antilisterial bacteria were isolated from different types of mushroom growth substrates and were identified using 16s rRNA sequencing, while colony MALDI-TOF mass spectrometry was used to identify the bacteriocins produced. Antilisterial activity of bacteriocins produced was tested using the cell-free supernatants from the *Lactococcus lactis* strains on 72-h biofilms of *L. monocytogenes* in microtitre plates. Competitive exclusion activity of *L. lactis* strain Ca55 was then tested in mixed-biofilm conditions with *L. monocytogenes* on stainless steel coupons for 72 h.

Results: *L. lactis* subsp. *lactis* strains with antilisterial activity were found to be Nisin Z producers and naturally present in the mushroom production environment. Growth of *L. lactis* subsp. *lactis* using mixed-biofilm conditions with *L. monocytogenes* on stainless steel resulted in a significant ($P < 0.05$) 4-log reduction of *L. monocytogenes* cell numbers.

Significance: *L. lactis* has a generally recognized as safe (GRAS) status and therefore has potential for use as an environmentally benign solution to control *L. monocytogenes* in order to prevent product contamination and to enhance consumer confidence in the mushroom industry.

T1-04 Identification, Spoilage and Biofilm-forming Properties of Microorganisms from Surface Contamination in Food Companies

SHARON MAES¹, Thijs Vackier², Hans Steenackers³, Alex Verplaetse², Marc Heyndrickx⁴ and Koen De Reu¹

¹Flanders Research Institute for Agriculture, Fisheries and Food (ILVO), Melle, Belgium, ²KU Leuven, Gent, Belgium, ³KU Leuven, Leuven, Belgium, ⁴ILVO - Flanders Research Institute for Agriculture, Fisheries and Food, Technology and Food Science Unit - Food Safety, Melle, Belgium

Introduction: The importance and role of biofilms in persistent infections with spoilage organisms and pathogenic bacteria is still insufficiently known. Research about the characterization of biofilms in the food industry can help to provide new insights.

Purpose: The aim of this study was to sample surfaces in different food companies and to characterize (identification, spoilage potential, and biofilm-forming properties) the microbial population present after cleaning and disinfection (C&D).

Methods: Surfaces in seven food companies were sampled after C&D. Different microbiological enumerations were performed on the samples and the dominant bacteria were identified using (GTG)₅ clustering, followed by 16S rRNA gene sequencing. The possibility of the collected dominant bacteria to form biofilm under lab conditions was evaluated, as well as their spoilage potential.

Results: Identification of the collected isolates showed a wide diversity, but the most common identified genera collected from total aerobic count ($n=327$) were *Pseudomonas* (20.5%), *Microbacterium* (12.2%),

Stenotrophomonas (9.2%), *Staphylococcus* (7.6%), and *Streptococcus* (5.8%). For isolates that were classified to the tentative species level ($n=247$), 8.9% were identified as *Stenotrophomonas maltophilia*, 4.5% as *Staphylococcus warneri*, and 4.0% as *Microbacterium flavum*, *Microbacterium lacticum*, or *Rothia marina*. Identified genera and species were in 60 and 84.7% of the cases company-specific. Yet, *Stenotrophomonas maltophilia* was present in five out of seven food companies. Regarding biofilm-forming properties, microorganisms with the strongest possibility to form biofilms belonged to the genera *Pseudomonas*, *Acinetobacter*, and *Stenotrophomonas*, but species and even strain differences were observed. Of those that formed strong biofilms, 86% showed spoilage potential, mostly by lipolysis.

Significance: Detection and characterization of biofilms in the concerned food companies gave useful insights into the potential to cause food spoilage and foodborne infections and offered a basis for the development of more efficient C&D procedures.

T1-05 Evaluation of *Bacillus thuringiensis* Strains Abts-351 and Abts-1857 within a Simulated Gut Environment

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Introduction: Trends in agriculture and environmental regulation have begun to favor biopesticide adoption over conventional, synthetic pesticides; however, the intersection between *Bacillus thuringiensis*-based biopesticides and *Bacillus cereus* species has raised concerns about the safety of *B. thuringiensis* in food. Novel research utilizing the Simulator of the Human Intestinal Microbial Ecosystem (SHIME) supports the low risk of commercial *B. thuringiensis* products and highlights a need for more discerning regulations.

Purpose: The SHIME system was employed as a novel model for analyzing the behavior of commercial *B. thuringiensis* strains in the gastrointestinal tract and relative impact on colonic microbiota and simulated host gut wall.

Methods: A TripleSHIME consisting of stomach and small intestine reactors, proximal colon reactors, and distal colon reactors was treated with clindamycin to disturb the stable microbial community and mimic symptoms of an immunocompromised individual. Commercial *B. thuringiensis* strains were fed into parallel arms of the SHIME at 4.9×10^8 CFU/day for two weeks. An *in vitro* human cell culture model, consisting of a co-culture of Caco-2 cells and THP-1 macrophages, was exposed to spore-free extracts of the model to measure the impact of secondary metabolites on the inflammatory response induced by *Escherichia coli* lipopolysaccharides.

Results: Neither *B. thuringiensis* strain germinated in the upper and lower intestine; however, gastric passage undermined thermotolerance of *B. thuringiensis* spores, resulting in a 1 log reduction following the standard pasteurization procedure (20 sec at 80°C) for quantification. *B. thuringiensis* treatments had minor beneficial effects on recovery of the microbiota upon antibiotic-induced dysbiosis, e.g., slower increase in ammonium production and strong stimulation of the butyrate-producing *Faecalibacterium prausnitzii*. Finally, spore-free extracts of the SHIME media improved cell integrity (TEER).

Significance: The SHIME system can be used to understand the behavior of *B. cereus* species during digestion. Furthermore, these data support the low risk of commercial *B. thuringiensis* products and highlight the need for more discerning detection methods.

T1-06* Butyrate Effect on Extracellular GABA Production in *Listeria monocytogenes* 10403s WT and Lactic Acid Bacteria Isolated from Cheese and Gut

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Introduction: Butyric acid is present in butter and cheese and is extensively used in the food, beverage, pharmaceutical, and chemical industries. Butyrate is also produced by bacterial fermentation of carbohydrates in the colon. Gamma-aminobutyric acid (GABA) is an inhibitory neurotransmitter in the brain, and several studies have associated GABA with health benefits, including decreased anxiety, hypotension, and autoimmune inflammation suppression. Different bacteria have been reported to produce GABA as a product of the glutamate decarboxylase system, a mechanism that helps bacteria survive in acidic environments.

Purpose: This work aims to evaluate the effect of sodium butyrate on extracellular GABA (GABAE) production of isolates from cheese, the gut, and *Listeria monocytogenes* 10403s WT.

Methods: Overnight cultures of 11 cheese and 16 gut isolates were sub-cultured in 20 ml of MRS broth enriched with 3g/L glutamate, supplemented with 40 mM sodium butyrate with pH adjusted to 6. Cultures were anaerobically incubated at 37°C for 48 h and samples taken for GABAE analysis. *L. monocytogenes* 10403s WT overnight culture was inoculated in 20 ml of BHI broth supplemented with 20 mM sodium butyrate either in 250 ml conical flasks and incubated at 37°C for 24 h at 120 RPM, or in 50 ml falcons and incubated anaerobically at 37°C for 24 h. The pH was reduced to 4.3 (HCl 1 M) for GABAE production analysis or to 3 for acidic conditions. Cultures were incubated at 37°C with or without shaking. Viable cell counts were assessed every 20 min and GABAE samples were obtained after 1 h.

Results: All cheese and 92.59% of gut isolates presented reduced GABAE production after growth on media supplemented with sodium butyrate. *L. monocytogenes* 10403s WT grown with 20 mM of sodium butyrate in aerobic and anaerobic conditions showed a decrease in GABAE production, and was also more sensitive during acid stress.

Significance: GABAE production is negatively impacted by butyrate under these experimental conditions.

Methods: The heat treatments were carried out with the strain of *A. acidoterrestris* AD-746 in BAT medium using the capillary method. The pH ranges of the treatment and recovery media were from 2 to 7.

Results: The determined heat resistance, $D_{100^\circ\text{C}}$ value, of the studied strain at pH 4.5 was 0.37 min, while the thermosensitivity parameter value (z -value) was 6.6°C. The pH of the heating medium slightly reduced the heat resistance value of $D_{90^\circ\text{C}}$ from 13.8 min at pH 7 to 9.07 min at pH 2. However, the pH levels of the BAT agar recovery media after heat treatment strongly affected the apparent heat-resistance of this strain. For a heat treatment in pH 4.5 medium, the maximum apparent heat resistance was observed at a recovery medium of pH 4.75 ($D_{90^\circ\text{C}}=12.2$ min). On the one hand, the D -value decreased when the pH of the recovery medium decreased to pH 3, which was the minimum pH of growth. On the other hand, the D -value also decreased when the pH of the recovery medium increased to pH 5.5, which was the maximum pH of growth.

Significance: These physiological parameter values allow for optimized thermal treatments in order to reduce the risks of *A. acidoterrestris* spoilage in fruit juice.

T2-02* Growth/No-growth Boundaries of Heat-resistant Molds as a Function of Temperature and Degrees Brix

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Introduction: The thermal resistance of heat-resistant molds (HRMs) and their ability to grow in a broad range of conditions poses great challenges to processors of fruit-based products. Therefore, the study of factors (conditions) that may inhibit the growth of HRMs is very important in order to prevent spoilage of these food products.

Purpose: The aim of this study was to assess the growth/no growth boundaries of HRM as a function of degrees Brix (°Bx) and temperature.

Methods: To assess the effect of degrees Brix, suspensions of HRMs ascospores belonging to *Byssoschlamys* sp., *Neosartorya* sp., and *Talaromyces* sp. were spread plated (± 100 spores) on acidified potato dextrose agar (aPDA, pH 3.5) whose Brix was adjusted with fructose-glucose to 44 to 56 °Bx, followed by incubation at 30°C. To assess the effect of temperature, aPDA were inoculated as described above and incubated at 7, 8, 10, 12, and 14°C. Three replicates (3 aPDA plates) were prepared per condition evaluated. The plates were checked daily over two months for the development of visible colonies. The data obtained was used to establish growth/no growth boundaries as well as the distribution of the time to visible growth.

Results: With regards to temperature, *Byssoschlamys nivea* was the most temperature-sensitive, as it had the least ability to germinate and form visible colonies as temperature was decreased. On the other hand, *Neosartorya* species were less sensitive to reduced temperatures, as they were able to grow out at the lowest temperature evaluated. Likewise, *B. nivea* was the most sensitive to increase in degrees Brix, whilst *Neosartorya udagawae* was the least sensitive, as it was able to grow out at the highest degrees Brix evaluated.

WEDNESDAY, 25 APRIL — 14.00 – 15.30

T2 Technical Session 2 – Intervention Strategies and Modeling

T2-01 Quantifying the Heat Resistance of *Alicyclobacillus acidoterrestris* at Different pH in Heating and Recovery Media

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UMT14.01SPORE RISK, Quimper, France

Introduction: *Alicyclobacillus acidoterrestris* is a sporulating, acidophilic bacterial species which spoils acidic beverages such as fruit juices. To control its development and optimize the application of heat treatments, it is necessary to characterize its growth and heat-resistance dynamics in relation to environmental factors such as pH and particularly its recovery following heat treatments.

Purpose: The objective of this study was to determine the heat resistance of *A. acidoterrestris* spores and their recovery capabilities as a function of media pH.

Significance: The results will be used to develop probabilistic predictive models and Quantitative Microbiological Risk Assessment (QMRA) studies to prevent the spoilage of pasteurized fruit products.

T2-03 *Thermoanaerobacterium* Species as Emergent Spore-forming Bacteria Involved in Spoilage of Canned Food

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Introduction: *Thermoanaerobacterium* spp. are anaerobic thermophilic species which form highly heat-resistant spores as *Geobacillus* or *Moorella* encountered in spoiled cans.

Purpose: These bacteria represent a new major microbiological risk for canned food industries, which need to better understand how to control these species on food processing lines.

Methods: In France, hundreds of spoiled cans have been analysed since 2007. Physiological studies were performed concerning (i) growth parameters, (ii) sporulation ability, and (iii) spore heat resistance.

Results: The number of 55°C-incubated canned foods spoiled by *Thermoanaerobacterium* spp. increased significantly and reached the same occurrence as *Geobacillus* and *Moorella*. Nowadays, 25% of 55°C spoiled cans contains *Thermoanaerobacterium* spp. Occurrence of *Thermoanaerobacterium* spp. has also increased significantly for spoiled canned foods incubated at 37°C. All food matrices are affected by spoilage with these species such as vegetables, fish, and meat products, but also some acid products and duck fat. In food products, we identified 13 different species belonging to *Caldanaerobius*, *Thermoanaerobacterium*, and *Thermoanaerobacter* genera, considered as *Thermoanaerobacterium* group detected by a molecular tool (SporeTraq). *Caldanaerobius polysaccharolyticus* and *Thermoanaerobacterium thermosaccharolyticum* were the most frequently encountered in spoiled cans.

Temperatures for growth were studied between 30 and 70°C and pH from 3.5 to 7.0. Growth at 37°C was observed for 75% of strains (14). The growth at 30°C was only observed for two out of eight strains studied. No strain was able to develop at 70°C or at a low pH of 3.5. However, five strains developed at pH 4.0 and 55°C. Spore heat resistance was determined between $D_{115^{\circ}\text{C}} = 0.7$ min and $D_{120^{\circ}\text{C}} = 5.9$ min.

Significance: The data show that *Thermoanaerobacterium* species are able to adapt to large environments: mesophilic and thermophilic conditions, various substrates, and even acidic conditions. Recommendations along food processing lines can be given to canners.

T2-04 Quantifying the Food-safety Risk of Federally Registered Dairy Establishments in Canada Using the Canadian Food Inspection Agency's Establishment-based Risk Assessment Model (2016–2017)

Sylvain Quesy¹, MANON RACICOT², Alexandre Leroux³, Raphael Plante², Sunny Ng³, Romina Zanabria³, Hargun Chandhok³, Genevieve Comeau², Suzanne Savoie⁴ and Anna Mackay³

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Introduction: The Canadian Food Inspection Agency (CFIA) has developed a quantitative risk assessment model to help inform the allocation of inspection resources for food establishments. A pilot, conducted in 2014, assessed

the model's performance within 29 dairy establishments and resulted in a Spearman correlation coefficient of 0.60 ($P < 0.001$) between the model outputs (estimated as the annual number of disability-adjusted life years [DALYs]) and the risk assessment performed by senior inspectors.

Purpose: To quantify the food safety risk of dairy establishments in Canada using the CFIA's establishment-based risk assessment model.

Methods: Over a one-year period, inherent and mitigation data were collected using a questionnaire shared with 273 establishments and completed by inspectors. This information was analysed along with compliance data obtained from CFIA data systems by the model algorithm.

Results: Six establishments did not distribute any dairy products during the assessment and were not included in the analysis. Results show that 31% of establishments distributed products to a vulnerable population. All establishments reported applying at least one mitigation measure to control the inherent risk and 75% implemented at least one food safety certification scheme. Based on the model, ten establishments were responsible for 50% of the total risk, while 80% of establishments represented 10% of the total food safety risk related to dairy products. Furthermore, establishments were categorized in four groups based on individual contribution to the overall food safety risk in the dairy sector (1, 18, 72, and 174 for category 1 to 4 respectively, where 1 represents the highest risk and 4 the lowest). They were correspondingly responsible for 12, 55, 29, and 3% of the total risk.

Significance: The model outputs could be used to proportionally allocate inspection resources based on the establishment risk contribution.

T2-05* The Transfer Rate of *Salmonella* Typhimurium from Contaminated Parsley to Other Consecutively Chopped Batches Via Cutting Boards under Different Food-handling Scenarios

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Introduction: In many Mediterranean and Middle Eastern countries, leafy green parsley is typically eaten raw and prepared by finely chopping several batches. It is becoming more evident that *Salmonella*-associated outbreaks are not limited to contaminated foods of animal origin; they are periodically linked to consumption of fresh produce, including parsley.

Purpose: This study aimed to quantify the transfer rate of *S. Typhimurium* across all chopped batches in scenarios that resemble normally occurring operations in restaurants and home kitchens.

Methods: Fresh parsley leaves were inoculated at concentrations of 6 and 3 log CFU/g and chopped on a polyethylene cutting board (CB). Uninoculated parsley leaves were sequentially chopped in individual batches on the same cutting surface: (1) instantly (CB Instant); (2) after washing in water and holding at 30°C for 24 h (CBWW) and (3) after washing in soapy water, sponge scrubbing, and holding at 30°C for 24 h (CBSW).

Results: Using the high inoculum levels, the mean *S. Typhimurium* was 0.012±0.04, 0.014±0.02, and 0.010±0.008 via CB Instant, CBWW, and CBSW, respectively. Comparatively, the *S. Typhimurium* mean values were significantly higher with the low inoculum levels: 0.60±0.65 and 0.64±0.46 via CB Instant and CBWW, respectively, and transmissions of *S. Typhimurium* significantly decreased across consecutively chopped batches on both washed CBs ($P < 0.05$). These results demonstrated continuous transfer of *Salmonella* cells, from contaminated parsley to cutting boards and

subsequently re-contaminating up to six batches of parsley chopped consecutively on the same surface. A greater cross-contamination rate was recorded during the initial phases of chopping and remained for 24 h at 30°C.

Significance: Vigilant cleaning and sanitation procedures on cutting surfaces should be a fundamental requirement after use with fresh produce, particularly if there is a likelihood of insufficient food safety measures at harvest and post-harvest stages.

T2-06 *Listeria monocytogenes* is Prevalent in Retail Grocery Produce Environments and is Influenced by Infrastructure, Sanitation, and Management Practices

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Introduction: There has been a notable increase in produce-related listeriosis in the United States. Our previous studies definitively show retail deli environments may cross-contaminate foods with *Listeria monocytogenes*. However, environmental prevalence of *L. monocytogenes* in retail produce systems was largely uncharacterized.

Purpose: The purpose of this study was to determine the prevalence of *L. monocytogenes* in retail produce environments, to elucidate ecological niches, and identify other factors (e.g., management) that may be risk factors for INCREASED prevalence.

Methods: Thirty environmental samples (17 food contact surfaces [FCS], 13 non-food contact surfaces [NFCS]) were collected monthly for six months in duplicate in 30 retail produce departments in seven states during daily operation. Samples were tested for *L. monocytogenes* using the AOAC-validated ROKA Atlas LmG2 assay with secondary enrichment and plating confirmation. Each store manager completed a 110-question survey on infrastructure, sanitation, and management practices. Pearson correlation and analysis of variance were used to identify significant survey variables associated with *L. monocytogenes* prevalence. Tukey pairwise comparison elucidated significant effects ($\alpha=0.05$).

Results: A total of 4.4% (226 of 5,112) environmental samples tested positive for *L. monocytogenes*; *L. monocytogenes* was present on 8.1% (178 of 2,205) NFCS and 1.6% (48 of 2,907) FCS surfaces. Four of 30 stores had high overall prevalence (>10%). Water pooled near case drain covers ($P=0.0125$) and bottom shelves inaccessible for cleaning ($P=0.0217$) were associated with overall increased *L. monocytogenes* prevalence. Changing gloves after handling different produce types ($P=0.0114$), employees with food safety role models ($P=0.0128$), proper glove use ($P=0.0015$), and restricting employee traffic from other departments ($P=0.0394$) correlated strongly with reduced prevalence.

Significance: The data indicate that retail produce environments may be a significant source of *L. monocytogenes*, which may result in cross-contamination of produce. This is the first longitudinal study to identify facility design and management practices that may influence *L. monocytogenes* prevalence in retail produce.

WEDNESDAY, 25 APRIL — 16.00 – 17.30

T3 Technical Session 3 – Pathogens and Antimicrobials

T3-01* Lessons from Growing *Listeria monocytogenes* in a High Oxygen Environment – Detaching the Exponential Phase from Anaerobicity

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Introduction: During its life cycle, *Listeria monocytogenes* is exposed to a wide range of environmental conditions, such as oxidative stress (OS). We observed that during the exponential phase, the dissolved oxygen (DO) is close to anaerobic levels for four hours. By growing *L. monocytogenes* in a bio-fermenter, for the first time we were able to keep the levels of DO high during the exponential phase and dissociate the stage of growth from the levels of DO. We hypothesised that the DO, rather than the actual phase of growth, could be modulating *L. monocytogenes*' ability to cope with oxidative stress.

Purpose: Investigate *L. monocytogenes*' response against oxidative stresses and how the levels of oxygen modulate this response during growth.

Methods: *L. monocytogenes* 10403S, wild type (WT), grown at 37°C either in hyper-aerophilic conditions in a 5-L bio-fermenter with constant stirring (250 rpm) and aeration (3 L/min) or in a 250-mL conical flask, were challenged with 1% (volume/volume) of H₂O₂ after 6 and 10 hours of growth. Survival against H₂O₂ was assessed during the 60-min challenge. In parallel, the DO and the catalase activity were determined.

Results: After 6 hours, cells grown in the bio-fermenter maintained levels of DO above 30% of the initial, while DO lowered to 7 to 14% in cells grown in the flask. The differences in DO correlate with an increased resistance against H₂O₂ (5.32 log difference, $P<0.005$) in the bio-fermenter compared to the flask. After 10 hours, the flask showed levels of DO similar to the bio-fermenter (~50%); however, it retained the lower resistance phenotype against H₂O₂. The catalase activity was higher in the bio-fermenter during the whole course of the experiment.

Significance: The present study shows the importance of taking into consideration the environmental conditions where the pathogen grew before extrapolating the conclusions of previous research to a "real-world" scenario.

T3-02 Characterization of the Virulence Potential of Environmental Shiga Toxin-producing *Escherichia coli* Reveals a Novel Plasmid-encoded Biomarker

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Introduction: Shiga toxin-producing *Escherichia coli* (STEC) O157 and non-O157 are foodborne and waterborne pathogens that are responsible for outbreaks of human gastroenteritis with diverse clinical spectra. Expression of Shiga toxins (Stx) is attributed to the development of severe disease symptoms in humans such as the hemolytic uremic syndrome.

Purpose: The objective was to employ molecular, cellular, and biochemical techniques to better characterize the virulence potential of STEC strains recovered from various environmental sources in a major produce production region in the United States.

Methods: STEC O157 and non-O157 strains were recovered from livestock, wildlife, soil, water, and produce samples, which were subjected to enrichment and immunomagnetic separation steps. Suspect STEC colonies were isolated on chromogenic selective solid agar. *Stx* expression was induced for 24 h at 37°C in Luria-Bertani (LB) agar supplemented with 1200 ng/mL mitomycin-C. Supernatants from lysed cells were analyzed on a MALDI-TOF-TOF mass spectrometer. The activity of *Stx* in mammalian cells was assessed in the STEC strains grown overnight in LB broth, and the cell-free culture supernatants were added to a Vero-d2EFGP fluorescent cell line. Next-generation genome sequencing was performed using Pacific Biosciences and Illumina platforms.

Results: Genotypic analyses of a subset of 50 recovered STEC strains revealed that they harbored either *Stx2a* or *Stx2c* subtype and belonged to the clinically relevant serotypes O113:H21, O121:H19, and O157:H7. Results from the cell-based assays indicated that the strains expressing the *Stx2a* subtype were more effective at inhibiting protein synthesis than those expressing *Stx2c*. Among those expressing *Stx2a*, serotype O113:H21 strains from cattle, feral pigs, and water were the most effective at inhibiting protein synthesis. Mass spectrometry revealed that serotype O113:H21 strains also produced high levels of a novel plasmid-encoded biomarker with a mass of 7,838±8 Daltons.

Significance: Identification of a novel plasmid-encoded factor in STEC O113:H21 strains expressing *Stx2a* under antibiotic stress.

T3-03 Comparison of Regional Residue Assessments Evaluating the Safety of Anti-microbials and Cleaners in Food Contact Applications

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Introduction: Evaluating the safety of food-related antimicrobial and cleaning chemicals differs across the globe. This makes it challenging for stakeholders to consistently address their safety.

Purpose: While food safety management systems, including CODEX, Global Food Safety Initiative guidance, and U.S. Food and Drug Administration regulations are well-established, there is a need for joint management of new residue challenges to enable seamless partnerships across industries that satisfy consumer expectations of “zero” risk. The presentation will illustrate the gaps and commonalities of how different regions assess safety of food contact chemicals.

Methods: Using expert elicitation, a comparative assessment of terminology, methods, and regulations used to conduct residue assessments in food for cleaning and antimicrobial chemicals was performed for countries in different areas of the world, e.g., Europe, United States, Latin America, Africa, and Asia.

Results: The analysis highlighted the largest gaps in how food contact cleaning and antimicrobial products are evaluated and managed from a food safety standpoint. Guidance and best practices for residue assessment in food around the globe vary, driven by different regional regulatory approaches. In comparison regions, e.g., the European Union and the United States, there are clear methods and regulatory requirements for evaluating the safety of residues from antimicrobials in contact with food, whereas the process for evaluating cleaning chemistries is less prescriptive.

Significance: Recent interest in assessing risks from sanitation chemical residues in food streams reinforces the need to critically assess global approaches. The presentation educates and shows possible future solutions highlighting opportunities for greater alignment.

T3-04 The Absence of *N*-Acetylglucosamine in Wall Teichoic Acids of *Listeria monocytogenes* Modifies Biofilm Architecture and Tolerance to Cleaning and Disinfection Procedures

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Introduction: *Listeria monocytogenes* is able to form biofilms composed of cells and extracellular matrix. In 2015, Brauge et al. demonstrated that the major polysaccharide present in the extracellular matrix of the *L. monocytogenes* biofilm was teichoic acid. Moreover, around 50% of *L. monocytogenes* strains (out of 93 strains) carried a mutation of the *lmo2550* gene involved in the GlcNAcylation of this teichoic acid.

Purpose: In order to better characterize biofilms *L. monocytogenes*, we evaluated the impact of the absence of the GlcNAc residue on adhesion and 48-h biofilm development, as well as biofilm-related phenotypes.

Methods: We studied the wild-type EGD-e strain and two mutants, EGD Δ *lmo2549* and EGD Δ *lmo2550*, inactivated respectively in the *lmo2549* and *lmo2550* genes encoding glycosyltransferases, involved in the GlcNAcylation of teichoic acid in *L. monocytogenes*. First, we evaluated the impact of these mutations on adhesion, formation of *L. monocytogenes* biofilms by epifluorescence microscopy, and by counting the viable cultivable population on agar media. Second, we studied their further detachment after mechanical and chemical actions by qPCR and PMA-qPCR assays.

Results: The mutation of the *lmo2549* or *lmo2550* genes caused a decrease in bacterial adhesion to stainless steel during the adhesion step. Bacterial population was not significantly different after 24-h biofilm formation. The biofilm architecture was different between the wild-type strain and the two mutants with the presence of bacterial micro-colonies for mutants, which were not observed in the wild-type EGD-e strain biofilm. Upon a water flow or cleaning procedure at a shear stress of 0.16 Pa, the mutant biofilms showed a higher detachment rate compared to wild-type strain. Meanwhile, an increase in the amount of residual viable but non-culturable population on stainless steel was recorded in the two mutants.

Significance: Our data suggest that the GlcNAc residue of teichoic acid played a role in adhesion and biofilm formation of *L. monocytogenes*.

T3-05 Chitosan Coating as an Alternative Seed Disinfection Treatment for Alfalfa and Leek Seeds Intended for Sprouting

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Introduction: Sprouts are considered healthy ready-to-eat foods and are consumed raw or minimally processed. Still,

many outbreaks occur and the bacterial pathogens most frequently associated are *Salmonella* and Shiga toxin-producing *Escherichia coli* (STEC). The batch of seed used for sprouting has been identified as the most likely source of contamination. Effective seed decontamination is, therefore, a key step for a safer production process.

Purpose: The aim of this study is to assess the potential of a chitosan coating in reducing the microbial contamination on alfalfa and leek seeds intended for sprouting.

Methods: A 0.5% weight/volume chitosan solution was prepared in 0.5% volume/volume lactic acid. Seeds were treated for 10 min or 1 h and dried. Two chitosan powders with a different molecular weight were tested (100 to 150 or 300 to 340 kDa). The effect was evaluated on seed germination percentage and by determining the aerobic mesophilic colony count (ACC) and *Enterobacteriaceae*. The most promising treatment was tested on artificially contaminated seeds with two *E. coli* O157 and two *Salmonella* strains. For each seed type, three seed batches were examined.

Results: The main effect of chitosan seemed to be the prevention of the multiplication of the natural background microbiota during the drying step. When the treatment (300 to 340 kDa, 1 h soaking), was tested on pathogen inoculated seeds, similar reductions were obtained for all the strains (average reduction between 1.9 and 2.4 log units). Also the effect on the germination rate was evaluated. Overall, the germination rate of the coated seeds were least affected by the 1-h treatment, but seeds were more stuck together.

Significance: Our study showed that a chitosan coating should be further optimized in order to be used as an antimicrobial treatment for seeds intended to be used for sprouting.

T3-06* Development and Validation of Growth Models for *Listeria monocytogenes* in Mediterranean Fish Species from Aquaculture Production

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Introduction: *Listeria monocytogenes* is a foodborne pathogen frequently associated with minimally processed fish products, posing a major food safety concern for the fish industry. Predictive microbiology is proposed as a suitable method to predict the behaviour of pathogenic bacteria during processing and storage. Over the last decade, numerous studies have evaluated and modelled the potential growth of *L. monocytogenes* in minimally processed fish products commonly consumed in Nordic European countries (e.g., smoked salmon). Fish species from aquaculture production, such as sea bream and sea bass, are of special interest given their added value and relevance in the diet of Mediterranean countries. In a previous work, the growth of *L. monocytogenes* was evaluated in fish-based juice (FBJ) under two environmental conditions (reduced oxygen and aerobic atmosphere) and secondary models were generated describing the effect of temperature on pathogen growth.

Purpose: The objective of this study was to validate the generated models in FBJ through challenge tests with sea bream and sea bass fillets under constant and dynamic storage conditions.

Methods: Experiments were carried out at three constant storage temperatures (4, 9, and 16°C) and with a dynamic temperature profile simulating cold-chain distribution and storage of fishery products in Spain (ranging from 2.6 to 8.9°C).

Results: The performance of the model generated under reduced oxygen conditions resulted in a bias and accuracy factor of 1.15 and 1.25, respectively, demonstrating its applicability to adequately predict *L. monocytogenes* growth in the studied Mediterranean fish species. In aerobic conditions, the model significantly overestimated growth rates of *L. monocytogenes* on the studied fish products.

Significance: The validated models for *L. monocytogenes*, together with published growth models for specific spoilage bacteria in sea bream, were incorporated in the predictive software tool MicroHibro to predict the effect of both constant and changing temperatures on the shelf life of Mediterranean fish products.

T4 Technical Session 4 – Risk Assessment

T4-01* Integrating WGS Data into Quantitative Microbial Risk Assessment: Refinement of the *Listeria monocytogenes* in Cold Smoked Salmon Model

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Introduction: Recent developments in genome sequencing open new opportunities for explaining the intraspecific variability of phenotypes (e.g., virulence and growth behavior). Successful association between whole genome sequencing data and specific phenotypes is thought to contribute to better predicting microbial behaviors. Implementing this information in hazard identification, exposure assessment, and hazard characterization processes will refine quantitative microbial risk assessment (QRMA) models.

Purpose: The aim of this study was to explore the refinements in QRMA studies when considering pheno- and genotype associations for hazard properties, particularly related to growability at low temperature (minimal growth temperature [T_{min}]) and virulence.

Methods: The QMRA model was previously developed in order to assess the number of listeriosis cases associated with cold smoked salmon in France. The global prevalence in the existing model was replaced by the specific prevalence for each genotypic subgroup (clonal complex [CC]). In order to describe the variability of *Listeria monocytogenes*' growth characteristics more accurately, two different distributions of T_{min} were implemented (according to the presence and absence of a biomarker).

For risk characterization, different groups of virulence were created according to the CCs. The 294 strains were included in groups of virulence associated to their CC. According to the groups, the dose-response was adjusted in the model.

Results: The CCs that contributed the most in exposure were not those that contributed the most to listeriosis cases. The most prevailing CCs led to few listeriosis cases, unlike uncommon virulent strains which were responsible for many cases. Similarly, the group of strains with high T_{min} was approximately two times less implicated when considering human listeriosis in comparison to food contamination.

Significance: Considering genotypic data in QMRA opens the way for the establishment of risk-based measures specific to genetic elements.

T4-02 The Canadian Food Inspection Agency Establishment-based Risk Assessment Model for Hatcheries: Principles and Algorithm

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Introduction: The Canadian Food Inspection Agency (CFIA) has developed a risk assessment model specifically designed to quantify the food safety risk associated with Canadian hatcheries by estimating their potential impact

on consumers' health in Canada. The intent is to allocate inspection resources to areas representing a higher risk for food safety.

Purpose: To present the principle and algorithm of the CFIA Establishment-based Risk Assessment model for hatcheries.

Methods: In designing the model algorithm, the identification and selection of significant risk factors and associated assessment criteria were critical steps that were completed through an elicitation involving Canadian hatchery experts in 2017. Through a face-to-face Delphi approach, experts ($n=11$) were asked to estimate the relative contribution of each assessment criterion ($n=81$) to the food safety risk of a hatchery.

Results: The total health impact, expressed as disability-adjusted life years (DALYs), remains constant within a period of time, but the allocation estimated by the algorithm to individual hatcheries is fluid. The total health impact value combines the annual number of human cases of *Salmonella* spp., its associated burden, and the attribution to the poultry and egg commodities with the hatchery being the focal point of those commodities considered in the current model. In a first instance, the health impact is individually allocated to hatcheries based on their production volume and then adjusted according to the presence or absence of specific food safety risk factors considered in the model.

Significance: The model was tested with 30 Canadian hatcheries and its performance will be assessed by comparing the model outputs with the results of a risk assessment done by senior inspectors on those same hatcheries. By quantitatively assessing the food safety risks represented by each of the 96 hatcheries under CFIA's jurisdiction, this new risk assessment model will help CFIA to appropriately allocate inspection resources and will further improve the protection of public health in Canada.

T4-03 Development of a Software Tool for Risk Assessment of *Listeria monocytogenes* in Selected Ready-to-Eat Food Categories in the European Union

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Introduction: The development of predictive microbiology software tools has gained interest from academia and practitioners, which has resulted in an increasing number of applications over the last few years (e.g., Pathogen Modeling Program, Combase, FSSP, etc.). However, few attempts have been made to incorporate probabilistic microbial risk assessment methods, which limits their applications for end users.

Purpose: To design a user-friendly tool to perform probabilistic risk assessment of *Listeria monocytogenes* in different food categories and develop a standardized risk model annotation and metadata enabling an easy update of models and their applications.

Methods: An Excel add-in, called *Lis-RA*, was developed with Visual Basic for Applications and included a customized ribbon interface. The simulation capabilities were built upon functions from @Risk software. The application was optimized for Excel 2016 and @Risk 7.0 and 7.5.

Results: The software tool allows users to load risk model spreadsheets, select scenarios, and define model inputs and simulation settings in an easy and intuitive way. The tool can be downloaded for free at the European Food Safety Authority community for food safety tools, Knowledge Junction, which is built in the Zenodo research-sharing platform. The model application was used to estimate the annual number of listeriosis cases in the European Union population, considering different susceptible groups. The burden of listeriosis cases estimated with the application was 2,318 (95 confidence interval: 1,450 to 3,612) which

resembles the actual cases reported by the surveillance system. Cooked meat and sausage presented most cases (median of 863 and 541, respectively).

Significance: The risk assessment tool developed in this work, *Lis-RA*, represents a relevant advance in the field of microbial risk assessment and risk model standardization. The application *Lis-RA* can be updated to consider other foodborne pathogens and food products based on the existing standardized model annotation designed in this work.

T4-04 Application Potentials of Network Science to Food Chain safety Risk Analysis

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Introduction: The increasing complexity of food production and trade poses an increasing challenge to governmental stakeholders in their efforts to protect consumers. On the other hand, exponential growth of data available for food products and commodity chains provides the potential for better-informed decisions.

Purpose: Network science may have an important role in enhancing the safety of consumers and the supply chain itself and could easily be adopted to support crisis prevention, risk-based control, early warning, and predictive systems. The presentation focuses on the possible applications of network analysis to food chain safety and on a case study aiming to develop a network-based assessment methodology for Hungarian cattle holdings.

Methods: We have built up a cattle trade network using the database of the Hungarian cattle identification system; we then applied network analysis methods to gain insight into the network and its most critical elements. We have calculated static and dynamic measures of the network, as well as centrality measures showing the important role a holding plays.

Results: It was possible to determine the highest risk flows in the system and construct different models for the cattle network. Based on these models, we have been able to determine the most important centers of the network, which is important, as the most critical points are not necessarily the largest hubs. The various statistical algorithms provided a characterization of the cattle movement system, exploring both its structural and dynamical properties.

Significance: Data are increasingly becoming available thanks to traceability systems put in place in the European Union. By using network science methodology, it is possible to analyze the dynamic system of cattle movements, going beyond static and simple approximations. An important aim of this work is to share the methodology and algorithms with the network science and food chain safety community to enhance the capacity building process.

T4-05 Exposure Assessment of Process-related Contaminants in Food by Biomarker Monitoring

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Introduction: In risk assessments, exposure assessment can often present a number of challenges and uncertainties. This problem is especially relevant for contaminants formed upon heating of food, which may be reactive and/or volatile, hampering their analysis in food.

Purpose: To accurately assess consumer exposure and thus better inform the risk assessment, novel approaches for exposure assessment are essential. The purpose of the current study was to present an overview of the state of the art with respect to the use of biomarker-based methods to estimate exposure for the process contaminants acrylamide, 3-MCPD esters, glycidyl esters, furan, and acrolein.

Methods: A scientific literature evaluation was conducted by an expert group of the European branch of the International Life Sciences Institute and coordinated by the Process-Related Compounds and Natural Toxins Task Force.

Results: The evaluation revealed that biomarker monitoring to assess human exposure to process-related contaminants in food is a promising and strongly developing field. Data gaps and challenges for the future may include: (i) using duplicate diet studies and physiologically based pharmacokinetic modelling to establish, preferably in humans, correlations between external exposure and biomarkers, (ii) obtain better insight in the possible endogenous formation of the process-related contaminants and the resulting biomarker levels, (iii) characterise inter-individual variations and how these affect the biomarker-based exposure predictions, (iv) identification and correction for confounding factors, (v) the value of the different biomarkers for exposure and risk assessment, and (vi) possible novel methodologies.

Significance: In spite of the challenges that remain to be solved, it can be concluded that more accurate assessment of consumer exposure to process-related contaminants in food, and thus to improve risk assessment biomarker-based exposure assessments, provides a unique opportunity.

T5 Technical Session 5 – Molecular Characterization and Risk Assessment

T5-01 16s Metagenomic Sequencing of a Facility to Determine Point Source Contamination of Products

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Introduction: Next generation sequencing (NGS) has created new applications for DNA sequencing, including uses for a variety of biological investigations. These applications empower users with new ways to address microbiological problems and understand the relationship of microbes to spoilage and organoleptic properties of food, contamination sources, and transmission patterns of microbes in production operations.

Purpose: An application of NGS, metagenomic 16s rRNA gene sequencing, can be used by sanitarians and food producers to obtain accurate taxonomies of microorganisms from microbial communities without enrichment. 16s sequencing was used for determining point source contamination of a client's product and environmental samples.

Methods: The client supplied samples from the environment and product, which were sent to our genomics lab. Total DNA was extracted and the 16s V4 region PCR amplified from an aliquot of each sample. The PCR reactions were sequenced on an Illumina MiSeq. Sequence data was processed through taxonomic-binning and NGS data processing software to compute the microbial abundance profile of each sample.

Results: Several products were dominated by a single genus. However, other microbiota profiles were present, indicating multiple sources of contamination. The genus *Meiothermus*, a thermophile that can be resistant to biostatic cleaners, was found in a product sample, hot water bath, drain, and adjacent table. Subsequent sampling after removal of the bath found *Meiothermus* to be absent. In that study, environmental samples taken pre-cleaning with an alkaline cleaner were dominated by *Brevibacterium*, *Staphylococcus*, and *Lysinibacillus*. The post-cleaning samples were abundant in *Brevibacterium*, which is not surprising as it is alkaline tolerant.

Significance: The data allows for multiple issues to be addressed, including frequency of rotation of cleaners and sanitizers and adjustment of sanitation SOPs to prevent contamination. Addressing these provides an understanding of the microbiome of a facility, the potential impacts on food safety and quality, and ways to design more effective cleaning and sanitation practices.

T5-02 Whole Genome Sequencing for Source Tracking: A Validation Approach of the End-to-End Workflow for *Listeria monocytogenes* and *Salmonella enterica*

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Introduction: Whole genome sequencing (WGS) is increasingly being used for source tracking, pathogen surveillance, and outbreak investigation due to its high

discriminatory power. In the food industry, WGS used for source tracking is beneficial in order to support contamination investigations. Despite its increased use, no standards or guidelines are available today for the use of WGS in outbreak and/or trace-back investigations. The differences between genomes identified by WGS need to be trusted and a validation of all steps of the WGS workflow is therefore recommended.

Purpose: Here, we present a validation of an end-to-end WGS workflow for *Listeria monocytogenes* and *Salmonella enterica*.

Methods: The WGS workflow consisted of subculturing the isolates, DNA extraction (QIAamp DNA Mini kit, Qiagen), sequencing (Illumina MiSeq), and bioinformatics analysis (CFSAN SNP Pipeline v.1.0.0 and U.S. Food and Drug Administration). The validation was done by the assessment of the performance criteria: stability, repeatability, reproducibility, discriminatory power, and epidemiological concordance.

Results: The current study showed that few single nucleotide polymorphisms were observed for *L. monocytogenes* and *S. enterica* when comparing isolate sequences derived from the same subculture and between isolates after ten subcultures. Consequently, the stability of the WGS workflow for *L. monocytogenes* and *S. enterica* was demonstrated, despite the few genomic variations that can occur during subculturing steps, as well as repeatability and reproducibility. The WGS workflow was shown to have a high discriminatory power and has the ability to show genetic relatedness. Additionally, the WGS workflow was able to reproduce published outbreak results, illustrating its capability to show epidemiological concordance.

Significance: The current study proposes a validation approach comprising all steps of a WGS workflow and demonstrates that the workflow can be applied to *L. monocytogenes* or *S. enterica*. This work is one of the first steps in the harmonization of WGS methodologies for source tracking.

T5-03 Development of Abiotic Bacterial Surrogates for Various Sanitation Processes

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Introduction: Proper sanitation is a vital part of an integrated food safety and quality system. Existing sanitation verification methods have significant challenges that impede their effectiveness and create an opportunity for a novel approach. Advances in bioengineering have produced a material that can enable an efficient, effective, and low-cost sanitation verification method.

Purpose: This study developed abiotic bacterial surrogates to be used in the verification of sanitation processes.

Methods: Abiotic bacterial surrogates were developed by encapsulating short, unique, and naturally occurring DNA sequences (derived from *Micromonas commode* and *Ostreococcus lucimarinus*) into food-grade materials. The resulting particles were tuned to match various properties (size, hydrophobicity, etc.) of the pathogens studied (*Escherichia coli* O157, *Shigella dysenteriae*, *Salmonella* Enteritidis, and *Listeria monocytogenes*). The stability in sanitizing solutions (water supplemented with different level of chlorine and various commercial products) and attachment to stainless steel surfaces were compared to the behaviors of the pathogens. The bacterial counts were assessed using traditional microbial methods, while the surrogate counts were assessed by qPCR.

Results: The particle size measurements revealed that the surrogates had similar sizes to bacteria. Their stability under various sanitation processes was also found similar

to that of bacteria. The washing of the inoculated stainless steel surfaces also revealed that all evaluated sanitizers reduced the concentration of both surrogates and bacteria similarly. The surrogates were detected and quantified in approximately 15 minutes, while the bacterial tests took 24 hours.

Significance: The behavior of these abiotic bacterial surrogates under sanitation of stainless steel surfaces can be used to predict the lethality of the wash on the bacteria tested. The surrogates can be used to develop a sanitation verification solution that is specific, rapid, and overcomes sampling errors. Future work will focus on their use in equipment and facility sanitation, as well as various produce wash systems.

T5-04 Surveillance of *Salmonella* Prevalence in Animal Food and Characterization of the *Salmonella* Isolates by Serotyping

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Introduction: Animal food (pet food, animal feeds, and ingredients) can become contaminated with *Salmonella*. This contaminant may present a hazard to animal health by consumption of the animal food and to human health by consumption of animal-derived human food or by exposure to a contaminated animal food.

Purpose: The U.S. Food and Drug Administration's (FDA) Center for Veterinary Medicine conducts surveillance of *Salmonella* contamination in animal food. The objective of surveillance is to monitor the trend and sources of *Salmonella* contamination.

Methods: The FDA randomly collected samples of pet food, pet treats, nutritional supplements for pets, finished complete animal feeds for poultry and livestock, and ingredients at manufacturers, distributors, wholesalers, or retailers in the United States or at United States ports of entry. The FDA tested the samples for the presence of *Salmonella* and serotyped the *Salmonella* isolates isolated from the *Salmonella*-positive samples.

Results: Of the 2,058 samples collected between 2002 and 2009, 259 were positive for *Salmonella* (12.5%), while of the 2,963 samples collected between 2010-2014, 151 were positive for *Salmonella* (5.1%). Based on these findings we were able to conclude the following: Pet food and ingredients had the most significant *Salmonella* reduction, and the most common *Salmonella* serotypes found in humans were seldom found in animal food, although some *Salmonella* serotypes found in animal food were found in humans.

Significance: It is our hope that the information provided in the reports generated from these two time periods can be used by the regulated industry to address *Salmonella* contamination problems in manufacturing facilities and can be used as a source of educational information for pet owners on handling of pet food and treats at home to prevent salmonellosis.

T5-05 The Effect of Phage Presence on PFGE and WGS in *Listeria monocytogenes*: Who is Right?

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Introduction: Though *Listeria monocytogenes* is involved in only a minority of foodborne outbreaks, the number of reported deaths in Europe is much higher than other foodborne pathogens. The typing of *Listeria* in food and clinical isolates is therefore very important, and most public health labs are switching from pulse field gel electrophoresis (PFGE) to whole genome sequencing (WGS) methods. During this transition, both methods are analyzed next to

each other and sometimes discrepancies between both methods are detected.

Purpose: Isolates with different PFGE types and identical WGS type are difficult to explain. Which is correct in these cases? Can the isolates be considered as originating from the same source or not?

Methods: At the Dairy Science Laboratory in Celbridge, Ireland, all *Listeria monocytogenes* isolates are typed with both PFGE and whole genome multi locus sequence typing (wgMLST) and all results are analyzed and stored in a database using BioNumerics 7.6.2. During an outbreak investigation, six isolates were found to belong to two PFGE types, though they were indistinguishable with wgMLST. We further investigated these isolates using the following tools: SNP (single nucleotide polymorphism) analysis, an alignment of the de novo assemblies, and a cluster analysis based on absence or presence of gene content.

Results: The results of the additional analyses showed the presence or absence of a phage correlated with the PFGE type. This phage caused a shift of one of the bands on the profile, resulting in a difference of two bands between both profiles.

Significance: The presence of a phage has a much higher influence on PFGE than on most WGS-based typing methods and as a result, potential outbreaks with differential presence of a phage can be easily missed with PFGE alone. The additional information provided by WGS may help solve the case.

T5-06 A Bi-Phasic Model to Predict Heat Inactivation of *Salmonella* in Low-moisture Foods

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Introduction: Whilst *Salmonella* is not considered to be a heat-resistant organism in high-moisture foods, its resistance to heating is greatly increased in low-moisture environments.

Purpose: Predict the time to 6-log reduction of *Salmonella* in low-moisture foods in order to give guidance to food process managers for a proper heat-treatment regime.

Methods: A total of 31 design products were tested for heat inactivation of *Salmonella* Napoli. A non-linear bi-phasic Bayesian model was fitted to the data to predict the concentration of *Salmonella* over time. The model includes four parameters: one for the initial concentration, two time constants for decay, and accompanying fractions for decay. Both biological variability and experimental uncertainty in *Salmonella* heat inactivation are included in the Bayesian model set-up.

Results: The results show a non-linear effect on pH in combination with a_w, sucrose-, NaCl-, and oil concentrations at different temperatures.

Significance: Give accurate predictions for heat-treatment regimes in factories where low-moisture foods are produced.

THURSDAY, 26 APRIL — 13.30 – 15.00

T6 Technical Session 6 – Intervention Strategies and Management

T6-01 Innovative Strategy to Improve the Food Safety Standards in the Emirates of Dubai – Happiness Inspection Team

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Introduction: There are more than 17,000 food establishments that serve more than 200 different cuisines in Dubai. The Food Safety Department of Dubai Municipality regularly inspects these establishments and evaluates the businesses based on food safety standards. In 2016, the Food Safety Department evaluated 491 food companies and outlets as "Excellent", while 4,751 others were evaluated as "Very

Good", 1,142 as "Unsatisfactory", and the remainder as "Acceptable". Furthermore, 450 "Unsatisfactory" food establishments were given the same status in the previous three consecutive years and required special intervention to meet regulatory standards.

Purpose: The main purpose of this study was to evaluate the effectiveness of the "Happiness Inspection" initiative in improving food safety standards of establishments evaluated as "Unsatisfactory".

Methods: Initial data was retrieved from Dubai Municipality Food Inspection System (Accela) for 450 locations to determine the nonconformities. Following data analysis, these locations were visited to establish the root cause of the nonconformities. Regular meetings and frequent follow-ups with the establishment management resulted in gradual improvement in hygiene practices. After achieving the required level of compliance, locations were scheduled back to regular routine inspections.

Results: Two-hundred premises were inspected in the first six months. The data was statistically analyzed (Student's *t* test) and a significant improvement ($P < 0.05$) was observed in 96 locations, with food safety standards, i.e., cross-contamination, cleaning, disinfection, and pest control, being considerably improved. Post-training, routine inspection reports showed significant improvement in the grade and color cards issued to these locations. In addition to the improvements, the initiative also helped to understand and improve shortcomings in the regular routine inspection system.

Significance: The initiative assisted regulatory authorities in planning new specialized training to improve the inspection approach and enhance food hygiene practices inside establishments by proper coaching and guidance for establishment management, restructuring the layout, and reduction in menu items as per the capacity of operations.

T6-02 FSMA Regulatory Audit: A European Experience, in Italian Manufacturers

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Introduction: From May 2016 to January 2018, ITA Corporation's Food Safety Modernization Act (FSMA) Regulatory Auditors have performed 12 FSMA regulatory audits in Italy on different Italian food manufacturers of cheese, olive oil, canned vegetables, ice cream powder, coffee powder, and flour.

Purpose: The purpose of each regulatory audit was to verify the real understanding and implementation of FSMA and other U.S. Food and Drug Administration (FDA) food safety regulations.

Methods: We developed a specific checklist software, following the standardized curriculum created by the Food Safety Preventive Controls Alliance (FSPCA) and the 62 different sectors linked to food categories covered by FDA jurisdiction, as indicated by ANSI. The regulatory audit was one to one-and-a-half days long, based on the complexity of the manufacturer's plant.

Results: We obtained a statistical index of compliance with FSMA and FDA regulations. A full 100% of all companies audited did not know the Hazard Analysis Critical Control Points (HACCP) and Current Good Manufacturing Practice (c-GMP) regulations introduced by 21 CFR Part. 117; 65% of audited companies developed an inaccurate Hazard Analysis and Risk-based Preventive Controls (HARPC) plan; and 50% were not in compliance with Food Allergen Labeling and Consumer Protection Act (FALCPA) regulations.

Significance: Foreign food companies need a better understanding of the new c-GMPs introduced by the Preventive Controls for Human Food Rule, and also of the FDA HACCP regulation as applied from the European point of view. The official training offered the FDA FSMA Training Network is

a very important tool. Thanks to this regulatory audit, four companies investigated in November 2017 by an official FDA investigator received zero non-conformities.

T6-03 Development of Food Safety Interventions Using a Patient-centred Approach to Reduce the Risk of Foodborne Illness among Patients Receiving Chemotherapy Treatment

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Introduction: Chemotherapy patients have an increased risk of foodborne illness due to immunosuppression; indeed, risk of listeriosis is five times greater in chemotherapy patients. Consequently, ensuring food safety at home is essential. However, it is suggested that limited food-safety information is available to chemotherapy patients/family-caregivers in the United Kingdom (UK), and data on food-safety practices during chemotherapy are particularly lacking.

Purpose: To explore current food-safety practices and preferences to inform the development of targeted food-safety interventions to aid patients/family-caregivers in the implementation of risk-reducing food safety practices.

Methods: A review of UK food-safety information, along with a consumer-oriented approach involving interviews, self-complete questionnaires, and focus groups, allowed for the design, development, and evaluation of a targeted food safety education strategy.

Results: A review of food-related information available to chemotherapy patients obtained from 42 of 141 National Health Service chemotherapy providers established that many failed to highlight the importance of food safety in preventing infection; considerable gaps exist and information varied between sources. In-depth interviews ($n=15$) determined food-safety information during chemotherapy was considered to be inconsistent, insufficient, and particularly sought-after. Self-complete questionnaires ($n=172$) determined that despite increased awareness of the importance of food safety, malpractices were reported and perceived risks were underestimated, particularly among patients. During chemotherapy, information on 'keeping active' and 'healthy eating' were significantly ($P < 0.05$) more available than on 'food safety'. Focus groups ($n=23$) enabled the design, development, and evaluation of food-safety education interventions. Evaluation by patients/family-caregivers determined the interventions to be acceptable, and they were effective in increasing knowledge and improving attitudes regarding food safety during chemotherapy.

Significance: This project has informed the design, development, and evaluation of targeted food-safety interventions using a patient-oriented approach. This, alongside input from food-safety experts, has resulted in tailored resources that may help to reduce the risk of foodborne illness among patients undergoing chemotherapy treatment. The interventions require piloting in healthcare environments to assess acceptability and effectiveness.

T6-04* The (FAO/WHO) International Food Safety Authorities Network (INFOSAN): How Responsive Were Members during Food Safety Emergencies between 2010 and 2016?

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Introduction: The International Food Safety Authorities Network (INFOSAN) is the global network of national food safety authorities, managed jointly by the Food and Agriculture Organization of the United Nations and the

World Health Organization (WHO). One of the main aims of INFOSAN is to facilitate rapid exchange of information across borders when internationally traded food products are identified as unsafe for human consumption. The INFOSAN Secretariat regularly engages with INFOSAN Emergency Contact Points (ECPs), urgently requesting information about potential food safety events of international concern.

Purpose: The purpose of this study was to examine how responsive INFOSAN ECPs have been to requests for information from the INFOSAN Secretariat at WHO from 2010 to 2016.

Methods: The INFOSAN Secretariat reviewed all requests for information that were sent between 2010 and 2016 and calculated the duration between the date of request and the date of acknowledgement, as well as between the date of request and the date that an answer was provided.

Results: INFOSAN ECPs acknowledged receipt of the request within the stated deadline of 24 hours for 42% (184 of 383) of requests between 2010 and 2016. More importantly, INFOSAN ECPs provided a detailed answer in response to the request for 69% (265 of 383) of requests made between 2010 and 2016. The average number of days elapsed between initial request for information and receipt of the answer was seven days. However, in 2016, this response time decreased to three days. Variations across the six WHO regions were also documented.

Significance: Timely communication and application of risk-management measures is extremely important during international food safety events in order to prevent foodborne illness. This study demonstrated the responsiveness of INFOSAN members during international food safety events, highlighting strengths of the network and areas for improvement. Regionally targeted efforts of the INFOSAN Secretariat to improve responsiveness and strengthen the INFOSAN Community of Practice may account for regional variations.

T6-05 1H NMR-based Metabolomics Approach to Study the Toxic Effects of Phoxim on Crucian Carp (*Carassius auratus gibelio*)

Hong Li, XIAOYU LIU and Si Li
Huazhong Agricultural University, Wuhan, China

Introduction: Phoxim, one of the most widely used agricultural insecticides, has been reported to have high toxicity to aquatic animals.

Purpose: In this study, a ¹H nuclear magnetic resonance-based (¹H-NMR) metabolomics approach was applied to investigate the toxic effects of phoxim on crucian carp (*Carassius auratus gibelio*).

Methods: ¹H-NMR profiling combined with three kinds of multivariate statistical pattern recognition methods such as orthogonal partial least squares discriminant analysis, partial least squares discriminant analysis, and principal component analysis was developed to discern metabolite changes occurring after two weeks of phoxim exposure in crucian carp.

Results: Compared with the control group, the metabolites in crucian carp serum sampled from the phoxim group presented a significantly increased concentration of 3-hydroxybutyrate, glucose, and phenylalanine, and a significant decrease of glutamic acid, alanine, asparagine, histidine, lactic acid, glycerol phosphate of choline, phosphoric acid, choline, choline and unsaturated fatty acids, lipids, guanine nucleoside, and inosine ($P < 0.05$). The metabolic changes that were related to the toxic effects of phoxim including oxidative stress, disordered energy and amino acids metabolism, and disturbance of neurotransmitter balance.

Significance: This integrated metabolomics approach provided a molecular basis underlying the toxicity of phoxim and demonstrated that metabolomics was a powerful and highly effective approach to elucidate the toxicity of herbicides and pesticides, which will be applicable to their risk assessments.

T6-06* Effects of Food Safety Training on Achieving Food Safety Knowledge and Practices in Restaurants in the Emirates of Dubai

ABDUL AZEEZ MULLATTU EBRAHIM

M R S International Food Consultants, Dubai, United Arab Emirates

Introduction: Food safety is a public health priority due to the increasing number of meals eaten outside the home. Based on foodborne illness data, prevention is a significant concern and a public health priority in the United Arab Emirates (UAE). A significant proportion of foodborne illness cases are traced back to restaurants.

Purpose: The purpose of the study was to evaluate the effectiveness of using demonstrations in training sessions to improve food safety knowledge and food-handling practices. As part of this goal, the study also evaluated the current food hygiene practices in Dubai restaurants based on well-established current good practices (cGPs).

Methods: A quantitative research design was adopted. The population for this study was food handlers in commercial independent and chain restaurants licensed to sell food in Deira and Bur Dubai. Using a systematic random sample, all restaurant employees were invited to participate in the study. A control group and an intervention group were used to test the internal validity of the training effectiveness. Six independent restaurants were invited to participate voluntarily as the control group, which did not receive food safety trainings. Eight other independent restaurants were also invited as the intervention group and also did not attend any food safety trainings.

Results: The study result shows that training is an effective way to improve compliance with food safety guidelines. Gaps in restaurant food safety include: (i) time-temperature control, (ii) improper hygiene, and (iii) cross-contamination.

Significance: This study evaluates the effectiveness of demonstrations involving use of thermometers in hand washings, among other hands-on hygiene activities, during food safety training programs for food handlers. This study could lead to improved attitudes towards food safety and improved good hygiene practices of employees in food service and food safety-related paths.

THURSDAY, 26 APRIL— 15.30 – 17.00

T7 Technical Session 7 – Meat and Poultry, Seafood, Epidemiology

T7-01 Does Sodium Reduction Compromise the Food Safety of Cooked Ham?

Cristina Serra-Castelló¹, Anna Jofré²,
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¹IRTA, Food Safety Programme, Monells, Spain,
²IRTA, Food Safety Programme, Monells, Spain

Introduction: The reduction of sodium in order to nutritionally improve food products is an increasing trend in the meat industry, though it may compromise food safety because an antimicrobial hurdle is minimised.

Purpose: The aim of the study was to assess the safe shelf life of cooked ham, comparing standard and sodium-reduced products from four different commercial brands.

Methods: The growth rate of *Listeria monocytogenes* was empirically quantified through challenge tests (with either *L. monocytogenes* 12MOB089LM or *L. monocytogenes* CTC1034) in sliced products stored vacuum packaged at 7°C for up to two months. Additionally, commercial products were characterised for pH, a_w , moisture, sodium, and organic acid concentrations and the growth rate predicted by the Food Spoilage and Safety Predictor (FSSP) model.

Results: The growth of *L. monocytogenes* was highly affected by the brand formulation and the strain. The 12MOB089LM growth rate (μ_{max} from 0 to 0.059 h⁻¹) was 30 to 50% faster in sodium-reduced cooked hams in comparison with their standard counterparts, with the consequent safe shelf-life reduction. The CTC1034 strain was able to grow

in all products (μ_{max} from 0.014 to 0.060 h⁻¹), being less sensitive to salt and organic acids. The predictions provided by the FSSP were in agreement with the results obtained with the 12MOB089LM in standard formulated cooked ham (only 6% bias), whereas a considerable underestimation of the growth of *L. monocytogenes* was recorded in sodium-reduced cooked ham (65% bias).

Significance: The work highlights the importance of the strain selection to perform challenge tests and the proper product characterisation to enable accurate predictions. Furthermore, a calibration factor could be applied in order to use the predictive tool for the design of safe reduced-salt formulations for cooked meat products.

T7-02 Inter- and Intra-generic Interaction between Meat Plant Environmental Bacteria and *Escherichia coli* O157:H7 in Co-culture Biofilms

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Introduction: Bacterial isolates belonging to very diverse genera have been isolated from meat plant environments, many of which may form biofilms and in turn affect the persistence of pathogens like *Escherichia coli* O157:H7.

Purpose: To examine the influence of meat plant environmental bacteria recovered from beef packing plant equipment on biofilm formation of *E. coli* O157:H7.

Methods: Biofilm formation of bacterial isolates belonging to 41 genera consisting of 18 gram negative aerobes (GNA), eight gram positive aerobes (GPA), five lactic acid bacteria (LAB), nine *Enterobacteriaceae* (ENT), and ten generic *E. coli* (GEC) was tested alone or in co-cultures with *E. coli* O157:H7 at 15°C for six days. Biofilms were quantified using crystal violet (CV) staining on days 2, 4, and 6. Numbers of *E. coli* O157 in mono- or co-culture biofilms with *Acinetobacter* sp., *Sphingopyxis* sp., *Carnobacterium* sp., and *E. coli* genotype 136 were obtained by plating on selective agar.

Results: *E. coli* O157:H7 did not form detectable biofilm as determined by CV staining for up to 6 days, while ≥ 61 , ≥ 12.5 , ≥ 20 , ≥ 56 , and 80% GNA, GPA, LAB, ENT, and GEC, respectively, formed biofilms either alone or when cultured with *E. coli* O157:H7. The frequency of no, synergistic, or antagonistic effect in co-culture biofilms varied significantly ($P < 0.05$) within, as well as between, each group of these organisms. In monoculture biofilm, the numbers of *E. coli* O157:H7, *Acinetobacter* sp., *Sphingopyxis* sp., *Carnobacterium* sp., and *E. coli* genotype 136 were 7.1, 6.3, 7.2, 6.8, and 7.7 log CFU at day 4. In co-culture biofilms, *E. coli* genotype 136 significantly ($P < 0.05$) reduced the number of *E. coli* O157:H7 by 1.7 log CFU, while others did not affect the numbers of this pathogen.

Significance: The finding shows that generic *E. coli* may outcompete *E. coli* O157:H7 while establishing biofilm in meat processing environments.

T7-03 Two Studies Show Association between Consumption of Dry Raw Pork Sausages and Hepatitis E in The Netherlands

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Introduction: The number of hepatitis E patients has recently increased in The Netherlands, as in other European countries. Although swine a known reservoir for hepatitis

E virus (HEV), causes of the (re-)emergence of HEV and exact transmission routes of HEV are currently unknown.

Purpose: To identify risk factors for HEV exposure and infections in the Netherlands, two studies were performed among: (i) acute hepatitis E patients, and (ii) blood donors.

Methods: A questionnaire on potential risk factors for HEV exposure, health, and socio-demographic characteristics was completed by: (i) 283 patients with laboratory-confirmed acute hepatitis, enrolled through 23 medical microbiological laboratories (June 2015 through June 2017) and 972 control persons matched for age, gender and region of residence; and (ii) 1,562 healthy blood donors from all over The Netherlands (March through May 2016) aged 18 to 70 years whose plasma samples were tested with Wantai EIA for anti-HEV IgG antibodies.

Results: Hepatitis E infection in The Netherlands was associated with consumption of traditional Dutch dry-fermented raw pork sausages, which are generally consumed sliced unheated on bread. According to multivariate analyses adjusting for age and gender: (i) Patients with acute hepatitis E were more likely than population controls to report consumption of "sliced sausage" (aOR 2.6; 95% confidence interval [CI], 1.6 to 4.3), "farmer sausage" (aOR 2.4; 95% CI, 1.5 to 3.6), or "cervelaat" (aOR 2.3; 95% CI, 1.5 to 3.4); (ii) HEV-IgG-seropositive blood donors were more likely to report consumption of dry sausages called "cervelaat", "fijnkost", "salami", and "salametti" (combined aOR 1.5; 95% CI, 1.2 to 1.9). HEV-IgG-seroprevalence was 31% and increased with age.

Significance: Two studies show that several dry raw pork sausages are associated with HEV exposure and infections in The Netherlands. The prevalence and infectivity of HEV in these products should be investigated, as well as the production methods and possible origin of HEV-contamination within these sausages, e.g., small amounts of pork liver.

T7-04 Don't Wash My Chicken!? Identifying Barriers to Consumers Adopting the Practice of Not Washing Raw Poultry

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Drexel University, Philadelphia, PA

Introduction: Efforts to educate consumers against washing raw poultry found that some consumers practice this risky behavior in spite of such education. There is a need to better understand why consumers do not accept the message to not wash raw poultry.

Purpose: The purpose of this research was to identify perceived barriers that exist which prevent consumers from adopting the appropriate behavior of not washing raw poultry.

Methods: A thematic analysis was performed using consumer comments on websites which promoted the "Don't Wash Your Chicken!" campaign. Three search engines were queried for sites where the message was promoted. Consumer comments from 50 websites were analyzed from the 161 websites identified. Comments were thematically analyzed using both traditional thematic analysis and NVivo qualitative analysis software. More than 4,000 comments were collected and cleaned to eliminate usernames, icons, numbers, etc. Comments were categorized into four groups which included: (1) Agreed with Message, (2) Disagreed with Message, (3) Irrelevant Comments, and (4) Unclear Opinion.

Results: Thematic analysis of data indicated that comments that "disagreed" with the message outnumbered comments that "agreed" with the message. Analysis identified as themes among those who "disagreed" with the message: (1) the need to "clean" something off the chicken, and (2) the practice of washing poultry with acid rinsing solutions. Analysis with NVivo qualitative software identified top contaminants consumers thought they needed to clean off raw poultry as blood, feces, fat, slime, feathers, and dirt. Common solutions that consumers reported using to wash their raw chicken included vinegar, lemon, lime, or alcohol.

Significance: It is anticipated that the identified themes provide a better understanding of barriers to consumers adopting the practice of not washing raw poultry and may inform future education campaigns around this topic.

T7-05* Deriving Personalized Recommendations for Fish Intake Using Mathematical Optimization Methods

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Introduction: Consumption patterns in a population often vary greatly and national dietary guidelines may differ too much from individual preferences to be realistic. Providing personalized recommended intakes may be a step towards increasing adherence to dietary guidelines.

Purpose: We developed a method for modelling achievable individual dietary recommendations based on personal preferences. The method is applied in a model on fish intake in Denmark.

Methods: A mathematical optimization model that applies quadratic programming was developed to model personalized recommendations for fish intake, fulfilling criteria on nutrients and contaminants, while simultaneously deviating as little as possible from observed individual intake. Model constraints ensured that modelled fish intake levels met the recommendations for EPA, DHA, and vitamin D without violating the tolerable intake recommendations for methyl mercury, dioxins, and dioxin-like PCBs. Recommended intakes for 11 species were generated for each individual in a group of 3,016 Danes (1,552 women and 1,464 men, aged 18 to 75 years) whose fish intakes and body weights were recorded from a national dietary survey. Background intakes of the nutrients and contaminants in question from foods other than fish, supplements, and environmental exposure were analyzed.

Results: Our results on the fish intake case suggest that 2% of the 3,016 Danes should be recommended to decrease their fish intake, 55% should increase their fish intake with up to 184 g/week, and 24% should increase their fish intake with more than 100 g/week. These recommendations were different from the observed intakes ($P < 0.05$) according to the Wilcoxon matched-pairs signed-rank test. The results appeared to be specifically sensitive to the uncertainty on vitamin D levels due to the effect of exposure from the sun.

Significance: Mathematical optimization methods could be used to provide more realistic and achievable dietary guidelines that use data from nutrition science and account for personal preference.

T7-06 Attributing STEC Infections to Food Sources to Inform International Food Standards: Results of an International Consultation and Evidence Synthesis

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Introduction: Shiga toxin-producing *Escherichia coli* (STEC) infections pose a substantial health burden worldwide. Circa 2010, STEC infections transmitted via food (as opposed to

other routes of transmission) caused more than 1 million human illnesses and nearly 13,000 Disability Adjusted Life Years (DALYs) globally. To appropriately target interventions to prevent these infections, it is important to determine the types of foods leading to these illnesses.

Purpose: Our objective was to determine the food sources of STEC infections, both globally and for the six World Health Organization (WHO) regions (i.e., the Americas, Europe, the Eastern Mediterranean, Africa, Southeast Asia, and the Western Pacific).

Methods: We used data from outbreaks and case-control studies of sporadic infections to estimate the fractions of STEC infections that can be attributed to specific foods. We sought data on all outbreaks of STEC that have occurred globally, via WHO points of contact within regions and member states, and conducted a systematic review to identify case-control studies of sporadic infection in both the peer-reviewed and grey literature. The two data sets were modelled using a stochastic model and a meta-analysis, respectively.

Results: Our results show that produce, beef, and dairy products were the most important sources of STEC globally. While beef was identified as the most frequent food category attributed in the African, Americas, European, and Eastern Mediterranean regions, analysis of the outbreak data indicated that fresh produce (i.e., fruits and vegetables) were almost as frequent in North America and Europe.

Significance: Our results show that, while beef as a food remains a key source of STEC illness, other commodities such as vegetables and dairy are also important. Addressing the potential for STEC infection to be transmitted by these foods products will be important to lowering the global burden of foodborne STEC infections.

FRIDAY, 27, APRIL — 8.30 – 10.00

T8 Technical Session 8 – Detection and Typing Methods

T8-01 Genome Diversity of *Salmonella enterica* subsp. *enterica* serotype Derby from Animal to Human: Sporadic Cases Source Attribution Study and Specific Host-association Identification

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Introduction: In the European Union (EU), *Salmonella enterica* subsp. *enterica* serotype Derby is the most abundant serotype isolated from pork meat. In France, this pathogen is mainly isolated from pork and poultry meat and since 2000 has ranked between the 5th and 8th most frequently isolated serotypes in humans. Despite this significant threat to human health, few studies have focused on the genetic diversity of this pathogen and all suffer from the low discriminatory power of the sub-typing method used or the lack of strains.

Purpose: This study aims to characterize *S. Derby* diversity within a collection of 442 strains comprising all human cases recorded in France between 2014 and 2015 ($n=302$) and a representative selection of the pork and poultry sectors ($n=140$).

Methods: Phylogenetic analysis was conducted by single nucleotide polymorphism (SNP) and Roary analysis on the core and accessory genome, respectively. Antimicrobial resistance profiles were investigated at both genomic

and phenotypic levels. *Salmonella* pathogenicity islands (SPI) and *FimH* gene sequences were compared using BioNumerics software.

Results: Derby serotype is shown to be polyphyletic with four genomic lineages (ST40, ST39, ST71, and ST682) at a distance of 15,000 SNP on average. The lineage ST40, characterized by resistance to aminoglycosides, sulfonamides, and tetracyclines, and the presence of SPI-23, contains 71% (213 of 302) of the human strains. The human cases were associated with food sources by statistical analysis, revealing the spread of this major lineage both geographically and throughout the pork sectors. Only lineage ST71 was associated with the poultry sector and included only 2% (6 of 302) of the human cases.

Significance: Recognition of these four lineages is of crucial importance for epidemiological surveillance along food production chains and is the first step in refining, monitoring, and preventing dissemination of this pathogen.

T8-02 Rapid *Salmonella* Serotyping Via Targeted Amplicon Sequencing

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Introduction: *Salmonella* serotyping is the “universal language” of *Salmonella* among diagnostic laboratories, public health, and regulatory agencies. While *Salmonella* test kits (molecular, phage, antibody, etc.) only screen for the presence of over 2,600 known *Salmonella* spp., traditional serotyping allows for specific identification of *Salmonella* using agglutination reactions. Serotyping is an important tool in a food safety program to identify and eliminate sources of *Salmonella* in a plant and is also used by government agencies in outbreak investigations. Serotyping can also help a food plant determine if a strain is resident or transient, as it may require different root cause investigations and remediations.

Purpose: To circumvent issues with expensive and labile reagents, subjective results, and possible lack of protein expression, a molecular approach, NeoSeek *Salmonella* serotyping, was collaboratively developed using general *Salmonella* targets, a specific region identified by Dr. Jean Guard (United States Department of Agriculture – Agricultural Research Service) and bioinformatics pipelines and algorithms developed by MetaGenome Analytics.

Methods: The method utilizes targeted amplicon sequencing on a four-region MiSeq within the *Salmonella* genome of isolates. Assembled sequences are compared to a curated database of whole-genome and individual-target sequences. Serotype assignment uses a sophisticated scoring matrix for serotype determination using the Kauffmann-White scheme.

Results: Serotype call accuracy was first examined in a single-blind study with strains that included, but were not limited to, the Centers for Disease Control and Prevention’s “Top 30” and was 80% accurate ($n=70$), while the traditional antibody-based agglutination method was 88% accurate. Optimization of the scoring algorithm increased the accuracy to 96%. A blinded sample set from a collaborator ($n=25$) was submitted for validation and, again, showed 96% accuracy versus 88% accuracy for the traditional method.

Significance: This method accurately reports *Salmonella* serotypes from isolates using targeted amplicon sequencing. The shorter time to result, 4 to 5 days versus weeks, provides for faster responses that lead to quicker interventions than traditional methods.

T8-03 Comparison of Typing Reference Methods and Whole Genome Sequencing Analyses for the Characterization of *S. aureus* Strains Isolated from Food Outbreaks

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¹Anses, Maisons-Alfort, France, ²ANSES, Laboratory for Food Safety, University of Paris-Est, Maisons-Alfort, France, ³Université Paris-Est, Anses, Maisons-Alfort, France, ⁴Université Paris-Est, ANSES, Maisons-Alfort, France

Introduction: Due to its ability to produce enterotoxins, *Staphylococcus aureus* is a leading cause of staphylococcal food poisoning outbreak (SFPO). Absorption of these toxins could induce severe diarrhea and vomiting. The European Union Reference Laboratory (EURL) has been involved in the development of methods for SFPO investigation. Currently, the detection of five toxins is possible from the food matrix and the detection of 11 enterotoxin genes from the individual strains. Both detections confirm the involvement of the isolated strains in the SFPO. Outbreak source tracking is performed through pulsed field gel electrophoresis (PFGE); it is supposed that strains sharing the indistinguishable PFGE profile are probably related.

Purpose: The development of whole genome sequencing (WGS) allows access to a lot of data and could replace current typing methods (enterotoxin gene detection and PFGE). Comparisons of results obtained by these different methods were performed.

Methods: From a selection of reference strains and strains isolated from outbreaks, 143 genomes have been sequenced using illumina Technology. Firstly, toxic profiles obtained by real-time PCR were compared to those obtained with a blast tool as part of an in-house workflow. Secondly, correlation between PFGE profile families obtained with BioNumerics and core genome obtained with Roary was studied. For this, a phylogenetic tree was built based on maximum likelihood and using RAxML.

Results: Our results showed that WGS approaches and EURL reference methods were consistent. Toxic profiles were similar with blast approach and real-time PCR. PFGE profile families seem to follow the clustering obtained by core genome analysis.

Significance: The results are encouraging to validate the development and the use of new WGS tools for foodborne strain characterization. Furthermore, the WGS results highlighted the robustness of the reference methods, more easily applicable by the laboratories of the EU Members States network for the investigation of food poisoning outbreaks.

T8-04 Bacteriophage Receptor Binding Proteins for the Isolation of *Yersinia enterocolitica* in Foods

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Introduction: The isolation of *Yersinia enterocolitica*, a zoonotic agent causing self-limiting gastroenteritis known as yersiniosis in humans, poses a major challenge due to poor sensitivity of culture methods.

Purpose: Since bacteriophage receptor binding proteins (RBPs) mediate specific attachments to bacterial host surfaces, this study investigated their use as capture

molecules for selective isolation of epidemiologically significant *Y. enterocolitica*.

Methods: RBPs Gp47, Gp17, and Gp37 with affinity for serotypes O:3, O:5,27, O:8, and O:9 were identified through characterization of *Y. enterocolitica* bacteriophages ϕ 80-18, vB_YenP_AP5, and vB_YenM_TG1 based on genome sequences, host range, and receptor specificity. RBPs produced by expression in *Escherichia coli* were purified and functionalized onto microparticles for use as capture ligands in magnetic separation. Target-bead complexes were collected and plated onto CIN and CAY agars (30°C for 48 h) followed by MALDI-TOF mass spectrometry for rapid identification of suspect colonies (RBP-MS). As a proof of concept, RBP-MS was applied to suspensions of 160 *Yersinia* sp. and 20 non-*Yersinia* sp. strains. Also, inoculated ground pork, mixed salad, and milk samples were tested with and without RBP-MS to determine the impact of RBP-MS on *Y. enterocolitica* isolation rates.

Results: Simultaneous capture of *Y. enterocolitica* O:3, O:5,27, O:8, and O:9 was attained using a mixture of Gp47 and Gp37 coated microparticles. Notably, RBP-MS in combination with CAY agar applied to the above 180 strains achieved 100% sensitivity and 95.7% specificity for virulent *Y. enterocolitica*. Also, isolation rates of *Y. enterocolitica* O:3, O:5,27, O:8, or O:9 from inoculated food samples were substantially higher with than without RBP-MS: ground pork, 17 of 36 versus 1 of 36; mixed salad, 20 of 36 versus 1 of 36; and milk, 26 of 36 versus 8 of 36.

Significance: This research demonstrates that use of bacteriophage RBPs as capture molecules represent an alternative approach for bacterial concentration as a preparative step to improve the isolation of pathogenic *Y. enterocolitica* from foods.

T8-05 Monitoring of Foodborne Viruses in Berries and Considerations on the Use of RT-PCR Methods in Surveillance

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Introduction: Foodborne viruses such as norovirus (NoV) and hepatitis A virus (HAV) are an important food safety concern and have been associated with many recent foodborne illness outbreaks linked to fresh and frozen berries worldwide. The standard method for qualitative detection of hepatitis A virus and norovirus from foods has been published in 2013 (ISO/TS 15216-2). Since then, multiple laboratories have been offering foodborne virus detection in foods (including berries) using methods based upon the ISO standard method.

Purpose: To report data on foodborne virus monitoring in berries and to discuss some remaining pitfalls for testing and interpretation of results during this type of monitoring program.

Methods: A wide range of frozen berries (2,015 samples in total) were collected by Nestlé and the European Association of Fruit and Vegetable Processors in the period between 2009 and 2016 and tested for NoV and HAV by 11 service laboratories. Additionally, in order to gather data on the potential use of human adenovirus (HAdV) as “index viruses” of human fecal contamination for monitoring purposes, 632 berry samples which had previously been tested for NoV and HAV were also tested for HAdV by Nestlé.

Results: Seven positive signals (NoV and HAV) were identified from 2,015 samples analyzed in total (0.3% [95% confidence interval: 0.2 to 0.7%]). Six HAdV positives were recorded, corresponding to a prevalence of 0.9% (95% confidence interval: 0.4 to 2.1%). Not a single case was noted in which the positives of HAdV and pathogenic viruses (NoV and HAV) were noted in the same sample.

Significance: This study provides elements to show that monitoring programs – even if providing numerous negative test results – are a useful tool to obtain baseline data, to increase awareness on this food safety issue with stakeholders, and may contribute in providing data for risk assessments.

T8-06 Method Validation for Staphylococcal Enterotoxin Detection in Food Matrices from Various Food Poisoning Outbreaks

YACINE NIA¹, Isabelle Mutel¹, Berivan Boran², Joan Ojenima² and Jacques-Antoine Hennekinne¹
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Introduction: Staphylococcal enterotoxins (SEs) represent the primary cause of food poisoning outbreaks due to bacterial toxins, according to the European Food Safety Authority. Today, two commercially available immunoassays are validated for the detection of SEs (SEA to SEE) in foods in the frame of the European Screening Method (ESM) and the new standard ISO 19020:2017. These detection assays are able to qualitatively detect five SEs (SEA to SEE), but they are able to identify neither their type nor their concentration.

Purpose: As it is well-known that interferences may occur in enzyme-linked immunosorbent assay-based (ELISA) methods, confirmation using another principle is recommended. The scope of the study was the validation of a specific in-house ELISA-based method for SE detection in food matrices according to ISO 16140-2. This method is able to detect the type of SEs present in the food.

Methods: Samples were spiked and prepared according to the ESM. Analysis of five SEs was performed using a specific indirect sandwich-type ELISA. Specific commercial antibodies were used as coating and probing antibodies. The presence of SEA-SEE was revealed by immunoglobulins coupled with horseradish peroxidase and determined by a colorimetric measurement. About 30 matrices from five food categories were tested.

Results: Sensitivity and specificity were evaluated at >90%. For application, more than 20 real samples issued from several outbreaks were analysed using the validated in-house ELISA. Satisfactory agreement was established between SE genes identified in *Staphylococcus aureus* isolates and SE type detected in the samples. However, some outbreaks were only partially characterized due to the absence of diagnostic tools allowing the detection of other types such as SEG, SEH, and SEI.

Significance: The validated method can be used as confirmatory method for SEA-SEE detection in food matrices. However, new analytical tools are needed to enlarge the scope of the methods.



POSTER ABSTRACTS

25-27 April 2018 – Stockholm, Sweden

POSTER ABSTRACTS

* Student Award Competitor

WEDNESDAY, 25 APRIL — 10.30 – 16.00

Poster Session 1 – Beverages and Water, Communication Outreach and Education, General Microbiology, Meat and Poultry, Microbial Food Spoilage, Non-microbial Food Safety, Risk Assessment, Sanitation, Seafood

P1-01 Microbiological Inspection of Mineral Water by Redox Potential Measurement

ORSOLYA ERDŐSI, Katalin Szakmár, Zsuzsanna Szili, Géza Szita and Péter Laczay
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Introduction: The redox potential measurement as a validated method is suitable for rapid microbiological testing of mineral water. The time needed for a reliable detection of microorganisms is of key importance: In the water industry, the real-time (or at least as fast as possible) monitoring of the microbiological properties of the production is indispensable. In the public water supply, the essential basis of epidemiological and public health measures is fast and reliable results of microbiological inspections.

The principle of the measurement is that the redox potential of the medium is detectably reduced over a certain microbe concentration due to the energy-producing biological oxidative processes of the bacterial growth. The selectivity depends on the selectivity of the nutrient broths.

Purpose: The microbiological requirements of mineral water are referring to the time of production. The aim of the work was to examine the changes of the microbiological status of mineral waters during storage. Still and sparkling mineral waters in different price categories were examined. The experiment covered a six-month period.

Methods: The most frequently tested contaminant microorganisms in mineral water productions are: coliforms, *Escherichia coli*, *Pseudomonas aeruginosa*, *Enterococcus faecalis*, and total count (22 and 37°C). Bottled mineral waters were examined by redox potential measurement ($n=180$, 90 still and 90 sparkling).

Results: Compared to the detection time of the conventional culture method, the microbes can be detected within 2 to 24 h using the redox potential measurement. There was a difference between the microbiological status of the still and sparkling waters.

Significance: The redox potential measurement is especially suitable for the evaluation of membrane filter methods with the use of every nutrient broth. The microbiological status of the mineral waters changes during storage, which can result in food safety problems.

P1-02* Foodborne Outbreak Communications between Government Public Health Agencies

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Introduction: Information exchange and dialogue between public health agencies is essential for rapid response to foodborne outbreaks, whether the origin is naturally occurring, food fraud, or an act of terrorism. To adequately coordinate foodborne outbreak response across multiple

areas of responsibility, clear methods of communication should be defined and followed by all agencies involved.

Purpose: Identifying processes for inter- and intra-agency communications during foodborne disease outbreaks lays a foundation for gap analysis and performance comparison and highlights areas for functional improvement. Addressing these issues can improve the two-way flow of information for food protection issues and may be applicable to other areas where federal, state, and local agencies need interactive communication.

Methods: A comparative analysis of 21 states' general operating procedures was conducted to: (i) ascertain lines and modes of communication related to foodborne outbreaks; (ii) identify references and procedures currently in use for responding to intentional contamination incidents; (iii) determine procedural commonalities; (iv) identify potential barriers; and (v) recommend enhancements for multi-directional information exchanges between health agencies.

Results: Written protocols for inter- and intra-agency coordination during food emergencies vary considerably among states. Few include provisions for intentional contamination or coordination with law enforcement. Secure, collaborative, web-based networks are not widely identified as tools for response coordination. Plans for information exchange between responding agencies and references to the Incident Command System (ICS) are limited.

Recommendations for systemic improvement include expanding rapid response teams to all states, formalizing inter- and intra-agency communication plans in every outbreak protocol, assuring response plans stipulate ICS, and enhancing funding to assure multi-agency collaborations.

Significance: The public health system needs to collaborate, conceptualize, and develop a new and enhanced aptitude for addressing the threat of foodborne outbreaks and food terrorism and integrate an enhanced communications framework. This study provides foundations for establishing coordinated policies and improving inter- and intra-agency communications in foodborne outbreak response.

P1-03 Raising Awareness of Global Food Safety Scheme Audit Requirements with Regard to Cleaning Tool and Utensil Selection and Use

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Introduction: Food industry cleaning tools and utensils have long been identified as a major source and vector of cross-contamination. Data from a study funded by the government of the United Kingdom was used to establish food industry guidance on microbiological sampling and showed that 47% of cleaning tools tested were positive for *Listeria monocytogenes*. In 2017, Schäfer determined that 67% of equipment and utensils used in a poultry processing plant were contaminated with *L. monocytogenes*, even after cleaning. Nevertheless, cleaning tools and utensils are rarely considered in relation to food safety. Fortunately, GFSI-approved food safety schemes, including those operated by BRC, FSSC, and SQF, now each draw attention to them specifically.

Purpose: To review Global Food Safety Initiative (GFSI) food safety schemes with regard to the selection and maintenance of cleaning tool and utensils, to summarise this information, and to share it with those involved with food safety.

Methods: GFSI scheme standards were reviewed and key points relating to cleaning tool and utensil selection and maintenance were summarised. Using the information obtained through various articles, training/information presentations have been produced in order to share this valuable food safety knowledge.

Results: Articles on the hygienic design and food contact compliance of cleaning tools and utensils have been published and the information presented at scientific and technical events. A training/information presentation entitled "Selection, use and maintenance of cleaning tools" has been developed and presented to food businesses, and a white paper entitled "Food safety through good cleaning tool maintenance" has been produced. This latest presentation summarises current and proposed GFSI scheme audit requirements regarding cleaning tool and utensil selection and maintenance.

Significance: The use of cleaning tools and utensils is ubiquitous in the food industry. Given their proven role as a major source and vector of contamination, knowledge sharing of ways in which they can be controlled is essential to promote food safety.

P1-04 An Identification of Potential Food Safety Risks to Athletes

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Introduction: Food safety is essential for athletes as they are reported to be susceptible to infection after acute exercise. The incidence of foodborne illness among athletes participating at events has frequently made media headlines in recent years. However, to reduce incidence, there is a need to identify the food-safety risks to athletes.

Purpose: To identify potential food-safety risks from athletes' food preparation and consumption habits.

Methods: Sports nutritionists participated in a semi-structured discussion group ($n=4$).

Results: Food-preparation and consumption habits unique to athletes that may increase the risk of foodborne illness were identified. Advanced preparation, cooking and prolonged storage were discussed in relation to limited access and awareness regarding appropriate refrigeration/re-heating facilities when training: "once the food is cooked then what to do with it... how to store it, how long these cooked foods should be stored for." The age of athletes, their level of competence and independence in food preparation and time to commit to food preparation were also identified as potential food-safety risks. Concerns regarding food provision while competing and travelling abroad were discussed: "They can be in a self-catered apartment in which case they'd be travelling to that competition or training camp, buying the food, and doing the self-catering themselves." While travelling or competing at events abroad, independence in food choices and language were identified as potential barriers to ensuring food safety. The role of sports nutritionists to inform athletes of food-safety risks and enable risk-reducing behaviours was considered: "Something that I try to instill in the athletes is just to make sure that cold food is meant to be served cold then it actually is... if it's meant to be hot then it actually is hot."

Significance: Two key areas of risk have emerged from the research that require further exploration with athletes: food preparation, storage and consumption practices during training, and food safety awareness when travelling to overseas competitions.

P1-05 Novel Approaches to Reduce Risks of *Campylobacter* Infections among Consumers

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Introduction: *Campylobacter* causes an estimated 1.3 million infections annually in the United States. Most illnesses occur due to consumption of raw or undercooked poultry. Illnesses increased by 13% in 2014, when compared to 2006 through 2008. Yet, consumers know little about *Campylobacter* control. In fact, very few consumers, including at-risk populations, have even heard of *Campylobacter*.

Purpose: The objective of this study was to use educational intervention to: (i) increase awareness and knowledge about *Campylobacter* among parents of young children, and (ii) improve food safety behaviors related to control in domestic kitchens.

Methods: Theory of planned behavior was used as a model in this study. Educational intervention was delivered over two weeks to parents of young children using: (i) traditional outreach education (systematic information processing), and (ii) social media outreach (SMO) education (heuristic information processing). Each group was randomly assigned treatments and completed a pre-post and surveys after each lesson online. Awareness, knowledge, practices, attitudes, normative beliefs, and perceived behavioral control were measured. Expanded Food and Nutrition Education Program recipients were used as a control group.

Results: *Campylobacter* risk awareness increased in both groups ($P<0.001$). Participants rated their knowledge 57.73 ± 21.88 out of 100 before and 76.63 ± 12.53 after intervention ($P<0.05$). Increase in understanding of *Campylobacter* control strategies was observed (cooking of ground meat and poultry $P<0.05$; cross-contamination from raw poultry to fresh produce $P<0.001$; preparation of turkey hot dogs, $P<0.005$). Perceived behavioral control decreased in SMO group while the concern over the disease increased ($P<0.05$). Food safety attitudes improved only in SMO group indicating higher likelihood of behavior change.

Significance: Food safety education intervention improved awareness and increased knowledge about *Campylobacter* and the adequate practices to control the risks. SMO intervention is more likely to lead to behavior changes, but has to be implemented with caution due to the possibility of lowering perceived control among parents of young children.

P1-06 Perceptions of the Welsh Food and Drink Manufacturing and Processing Industry Regarding the Potential Impact of the UK Leaving the European Union (Brexit)

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Introduction: Over the past 41 years, membership of the European Union (EU) has allowed free movement of citizens and enabled the United Kingdom (UK) to import and export products within a single market. Given that the value of food, feed and drink exports from the UK is $>£18$ bn, the Welsh Food and Drink Industry (WFDI) largely employs non-UK EU nationals as its workforce, and that UK food-related policies are derived from EU food legislation, there is a need to explore the perceived impact of Brexit in the Welsh food industry.

Purpose: To explore the perceived impact of Brexit upon food and drink manufacturing and processing businesses (FDMPBs) and the WFDI.

Methods: Online, self-completed questionnaires were distributed to Welsh FDMPBs in two phases – pre-referendum ($n=32$) and post-referendum ($n=56$), collating both quantitative and qualitative data to determine perceptions regarding Brexit.

Results: Overall, the majority of FDMPBs perceived that Brexit would have a negative impact upon FDMPBs and the WFDI. FDMPBs indicated optimistic bias by perceiving Brexit would have a greater negative impact on the WFDI than their FDMPB, as 53% perceived their company would be weaker out of the EU, whereas 67% believed the WFDI would be weaker. Comparison of findings indicated greater uncertainty that Brexit would “not have an impact” upon their FDMPB, reducing from 27% (pre-referendum) to just 6% (post-referendum) and those indicating they “did not know” what impact Brexit would have increasing from 17% (pre-referendum) to 38% (post-referendum). Qualitative data indicated that Brexit would have a potentially negative impact upon job creation, retention of staff from the EU, funding, and import/export activities.

Significance: The majority of FDMPBs anticipate a negative impact. Findings indicate a need for information provision for FDMPBs regarding the potential impact of changes to the free movement of EU citizens and trade. Additionally, further investigation regarding export opportunities to non-EU countries is required.

P1-07 Assessment of Trainee-Dietitians' Food Safety Awareness and Training Experiences in Lebanon

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Introduction: Dietitians are key gatekeepers that provide food-related information to patients and are perceived as trusted sources for food-safety information by patients; however, gaps in registered dietitians' food-safety knowledge have previously been identified. Appropriate knowledge and skills to deliver food-safety information to vulnerable patient groups can be obtained during dietetic training. Currently information detailing the training experiences and awareness of trainee-dietitians in Lebanon regarding food-safety is lacking.

Purpose: To assess trainee-dietitians' knowledge, attitudes and training experiences regarding the provision of food-safety information in Lebanon.

Methods: Paper-based questionnaires were completed by trainee-dietitians ($n=25$) at the School of Health Sciences at the Modern University for Business and Science, Beirut, Lebanon.

Results: All trainee-dietitians reported studying food-safety during their degree. Although 84% felt they had received sufficient food-safety training, 43% believed they still had more to learn to enable them to inform patients about food-safety in the home. Ninety-two-percent indicated knowledge of recommended refrigeration temperatures ($\leq 5^{\circ}\text{C}$) and 96% expressed positive attitudes towards refrigeration. Conversely, although 100% believed they knew the recommended temperature that should be achieved when cooking meat/poultry, only 24% stated the correct temperature ($>75^{\circ}\text{C}$), and 25% did not think using a meat thermometer was required while cooking. Confusion regarding date labelling was indicated with only 32% aware the “use-by” date to be the best indicator of food-safety. The majority had positive attitudes toward the role of dietitians in the provision of food-safety information to immunosuppressed patients. Positive attitudes towards the role of dietitians in reducing the risk of foodborne infection among vulnerable patients were expressed and many indicated the desire to learn more.

Significance: Although trainee-dietitians attended food-safety lectures, knowledge of key food-safety recommendations was lacking. However, knowledge of food safety does not equate to the ability to disseminate food-safety advice. There is a need for specifically targeted training for trainee-dietitians to inform patients regarding food safety.

P1-08 Food Safety Knowledge and Self-reported Practices of Parents with Young Children

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Introduction: Due to immature immune systems, young children (< 5 years) have an increased risk of foodborne illness and are subsequently associated with increased incidence. Consequently, given the association of the domestic kitchen with sporadic foodborne illness, implementation of safe-food handling and storage practices is essential when preparing food for young children.

Purpose: To determine the food-safety knowledge and self-reported practices of parents and determine preferences and trusted sources of food safety information.

Methods: Online self-completed questionnaires distributed using social media were completed by parents of children (aged < 5 years, $n=78$).

Results: Although knowledgeable in some key areas of food safety, 28% indicated they were more concerned about nutrition than food safety, and some indicated malpractices may be implemented when preparing formula: “I'd make a day's worth and store in the fridge, getting one out to room temperature after the last feed.” It was determined that 31% believed powdered infant formula to be a sterile product. Although 78% were aware of recommended refrigeration temperatures (0 to 5°C), 54% reported to ‘never’ use a thermometer to check, and only 40% reported being aware of their refrigerator temperature. Of which 87% reported to be within the safe range. Only two-thirds (67%) were aware that a ‘use-by’ date referred to the last date food may be safe to consume, and only 35% reported ‘always’ following ‘use-by’ dates; however, 91% were more likely to adhere to ‘use-by’ dates if the food was intended for a child than themselves. Concerningly, 21% indicated that raw chicken may be washed before cooking.

Significance: Although parents are knowledgeable of some key aspects of domestic food safety when preparing food to be consumed by young children, gaps exist and food-safety malpractices are reportedly implemented. Given its increased popularity, there is a need to explore the role of online parenting communities in the provision of food-safety information to parents.

P1-09 A Narrative Review of Food Safety Research Studies of Professional Food Handlers in Catering and Manufacturing Environments

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Introduction: Foodborne illness outbreaks associated with manufacturing and catering environments remain a public health concern. Consequently, the need to assess the food safety of food handlers in such food production environments is of utmost importance. However, utilised methods influence the type of data that can be measured. There is a need to consolidate international food handlers food safety data to assess cognition and behaviour.

Purpose: To review the methods and measures utilised in research studies to assess food-safety awareness and practices of professional food handlers in catering and manufacturing environments.

Methods: Professional food handler food-safety research studies ($n=20$) were identified and reviewed and the findings summarised according to assessment of knowledge, attitudes, self-reported practices, and observed behaviours, relating to key components of food safety.

Results: The majority of studies (60%) were published between 2013 and 2017 and included North America, Europe, Asia, and Africa. Although all studies focused upon professional food handlers, the majority (70%) were from catering and retail establishments; fewer studies were conducted in manufacturing and processing environments (10%). Survey methods of data collection were widely applied, including self-completed questionnaires (80%) and interviews (35%). Observation of behaviour was less frequently used (30%); the majority of findings were based on self-reported practices and knowledge. The most frequently covered topics in reviewed studies included awareness of when hand hygiene should be implemented and practices relating to cross-contamination. Analysis of data also determined that gaps in knowledge of some key food safety practices may exist among professional food handlers and malpractices are reported.

Significance: Completion of this narrative review has identified the need for in-depth systematic literature review to further explore the topic. Given this study identified a lack of observational data, there is a need for research, particularly in manufacturing environments, to observe the food-safety behaviours of professional food handlers; such methods can also be used to evaluate the impact and effectiveness of food-safety training.

P1-10 Assessing the Feasibility of Using Video Observation to Evaluate Food-handler Hand-hygiene Practices in a Food and Drink Manufacturing and Processing Businesses

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Introduction: Food-handler hand hygiene is a significant factor for foodborne illness; hand hygiene is the most effective method for preventing cross-contamination. Although informative, food-safety cognitions are not indicative of actual practices and may be subject to biases; thus, observation of behaviour is required. During observations, researcher presence can increase reactivity, whereas video observation provides a more comprehensive analysis over a sustained period where familiarity reduces reactivity. Previous video observation research has assessed food retail and catering food-handler hygiene behaviours; however, this method of assessment has been underutilised in food and drink manufacturing and processing business (FDMPB) environments.

Purpose: To assess the feasibility of using video observation to evaluate food-handler hand-hygiene practices in FDMPBs.

Methods: In-depth interviews with FDMPB managers and technical supervisors ($n=11$) identified hand-hygiene protocols, training procedures, and perceptions of video-observation to assess hand-hygiene compliance. One FDMPB was selected, from which footage (24 h) was reviewed to evaluate hand-hygiene compliance. Detailed observations were recorded specifying hand decontamination component actions.

Results: FDMPBs had unique hand-hygiene protocols with variable details. Interviews identified positive attitudes towards using video-observation to assess hand-hygiene compliance. Although FDMPBs had cameras, none had the resources or time to conduct frequent or structured observation of footage. Observational findings indicated that of 674 instances when food-handlers entered production, 70 failed to attempt hand-hygiene practices. Of 604 attempts to implement hand-hygiene practices, only 2% complied with FDMPB protocol. Although 78% utilized soap, only 45% wetted hands first. Less than half (42%) utilized sanitiser. Malpractices included drying hands on overalls (9%). Hand-hygiene duration ranged from 1 to 69 s (median 17 s).

Significance: Video-observation data provided in-depth insight into hand-hygiene compliance when entering production and illustrated a valuable and useful resource for FDMPBs. Observed extensive hand-hygiene malpractices

contrary to FDMPB policy that may compromise food safety during food production. The case study identified site-specific issues to inform the development of an intervention to improve hand-hygiene practices.

P1-11 An Exploration of Consumer Food-safety Concerns in Lebanon

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Introduction: Food safety in Lebanon is a major public health issue potentially associated with the country's unique public health infrastructure and political challenges regarding policy and strategy. Foodborne illness is reportedly widespread, but the absence of a proper disease-reporting mechanism makes incidence difficult to quantify. Consumer food safety is critical to reduce risk to health. However, little is known about consumer food-safety concerns in Lebanon.

Purpose: To determine and explore Lebanese consumers' risk perceptions, concerns, and perceived adequacy of food-safety information.

Methods: Qualitative face-to-face interviews ($n=43$) were conducted with consumers who approached a Lebanese University (MUBS) Health Day stand held in a shopping mall in Beirut, Lebanon. Interviews enabled exploration of food-safety perceptions and concerns amongst consumers.

Results: The majority perceived overall personal risk of foodborne illness to be 'very high' and 'greater' when eating outside of the home. Some respondents reported avoiding consuming food prepared outside of the home due to perceived lack of food safety guidelines and audits: "Restaurants...do not abide by the necessary guidelines of food safety." Consumers were concerned regarding accuracy and adherence of expiry dates: "The expiry dates are changed on some products; they even change the food source on the label." Concerns unique to Lebanon included electricity interruptions which were believed to be associated with unsafe food storage practices: "We do not have electricity 24 out of 24 hours and this is a huge defect when storing food in fridges because the temperature of those fridges goes up." Other food safety concerns included water safety, food contamination, and crop irrigation. A lack of food-safety information was reported with a desire for more information.

Significance: The study highlights food-safety concerns which are particularly unique to Lebanon and has identified the need for further research to determine Lebanese consumers cognitive and behavioural influences related to food preparation and storage, as well as food-safety education.

P1-12 Foodborne Outbreaks: High School Education for Prevention

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Introduction: Foodborne outbreaks (FBO), caused by consumption of microbiologically contaminated foods, can be prevented by teaching good food-safety practices to student populations. The educative school material can be evidenced by identification of FBO risk factors in scientific publications and epidemiologic analysis of information of food occurrence hazards and FBO surveillance, shared by European Food Information Networks.

Purpose: Develop food safety educational materials for high school student populations.

Methods: (i) Analysis of scientific publications about microbiological risk analysis associated to food consumption

to understand the best practices for prevention of FBO and the concepts related to food safety: food, hazard, risk, FBO and its epidemiology, surveillance, and control; (ii) Identification of the food hazards and bad practices representing contributing factors to FBO occurrence by epidemiologic analysis of information from (1) FBO occurring in Portugal, (2) European Union summary reports on trends and sources of zoonoses, zoonotic agents, and foodborne outbreaks, and (3) the Rapid Alert System for Food and Feed report from recent years; (iii) Identification of good practices for FBO prevention; and iv) Elaboration of school educative material, adapting all the scientific content to the high school curricula and making it available to schools on the website of the National Institute of Health Doutor Ricardo Jorge IP.

Results: Power point presentation 1: Theoretical foundation of concepts regarding FBO prevention (food, hazard, risk, FBO and its epidemiology, surveillance, and control).

Power point presentation 2, flyer, poster and learning assessment questionnaire of students after class: Consumer good practices for FBO prevention, from buying food to its consumption.

Significance: Analysis of data from food hazards and FBO surveillance systems is useful for producing scientific evidence on existing FBO risk factors, which can guide the production of materials that support interventions targeted to students in order to promote good food-safety practices for FBO burden minimization.

P1-13 Food-safety Education Makes a Difference

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Introduction: Earlier findings from a Swedish investigation of food-safety teaching in Home and Consumer studies classes suggest that students 15 to 16 years old might leave compulsory school without having learned basic food-safety rules. It was shown that food-safety teaching in Home and Consumer studies classes depends on the teachers' subjective didactic choices and that one of the most trusted sources of food-safety knowledge is the mother. Future responsibilities for young adults may be to cook for small children and older relatives. They could also experience pregnancy and other vulnerable categories in private food-handling situations. International studies have shown limited food-safety knowledge, attitudes, and behavior among university students. As far as the authors know, there has been no previous investigations targeting university students in Sweden.

Purpose: To investigate self-reported food-safety attitudes, knowledge, and behavior among university students in Sweden.

Methods: A nationwide web-based questionnaire (28 questions) targeting university students in Sweden was distributed through social media, email, and various university contacts.

Results: Among the 606 respondents, 36% reported earlier attendance in food-safety courses in gym classes or university. The average number of correct answers regarding knowledge was 7.6 out of 12.0. However, students reporting education as their primary source of knowledge scored 9.6 correct answers in general. Food-safety education correlated with higher knowledge ($P < 0.05$). More unfavorable behaviors were reported among those with fewer correct answers on the knowledge questions. A majority (62%) of the respondents reported hand washing as very important,

while 27% stated that it was very important (32% said rather important) to cool leftovers within four hours.

Significance: Food-safety education correlated with better knowledge in the subject and indicated safer self-reported food handling. The attitudes, however, did not always reflect reported behaviors.

P1-14 The FSVP Audit On-site Checklist: A Tool for Regulatory Auditors

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Introduction: We developed an innovative Foreign Supplier Verification Program (FSVP) "Audit on Site" software checklist following the ANSI 62 sectors of U.S. Food and Drug Administration food categorizations, the requirements of 21 CFR Part 1 Subpart L and 21 CFR 117, plus the Food Safety Preventive Controls Alliance (FSPCA) standardized curriculum to help FSVP importers to implement this new kind of verification activity with foreign food manufacturers and suppliers.

Purpose: Carry out a new regulatory audit required by FSVP and Third Party Certification rules to verify that "serious adverse health consequences or death to humans or animals" hazards and other relevant hazards are well-controlled by foreign food suppliers.

Methods: Our checklist is divided in 15 different sections following the FSPCA Preventive Controls for Human Food standardized curriculum, and each section can generate four different results: "C" (conformity), "NP" (not present), "R" (recommendation), or "AB" (absence).

Results: Thanks to a simple compilation of all the software sections, in the end we will get a percentage of documentation compliance and correctly implemented good manufacturing practices, with all FSMA requirements linked directly to the audited foreign facility.

Significance: A very important new tool, which can be used to perform activities related to FSVP verification, the Voluntary Qualified Importer Program, and Third Party Certification.

P1-15 Developing and Maintaining Food Safety Culture through Implementation of Global Food-safety Bench Marked Standards – A Success Story

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Introduction: Access to safe and high-quality food is of paramount importance and an essential requirement for consumers to maintain their health and well-being. The meticulous efforts of food producers to demonstrate their commitments to food safety and fulfill customers' preferences and expectations can gain more attention if organizations demonstrate well-established quality and food safety cultures. Top management commitment and involvement is mandatory to embed positive food safety culture at all levels in the organization.

Purpose: The purpose of this study was to depict adoption of innovative ideas and reflection of collective attitudes, beliefs, and behaviours of organizations' top management, managers, supervisors, and food handlers towards resolving food safety and hygiene issues and setting contemporary

trends, leading to the transformation of existing food safety practices into a more sophisticated and regimented food safety culture.

Methods: In the present study, a quarterly survey of food manufacturing units and distribution centers in the Mawarid Food Company was conducted in the Kingdom of Saudi Arabia.

Results: Results of this study showed that appropriate training, empowering employees to share their ideas, motivations, and strong commitments from top management lead to transforming existing food safety practices into a more sophisticated and regimented food safety culture.

Significance: This study is quite helpful for food producers and retailers, showing them how they can turn their dreams into reality when they successfully attain certification of their food facilities against the benchmarked standards of Food Safety System Certification 22000, a prestigious Global Food Safety Initiatives approved certification body.

P1-16* Investigating the Ability of *Listeria monocytogenes* to Form Biofilm on Surfaces Relevant to the Mushroom Production Environment

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Introduction: *Listeria monocytogenes* poses a threat to all fresh fruits and vegetables due to its ubiquitous presence in the natural environment, including mushrooms, which are Ireland's largest horticultural crop. Although mushrooms (*Agaricus bisporus*) have not been linked with listeriosis outbreaks, the organism still poses a threat to the industry due to its presence in the environment and its ability to form biofilms. This threat is highlighted by studies demonstrating that *L. monocytogenes* is present in the mushroom production environment and that it can form biofilms on surfaces used in the food industry.

Purpose: The aim of this study was to investigate the biofilm formation potential of *L. monocytogenes* strains, isolated from mushroom production environment, at temperatures and on surfaces that are relevant to the mushroom industry.

Methods: Preliminary assessment of biofilm formation of 44 mushroom industry isolates of *L. monocytogenes* was carried out using a crystal violet assay on polystyrene microtitre plates at 18 and 25°C for 72 h. Strains were then selected according to their biofilm forming ability and were assessed for their biofilm formation on different surfaces (stainless steel, aluminium, rubber, polycarbonate, polypropylene, and concrete) using the CDC biofilm reactor at 25°C for 72 h.

Results: The crystal violet assay showed that the mushroom industry isolates were able to form various levels (weak, moderate, or strong) of biofilm on microtitre plates under industry relevant temperatures. Stainless steel, aluminium, rubber, polypropylene, and polycarbonate were all found to be able to support biofilm levels ranging log₁₀ 4–4.9 CFU/cm², for seven different *L. monocytogenes* strains, with no significant difference ($P>0.05$) between them. On the other hand, concrete supported log₁₀ 7.7 CFU/cm² of biofilm from the same strains.

Significance: These results indicate that *L. monocytogenes* can readily form biofilms on industry relevant surfaces, and additionally identifies areas of specific concern where rigorous cleaning and disinfection is required.

P1-17* Survival of *Escherichia coli* and *Listeria innocua* on Lettuce after Irrigation with Contaminated Water

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Introduction: Leafy green vegetables are associated with a large number of microbial disease cases and outbreaks. These foodstuffs are susceptible to microbial contamination via a number of different pathways, such as contaminated manure or irrigation water. Contamination can be caused by pathogenic strains of *Escherichia coli* or *Listeria monocytogenes*, among other microbial pathogens. Once present on the plant, these bacteria may persist for different periods of time, which represents a risk for consumers' health.

Purpose: The objective was to analyse the survival of *E. coli* and *Listeria innocua* in lettuce plants inoculated with contaminated irrigation water via a single overhead spray irrigation event, in typical Irish winter glasshouse growth conditions.

Methods: Three adjacent plots of seven by four lettuce plants (*Lactuca sativa* var. *capitata*) were inoculated with water spiked with streptomycin resistant *E. coli* FA7 Lys9 or *L. innocua* ATCC 51742. Each plant was inoculated with 300 mL of water with 10⁷ CFU/mL of either strain. Two additional plots were irrigated with uninoculated water. Three plants from each plot were removed at eight sampling points, from day zero to day 28. Samples of 25 g from each plant were analysed for the presence of *E. coli* or *L. innocua* via direct colony enumeration and enrichment.

Results: Survival of both strains was observed in lettuce plants up to 28 days after inoculation, at which point plants were harvest ready. Direct quantification showed a 4-log decrease in the concentration of *E. coli* 14 days after inoculation and a 3-log decrease in the concentration of *L. innocua* 10 days after inoculation.

Significance: These results demonstrate that *E. coli* and *Listeria* strains are able to persist in lettuce plants after a single contamination event up until the plants were ready for harvest. This demonstrates that irrigation water can be an important contamination vector on the production of leafy greens and vigilance is needed.

P1-18 Effect and Comparison of Swabbing for *Listeria* Species Using Hygiene InSite Swabs and Other Swabs Compared to Contact Plates Using Residual Bacterial Method

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Introduction: To test swabbing efficiency: A superior method looks at what is left behind on a surface rather than attempting to measure recovery from swabs. The binding of material or bacteria onto swabs can bias results.

Purpose: To test how effectively Hygiene's swabs recover viable dried bacteria from surfaces compared to routinely used standard environmental contact plates.

Methods: 100 µL of *Listeria monocytogenes* dilutions (Dil -1, -2 and -3) were pipetted onto sterile coupons. Dilutions were dried on coupons measuring 4 in by 4 in and 12 in by 12 in. The surface was left to dry for 24 h until visibly dry. Contact plates and three swab types (Dacron bud, small foam, and large foam InSite swabs) were used to collect the dried bacteria from the surface, and the swabs were

moistened with 100 to 400 µL of maximum recovery diluent (MRD) depending on swab size. After sample collection, the coupons were put in Whirl-Pak bags and 50 ml of MRD was added to the bag; each coupon was mixed thoroughly to remove remaining bacteria. Each run used five replicate coupons, both control and both tests (swab and contact plate). The unswabbed counts from the controls are considered 100% for comparison.

Results: The comparison of swabbing with any swabs proved that the pick-up was superior to contact plates. The 4 in by 4 in coupons swabbed with Dacron bud swabs, small foam, and large foam removed mean 91%±6% (4 in by 4 in), 98%±1% (4 in by 4 in), and 91%±3 (12 in by 12 in) compared to contact plates, which removed 72%±16%, 71%±7%, and 50%±12%. Comparing 4 in by 4 in to 12 in by 12 in coupons reduced the efficiency slightly, but the pick-up was still superior to contact plates for *Listeria monocytogenes*.

Significance: The efficiency of swabbing devices can be easily measured by using the residual bacteria method with proper controls.

P1-19 Real-time Monitoring of TVC Using Non-invasive Bioluminescence Growth Media

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Introduction: This study introduces a new technology from Hygiena, MicroSnap Surface Express, for the real-time monitoring for non-specific viable bacteria from surfaces. The collected samples begin to produce light as they metabolise a new substrate, illustrating by the emission of light per bacteria a new method to both enumerate and detect at low CFUs. The collected CFU burden proved that the method can be used as a disaster test to indicate very rapidly if a heavy bioburden of viable bacteria was swabbed.

Purpose: To demonstrate performance of MicroSnap Surface Express Total in the field as an easier and simpler method for detecting viable bacteria from environmental samples in a single, self-contained device.

Methods: Forty-seven sites within a food plant were sampled and split into two portions; one portion was incubated with the real-time viability growth media and the second portion was inoculated onto agars to count total viable plate count (TVC). MicroSnap Surface Express devices were incubated at 30°C and measured for bioluminescence each hour for 12 h. At 24 h, the TVC plates were counted and compared to each hour in the bioluminescent growth cycle.

Results: The results show that increasing bacteria from the surfaces produced detection in shorter incubation times. The detection was inversely proportional to the CFU, with shorter incubation periods of 1 to 5 h detection in the first bin (> 5000 CFU); as levels of bacteria swabbed decreased, the time to result increased. The following are mean time ($n=5$ to 10): 1,001 to 5,000 CFU, time to result < 6 h; 101 to 1,000 CFU, time to result < 7 h; 11 to 1,000 CFU, time to result < 8 hours; and < 10 CFU, time to result 9 h.

Significance: The new method equips food processors with an ultra-rapid tool for identifying viable bacteria as part of a sanitation monitoring program.

P1-20 Rapid Detection of *Enterobacteriaceae* from Food Preparation Surfaces Using Simple Bioluminogenic Device

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Introduction: The rapid detection of *Enterobacteriaceae* from food preparation surfaces is particularly important to the cleanliness of food manufacturing facilities.

Purpose: The Hygiena MicroSnap Surface Express system allows collection, detection, and enumeration of *Enterobacteriaceae* from surfaces in the same working shift.

Methods: MicroSnap Surface Express is an all-in-one device containing a surface swab and ready-to-use, patented media. Upon sample collection, the entire device is incubated from 2 to 6 h to detect as low as < 10 CFU per swab. The device is then activated. The reagent bathes the growing sample and light is produced proportionally to the level of bacteria present (CFU). The system is self-sterilising; after activation, the device and bacteria collected are rendered non-viable. In this study, *Salmonella enterica* ATCC 13076 and *Escherichia coli* ATCC 8739 were grown overnight and diluted in diluent, inoculated onto the swab, and incubated at 37°C for 1, 2, 3, 4, 5, 6, 7, and 8 h. The devices were activated at each time point and measured in Hygiena SystemSURE Plus and EnSURE luminometers. Reference counts were run on traditional pour plates and read at 24 h.

Results: *S. enterica* and *E. coli* started to be detected in 2 h with confirmed detection in both luminometers in 4 h. The relative light units signal to noise ratio (S/N), which uses the non-inoculated background as a baseline, was recorded. For *E. coli* and *S. enterica*, the S/N at each time were: 1 h (1:1), 2 h (2:4), and 3 h (9:30).

Significance: This rapid method will allow microbiological cleanliness to be run in-house by more food manufacturers, allowing better surveillance and control of surface-borne cross-contaminating pathogenic *Enterobacteriaceae*.

P1-21 Study to Demonstrate the Detection of Cross-contamination of Surfaces with Raw Chicken Juice Using Simple Acid Phosphatase Detection Device

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Introduction: The use of a rapid detection method for cross-contamination has been highlighted as a possible method for the reduction of *Campylobacter* infections from store-purchased chickens. These potential sources of *Campylobacter* require an easy method to quickly decide if a surface has been contaminated or requires better sanitation.

Purpose: This study highlights a simple detection device (Hygiena CrossCheck) that will detect raw chicken juice dried onto a surface that has been transferred multiple times through contact and still remains detectable. The viability of a contaminant through a hand-contact environment highlights the need for rapid detection.

Methods: The juice from grocery store packaged raw chicken was decanted. The chicken juice was then inoculated onto a surface and allowed to dry overnight at ambient temperature. The chicken juice was also rubbed onto gloved hands which then touched 20 sterile squares of stainless steel in sequence. Using the CrossCheck device, each square was tested in turn. The assay consists of swabbing the surface and then activating the device. After a five-minute incubation at 37°C, the level of acid phosphatase is indicated by the level of relative light units (RLUs) on a luminometer. The assay is extremely sensitive and will detect acid phosphatase down to nanogram levels.

Results: The acid phosphatase in the raw chicken from the initial drying was 34,000,000 RLUs; the background level of acid phosphatase was 24,000. This initial signal dropped with sequential transfers until ACP through the gloved hands became insignificant, i.e., less than the background after seven transfers onto sterile squares.

Significance: The use of rapid acid phosphatase detection is a useful tool in the detection of cross-contamination and is suggested as a proxy measurement to aid reduction of *Campylobacter* contamination.

P1-22 Simultaneous Detection of Coliforms and *Escherichia coli* Using a Combined Bioluminescent and Fluorescent Detection System

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Introduction: This study introduces simultaneous detection of coliforms and *Escherichia coli* using a combination of bioluminescence and fluorescence biochemistries in a self-contained device.

Purpose: The simultaneous detection system for coliforms and *E. coli* is designed to make surface evaluation easier for food manufacturers by combining both detections in one simple device. The use of both indicator organisms can triage the likelihood for true faecal contamination in one test. The measurement of two samples in the same device also eliminates parallel sampling using two separate chemistries.

Methods: *E. coli* ATCC 8739 was grown in TSB overnight at 37°C and serially diluted in sterile diluent. 10 µL of dilutions -4, -5, -6, and a blank were added to a tube containing 500 µL selective enrichment media. Tubes were set up to be incubated for 6 h and read every 2 h using 500 µL of bioluminescent coliform detection reagent that was added, mixed, and read on a luminometer. If the relative light units are above a pre-set threshold, it signifies a presumptive positive for coliforms. The positive devices were incubated longer and visible green fluorescence measured using a 365 nm UV lamp indicating positivity for *E. coli*.

Results: All dilutions were detected using bioluminescence and fluorescence at 12 h. Limit of detection for coliform detection through bioluminescence in this study was 10⁶CFU/mL with 6 h of incubation. The limit of detection for *E. coli* through fluorescence after confirmed coliform detection was 10⁶CFU/mL at 10 h incubation.

Significance: The simultaneous detection of both coliforms and *E. coli* in a single system using two technologies offers an advantage in a food protection program. This means that food processors can act quickly and decisively to eliminate potential surface contamination and then react to the confirmation step for *E. coli*. With further work on media, this technology could be applied further for strain identification for Shiga toxin-producing *E. coli* using PCR.

P1-23 Population Genetic Structure of *Listeria monocytogenes* Strains Isolated from the Pig and Pork Meat Production Chain in France

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Introduction: *Listeria monocytogenes* is a pathogenic bacterium, transmissible through contaminated food consumption. The pork meat sector has been hit hard by *L. monocytogenes*-related outbreaks in France. Numerous studies have been carried out to investigate the genetic diversity of *L. monocytogenes* pork strains from food or some food factories, but never from the entire production chain.

Purpose: This study aims to identify the contamination routes of this bacterium through the global description of the strains' genetic diversity in the pork meat production chain.

Methods: We analysed the population genetic structure of 687 strains isolated over 20 years, throughout France and in three compartments: pig farming (PF), food processing environment (FPE), and finished food products (FFP). Clonal

complexes (CCs) were obtained by mapping the PFGE profiles of the strains. Their distribution was compared firstly between the three compartments, and then with CCs obtained from 1,106 strains isolated from other food production sectors.

Results: The major CCs were not equally distributed among the three compartments. CC37, CC59, and CC77 strains, rarely found in the FPE and FFP, were prevalent in the PF compartment. The two most prevalent CCs in the FPE and FFP compartments, CC9 and CC121, were rarely or not found in the PF compartment. No CC was exclusively associated with the pork sector. CC5, CC6 and CC2 were found in comparable proportions in all the sectors. The two most prevalent CCs in all the sectors were CC121 and CC9, but their distribution was disparate. CC9 was associated with meat products, while CC121 was not associated with a given sector.

Significance: The distribution of this ubiquitous bacterium appears largely influenced by steps in the production chain. This study provides indications on the ecological decline or success of *L. monocytogenes* population along the pig and pork meat production sector.

P1-24* A Pilot Study of Ultrasonication Pre-treatment and High-pressure Processing Affecting Microbial Inactivation and Colour Attributes of Liquid Whole Egg

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Introduction: Ultrasonic processing of a variety of liquids, drinks, and beverages has generated much interest, with published papers increasing within this area in recent years. A combination of ultrasonication and high-hydrostatic pressure could be used as a combined non-thermal, minimal processing technology for fluids.

Purpose: This work investigated the impact of ultrasound (US) pre-treatment combined with high-hydrostatic pressure (HHP) on colour and microbial inactivation in liquid whole egg (LWE).

Methods: Homogenized LWE was pre-treated with US (12.50±0.31 W and 55, 65, and 75% amplitude) for 30 min prior to HHP treatment (300 and 350 MPa, 5 min).

Results: Our results revealed that colour variation of LWE was statistically significant (one-way analysis of variance, $\alpha = 0,05$), but nevertheless colour difference ΔE_{ab} showed maximum visible differences. Increasing treatment parameters caused decreased microbial cell count; increased microbiological inactivation was significant and caused by different treatment parameters.

Significance: Summarising our results shows that US pre-treatment combined with HHP can provide a microbiologically safe product while protecting colour stability.

P1-25 The First Characterization of the Diversity of *Staphylococcus* Strains Using MALDI-TOF MS and Detection of MRSA Strains Isolated from Raw Sausage in Algeria

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Introduction: Staphylococci, especially *Staphylococcus aureus*, is one of the most economically devastating types of foodborne bacteria, with high concern for public health and food safety. It has adjusted to all antimicrobial agents that have been developed, including developing resistance to methicillin.

Purpose: Our objective was to investigate the diversity of staphylococci species isolated from raw sausages in Algeria and their susceptibility against 16 antimicrobials, including B-Lactamines and Vancomycin, with a special search for methicillin-resistant *Staphylococcus aureus* (MRSA).

Methods: Meat samples from 325 Algerian butchers were identified using MALDI-TOF MS and confirmed as MRSA by examining the presence of the *mecA* gene by PCR in real time and confirmed by PCR standard. Antibiotic resistance was determined by the antibiotics disk diffusion method.

Results: A total of 88 strains were identified by MALDI-TOF mass spectrometry. We have identified 71 strains of *Staphylococci*, including *S. saprophyticus* (24); *S. sciuri* (8); *S. xylosum* (6); *S. vitulinus* (3); *S. equorum*, *S. lentus*, *S. haemolyticus*, *S. gallinarum* with two strains each; and *S. warneri* (1). The rest represented *Macrococcus caseolyticus* strains. The main spectrum projection dendrogram revealed four distinct clusters according to an arbitrary cut off at the distance level of 500. All *S. aureus* were severely resistant against B-Lactamines, as were *S. vitulinus*, *S. sciuri*, *S. xylosum*, and *S. equorum*. All *S. saprophyticus* strains were only severely resistant to oxacillin. There was no resistance to vancomycin. We also detected the *mecA* gene in five methicillin-resistant strains confirmed by PCR. Comparing our strains with previously confirmed human strains of *S. aureus*, results suggest human contamination.

Significance: The findings suggest that when present, *S. aureus*, and staphylococci in general, were considered a potential hazard for consumers and require enforcing hygienic practices. We aim to support the role of the human and animal as reservoirs of bacterial resistance, driving us to study staphylococci contamination of raw sausage from farm to fork.

P1-26 The Occurrence of Shiga Toxin-producing *Escherichia coli* and Antimicrobial Resistance in *E. coli* in Selected European Beef and Sheep Abattoirs

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Introduction: Shiga toxin-producing *Escherichia coli* (STEC) and antimicrobial-resistant (AMR) *E. coli* are two zoonotic pathogens that may be transmitted through the meat production chain. It is important to have knowledge of the occurrence of these in abattoirs.

Purpose: The aim was to study STEC and AMR in selected European beef and sheep abattoirs.

Methods: A total of five composite carcass swab samples (before chilling) from each of 11 sheep slaughter lines in Norway, UK, and Spain, and nine for beef in Norway, Denmark, and Germany were analysed for STEC and AMR, giving a total of 100 samples. Samples were analysed for STEC using a modified ISO TS 13136, *stx*_{2A} using a specific real-time PCR, and for AMR using methods described by EURL-AR for extended spectrum cephalosporin-resistant (ESC) *E. coli* and carbapenemase-producing *E. coli* (CPE), and a selective method for the isolation of quinolone resistant *E. coli*.

Results: STEC from the “big five” serogroups were not isolated from any of the samples tested, but the majority of the samples were positive for one or more of the genes *stx*₁, *stx*₂, and *eae* in screening of the enrichment broths. *E. coli* harbouring *stx*_{2A} of unknown serotype were isolated from three cattle slaughter lines. *E. coli* resistant to quinolones were detected in 17 samples from six abattoirs. Preliminary results indicate that the resistance is due to chromosomal mutations, but for one isolate the MIC values for ciprofloxacin and nalidixic acid indicates plasmid-mediated resistance

mechanisms. Further, ESC- or CPE-resistant *E. coli* were not detected using selective methods.

Significance: The results suggest a low occurrence of STEC from the “big five” serogroups, and AMR *E. coli*. *E. coli* harbouring *stx*_{2A} genes were isolated, which may indicate potential for severe human infection. The presence of possible plasmid-mediated resistance is a challenge due to potential horizontal transfer to other bacteria.

P1-27 Prevalence and Location of *Listeria monocytogenes* Contamination on Beef Carcasses in Belgian Slaughterhouses

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Introduction: Despite the efforts already made by the sector, slaughterhouses are still confronted with the food-borne pathogen *Listeria monocytogenes* in the production environment and on carcasses. The presence of the pathogen on carcasses may result in contamination of the meat, which may lead to economic losses and a risk for public health.

Purpose: This study on the prevalence and location of *L. monocytogenes* on beef carcasses after slaughter should be the basis for a better understanding of the complex introduction, persistence, and variable contamination sources and routes of this pathogen.

Methods: The sampling of 90 carcasses was carried out in three beef slaughterhouses. A total of 720 cattle carcass samples were taken just before cooling. *L. monocytogenes* was detected and enumerated according to ISO 11290. Carcass locations assumed to have an increased risk for contamination were swabbed. The eight following sites (400 cm² each) were sampled: pelvic duct, split surface of neck, inside throat region, hind leg (medial side), flank (medial side), brisket, inside foreleg, and shoulder region.

Results: *L. monocytogenes* was detected in 10% (71 of 720) of the swab samples. Overall, 47% (95% confidence interval from 36 to 57%) of the carcasses with a variation from 23 to 73% for the three slaughterhouses were found to be positive for *L. monocytogenes* for at least one of the eight sites. Among the different locations, the inside hind leg and the inside foreleg represented the highest contamination frequencies (13 and 12%, respectively). However, the contamination rate of the different sampling sites was not significantly different ($P > 0.05$). Across all samples, only five samples exceeded the lower limit of enumeration (above $-1.3 \log_{10}$ CFU/cm²).

Significance: The high prevalence of this pathogen highlights the need for identifying contamination sources and routes. The knowledge of the most contaminated carcass sites is useful in this context.

P1-28 Foodborne Pathogens: Inactivation in Protected Designation of Origin Coppa and Pancetta Piacentina during the Process and after High-pressure Processing

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Introduction: Foodborne pathogens such as *Escherichia coli*, *Listeria monocytogenes*, and *Salmonella* spp. may occur in dry cured meat products via contaminated raw meat, ingredients, processing equipment, and/or as a consequence of post-processing contamination, and they have also been shown to survive during manufacturing processes.

Purpose: The objective of this study was to evaluate pathogen inactivation in two typical Italian protected designation of origin meat products, Coppa and Pancetta Piacentina. We also validated the integrated lethality of high-pressure processing (HPP) as a post-processing intervention to eliminate pathogens in meat cured products.

Methods: Three multi-strain cocktails of *E. coli* O157, *Listeria innocua*, and *Salmonella* Typhimurium were used to contaminate separately a total of 36 Coppa samples (on the surface) and 30 Pancetta samples (in the core and on the surface) just before putting them into the casings for curing (180 days for Coppa and 120 days for Pancetta). Control products were inoculated with sterile solution to evaluate the physical-chemical changes during the process. HPP treatment (600 MPa for 5 min) was applied after curing on six replicates. Microbial survivals were estimated by plate count method.

Results: The long curing times, combined with unfavorable conditions, allow for pathogen reduction during the process. With HPP treatment, the results showed that on Coppa it was possible to achieve a total reduction of about 6.2, 5.5, and 7.0 log CFU/g for *E. coli*, *L. innocua*, and *Salmonella* spp., respectively. For Pancetta, it was possible to achieve a total reduction of about 7.0, 4.7, and 5.3 log CFU/g (on the surface) and 6.9, 4.6, and 5.1 log CFU/g (in depth) for *E. coli*, *L. innocua*, and *Salmonella* spp., respectively.

Significance: These data showed that adding HPP to the production process could be used to reach the performance standards for traditional Italian dry-cured meat products.

P1-29 First Report of *Sarcocystis bovifelis* in Italy

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Introduction: *Sarcocystis* spp. are usually characterized by a two-host life cycle, with herbivores as intermediate hosts and carnivores as definitive hosts. Cattle are intermediate host for multiple species with different definitive hosts, causing lesions that lead to carcass condemnation and public health issues. Until recently, only three species were officially recognized, with canids, humans, and felids as definitive hosts, respectively: *S. cruzi*, *S. hominis*, and *S. hirsuta*. This latter name came into use as a substitute for the original *S. bovifelis* in the mid-1980s and since then has been used in reference to all cat-transmitted *Sarcocystis* spp. in cattle. However, the analysis of mitochondrial cytochrome oxidase subunit 1 (COI) led to the reintroduction of the name *S. bovifelis* with reference to a different *Sarcocystis* species in cattle transmitted by felids.

Purpose: This study reports the first evidence of *S. bovifelis* in Italy.

Methods: Samples of cattle muscle tissues harbouring *Sarcocystis* spp., previously identified as *S. hominis* by amplification of the 18S rRNA gene (18S rRNA), were subjected to a specific PCR performed with newly designed primers targeting the COI gene and subsequently sequenced.

Results: The amplification of the expected size fragments demonstrated the presence of *S. bovifelis* DNA in four of the ten *S. hominis* positive samples. Sequences of about 400 bp were obtained by Sanger sequencing and 100% homology with the sequences of *S. bovifelis* deposited in the Genbank of the National Centre for Biotechnology Information using the Basic Local Alignment Search Tool was found.

Significance: This study supports previous findings of cattle being the intermediate host for at least a second cat-transmitted *Sarcocystis* species, and the results also indicated the higher discriminative power of COI mitochondrial gene, compared to the 18s rRNA gene, for the taxonomy clarification of *Sarcocystis* species, which will allow an appropriate risk assessment of public health issues arising from consumption of contaminated beef.

P1-30 Use of Probiotics in the Primary Chain Production of Swines for the Reduction of *Salmonella* Resistant to Antibiotics

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Introduction: *Salmonella* is one of the main causes of zoonotic diseases present in foods of porcine origin, and many *Salmonella* serovars are resistant to antibiotics. Given the appearance of these phenomena, the implementation of probiotics represents a good alternative to consider within primary production in the swine industry.

Purpose: Probiotics supplied in food have been an alternative to reduce the prevalence of *Salmonella* in the primary chain of swine production. The main objective of this work was to research a commercial probiotic as an alternative to control strains of antibiotic-resistant *Salmonella* isolated from pigs.

Methods: The growth in co-culture of a commercial probiotic was evaluated along with a control strain of *Salmonella* (*S. enterica* ATCC113076) and five strains of *Salmonella* resistant to antibiotics isolated from the swine industry during 48 h in which plate count was performed (expressed in log CFU/mL in XLD and SPC media), as well as measurement of OD and pH.

Results: A reduction in the growth of *S. enterica* ATCC 113076 was obtained in co-culture with the probiotic at 5.6 logarithmic units (LU) at 24 h, 10.5 LU at 30 h, and 3.63 LU at 36 h. The probiotic co-incubated with each of the antibiotic-resistant strains of *Salmonella* promoted the reduction of the growth of all strains from 18 to 36 h, obtaining reduction values of up to 11 LU.

Significance: Reduction of antibiotic-resistant *Salmonella*, improving the safety of porcine meat and the use of antibiotics in pigs.

P1-31 Dynamics of Modified Atmosphere Packing Melon Microbiota

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Introduction: Several outbreaks of foodborne illness associated with the consumption of fresh-cut melons have been reported. However, microbiota of cube-cut melon packed in modified atmosphere (MAP) has never been deeply analyzed.

Purpose: The objective of this work was to determine bacterial and fungal microbiota composition of MAP cube-cut melon, both at packaging and at time of use, after eight days of storage at 7°C.

Methods: Cultivable bacteria and fungi of four samples were enumerated on PCA, MRS, MacConkey, ALOA, and PDA plates at delivery and after eight days at 7°C. The dominant stains isolated in MRS and PDA plates at the end of the shelf life were genotyped by RAPD-PCR fingerprinting, and taxonomic diversity was estimated by partial sequencing of 16S rRNA genes or ITS. A comprehensive picture of bacteria and fungi present in the samples was obtained by metataxonomic analysis of Illumina 16S sequences.

Results: PCA initial charge was generally in the magnitude of 5 log (CFU g⁻¹), and it increased up to 8 log after eight days. At packaging, coliform load ranged between 0.8 and 2.8 log (CFU g⁻¹) and linearly increased up to 2.7 (CFU g⁻¹). *Listeria* was rarely present. On average, fungi started at 2.8 log and reached approximately 7.4 log. Most of the MRS isolates were ascribed to the genus *Leuconostoc*, with *L. citreum* being the most recurrent species. Yeast isolates were much more heterogeneous. Metataxonomic analysis showed that dominant microbiota of S samples was associated with animal gut bacteria. Each E sample

was dominated by different bacteria: *Leuconostoc*, *Serratia*, *Pseudomonas fragi*, or *Pantoea*. The dominant fungus at packaging was *Wickerhamomyces anomalus*; at the end of the shelf life, it was *Candida sake*.

Significance: Knowledge on the evolution of microbiota of MAP cube-cut melon maintained at 7°C during the shelf life was provided.

P1-32 Changes in Microbiota of Fresh Sausage throughout Shelf Life

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Introduction: Fresh sausage is a perishable product easily colonized by spoilage bacteria. Microbial diversity of fresh sausage has been generally investigated using traditional cultural methods.

Purpose: The present study aimed to obtain an exhaustive description of the microbiota composition and diversity throughout the shelf life of fresh sausage.

Methods: Cultivable bacteria from ten batches of sausage provided by the same factory were enumerated on PCA, MRS, MacConkey, ALOA, and PDA plates, at delivery and after 12 days at 7°C. The dominant strains were genotyped by RAPD-PCR fingerprinting, and taxonomic diversity was estimated by partial sequencing of 16S rRNA genes. A comprehensive picture of bacteria was obtained by metataxonomic analysis of Illumina 16S sequences.

Results: The mean charge (log of CFU/g) of cultivable aerobic and aerotolerant bacteria started at 5.0 and reached 7.2 after 12 days at 7°C, with a concomitant drop of pH from 5.9 to 5.5. Lactic acid bacteria showed the highest increase starting from 4.1 and reaching 8.6 log, followed by staphylococci (4.0 to 6.5 log) and by *Enterobacteriaceae* (3.2 to 6.2 log). The majority of MRS biotypes belonged to *Lactobacillus curvatus*, followed by *Lactobacillus sakei*, *Leuconostoc carnosum*, and *Leuconostoc mesenteroides*. The most represented species in PCA was *Brochothrix thermosphacta*. Metataxonomic analysis revealed that at packaging, fresh sausage was characterized by bacteria generally ascribable to two diverse microbiota associated with gut microbes. At the end of the shelf life, *Firmicutes* dominated the microbiota, in some cases with prevalence of *Lactobacillales*. In other samples, *Listeriaceae* took over, with *Brochothrix* being the most represented genus. *Enterobacteriaceae*, generally ascribed to *Serratia*, were abundant in a few samples at the end of the shelf life.

Significance: This study provided a wide overview of microbiota evolution in fresh sausage, shedding light on meat hygiene and safety issues.

P1-33 Metagenomic Analysis of Spoilage Microbiota in Asian Seafood

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Introduction: Seafood products are generally subject to fast spoilage. The huge variety in packaging and processing methods likely results in a variety of dominant spoilage flora as well. Classic isolation and identification of spoiling microorganisms is a slow and labor-intensive process, and it is subject to biases. Metagenomics enables less expensive and less labor-intensive analysis of spoilage flora, inevitably leading to the establishment of product segment-associated microbiota.

Purpose: The current study is to analyze the dominant spoilage flora in seafood from Asian markets stored under "wet market conditions" (two days at 30°C) and at "refrigerator conditions" (30 days at 10°C).

Methods: Multiple samples (15 in total) were bought in Thailand, Singapore, and Indonesia. Of the samples, 85% were bought in the supermarket (65% from refrigerator section, 20% from the freezer) and 15% from the wet market. Samples were frozen after purchasing at -28°C. Frozen samples were shipped to the laboratory in The Netherlands and stored at -28°C until analyzing. Seven samples were stored for 30 days at 10°C and eight samples were stored for two days at 30°C. At the end of incubation time, the total plate count, pH, and water activity were measured. The microflora was analyzed by metagenomics.

Results: *Leuconostoc mesenteroides* was the most dominant species in seafood stored for 30 days at 10°C. *Bacillus paraflexus* was the most dominant species in seafood stored for two days at 30°C. Overall, the main flora of seafood obtained from the Asian market contained lactic acid bacteria, *Bacillus*, *Brochothrix*, and *Spirochaeta psychrophila*. There was no obvious difference in type of product or the history of the samples (refrigerated/frozen or wet market).

Significance: A better understanding of the dominant spoilage flora will lead to better and specific interventions to elongate shelf life.

P1-34 Selection of Bioprotective Starters for Cooked Ham

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Introduction: Spoilage microorganisms colonize cooked ham packaged in modified atmosphere, with a preferential growth of psychotropic lactic acid bacteria (LAB) such as *Leuconostoc*, *Lactobacillus*, and *Carnobacterium*. Depending on the strains, the colonization can result in premature spoilage, characterized by pH decrease, gas and slime production, discoloration, and/or formation of off-flavors.

Purpose: The purpose of the study was the isolation and identification of potential bioprotective starters. Using careful selection based on their physiological properties, starters will be designed to extend shelf life and to meet food safety requirements.

Methods: The first step of the project aimed to identify the main consortia of microorganisms in fresh and spoiled sliced cooked ham by microbiological analysis. The interaction between consortia was then studied in order to screen for strains able to inhibit spoiled consortia. The isolated strains were screened for their antimicrobial activity by agar-well-diffusion assay. Strains showing antimicrobial activity were identified by molecular methods and PCR was used to amplify the genes of the known bacteriocins.

Results: A total of about 150 strains were isolated and six of them, identified as *Lactococcus lactis*, were selected for their ability to inhibit a large number of spoiled consortia and for their ability to produce bacteriocins.

Significance: The results provide information on the use of microbial starter for the extension of shelf life in meat products such as cooked ham. The effectiveness of potentially protective starters will be evaluated by challenge tests analyzing sensorial and microbial profiles.

P1-35 Performance of MC-Media-Pad in Detecting and Enumerating Spoilage Microorganisms in Dairy Products

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Introduction: MC-Media-Pad is an AOAC- and MicroVal-approved culture media developed to provide easy enumeration of total aerobic count, *Escherichia coli*, coliforms, and yeasts/molds in food and beverage matrices. The ready-to-use device is comprised of dried culture

medium coated onto a pad protected by a transparent lid. Food samples of 1 ml each are inoculated and rehydrated, according to reference method sample preparation procedures. Enumeration follows 24 to 72 h incubation. Color indicators and coding enables quick differentiation and improved read out.

Purpose: Total aerobic count (TAC), coliforms, *E. coli*/coliforms, and yeasts/molds device performance is evaluated for microbial monitoring in dairy applications.

Methods: Flavored yogurt and three cheese matrices (mozzarella, pecorino, and brie) were artificially inoculated with *Escherichia coli* ATCC 25922; *Salmonella* Typhimurium ATCC 14028; and at 30 to 100 CFU per test for TAC, coliforms, and *E. coli*/coliforms detection. *Saccharomyces cerevisiae* ATCC 9763 was used for yeasts/molds enumeration. Sample preparation followed the appropriate ISO method, using 0.9% sodium chloride solution as diluent. Inoculation volume was set at 1 ml.

Results: *E. coli* and *S. Typhimurium* were detected respectively within 24 and 48 h at 35°C in the four matrices with the TAC device. Detection and enumeration was facilitated by the universal red color of colonies. *E. coli* and *S. Typhimurium* were detected with coliform and *E. coli*/coliforms devices within 24 h at 35°C. The *E. coli* coliforms device differentiates by color *E. coli* colonies (red-purple) from coliforms (blue). The 1:10 dilution of the matrices allowed for removal of the light blue background in case of high level of natural flora in the coliform device. *Saccharomyces cerevisiae*, colored in red, were detected within 48 and 72 h at 25°C with the yeast/mold device. Natural flora, also stained in red, was detected in addition to inoculated microorganisms.

Significance: Convenient culture media devices are a reliable alternative that reduces time and workload in food spoilage detection.

P1-36 Estimation of the Microbiological Quality of Minced Pork Using Fourier Transform Infrared Spectroscopy and Multispectral Imaging

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Introduction: Due to the limitations of traditional microbiological approaches, including their time-consuming, destructive, and tedious character, vibrational spectroscopy and multispectral imaging, in tandem with chemometrics, have been increasingly investigated for their use in the evaluation of food quality.

Purpose: The aim of this work was the estimation of the microbiological quality of minced pork through the use of Fourier transform infrared (FTIR) spectroscopy and multispectral imaging (MSI).

Methods: Portions of minced pork were stored aerobically under isothermal (4, 8, and 12°C) and dynamic temperature (periodic temperature changes from 4 to 12°C) conditions for a maximum time period of 14 days. At regular time intervals during storage, duplicate samples were analyzed using conventional microbiological approaches for the determination of the total mesophilic microbial populations, as well as FTIR spectroscopy and MSI. Two independent experimental replicates were conducted. The collected FTIR and MSI data were subjected to pre-processing, i.e., smoothing based on the Savitzky Golay algorithm and SNV transformation, respectively. Partial least squares regression (PLSR) was used to establish the correlation between spectral/imaging data and microbial counts, with the former constituting the input and the latter the output variables in the PLSR models.

Results: PLSR models were calibrated and validated with the data collected from the studied isothermal (170 samples) and non-isothermal (58 samples) conditions, respectively. The values of the coefficient of determination and the root

mean square error for the validation of the model based on FTIR data were 0.834 and 0.91, respectively, whereas the corresponding values for the model based on MSI data were 0.749 and 1.12.

Significance: FTIR spectroscopy and MSI appear to be promising rapid techniques for the quantitative monitoring of the microbiological spoilage of minced pork.

P1-37 Microbiological Spoilage of Cut Ready-to-Eat Pineapple during Storage under Different Temperature Conditions

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Introduction: Ready-to-eat fresh fruits, such as cut pineapple, are very popular among consumers.

Purpose: Evaluation of the microbiological spoilage of fresh-cut, ready-to-eat pineapple under different temperature conditions.

Methods: Trays with sliced pineapple were stored aerobically at different isothermal conditions (4, 8, and 12°C), as well as under a dynamic temperature profile (12 h at 4°C and 12 h at 12°C) for a maximum time period of ten days. At regular time intervals, pineapple samples were analyzed for the determination of the total mesophilic microbial populations, as well as of specific microbial groups including molds and yeasts, *Pseudomonas* spp., lactic acid bacteria, and bacteria of the *Enterobacteriaceae* family.

Results: The dominant microbial group in pineapple was yeasts, with their counts coinciding with the total mesophiles' counts. *Pseudomonas* spp. and bacteria of the *Enterobacteriaceae* family were below 2 log CFU/g throughout storage at all studied temperatures. Moreover, the population of lactic acid bacteria was very low and not easily detectable with common microbiological analyses due to the yeasts' dominance. The initial level of yeasts (mean±standard deviation, n=4) was 4.74±0.60 log CFU/g, while the final populations were 6.90±0.70, 7.63±0.20, and 7.64±0.40 log CFU/g during storage at 4, 8, and 12°C, respectively. The maximum specific growth rate of yeasts was estimated to be 0.058, 0.134, and 0.151 h⁻¹ for 4, 8, and 12°C, respectively, whereas no lag phase was observed at any of the studied temperatures. The growth monitored during storage at the dynamic temperature conditions resembled that at 8°C.

Significance: Although pineapple is a widely consumed fruit, there are very few studies on its microbiological spoilage under different temperature conditions.

P1-38* Sequencing the Food Factory: Environmental Microbiota Monitoring – the Smoked Salmon Case Study

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Introduction: Surface hygiene is considered a main part of the quality system in food processing plants. However, surface bacteria are commonly not identified and their roles in food spoilage and safety are generally unknown. These residential communities persist in food processing plants due to growth at low temperatures, biofilm formation, and tolerance to biocides. They may affect food quality through cross-contamination at each step of a process. Next-generation sequencing (NGS) technologies like metabarcoding 16S can be used to appreciate the potential implications of surface microbiota for food safety and quality

Purpose: The aim of this study is to develop a metabarcoding methodology to monitor bacterial communities in food processing plants. This could be useful in characterizing microbial reservoirs and improving targeted hygiene procedures, and may lead to improved shelf life and product quality.

Methods: Surface samples from a smoked salmon production plant and from products were analyzed using a polyphasic approach. Bacteria were plate counted and identified by MALDI-TOF MS and full 16S rDNA sequencing. DNA was extracted from samples using PCR to amplify the V3-V4 region of 16S rDNA and sequenced on Illumina MiSeq. Taxonomic classifications were obtained using FROGS pipeline, Silva 16S reference database, and RDP classifier.

Results: An environmental surface strain collection was identified. NGS were useful to evaluate bacterial community diversity and dynamics in the environment and on products. From 12 surfaces and product samples, 180 operational taxonomic units could be identified. Beta diversity enabled identification of a core community between the processing environment and products. This core microbiota is mainly composed of gram positive spoilage bacteria (lactic acid bacteria, *Brochothrix*) and gram negative (*Enterobacteriaceae*, *Psychrobacter*, *Pseudomonas*, and *Shewanella*).

Significance: A better understanding of microbial dynamics within processing environments could help to reduce contamination and spoilage. The development of a metabarcoding-based method allowed to improve cleaning and disinfection procedures could reduce food wastage and enhance product quality.

P1-39 Shelf Life of Functional Orange Juice and Inactivation of *Alicyclobacillus acidoterrestris* after High-pressure Processing and Thermal Treatment

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Introduction: As consumers' demand for functional foods has increased and new technologies are able to enhance food safety and quality, the development of a fruit juice processed with high-hydrostatic pressure (HHP) and enriched with probiotics is an interesting approach.

Purpose: To evaluate the potential of HHP to preserve the quality and natural freshness during the shelf life of a functional orange juice containing a potential probiotic *Lactobacillus pentosus* strain. Moreover, to study the inactivation of *Alicyclobacillus acidoterrestris*, a target spoilage microorganism in the fruit juice industry, and its possible interaction with the inoculated probiotic strain.

Methods: Two treatments were applied in fresh orange juice: thermal at 60°C for 10 min and HHP/thermal at 600 MPa and 60°C for 10 min, while fresh untreated juice was used as control. The samples were inoculated or not with spores of *A. acidoterrestris* Aac (10⁷ CFU/ml) before processing and/or with the potential probiotic *L. pentosus* E104 (10⁷ CFU/ml) after processing. Microbiological, physicochemical, and sensory analyses were performed during storage of the samples at 4°C for 1 month.

Results: The population of the probiotic *Lactobacillus* strain remained stable during the 1-month storage in all cases, while the levels of *Alicyclobacillus* remained stable in the control and heated samples, but reduced approximately

1.8 log cycles in HHP-treated samples. According to sensory analyses, the samples with *Lactobacillus* were acceptable for eight days and the samples containing both strains for ten days of storage in all treatments. The control and heated juices with *Alicyclobacillus* were acceptable for 12 days, while the HPP treated samples for 18 days. All non-inoculated samples were acceptable during storage.

Significance: The results showed that HHP may enhance the shelf life of fresh juice when contaminated with *A. acidoterrestris*, while a possible antagonistic activity with the probiotic *Lactobacillus* related to reduced production of off-flavours is evident.

P1-40 Antimicrobial Activity of *Saccharomyces cerevisiae* against Fungi Associated with Food Quality and Safety

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Introduction: Fungal species present in selected foods such as cereals, dried fruits, etc. are common members of the indigenous microbiota able to cause spoilage or produce toxins, depending on species characteristics.

Purpose: The aim of the present study was to assay *Saccharomyces cerevisiae* FMCC Y₃₃ as a control factor against fungal growth.

Methods: Competition experiments were conducted using 35 different fungal species belonging to *Penicillium*, *Aspergillus*, *Phoma*, *Fusarium*, *Mucor*, *Trichoderma*, *Rhizopus*, *Cladosporidium*, *Aureobaculum*, and *Alternaria* genera. Yeast and fungal purity was checked using Yeast Malt Agar and Malt Extract Agar cultured at 25°C for 2 and 7 days, respectively. *S. cerevisiae* was inoculated on MEA Petri dishes (approximately 10⁵ CFU/mL) and fungal species were spot inoculated at approximately 10⁵ conidia/mL. Control plates were also inoculated in every case study. Six replicates were prepared and the plates were incubated at 25°C for 14 days. Fungal growth measurements in terms of growth area of mycelium were taken at 0, 3, 5, 7, 10, 12, and 14 days.

Results: Regarding the genus *Aspergillus* (e.g., *Aspergillus carbonarius*), growth area was inhibited by 98.62% at 14 days. Similarly, for *Penicillium* spp. (e.g., *Penicillium commune*) and *Fusarium* spp. (e.g., *Fusarium oxysporum*), the inhibited area was 96 and 98.3%, respectively, at 14 days. Similar results were observed for all studied species.

Significance: *S. cerevisiae* FMCC Y₃₃ can be considered as a natural antimicrobial agent, able to suppress or limit the growth of many different fungal species which colonize food matrices and are considered as food spoilers or pathogens due to toxin production.

P1-41 Analysis of Aflatoxin M1 in Breast Milk and Its Association with Nutritional and Socio-Economic Status of Lactating Mothers in Lebanon

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Introduction: Aflatoxin B1 (AFB1) is the most potent of the dietary aflatoxins and its major metabolite, Aflatoxin M1 (AFM1), is frequently observed in the breast milk of lactating mothers.

Purpose: The aim of this study was to assess the occurrence and factors associated with AFM1 contamination of breast milk collected from lactating mothers in Lebanon.

Methods: A total of 111 breast milk samples were collected according to the guidelines set by the World Health Organization. Samples were analyzed using competitive

ELISA assay between December 2015 and November 2016. A survey was administered to determine the demographic and anthropometric characteristics of participating lactating mothers. Dietary habits were assessed using a semi-quantitative food frequency questionnaire.

Results: Mean concentration of AFM1 in the breast milk samples was 4.31 ± 1.8 ng/L, while 93.8% of samples contained AFM1 with a range of 0.2 to 7.9 ng/L. The mean level of AFM1 was significantly ($P < 0.05$) lower in fall and winter (4.1 ± 1.9 ng/L) compared to spring and summer (5.0 ± 1.7 ng/L). None of the samples exceeded the European Commission regulation (25 ng/L) for milk infant formula. AFM1 contamination was significantly ($P < 0.05$) associated with the daily consumption of white cheeses, but not meat or cereal products. No significant ($P < 0.05$) association was observed between AFM1 levels in breast milk and anthropometric, socio-demographic factors (age and level of education) or the governorate of residence of nursing mothers. The mean AFM1 estimated daily intake was found to be 0.69 ng/kg bw/day.

Significance: Although the incidence of AFM1 contamination was low, our first-of-its-kind study highlights the importance of more investigations on mycotoxin contamination in breast milk, in addition to protection strategies to tackle the exposure of infants to this potent chemical hazard.

P1-42 From HACCP to Global Standardisation of Hospital Food Safety and Hygiene Policies

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Introduction: The main reasons behind this study were the potential impact on the development of food safety and hygiene in Kingdom of Saudi Arabia (KSA) state hospital kitchens and continuous improvement in England's state hospitals, leading to further standardisation.

Purpose: The research aimed to identify the strengths and weaknesses of five National Health Service food safety policies in England (Lincolnshire, Northampton, Southampton, Nottingham, and Cumbria) and to establish a pilot policy for KSA hospitals.

Methods: Secondary data analysis in England's hospital policies identified the main similarities and differences, strengths and weaknesses. The primary research in the KSA state hospitals followed, involving 255 questionnaires.

Results: Questionnaire response rate was 72% (hospital catering managers/supervisors, 65%; supervisors employed by contracted catering companies, 5%; and contracted catering workers, 35%). The majority of female participants were in the role of managers, overall 42% (33) males and 58% (47) females. Also, older generations (more than 41 years old) are not represented in supervisory roles at all and qualifications vary as well. Knowledge of Hazard Analysis Critical Control Points (HACCP) and training findings are controversial, but they supported the design of the pilot KSA hospital food safety and hygiene policy. The results also showed that England's hospitals policies vary in structure, explanation conciseness, and user-friendliness. The current trend in England is to have consultations with patients, standardised training, explicit roles, responsibilities, clear processes, and use of centralised kitchens.

Significance: In the KSA, the main issues are lack of appropriate training and standardised qualifications, no existing food policy in addition to HACCP, and a high employee turnover rate. The importance of this paper is twofold: guidelines for improvement and standardisation of England's state hospital's policies, and transferable values are applicable in other hospitals, especially in developing countries that have adopted HACCP.

P1-43 Mitochontrakr: Mitochondrial Genome Assemblies of Insects Commonly Known to Infest Foods

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Introduction: Complete mitochondrial genomes are useful references for a wide variety of population genetic, phylogenetic, and comparative genomic analyses. There is a unanimous consensus that some insect species are important vectors of foodborne pathogens. Although targeted amplification techniques are being developed to identify insects in foods and food ingredients, there are always cases when more information is needed to distinguish between closely related insect contaminants. Thus, reference collections of fully sequenced mitochondrial genomes can provide new amplification targets, as well as sufficient information to distinguish between closely related species.

Purpose: To generate a high-quality reference collection of insect mitochondrial genomes for use with targeted and target-independent sequence-based detection methodologies.

Methods: Fifty-two insect species belonging to 19 families and six orders were used for genomic DNA extraction using the Qiagen DNeasy blood and tissue kit and sequenced on the Illumina NextSeq platform. Denovo assembly and iterative mapping were performed using NovoPlasty 2.6.2 and MITObim. Circularized assemblies were annotated using MITOS and Geneious.

Results: Fifty-two near-complete mitochondrial genome assemblies and contigs have been annotated and submitted to NCBI for future reference relatives.

Significance: This collection of complete mitochondrial genome sequences from insects and closely related insects known to commonly contaminate foods has been made publicly available at the National Center for Biotechnology Information (NCBI) under the header MITOchonTrakr. The publicly available data include annotated assemblies, raw sequencing data, and authenticated reference specimens. This collection should enable a variety of future target-based and target-independent detection assays.

P1-44* Quantitative Risk Assessment of Human Salmonellosis Attributable to the Consumption of Meat-based Meals in Kigali City, Rwanda

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Introduction: Human salmonellosis is one of the most noticeable foodborne diseases worldwide and meat constitutes one of the major vehicles for human *Salmonella* infection.

Purpose: The objective of this study was to assess the risk of developing *Salmonella* illness associated with the consumption of meat-based meals in Kigali, Rwanda inhabitants through a quantitative microbial risk assessment (QMRA) model and to determine through the model the efficacy of different interventions aimed at reducing the risk of *Salmonella* along the Rwandan meat production chain.

Methods: In the present study, the risk of *Salmonella* illness attributable to the consumption of meat-based meals was assessed through a QMRA model by using the Codex Alimentarius approach, and three main risk exposure pathways (namely beef consumption within the household, as well as beef and goat meat consumption outside the household) were considered.

Results: The number of human salmonellosis cases associated with the consumption of meat-based meals in Kigali was found to vary between 17,447 and 33,986 inhabitants per year, depending on the risk exposure pathway; females, as well as young adult consumers, appeared to be less exposed to the risk. The analysis of intervention scenarios aimed at reducing the risk of human salmonellosis within slaughterhouses, meat retail establishments, and the kitchens of households and collective catering establishments in Kigali showed a relative risk reduction ranging from 22.7 to 83.1%, and the risk reduction yield was significantly higher when different interventions were simultaneously applied at various stages of the meat chain.

Significance: Data gathered through this study would be helpful in monitoring the risk of *Salmonella* illness attributable to the consumption of meat-based meals in Rwanda and in other countries with comparable meat production chains.

signature, with shifts in the spectral bands corresponding to changes in membrane fluidity. Further analysis of the Amid bands permitted to identify this second group as spores in their early stage of germination.

Significance: This study shows for the first time reversible spore modifications by HP that could be used to design new sterilization processes.

P1-45 Reversible Initiation of Spore Germination by High Pressure at 20°C

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Introduction: Bacterial spores are extremely resistant life forms that play an important role in food spoilage and foodborne disease. High Pressure (HP) Process at mild temperatures (<50°C) is known to initiate the germination of a part of a spore population. In a preliminary work, we demonstrated that HP also induces reversible or irreversible nisin sensitization of another part of the spore population depending on the HP and T levels applied.

Purpose: The objective of the work is to investigate the nature of reversible and irreversible spore modifications induced by HP using Infra-red facilities in synchrotron Soleil. The changes of biochemical signatures of spore structures (lipids, proteins) were investigated both in situ and after HP treatments.

Methods: 8nL of a concentrated spore suspension of *Bacillus subtilis* PS533 in D2O were pressurized in a diamond anvil cell at 500 MPa, 20°C or 50°C for 10 min and IR spectra were recorded each minute thanks to in situ HP synchrotron source FTIR (Fourier Transform Infra-Red) spectroscopy. Experiments were repeated 3 times. Spores were also treated by HP in a small portatif HP vessel (one-mL samples at 108 CFU/mL). After treatment, individual IR spectra of spores were recorded using synchrotron radiation-based -FTIR microspectroscopy. 160 individual spectra were analysed by PCA (Principal Component Analysis) to identify spectral regions responsible for spectra differences. These measures were completed by plate count to determinate the inactivated and germinated spore fractions after treatments. Results were calculated from 3 independent experiments, and statistically analyzed through ANOVA (Analysis of Variance) and Tukey's HSD (Honest Significant Difference) test with R software.

Results: Our results showed that spores treated by HP at 50°C and germinated spores had similar spectral signatures involving same structural properties. However, after HP performed at 20°C, two groups of spores were distinguished; one of these groups was clearly identified as germinated spores. The second group displayed a unique spectral

P1-46 An Exposure Assessment of *Salmonella* in Poultry Product as a Tool for the Redesign of the HACCP Plan

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Introduction: It was reported that salmonellosis is one of the most common foodborne diseases in the world. There are many type of foods potentially contaminated with *Salmonella*, such as poultry meats due to cross-contamination. The situation is likely to get worse because of abusive time-temperature management in the food chain. As recommended by Codex Alimentarius, the Hazard Analysis Critical Control Points (HACCP) concept has been widely used as a tool to manage the safety of food in food companies, which remains a challenge for small- and medium-sized food companies.

Purpose: This research aimed to evaluate and redesign current HACCP plans in a poultry company by using quantitative microbial risk assessment.

Methods: Literature review was performed to collect relevant data related to prevalence and concentration of *Salmonella* in poultry meat. We collected the time-temperature profile in the poultry company by using a data logger. We also performed in-depth interviews and site visits to validate the obtained information. To evaluate the exposure, a Monte Carlo simulation was used with @Risk software with 10,000 iterations. Furthermore, a sensitivity analysis was performed to allow the selection of scenarios that can lead to effective intervention for reducing risk.

Results: Preliminary data showed that there were some serious stages of temperature abuse in the poultry chill chain. The overall average temperature was 12.0°C and the highest temperature went up to 29.7°C. The estimated exposure assessment in this study per package of fresh poultry was 2.64 log MPN. Sensitivity analysis indicated that initial contamination, temperature of distribution, processing and warehouse, and time of cutting have significant effects on bacteria load at the end of storage. Moreover, a new HACCP plan was proposed based on these outcomes.

Significance: These findings could help poultry meat and other associated food companies to identify, evaluate, select, and implement better risk-based food safety management.

P1-47 A Method for Estimating the Risk Level for Foodborne Illness Due to Individual Food Items Using Dynamic Time-temperature History Scenarios during Food Distribution

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Introduction: Statistics from the Centers for Disease Control and Prevention (CDC) show that most outbreaks of foodborne disease (FBD) occur when food is mishandled. One of the leading factors is inappropriate time-temperature conditions, in which food is maintained at improper holding temperatures. Food temperature can fluctuate during distribution, which is referred to as the non-isothermal dynamic time-temperature history (TTH).

Purpose: The main purpose of microbial risk assessment (MRA) is to estimate the risk of microbial hazards with the potential to cause foodborne illness in consumers. MRA can be performed to evaluate hypothetical scenarios for the occurrence of FBD resulting from ingestion of foods with non-isothermal dynamic TTH.

Methods: We estimated the risk of *Staphylococcus aureus* contamination in sandwiches as an MRA example. In addition, we compared the microbial risk estimation for sandwiches containing *S. aureus* under three scenarios with different dynamic TTH parameters.

Results: We developed a model for estimation of the risk of foodborne illness in various food distribution environments with dynamic TTH data and “what if” scenarios, using isothermal growth data to predict non-isothermal growth patterns. We found that sandwiches distributed under specific TTH conditions (e.g., distribution environments with large temperature spikes) are more likely to cause FBD.

Significance: Using MRA and dynamic TTH scenarios, this approach suggests a method for estimating the likelihood of FBD due to individual food items that can be used to develop preventive strategies and epidemiological surveys for FBD.

Introduction: Danish food-based dietary guidelines advise the Danish population to increase the consumption of fish while decreasing the consumption of red and processed meat due to evidence of association with nutrition-related diseases. However, how much would people gain by following these guidelines? And how does the presence of chemical hazards in these foods affect the risk-benefit balance of such a substitution?

Purpose: We quantified the overall health impact of substituting red and processed meat with fish in a Danish adult diet by means of a risk-benefit assessment.

Methods: We modeled the substitution of red and processed meat with fish for Danish adults (≥ 15 years) based on consumption data from the Danish National Survey of Diet and Physical Activity. We defined and compared four substitution scenarios based on varying chemical and nutrient exposures to the current consumption and quantified the overall nutritional and toxicological health impact of the substitutions in terms of Disability-Adjusted Life Years (DALYs).

Results: Approximately 150 DALYs per 100,000 individuals could be averted each year if the adult Danish population increased the consumption of fatty fish (-153.59 DALYs [95% uncertainty interval (UI): -192.98 to -115.70]) or a mix of fatty and lean fish (-148.99 DALYs per 100,000 [95% UI: -187.18 to -110.39]) to 350 g/week and correspondingly decreased the intake of red and processed meat. A lower beneficial impact of the substitution was estimated when substituting with lean fish (-79.85 DALYs per 100,000 [95% UI: -103.19 to -59.41]) and a marked health loss was estimated when substituting with tuna only (183.01 DALYs per 100,000 [95% UI: 75.75 to 326.31]).

Significance: Our results show an overall beneficial effect of substituting red and processed meat with fish to reach the recommended intake of fish if the proportion of large predatory fish is low. A larger benefit was estimated when substituting with fatty fish compared to only lean fish.

P1-48* Reinterpretation of the Mathematical Description of Variability in Bacterial Growth Using Stochastic Analysis of Individual Cell Division Times and Its Application Using a Computer Simulation

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Introduction: Conventional bacterial growth models describe changes in the average number of bacterial cells. However, because of individual cell heterogeneity, conventional kinetic models do not provide precise estimates of the time required for a population to reach a certain size. This makes it difficult to evaluate the risk of achieving adequate numbers of bacteria to cause foodborne illness or food spoilage.

Purpose: This study aimed to describe the variability in the time required to reach a certain population level via a mathematical formula and computer simulation.

Methods: We assumed that bacteria grow discretely and exponentially over time. Our target model was an exponential growth model without lag time. The variation in the division timing of individual cells was described as an exponential distribution. Then, we aggregated the division timing of individual cells to describe the time required to reach a certain population level. In addition, we simulated a stochastic growth model based on our mathematical formula. We set three different growth rate parameters to compare stochastic bacterial growth, and simulated growth from initial cell numbers of 1, 10, and 100 to a population of 10^4 cells.

Results: The required time to reach a certain population level could be calculated by the convolution of the exponential distribution, and our computer simulation revealed trends of stochastic bacterial growth. For example, as the number of initial cells decreased, the variance in time required to reach 10^4 cells increased. As the growth rate slowed, the time variance also increased. Our stochastic analysis enabled the calculation and visualization of the variability in time required to reach a certain population level, which has been described as point estimation.

Significance: Our work enables the stochastic calculation of variability in bacterial growth, which will be useful in determining the risk of the pathogenesis and spoilage.

P1-49* Risk-benefit Assessment of Substituting Red and Processed Meat with Fish in a Danish Diet

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P1-50 Survey of Home-handling Practices on Ready-to-Eat Products in Italian Households

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Introduction: Exposure assessment is a crucial part of microbial risk assessment. The lack of data about consumers’ habits at home related to food storage and preparation is often a great limitation. Ready-to-eat (RTE) products are particularly at risk of at-home bacterial contamination and/or proliferation, as they do not undergo any treatment to reduce microbial loads before consumption.

Purpose: The purpose of this study was to evaluate at-home, pre-consumption behaviours of Italian households that could impact the microbial safety of high-risk RTE foods.

Methods: A telephone survey was administered to 1,757 households evenly distributed throughout Italy. Interviews were carried out during a two-month period. Households were included in the survey only if they had purchased, during the previous 7 days, foods encompassed in one of the following high-risk RTE food categories: (1) meat products (722); (2) raw vegetables (528); or (3) soft and semisoft cheese (507). All questionnaires included from 15 to 18 questions related to demographic information, product characteristics, amount purchased, storage, preparation, and cooking habits.

Results: Interestingly, only 6% of interviewed people cooked meat products before consumption, while 69.7% washed vegetables even if they were explicitly labelled as “ready-to-eat.” Of the consumers, 37.7% did not remove cheese rind. All products were usually stored before consumption, but normally according to manufacturer’s instructions. Similarly, 57.8% did not remove salami or sausage casing.

Significance: It is known that a significant proportion of foodborne diseases arise from unsafe at-home practices. According to our results, high-risk behaviours (e.g., room

temperature food storage) are rare; however, potentially risky habits (e.g., eating cheese rind or salami casing or storing RTE perishable products for a long time) are quite widespread. Information related to the frequency of unexpected behaviours, such as cooking seasoned meat products or washing minimally processed vegetables, could be profitably added to risk models in order to get more precise results.

P1-51 **Campylobacter Control Measures in Indoor Broiler Chicken: Conservative Reassessment of Cost-utility and Analysis of Putative Barriers to Implementation**

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Introduction: Campylobacteriosis is one of the most important human foodborne infections, attributed to broiler chicken consumption in about 30% of cases.

Purpose: In a formerly published cost-utility model of potential *Campylobacter* control measures in the European Union (EU-27), disease burden and cost of illness estimates were driven by data from The Netherlands. The aim of our work was to gather country-specific cost-of-illness estimates, to generate a conservative estimate of *Campylobacter*-related disease burden expressed in quality-adjusted life years (QALYs), and to re-assess the cost-effectiveness of control measure options.

Methods: Data from The Netherlands on productivity loss and direct healthcare costs were corrected for country-specific gross average wages and total health expenditure per capita, respectively. Health burden due to acute gastroenteritis, Guillain-Barré syndrome, and reactive arthritis were estimated from published data. Inflammatory bowel disease and irritable bowel syndrome were omitted, in line with a World Health Organization opinion.

Results: Based on the adapted model, the EU-wide implementation of the available and acceptable measures against *Campylobacter* in indoor broiler chickens would be a dominant strategy as compared to the current practice, yielding 26,400 QALY gain and a cost savings of €85 million each year. The expected cost savings is due to the decrease of productivity loss (69%) and direct healthcare costs (31%). As the investigated strategy will not be reimbursed by healthcare payers, the societal perspective is justified. The poster also presents a strategic pricing exercise on bacteriocin treatment in the United Kingdom (UK) and in Hungary (an intervention option currently under development).

Significance: Implementation of the investigated control strategy would be dominant at the EU-27 level. The cost-effectiveness of add-on bacteriocin treatment in the UK could not be justified at the assumed price level. Application of health technology assessment methodology in food safety and nutritional policies could improve public health, and also the allocative efficiency of public health budgets.

P1-52 **Development of Safety Assessment Methods for the Usage of Sanitizers and Disinfectants on Food Contact Surfaces**

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Introduction: Nowadays, to ensure the hygiene and safety of food, many brands of chemical sanitizers and disinfectants are being used to sanitize and disinfect food contact

surfaces, and the kinds and types used are being gradually increased and diversified.

Purpose: In order to control the safety of these products for public health, we developed safety assessment methods for sanitizers and disinfectants according to risk assessment guidelines. The safety assessments evaluate new risk information based on the toxicological data and estimates of direct ingestion exposure to sanitizers and disinfectants that are applied to food contact surfaces and further determines the likelihood of harm to the human body in comparison with the acceptable daily intake or the acute reference dose based on estimated daily exposure.

Methods: Estimated daily intakes of sanitizers and disinfectants were calculated using food-consumption data for the Korean population derived from the Korea National Health and Nutrition Examination Survey and survey data based on the behavior of consumers in public eating places or homes that use sanitizers and disinfectants. These behaviors include how sanitizers and disinfectants are used (kinds of products, daily usage level, and preparation patterns) and how consumers prepare food on food-contact surfaces.

Results: The evaluation of risk level based on the daily exposure estimates of sanitizer and disinfectant revealed no health concerns for consumers.

Significance: Developed method is used as a decision-making tool to achieve improved public health and safety. Also, the results of this study will be re-evaluated if new information is confirmed in the future.

P1-53 **Comparison of Sanitary Inspection Results on Distribution Trays by the Type of Children's Food Service**

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Introduction: Since the establishment of Changwon Center for Children's Foodservice Management (CCFSM) in Korea in 2011, Center dietitians have been performing inspections on children's food service based on the Korea Food and Drug Administration's sanitary checklist. Children's food service can be divided between institutional food service, which is regulated by the Food Sanitation Act, and small food service, which is exempt. Distribution trays are most frequently sampled in sanitary inspections for food contact surfaces by CCFSM.

Purpose: The purpose of this study was to make a sanitary consulting strategy on distribution trays by comparing the results of simultaneously performed 'inspection by checklist', 'adenosine triphosphate (ATP) monitoring', and 'aerobic plate count (APC)' between the two types of children's food service.

Methods: Five dietitians visited 223 food service establishments (95 institutional, 128 small) to examine whether they performed 'keep distribution tray sanitarily by washing/sanitizing' and 'perform food distribution in a clean and appropriate way' as required. In this visit, dietitians swabbed a 100-cm² area of distribution trays two times, one for obtaining ATP measurements and the other for APCs using 3M Petrifilm Plates. Chi-square tests and *t*-tests were applied by SPSS 23.0.

Results: Mean of APCs from all inspections was $3.8 \times 10^2 \pm 2,102.0$ CFU/100 cm², and 208 (93.3%) trays were accepted by APC standard (less than 5.0×10^2 CFU/100 cm²). But in APCs, institutional food service establishments ($1.4 \times 10^2 \pm 600.0$) showed significantly ($P < 0.01$) higher rates than those of the small food service establishments ($5.5 \times 10^2 \pm 2,718.7$) and turned out to be performing better sanitary management. There was no significant difference between the two groups in ATP measurements (institutional: $766.4 \pm 2,704.3$ RLU/100 cm²; small: $647.5 \pm 2,194.4$) and in performance rate of two checklist items.

Significance: Compared to the 93.3% of APC adequacy from the total inspections, total ATP adequacy (standard: below 300 RLU/100 cm²) was only 71.7%. Therefore, more realistic guidelines should be prepared to conduct ATP hygiene monitoring for food contact surfaces, including distribution trays.

P1-54 Guideline on Bioluminescence ATP Hygiene Monitoring for Distribution Trays in Children's Food Service

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Introduction: Changwon Center for Children's Foodservice Management (CCFSM) has used adenosine triphosphate (ATP) bioluminescence to detect microbial contamination and food residues in children's food service facilities. It provides real-time estimates of total surface cleanliness, and dietitians use the results to complement inspection by sanitation checklists. 3M Guidelines (pass: below 300 reactive light units (RLU)/100 cm²; fail: above 300 RLU/100 cm²) are primarily used. Meanwhile, dietitians wanted to specify additional and more detailed limits to monitor food contact surfaces according to their types.

Purpose: This study was conducted to set a guideline for ATP hygiene monitoring for distribution trays in children's food service administered by CCFSM.

Methods: Five dietitians from CCFSM visited 214 food service establishments to swab a 100-cm² area of distribution trays using 3M Clean-Trace Surface ATP Test Swabs and measure ATP using a 3M Clean-Trace NG Luminometer. Data processing was handled by SPSS v. 23 to conduct graphical and statistical analysis. The raw data were transformed by a Box-Cox transformation. The mean +3 standard deviations or +2 standard deviations can be calculated based on process control procedures.

Results: Mean of 160 (71.7%) passing ATP measurements were 90.9±75.9 RLU/100 cm² (Min: 4, Max: 297); mean of 63 (28.3%) failed ATP measurements were 2,240.3±4,192.9 RLU/100 cm² (Min: 313, Max: 24,123). Transformed data showed normal distribution by Kolmogorov-Smirnov test. In graphical analysis, a high RLU value peak appeared at 150 RLU (55.4%), which was considered an ideal level. In statistical analysis 1,250 (=242.1) RLU (67.9%) was considered a satisfactory limit with reference to 2XSD. In statistical analysis 2,350 (=317.7) RLU (72.3%) was considered unsatisfactory limit with reference to 3XSD.

Significance: Distribution trays in between 250 and 350 RLU/100 cm² can be improved and able to pass 3M guidelines with a corrective measure of washing and sanitizing.

P1-55 Hygiene and Health Features of Edible Bivalve Molluscs Traded in Lombardy Region of Italy in 2017

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Introduction: Edible bivalve molluscs are frequently the cause of very serious outbreaks of food poisoning. Microbiological suitability for human consumption, according to European Union (EU) Regulation 2073, is based on two fecal bacteriological contamination parameters (*Escherichia coli* and *Salmonella*), but it is fundamental to consider other microorganisms which are naturally present within the marine environment and are potentially pathogenic, such as viruses.

Purpose: The object of this work was to analyze the data obtained from samples collected by public health officers in food stores of the Lombardy region of Italy in 2017, and to perform an overview of the hygienic condition of this food matrix.

Methods: A total of 307 samples (232 mussels, 48 clams, 25 oysters, 2 others) were collected in one- or five-sample units (s.u.). A total of 1,060 s.u. were analyzed. The detection of *Salmonella* with RT-PCR technique and further confirmation of positivity with culturing method was performed on 1,040 s.u., and 1,048 s.u. were analyzed for *E. coli* with most probable number (MPN) count. A total of 33 samples (41 s.u.) were also analyzed for the hepatitis A virus and for norovirus detection with RT-PCR technique.

Results: *Salmonella*, serotyped as *S. rissen*, was found in one s.u. Three samples had unsatisfactory results for *E. coli*, according to EU Regulation: one had a value > 700 MPN/100 g in one s.u., and two had a value between 230 and 700 in 2 of the 5 s.u.. Remarkably, 938 s.u. (89.5%) had < 18 *E. coli* MPN/100 g. One sample was positive for norovirus G1.

Significance: In general, the edible bivalve molluscs marketed in Lombardy showed a satisfactory level of hygiene, both for the microbial and the viral parameters. The low level of *E. coli* contamination suggests it would be profitable to replace the MPN count with the more cost-effective and less laborious pour plate count method.

P1-56 Development of Predictive Growth Model for Pathogenic *Vibrio parahaemolyticus* Strains in Cooked Shrimp

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Introduction: *Vibrio parahaemolyticus* has been the major threat of food poisoning in Taiwan, and a possible mechanism leading to illness is cross-contamination of cooked shrimp. *V. parahaemolyticus* is a gram negative, halophilic bacterium that occurs naturally in estuarine environments and is transmitted to humans primarily through the consumption of raw or mishandled seafood, such as oysters and shrimp. However, information is limited about the growth of pathogenic *V. parahaemolyticus* in cooked shrimp under commercially relevant storage conditions.

Purpose: This study aimed to produce a predictive model for the growth of pathogenic *V. parahaemolyticus* in cooked shrimp over a range of temperatures.

Methods: Six strains of *V. parahaemolyticus* isolated from years 1992, 1993, 1994, 1995, 1996, and 1997 were studied. Cooked shrimp were placed in sterile containers and stored at 10, 15, 20, 25, 30, and 35°C. At each sampling time, samples were analyzed by a direct-plating method for total *V. parahaemolyticus*. The populations of *V. parahaemolyticus* in cooked shrimp during storage were used to estimate the growth rates. A growth rate as a function of the storage temperature was produced.

Results: Preliminary results showed that at 35°C, the average growth rates of *V. parahaemolyticus* from 1992 and 1997 were estimated about 0.66, 0.75 log CFU/h. At 30°C, the average growth rates of *V. parahaemolyticus* from 1992 and 1997 were estimated about 0.56, 0.66 log CFU/h. A square-root model was developed for the growth rates of *V. parahaemolyticus* in oysters as a function of storage temperature.

Significance: The findings may provide useful information for measuring the risk of *V. parahaemolyticus* infection arising from cross-contamination of cooked shrimp with this pathogen.

Poster Session 2 – Antimicrobials, Applied Laboratory Methods, Dairy and Other Food Commodities, Epidemiology, Food Toxicology, Novel Laboratory Methods, Pathogens, Produce

P2-01 Antibacterial Activity of Nanostructured Chitosan/ Monolaurin Film Prepared by Sol-Gel Technique and Its Application on UF White Cheese

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Introduction: The advantage of using bilayer active packaging based on sol-gel method compared to other active packaging is to modulate the release of antimicrobial agents from films into food.

Purpose: In this study, cellulose-chitosan (CC) nano-structured double-layer films containing 0.5 and 1% monolaurin (ML) were prepared and their microstructures and antibacterial activity against *Listeria monocytogenes* were investigated *in vitro* and ultra-filter (UF).

Methods: Films were developed using sol-gel method. The antimicrobial effectiveness of CC- and monolaurin-incorporated films were assessed according to the disk surface spreading method, and the surface morphology of films were examined using a scanning electron microscope (SEM). The antimicrobial potential of films were also assessed on *L. monocytogenes* in UF cheese during storage at 4°C for 14 days.

Results: The results of antimicrobial activity revealed that the addition of ML significantly ($P < 0.05$) increased the diameter of the zone of inhibition. Moreover, CC film did not show inhibitory activity on *L. monocytogenes*. SEM images showed zinc nanoparticles of 20 to 100 nm size in the film. The addition of 0.5 and 1% ML into CC films made a 2.4- to 2.3-log reduction in *L. monocytogenes* population on UF cheese after 14 days of storage.

Significance: Bilayer active films developed by sol-gel method retain ML in the film for a long time, which is suitable for active packaging of cheese.

P2-02 Inhibitory Effect of Mushroom Extracts against Hepatitis A Virus

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Introduction: Hepatitis A virus (HAV) is a positive-sense single-stranded RNA virus of the *Picornaviridae* family. It causes acute liver infection through the fecal-oral route. Several antibacterial activities of mushroom extracts have been reported to date. However, anti-HAV properties of mushroom components have not been much-addressed so far.

Purpose: This study aimed to examine the inhibitory effect of ten mushroom extracts against HAV.

Methods: Ethanol extracts of *Auricularia auricula-judae*, *Cordyceps sinensis*, *Coriolus versicolor*, *Flammulina velutipes*, *Ganoderma lucidum*, *Grifola frondosa*, *Inonotus obliquus*, *Lentinula edodes*, *Phellinus linteus*, and *Pleurotus ostreatus* were prepared by following the previous study. CCK-8 assay was performed to determine the treatment concentration. The pre-, co-, and post-treatment effect of mushroom extracts against HAV was investigated at concentrations of 50, 100, 200, 300, 400, 500, and 1000 µg/ml on FRhK-4 cells.

Results: Compared with pre-treatment and post-treatment, the co-treatment of *C. versicolor*, *I. obliquus*, and *P. linteus* extracts with HAV significantly reduced HAV titer. The co-treatment of *C. versicolor* at 500 µg/ml and 1 mg/ml concentration showed 1.06±0.09 and 1.35±0.07 log reduction/ml, respectively. The co-treatment of *I. obliquus* at 500 µg/ml and 1 mg/ml concentration showed 0.84±0.09 and 1.02±0.03 log reduction/ml, respectively. *P. linteus* showed 0.76±0.34 and 0.98±0.03 log reduction/ml at 500 µg/ml and 1 mg/ml, respectively.

Significance: *C. versicolor*, *I. obliquus*, and *P. linteus* extracts are potential food components to inhibit HAV in a time- and concentration-dependent manner.

P2-03 Alternatives to Chlorine to Prevent and Reduce Bacterial Cross-contamination during the Washing of Ready-to-Eat Lettuce

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Introduction: Inadequate quality of water used in the washing of fresh-cut produce has the potential to be a direct source of contamination and a vehicle for spreading bacteria.

Purpose: Evaluate the potential application of different disinfectants in water used to wash fresh-cut produce in order to prevent cross contamination.

Methods: Fresh iceberg lettuce pieces (25 g) were washed in 1 L of simulated wash water inoculated with gentamicin-resistant *Salmonella enterica* (LFMFP 687, 10³ CFU/ml) under stirring at 260 RPM for 90 s at 4°C. *Salmonella* concentration was quantified in water and in the produce before and after the washing treatment. Antimicrobial properties of isothiocyanates (ITCs), isothiazolinones (CMIT/MIT), quaternary ammonium compounds (QACs – didecyldimethylammoniumchloride [DDAC] and benzalkonium chloride [BZK]), organic acids (malic and citric acid), bacteriocins (Nisin), and monoterpenes (carvacrol [CAR]) were tested in comparison with NaClO. Treated and untreated samples were kept under modified atmosphere for 14 days at 4°C. Colour, odour and microbial load were evaluated.

Results: Minimum inhibitory concentration (MIC) corresponded to 50 ppm (DDAC), 200 ppm (BZK), 50 ppm (CMIT/MIT), 300 ppm (CAR), and 10 ppm (free chlorine) against *Salmonella*. The combination of BZK (100 ppm) and CAR (200 ppm) was also effective. Nisin, ITCs, and organic acids did not show a MIC between 1 and 300 ppm or IU. These MIC values successfully inactivated *Salmonella* in wash water. CMIT/MIT and DDAC lead to 80% *Salmonella* inactivation in the produce. A concentration of 100 ppm reached 100% inactivation, but colour and odour were affected within 7 days of storage. Free chlorine, BZK and CAR, and CAR did lead to 100% *Salmonella* inactivation in produce, but sensory properties were affected after storage, except for free chlorine.

Significance: CMIT/MIT and DDAC displayed antibacterial activity in water and produce without compromising the produce shelf life, which may make them potential alternatives to chlorine during washing of fresh-cut produce.

P2-04 Antimicrobial Activity and Mode of Action of *Nigella sativa* against *Listeria monocytogenes*

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Introduction: *Nigella sativa* L. (black cumin) is a spicy plant. Thymoquinone (Thq), carvacrol (Car), and p-cymene (p-cy) are the main constituents of *N. sativa* essential oil (EO) and have a broad antimicrobial spectrum including gram negative and gram positive bacteria, viruses, parasites, and fungi.

Bacterial multidrug efflux pumps help bacterial populations to increase their resistance and survive the presence of antimicrobial substances. Two efflux pumps have also been described in *Listeria monocytogenes*. EOs and their compounds, besides antimicrobial activity, can increase antimicrobial activity of some antibiotics.

Purpose: The aim of our study was to investigate the antimicrobial and resistance-modifying activity of *N. sativa* EO and its active compounds against *L. monocytogenes*.

Methods: Broth microdilution, ethidium bromide accumulation, and LIVE/DEAD BacLight cell viability assays were used to evaluate *N. sativa* EO, Thq, Car, and p-cy antimicrobial activity, modulation of antimicrobial resistance, inhibition of antimicrobial efflux, and membrane integrity.

Results: Our results demonstrated a substantial susceptibility of *L. monocytogenes* toward *N. sativa* EO, Thq, and Car strains, while p-cy showed no inhibitory activity. Moreover, a significant reduction in minimum inhibitory concentrations of ethidium bromide and ciprofloxacin were noticed when tested in combination with *N. sativa* EO, Thq, Car, and reserpine. The ethidium bromide accumulation increased in the presence of *N. sativa* EO, Thq, Car, and p-cy and was comparable to reserpine; the membrane integrity was disintegrated in the presence of each compound.

Significance: *N. sativa* essential oil and two of its active compounds, thymoquinone and carvacrol, proved to be efficient modulators of antimicrobial resistance in *L. monocytogenes*, with at least two different mechanisms that contribute synergistically to this activity. Due to the promising modulation of the antimicrobial resistance, *N. sativa* essential oil, thymoquinone, and carvacrol have the potential to be further investigated for the control of antimicrobial resistance in *L. monocytogenes*.

P2-05 Amino Acid Decarboxylase Systems in Food Pathogens and Their Interaction with Dicarboxylic Acids

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Introduction: Fumarate has been used for long time as an antimicrobial preservative in foods. However, no mechanism is known for its activity under acidic conditions apart from that derived from its properties as a weak acid.

Purpose: To elucidate the antimicrobial mechanism of fumarate under acidic conditions.

Methods: Sodium fumarate (SF) fumaric acid (FA) salts demonstrated a high antimicrobial activity under acidic conditions against *Escherichia coli* K12 and *Listeria monocytogenes* 10403S possessing a glutamate decarboxylase system (GAD) and three strains of *Salmonella* possessing a lysine decarboxylase system (LDS). SF's effect on the GAD was determined by examining levels of γ -aminobutyric acid (GABA), the byproduct of the glutamate decarboxylation, by using gas chromatography-mass spectrometry (GC-MS) and enzymatic assays. SF's effect on LDS was assessed through monitoring pH recovery of strains and assessing the levels of lysine present following acid stress using GC-MS.

Results: FA has been observed in a number of studies to provide a high level of antimicrobial activity against pathogens. The presence of 10 mM of SF under low pH conditions (< pH 4) produced significant reduction in survival for *L. monocytogenes* (\pm logs), *E. coli* (\pm logs), and *Salmonella* Enteritidis (\pm logs) in paired Student's *t* test ($P < 0.05$).

GABA analysis indicated that SF influenced the GAD system. Differences were noted in the extracellular levels of GABA as a decrease was observed for *E. coli* and an increase for *L. monocytogenes*. We also found a negative effect of SF on LDS activity, which we will investigate further.

Significance: Elucidating the action of antimicrobials on pathogenic stress mechanisms could result in their efficient elimination from foods and the development of novel antimicrobials or the enhancement of the existing ones.

P2-06 Inhibition of Spoilage Lactic Acid Bacteria on Cured Ready-to-Eat Meats with Sodium Free and Clean-label Antimicrobial Ingredients

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Introduction: Contamination and spoilage by lactic acid bacteria is a major concern in refrigerated deli meats. In response to consumer demands, the portfolio of reduced-sodium and naturally derived antimicrobials are being expanded to provide the option to extend shelf life during refrigerated storage.

Purpose: To compare the inhibitory properties of Provian K (potassium acetate and diacetate blend) and Provian DV (neutralized dry vinegar) on the growth of lactic acid bacteria in cured RTE meats.

Methods: Five treatments of cured deli-style ham (72 to 74% moisture, 1.75 \pm 0.1% salt, and pH 6.2 to 6.4; 156 mg/kg sodium nitrite and 547 mg/kg sodium erythorbate) included a control without antimicrobials and different concentrations of Provian K (0.5 and 0.75%) and Provian DV (0.5 and 0.65%). Cooked products were surface-inoculated with 3-log₁₀ CFU/g of two lactic acid bacteria strains including *Carnobacterium divergens* and *Leuconostoc mesenteroides*, both isolated from spoiled cooked meat products. Inoculated slices (100 g per package) were vacuum-packaged and stored at 4 and 7°C for up to 4 weeks. Triplicate samples per treatment were assayed by enumerating twice on plate count agar (30°C, 48 h) and APT agar with bromocresol purple (27°C, 48 h), respectively.

Results: The control ham supported the increase of lactic acid bacteria to spoilage level (> 6 log) at 3 and 2 weeks storage at 4 and 7°C, respectively. In contrast, hams supplemented with 0.5% Provian K or 0.8% Provian DV showed complete inhibition of lactic acid bacteria for 4 weeks. For complete inhibition at 7°C, slightly higher concentrations of 0.75 and 1% were needed of Provian K and Provian DV, respectively.

Significance: Results from this study validate the efficacy of sodium-free antimicrobials and neutralized dry vinegar in inhibiting growth of lactic acid bacteria on cured ready-to-eat meats, which helps to extend the time to spoilage.

P2-07 Fitness Advantage of *Mcr-1*-Bearing IncI2 and IncX4 Plasmids In Vitro

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Introduction: Multiple mobile elements, especially plasmids, contribute to the rapid spreading of *mcr-1*.

Purpose: To assess the impact of diverse *mcr-1*-bearing plasmids on host fitness.

Methods: Forty-seven commensal *Escherichia coli* isolates recovered from the pig farm where *mcr-1* was first identified were screened for *mcr-1*. *mcr-1*-bearing plasmids were characterized by sequencing. The fitness impact of *mcr-1*-bearing plasmids was evaluated through *in vitro* competition assays.

Results: Twenty-seven (57.5%) *E. coli* isolates were positive for *mcr-1*. The *mcr-1* genes were mainly located in plasmids belonging to IncI2 ($n=5$), IncX4 ($n=11$), IncHI2/ST3 ($n=8$), IncFII ($n=2$), and IncY ($n=2$). IncHI2 plasmids also carried other resistance genes (*floR*, *bla*_{CTX-M1} and *fosA3*) and were only detected in isolates from nursery pigs. Sequences of the representative *mcr-1*-bearing plasmids were almost identical to those of the corresponding plasmid types reported

previously. An increase in the fitness of IncI2- and IncX4-carrying strains was observed, while the presence of IncHI2, IncFII, and IncY plasmids showed a fitness cost, although an insignificant fitness increase was initially observed in IncFII or IncY plasmid-containing strains. Acquisition of IncI2-type plasmid was more beneficial for host *E. coli* DH5 α than either IncHI2 or IncX4 plasmid, while transformants with IncHI2-type plasmid presented a competitive disadvantage against IncI2 or IncX4 plasmid-containing strains.

Significance: Increased fitness or co-selection by other antimicrobials might contribute to the further dissemination of the three epidemic *mcr-1*-positive plasmids (IncI2, IncX4, and IncHI2) in this farm and worldwide.

P2-08 Effect of High Pressure and Natural Antimicrobial on Bacterial Populations from Sea Bream Fillets

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Introduction: Consumers increasingly demand fresh foods that are ready-to-eat or ready-to-cook. Fresh ready-to-cook fish (such as fish fillets) is an appreciated protein source, but at the same time is highly perishable.

Purpose: The aim of the study was to determine the effect of high hydrostatic pressure and activated packaging, singly or in combination, on the preservation of sea bream (*Sparus aurata*) fillets.

Methods: Sea bream fillets were packed in polyethylene-polyamide films not activated with antimicrobials (controls) or activated with different combinations of the bacteriocin enterocin AS-48 (0.4 and 0.8 mg/ml) and thymol (0.25, 0.5, and 0.75%). Samples were pressurized at 200, 300, and 400 MPa for 5 min at room temperature. The treatments that had lowest impact on color and odor were applied in combination (0.5% thymol and 0.8 mg/ml bacteriocin, 300 MPa), and bacterial diversity was estimated by Illumina sequencing.

Results: Log reductions for total aerobic mesophiles ranged from 1.17 to 3.73 ($P < 0.05$) for high-pressure treatments and from 0.5 ($P > 0.05$) to 2.01 log cycles ($P < 0.05$) for the activated films. The combined treatment achieved the greatest reduction in counts (4.13 log cycles, $P < 0.05$) and delayed bacterial growth during refrigerated storage. The main bacterial group in controls (*Proteobacteria*) decreased during storage, while *Firmicutes*, *Bacteroidetes*, and *Actinobacteria* increased. *Firmicutes* were more abundant in pressurized samples towards the end of storage.

Significance: Activated films in combination with a mild high-pressure treatment improve microbial inactivation in sea bream fillets. Bacterial diversity in fillets is affected by storage and treatments.

P2-09 Evaluation of the Antibiofilm Activity of Maleic Acid against *Listeria monocytogenes*

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Introduction: Biofilm formation by foodborne pathogens on surfaces used in food-processing environments is an issue of major public health significance. Hence, the development of effective biofilm control strategies has been the objective of extensive research.

Purpose: Assessment of the antibiofilm activity of maleic acid against the foodborne pathogen *Listeria monocytogenes*.

Methods: The biofilm-forming ability of two *L. monocytogenes* strains was evaluated on stainless steel coupons; in brain heart infusion (BHI) broth with (50 mM) or without maleic acid; at pH values of 5 and 7; at 15°C; and for a total time period of 144 h. At 3, 24, 72, and 144 h of incubation, cells attached to stainless steel coupons were retrieved using the bead vortexing method and enumerated on BHI agar. Moreover, the disinfection efficacy of maleic acid was investigated by exposing mature biofilms of the pathogen, formed on stainless steel in its absence or presence, to different concentrations of this organic acid; the surviving biofilms were evaluated either directly through microbiological analyses or indirectly using conductance measurements.

Results: Maleic acid demonstrated a significant ($P < 0.05$) effect on the biofilm-forming ability of one of the tested strains at pH 5, an observation which, however, was not made at pH 7. The disinfection efficacy of maleic acid also appeared to depend on the tested strains, as well as on the previous pathogen's exposure (i.e., during biofilm formation) to the organic acid. In general, maleic acid concentrations higher than 20 mM were the most effective against *L. monocytogenes* biofilms.

Significance: Maleic acid exhibits an important antibiofilm potential against *L. monocytogenes*, with its exact activity, however, being considerably dependent on strain and environmental conditions. In addition, phenomena of bacterial adaptation and enhanced resistance to this weak organic acid may be encountered and should be taken into consideration.

P2-10 Predictability of Ionic Liquid Toxicity on Food-relevant Bacteria

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Introduction: Ionic liquids (ILs), a new class of solvents with unique and tunable physicochemical properties, were initially envisioned as working alternatives to traditional organic solvents. The availability of antibacterial activity data on ILs is still scarce.

Purpose: To assess and possibly predict IL-toxicity, a structure-activity relationship (SAR) approach was adopted using defined structural motifs. These included varied cationic alkyl side-chain lengths, cation lipophilicity, and diverse anion effects.

Methods: The predictive powers of such SARs with respect to antibacterial effects were compared using a total of 28 ILs on six gram negative and six gram positive food-relevant bacteria. Minimum inhibitory (MIC) and bactericidal (MBC) concentrations of the examined ILs were determined by the serial two-fold dilution microtiter plate method in tryptic soy broth. The MIC was defined as the lowest IL concentration where no bacterial growth could be observed within 24 h. The MBC was defined as the lowest IL concentration leading to 99.9% reduction of the initially inoculated CFU.

Results: Cationic alkyl side-chain effect: Chloride {Cl} anion ILs showed increased toxicity with increasing alkyl side-chain length. Tris (pentafluoroethyl) trifluorophosphate (FAP) anion ILs showed reverse side-chain effect on *Listeria* and *Enterococcus*; Cation lipophilicity: Gram-positive bacteria were generally more susceptible than gram-negative bacteria; Fluorinated anions: Except for FAP, there was no considerable influence on IL-toxicity. Results indicate that while certain limited IL-toxicity responses in bacteria can be predicted from SARs, they caution that predictions cannot be generalized across species.

Significance: This study demonstrates the complex species-specific nature of IL-toxicity and the current limitations of SAR predictability.

P2-11* Assessing the Adaptation Capacity of *Penicillium expansum* in the Presence of ZnO Nanoparticles

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Introduction: Zinc Oxide nanoparticles (ZnO NPs) have been shown to induce morphological aberrations in the fungal structures of *Penicillium expansum* spores and inhibit their proliferation, with ZnO minimum inhibitory concentration (MIC) and non-inhibitory concentration (NIC) of 1.8 and 9.8 mM, respectively. It is yet unknown how the successive application of ZnO nanoparticles can affect fungal adaptation in terms of growth and germination.

Purpose: The objective of this study was to quantitatively assess the potential for fungal spores to adapt to repeated exposure to ZnO nanoparticles.

Methods: Suspension of *Penicillium expansum* spores of a concentration of 10⁶ spores/ml was prepared. Potato extract glucose agar (PGA) plates with ZnO NPs suspension with final concentrations of 1.8, 9.8, and 15 mM were prepared and inoculated with 10 µL of the previously obtained spores' suspension. The plates were incubated at 25°C and two type of quantitative assessments were performed: (i) proliferation by assessing the mycelium diameter increase, and (ii) degree of germination by microscopic assessment of the single spores. A successive treatment at the same ZnO concentrations was also applied by re-harvesting the previous nanoparticle-exposed spores.

Results: The rate of proliferation of *Penicillium* varied between 0.0101 (per 1 h) and 0.0299 (per 1 h) for first and successive ZnO-treated samples. No significant difference was found between the first and second generation of the treated cells. A proportional increase of the lag phase with an increase in the nanoparticle concentration was reported. For the germination studies, it was evident that at the MIC, the *Penicillium* spores had a slower germination rate when compared with the NIC spores. Mild exposure to nanoparticles resulted in a more adaptive behavior of the spores.

Significance: The physiological adaptations of fungal spores may affect their proliferation in the presence of different antifungal factors. Further studies should focus on the impact of numerous sequential exposures to nanoparticle environment.

P2-12* *Yucca schidigera* Extract Enhances the Antimicrobial Efficacy of Organic Acid Blends against *Salmonella enterica* in a Laboratory Broth System

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Introduction: Organic acids are used to control undesirable microbes in foods and are commonly diluted with water to facilitate their application. However, water is a poor wetting agent for hydrophobic food surfaces where pathogens may be located. This problem can be circumvented by use of surfactants to aid better wetting. *Yucca schidigera* (Yucca) extract is a natural plant-derived surfactant that is approved in the United States for food use and contains saponins that have antimicrobial activity.

Purpose: The purpose of this study was to investigate the antibacterial efficacy of lactic-citric acid blend solutions alone or combined with Yucca extract against *Salmonella enterica* in a laboratory broth medium.

Methods: The antimicrobial efficacy of various concentrations of lactic-citric blend (1.5 and 2.5%) with Yucca extract (0.15 and 0.30% volume/weight) was evaluated individually

or combined against a five-serotype mixture of *S. enterica* (approximately 7.0 CFU/ml) by using a time-kill assay. Broth medium without organic acids and Yucca extract served as a control. Treatments were tested at 30, 60, and 90 s. The *S. enterica* survivors were determined by plating samples on tryptic soy agar with yeast extract (TSAYE) and counting colonies after incubation at 35°C for 24 h.

Results: Slight reductions in viability of the pathogen occurred with Yucca extract alone but were not significantly different from viability in control ($P > 0.05$). Lactic-citric acid and Yucca extract combinations exhibited significantly stronger antibacterial ($P < 0.05$) action than each component used individually. The treatment using 2.5% lactic-citric and 0.30% Yucca extract showed the greatest reduction in pathogen viability ($P < 0.01$) with numbers of survivors being approximately 4.5 CFU/ml after 90 s.

Significance: Yucca extract has good potential for enhancing the antimicrobial action of organic acids against *Salmonella* while acting as a surfactant. Results of this research can provide an additional strategy for better control of *Salmonella* on hydrophobic surfaces.

P2-13 Anthocyanin Compositions of Blackberry and Their Protective Effects on Human Umbilical Endothelial Cell Damage Induced by Hydrogen Peroxide

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Introduction: Blackberry anthocyanins were purified and characterized, and their protection effects on cell viability and inhibition of malondialdehyde (MDA) production, in comparison to ascorbic acid, and their regulation of cellular antioxidant enzymes and endothelial cell apoptosis induced by H₂O₂ were studied.

Purpose: The objective of the present study was to investigate protective effects of blackberry anthocyanins on human umbilical endothelial cells from H₂O₂-induced damage through the antioxidant properties.

Methods: Four anthocyanins from blackberries were identified as cyanidin-3-O-glucoside, cyanidin-3-O-arabinoside, cyanidin-3-O-malonyl-glucoside, and cyanidin-3-O-dioxalyl-glucoside by LC-DAD/ESI mass spectrometry. Then 25, 50, 100, and 200 µg/ml of blackberry anthocyanins were used to study their protective effects against damage in cultured human endothelial cells.

Results: All the concentrations of tested blackberry anthocyanins increased cell viability and decreased cell MDA levels significantly under oxidative conditions induced by H₂O₂. The peroxidase, superoxide dismutase, and glutathione S-transferase activities of endothelial cells pretreated with blackberry anthocyanins were also largely promoted. However, only 50 and 100 µg/ml of blackberry anthocyanins could inhibit the cell apoptosis induced by H₂O₂.

Significance: This paper characterized anthocyanins isolated from blackberries and indicated their mechanism of antioxidation *in vitro*. The conclusions in this paper also proved biological functions of blackberry anthocyanins that can be used as food ingredients.

P2-14 AOAC Oma Collaborative Study of the MALDI Biotyper to Confirm and Identify *Salmonella* spp., *Cronobacter* spp., and Other Gram Negative Organisms

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Introduction: Identification and confirmation of microbial isolates is crucial in the analysis workflow, providing decision-makers the information required to release or hold

products. Despite the difficulties in establishing a coherent definition for bacterial species and the long time-to-result, phenotypic and biochemical tests are still widely used and recognized. MALDI-TOF mass spectrometry has significantly changed the identification procedure and is now regarded as a reliable and accurate alternative to quickly identify or routinely confirm with systematic perspectives. The MALDI Biotyper is one of these MALDI-TOF technologies.

Purpose: AOAC Official Methods of Analysis Collaborative Studies were run to assess the performances of the MALDI Biotyper as a confirmation and identification method. The U.S. Food and Drug Administration Bacteriological Analytical Manual, Chapters 5 and 29; United States Department of Agriculture Food Safety Inspection Service Microbiology Laboratory Guidebook 4.09; and ISO 6579:2002 and 22964:2017 reference methods were used in parallel to the alternative method for comparison.

Methods: Fifteen European collaborators participated in the *Salmonella* study. Fourteen collaborators from the United States were involved in the *Cronobacter* study. Each data set was comprised of 24 blind-coded isolates, including 16 target strains (either *Salmonella* or *Cronobacter*) and eight additional gram negative bacteria. The following media were tested: TSA, XLD, and RAPID[®] *Salmonella* for *Salmonella* isolation, and TSA, ESIA, and CCI for *Cronobacter*.

Results: The MALDI Biotyper provided 100% correct identification of *Salmonella*, *Cronobacter*, and other gram negative strains, and thus 100% correct confirmation results from all of the tested culture media. For comparison, the reference procedures produced a correct identification rate of 96.6% for *Salmonella* and 95.5% for *Cronobacter*; for the non-*Salmonella* and non-*Cronobacter* organisms, percentages of 97.5 and 93.8% were obtained, respectively.

Significance: The studies demonstrate the reliability and robustness of the MALDI Biotyper for rapid confirmation and identification. The method is now an AOAC Official Method of Analysis under the reference number 2017.09.

P2-15 AOAC OMA Collaborative Study of the MALDI Biotyper to Confirm and Identify *Listeria* spp., *L. monocytogenes* and Other Gram-Positive Organisms

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Introduction: Identification and confirmation of microbial isolates is crucial in the analysis workflow, providing decision makers the information required to release or hold products. Despite the difficulties to establish a coherent definition for bacterial species and the long time-to-result, phenotypic and biochemical tests are still widely used and recognized. MALDI-TOF MS has significantly changed the identification procedure, and is now regarded as a reliable and accurate alternative to quickly identify or routinely confirm with systematic perspectives. The MALDI Biotyper is one of these MALDI-TOF technologies.

Purpose: An AOAC OMA collaborative study was run to assess the performances of the MALDI Biotyper as a confirmation and identification method. The FDA BAM Chapter 10, USDA/FSIS MLG 8.10 and ISO 11290 reference methods were used in parallel to the alternative method for comparison.

Methods: 17 collaborators received 36 blind coded isolates, including 16 *L. monocytogenes* strains, 12 *Listeria* spp. strains and 8 additional Gram positive bacteria. The following media were tested: TSYEA, Oxford, Ottaviani and Agosti Agar and RAPID[®] *L. mono*.

Results: The MALDI Biotyper provided a correct confirmation of 100% for *Listeria* spp and 99.5% for *L. monocytogenes*, and 97.8% of correct identification at the species level of the tested *Listeria* strains. For comparison, the reference methods provided a correct confirmation rate of 100% for *Listeria* spp. and 87.1% for *Listeria*

monocytogenes, and a correct identification rate of 86.5% of the tested the *Listeria* strains.

Significance: The study demonstrates the reliability and robustness of the MALDI Biotyper for rapid confirmation and identification. The method is now an AOAC Official Method of Analysis under the reference number #2017.10.

P2-16 Validation of a Confirmation Method According to ISO/DIS 16140-Part 6 (2017): A Pilot Study within MicroVal Using the MALDI Biotyper as an Alternative for *Salmonella* spp. Confirmation

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Introduction: The ISO 16140 standard provides technical and interpretation rules for method validation and verification and is comprised of six different parts. Part 6 is currently at the Draft International Standard (DIS) stage and describes the protocol for the validation of alternative (proprietary) methods for microbiological confirmation and typing procedures. The study design was set up in past years, and acceptability limits for the data interpretation were defined based on expert opinion, i.e., maximum number of positive or negative deviations between the reference and alternative methods.

Purpose: Are the defined technical rules sufficiently described for proper method comparison and inter-laboratory studies? Are the proposed acceptability limits (AL) fit for these purposes? Are these AL too restrictive as formulated by some experts in the field? A pilot study was coordinated by the MicroVal Organisation as a proof of concept.

Methods: The MALDI Biotyper was tested as an alternative to confirm *Salmonella* spp. from non-selective and selective agars. A method comparison and inter-laboratory study were realized. A total of 150 *Salmonella* spp. strains and 100 non-target strains were tested by two expert laboratories in the method comparison study. The collaborative study was run by involving a minimum of ten organizations to produce ten valid data sets with 16 target and eight non-target strains.

Results: The MicroVal reviewers and the expert laboratories encountered no specific difficulties in setting up the project, organizing the testing, and interpreting the generated data. The collaborating laboratories were able to easily understand the protocol of ISO 16140-6 and achieve the required tests. All the *Salmonella* spp. strains were correctly confirmed with the MALDI Biotyper on all tested media in the method comparison and inter-laboratory studies, passing the thresholds of the currently defined AL.

Significance: ISO/DIS 16140-6 provides valuable technical rules and interpretation concepts to validate confirmation methods.

P2-17 Evaluation of Alternative Microbiological Methods

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Introduction: There are many rapid and alternative methods for detection of pathogens and indicator bacteria in food and environmental samples. In order to use these methods in the context of official controls and for business operators, (European Commission Regulation No. 2073/2005 Microbiological criteria for foodstuffs), the methods need to

be validated against reference methods according to internationally accepted protocols, as well as assessed and certified by independent parties or authorised by a competent authority.

Purpose: In a third-party certification, the alternative method is compared to a reference method through extensive studies, reviewed by technical experts, and finally certified by an organisation like NordVal International. Hereby, it will be documented how well the methods perform, and if the alternative method will provide equivalent results to the reference method.

Methods: Independent expert laboratories conduct comparison studies and arrange interlaboratory studies according to ISO 16140-2:2016 Part 2: Protocol for the validation of alternative (proprietary) methods against a reference method. For qualitative methods, performance parameters such as sensitivity, selectivity, the relative level of detection and level of detection are determined. For quantitative methods; relative trueness, accuracy profile, limit of quantification and selectivity are studied. A considerable number of samples of different matrices and levels are required in the various studies.

Results: In order for the alternative method to be approved, the results of the studies need to fulfil certain acceptance criteria. If the validation and the criteria are fulfilled, it is documented that the alternative method provide equivalent result to the reference method.

Significance: Certified methods are beneficial for all laboratories and their business associates due to their higher quality and possible reduction in costs related to false or inaccurate results, as well as immense reduction in resources used by individual laboratories in performing extensive validations. Further, certification of alternative methods is required in the Microbiological Criteria in the EU Regulation.

P2-18 Optimisation of Separative Method Coupled with MS Detection for Staphylococcal Enterotoxin A, B, C, D, E and H Detection

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Introduction: Staphylococcal food poisoning outbreaks are a major cause of foodborne illnesses in Europe, and detection of their enterotoxins (SEs) in foodstuffs represents a potential hazard for human health. The Food Safety Laboratory (ANSES) has been appointed European Union Reference Laboratory (EURL) for coagulase-positive staphylococci (CPS). In this context, EURL is responsible for selecting, developing, and validating analytical methods for CPS, making them available to the National Reference Laboratories network.

Purpose: Among the 24 SEs reported in literature, only five (SEA, SEB, SEC, SED, and SEE) can be detected qualitatively by the official method and the new ISO 19020:2017, two enzyme-linked immunosorbent assay methods. Even these methods are highly sensitive; limits due to interference effect and specificity were observed. Therefore, specific and accurate analytical tools are required. In this work, a high resolution mass spectrometry-based method was developed.

Methods: Samples prepared from six SEs standards (SEA, SEB, SEC, SED, SEE, and SEH) were used for the set-up of the analytical protocol. Digestion method, sample concentration step, and ultra performance liquid chromatography mass spectrometry (UPLC-MS, quadrupole-orbitrap) parameters were optimised for detection of specific peptides for each toxin.

Results: Digestion and pre-concentration protocol using Trypsin Gold and speed vacuum were optimised. UPLC parameters (flow rate, mobile phases, and column type) were also optimised. Parallel reaction monitoring mode (PRM) was selected and optimised for detection of at least two specific

peptides, allowing the characterisation of each toxin. Skyline software was used for comparison and confirmation of the selected peptides.

Significance: Few LC-MS-based methods were published, but they focused only on two toxins: SEA and SEB. This optimised protocol on multi-SE toxin analysis in standard solutions can be used for the establishment of new protocol for SEs in food matrices. In this case, extraction and purification protocol must be optimised.

P2-19 EN ISO 19020:2017, Horizontal Method for the Immunoenzymatic Detection of Staphylococcal Enterotoxins in Foodstuffs

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Introduction: Staphylococcal food poisoning outbreaks are a major cause of foodborne illness in the European Union and notification of their occurrence has been mandatory since 2005. According to European Food Safety Authority data, the rate of staphylococcal food poisoning outbreaks increased from 5.2% in 2010 to 9.9% in 2015. Even if an official method was published and had been used by the European Union Reference Laboratory Network for coagulase-positive staphylococci, no performance criteria were standardized for the characterization and validation of methods dedicated to staphylococcal enterotoxin (SE) detection in foodstuffs.

Purpose: The aim of this work was to propose a horizontal method for the detection of SEs in foodstuffs. Inter-laboratory validation trials were organized on SE detection on eight matrices covering five food categories: ready-to-eat food, meat, milk products, pastries, and fish products.

Methods: The published European data on food poisoning outbreaks were used to determine the main food categories and the contamination levels to be used. For each trial, homogeneity and stability studies were performed before sending samples to 54 expert laboratories.

Results: Performances criteria in terms of sensitivity, specificity (>90%), and LOD50 (0.06 ng SEs/g) were established for immuno-enzymatic detection assays dedicated to SE detection in food matrices.

Significance: This work was published in June 2017 as ISO 19020 and will be included in the revision of the European Union regulation 2073/2005 dedicated to microbiological criteria for food.

P2-20 Validation of the BAX System Real-time PCR Assay for STEC Suite in Detecting Shiga Toxin-producing *Escherichia coli* in 25-g Raw Ground Beef Samples Enriched in BPW and mTSB

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Introduction: Shiga toxin-producing *Escherichia coli* (STEC) have natural reservoirs in ruminant animals such as cattle and deer, allowing for transmission to humans if meat or milk from infected animals is harvested. In 2016, haemolytic uremic syndrome (HUS) was most commonly reported from serogroup O26 in Europe, overtaking serogroup O157 for the first time. The low infectious dose and rise of serious health complications indicates the immediate need for a molecular-based, rapid method to detect STEC in raw meats.

Purpose: The purpose of this study was to assess the ability of a Real-time PCR assay suite to detect STEC in 25-g

ground beef samples, enriched with two common enrichment broths: buffered peptone water (BPW) and modified tryptone soya broth with casamino acids (mTSB+caa).

Methods: Raw ground beef ($n=40$) was artificially inoculated with *E. coli* O26 to obtain fractional recovery in 25-g portions and held at 4°C for 48 h to acclimate the target cells. Samples were enriched in either 225 ml pre-warmed (37°C) BPW ($n=20$) or 225 ml pre-warmed (37°C) mTSB+caa ($n=20$). Once homogenized, samples were incubated at 42°C for 10 to 24 h. After incubation, samples were analyzed at 10 and 24 h by PCR and confirmed using the United States Department of Agriculture Food Safety Inspection Service (USDA-FSIS) culture method.

Results: For samples enriched in BPW, real-time PCR assays for STEC detected 11 presumptive positives at both 10 and 24 h. For samples enriched in mTSB+caa, eight presumptive positives were detected at both 10 and 24 h. All PCR results agreed with the USDA-FSIS reference results.

Significance: This study demonstrates there is no significant statistical difference between the STEC real-time PCR method and the USDA-FSIS culture method to detect STEC in 25 g of raw ground beef. Moreover, the BAX System Real-Time PCR method is easy to follow, providing users with rapid, reliable single-shift results, compared to cumbersome IMS procedures in traditional culture methods.

P2-21 Comparison of the Sensitivity TRANSIA PLATE Staphylococcal Enterotoxins Kit for the Detection of Staphylococcal Enterotoxins in Raw Milk Cheese and Infant Formula with and without Dialysis/Concentration

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Introduction: The ISO 19020 standard for the detection of staphylococcal enterotoxins in foodstuffs requires the processing of dairy products using a dialysis/concentration protocol. Is the gain in sensitivity worth the extra work?

Purpose: To compare the POD of the extraction of various levels of SEC₂ from liquid infant formula and SEC₃ from raw milk cheese with and without the dialysis/concentration protocol outlined in the ISO 19020 standard.

Methods: Formula was contaminated directly with low levels of SEC₂. Portions of 25 ml each were tested either directly or processed according to ISO 19020, sections 8.3 through 8.5. Cheese was cut into pieces and 25-g portions were contaminated with low levels of SEC₃. Forty milliliters of water were added to each sample and masticated for two minutes, then acidified to pH 4 and centrifuged. The supernatant was neutralized and either tested directly or using the ISO 19020 protocol. Contaminations were done at concentrations that yielded fractional recovery (POD between 0 and 1). The formula was also tested at an independent laboratory.

Results: The recovery for infant formula using direct sampling was 11/20 at 0.075 ng/ml and 10/10 at 0.10 ng/ml. Recovery in the independent laboratory was 14/20 at 0.075 ng/ml and 10/10 at 0.10 ng/ml. The ISO 19020 protocol yielded recoveries of 3/20 at 0.015 ng/ml, 20/20 at 0.03 ng/ml, and 10/10 at 0.04 ng/ml. Independent laboratory recovery was 2/20 at 0.01 ng/ml, 9/20 at 0.02 ng/ml, and 10/10 at 0.03 ng/ml. For the cheese, recovery using direct sampling was 6/20 at 0.10 ng/g, 14/20 at 0.15 ng/g, and 10/10 at 0.20 ng/g. The ISO 19020 protocol yielded 7/20 positives at 0.03 ng/g, 14/20 at 0.04 ng/g, and 10/10 at 0.05 ng/g.

Significance: The ISO 19020 extraction yielded four to five times better sensitivity than testing the samples directly. Even without the laborious dialysis/concentration protocol, the sensitivity for each food was quite good.

P2-22 Comparative Validation Study to Demonstrate the Equivalence of an Alternate Next-day Enrichment Protocol for TRANSIA PLATE Salmonella Gold Method to Culture Methods for the Detection of Salmonella in Selected Foods and Environmental Surfaces

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Introduction: *Salmonella* is a significant pathogen and the causative factor of many foodborne illnesses and hospitalizations. BioControl Systems, Inc. has developed a protocol that allows next-day detection of *Salmonella* in foods and environmental surfaces using the TRANSIA PLATE *Salmonella* Gold (TPSG) enzyme-linked immunosorbent assay.

Purpose: To validate the equivalence of an alternate next-day enrichment method for TPSG to detect *Salmonella* in various foods and environmental surfaces versus a reference method using a proprietary enrichment media (mEHEC).

Methods: Various 25-g food samples were seeded with low levels (1 to 5 CFU/sample) of *Salmonella* and stabilized. Various surfaces were inoculated with *Salmonella* and allowed to dry overnight. The samples were enriched using two methods. Foods with low microbial background (roast beef and deli turkey) and environmental surfaces (steel, concrete, plastic) were incubated in mEHEC broth for 20 to 24 hours at 42°C. Foods with high microbial background (raw spinach, almonds, raw pasta, chicken rinsate) were incubated in mEHEC containing novobiocin (mEHEC+n) for 20 hours, then transferred to 10 ml of tryptic soy broth containing novobiocin for an additional six hours. Both 25 g and composite (325 to 375 g) samples were run. The samples were tested using TPSG. In all cases, the reference protocol (United States Department of Agriculture Microbiology Laboratory Guidebook or U.S. Food and Drug Administration Bacteriological Analytical Manual) was run for comparison. Inclusivity/exclusivity studies were also run.

Results: The comparative testing between the TPSG method and the reference method yielded statistically equivalent recovery data for each food and environmental surface tested. The TPSG method detected all 140 strains of *Salmonella*, representing all serogroups, and was negative for all 50 non-*Salmonella*.

Significance: Based on the results of the comparison studies and inclusivity/exclusivity studies, TRANSIA PLATE *Salmonella* Gold method is a viable next-day protocol for detection of *Salmonella* in foods. This data was also used for the certification of TPSG as an AOAC Performance Tested Method.

P2-23 Detection of Cronobacter spp. in Powdered Infant Formula and Cereals and Dried Milk Powder Using the Assurance Gds Cronobacter Tq Method

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Introduction: *Cronobacter* spp. are opportunistic pathogens for newborns and infants, and outbreaks and fatalities have been linked to contaminated powdered infant formula (PIF). Manufacturers are required to test PIF products and ingredients to prevent such illnesses.

Purpose: To develop a next-day method for enrichment and detection of *Cronobacter* spp. in PIF; infant cereals, with and without probiotics; and nonfat dry milk (NFDM) using the Assurance GDS *Cronobacter* Tq real-time PCR-based test.

Methods: *Cronobacter* spp. test strains were lyophilized into dry milk powder, stabilized, and diluted into test samples.

Powder samples of 25 or 375 g were incubated at 1:10 in buffered peptone water (BPW) at 37°C for 24 to 32 hours or 28 to 32 hours, respectively. For infant foods containing probiotic supplements, vancomycin was added (6 mg/ml) to BPW enrichments. For infant cereals, amylase was added (0.01%, 50 U/mg). NFDN was enriched in brilliant green water (0.002%). Seventy 25-g samples of infant formula or cereals plus probiotics, 18 25-g samples of NFDN, and 16 375-g samples of NFDN were tested with the Assurance GDS *Cronobacter* Tq method.

Results: The Assurance GDS *Cronobacter* method yielded statistically equivalent data compared to the paired microbial plate confirmation results.

Significance: The results demonstrated that the Assurance GDS *Cronobacter* Tq provided accurate detection of *Cronobacter* spp. in powdered infant formula, cereal containing probiotics, and dry milk powder.

P2-24 Identification of Plasmids and Virulence Markers of Shiga Toxin-producing *Escherichia coli* by MinION Nanopore Rapid Sequencing

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Introduction: Shiga toxin-producing *Escherichia coli* (STEC) of serotype O26:H11/- is the second most important hemolytic uremic syndrome-producing *E. coli* worldwide. Sequencing of STECs in outbreak situations is usually performed using Illumina MiSeq platform and Nextera XT for library preparation. However, due to the methodology, some portions of the genome are not included or are underrepresented in the final library. Because MiSeq is a shotgun sequencing technology resulting in draft assemblies encompassing >300 contigs for STECs, many regions of the genome are uncovered or fragmented.

Purpose: Here, we evaluated the use of MinION nanopore, an alternative sequencing method that produces complete closed genomes.

Methods: Using MinION sequencing technology, we sequenced the complete genomes of three STEC O26:H11 strains belonging to two different sequence types (ST21 and 29).

Results: We completely closed the genomes, consisting of chromosomes of 5.7 Mb and two plasmids of 95 and 72 Kb for ST21 and a single plasmid of 105 Kb for the ST29 strain. Using these data, we rapidly characterized the virulome as well as documented the presence of antimicrobial genes. We compared the results obtained by MinION against MiSeq-Nextera XT for accurate and inclusive determination of the virulome of these three strains and found that in every strain, the MiSeq-Nextera method failed to identify some virulence genes that were not missed by MinION sequencing. Some of those genes were in plasmids and some in the chromosome. Additionally, the results obtained with MinION were comparable to the information obtained using the PacBio data.

Significance: The correlation between the two long-read methods for determining plasmids, virulome, antimicrobial resistance genes, and phage composition in STEC O26 strongly indicates that the MinION sequencing technology is an excellent solution for rapidly determining STEC O26 closed genomes and comprehensive analysis of their genomics markers.

P2-25 A Novel Lysis Method for the Purification of Gram-positive and Gram-negative Bacterial Gdna from Food Samples

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Introduction: Food testing is performed by many organizations, including contract food testing labs, academic

researchers, and consumer protection agency labs. Molecular tests, such as qPCR and NGS, continue to gain widespread use in food safety testing. Using sub-optimal DNA purification methods for food samples can carry inhibitory compounds into the eluates, leading to inhibition. Therefore, the selection of a DNA purification method that provides amplifiable DNA of high quality is critical for accurate results.

Purpose: We report on a novel workflow that does not require enzymatic pre-treatment for the purification of both gram negative and gram positive bacterial DNA from raw and processed food samples.

Methods: Food samples were spiked with *Listeria monocytogenes*, *Salmonella enterica*, and pathogenic *Escherichia coli*, and enriched using a standard enrichment process. Bacterial DNA was then purified in the enriched samples using the Maxwell RSC PureFood Pathogen kit. For each extraction, bacterial DNA concentration was determined with fluorescence and qPCR using species-specific bacterial primers and probes.

Results: We detected low levels of bacterial contamination in food samples spiked with bacteria and enriched by culturing with the appropriate enrichment media. *E. coli* spiked into cilantro and strawberry samples was detected at a concentration as low as 4.1 CFU/g in the pre-enriched food culture. *S. enterica* spiked into roast beef cold cut meat and raw shrimp samples was detected at a concentration as low as 7.7 CFU/g in the pre-enriched food culture. *L. monocytogenes* spiked into soft cheese and canned chicken samples was detected at a concentration as low as 1.7 CFU/g in the pre-enriched food culture.

Significance: The Maxwell RSC PureFood Pathogen Kit is an automated protocol with few steps and has been validated for the purification of both gram negative and gram positive bacterial DNA, including *E. coli* O157:H7, *Salmonella enterica*, and *Listeria monocytogenes*, without the need for enzymatic pre-treatment.

P2-26 Occurrence of Deoxynivalenol in Cereals and Cereal Products in Hungary

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Introduction: Due to changing climate and weather conditions, Hungary has to contend with seriously diminished or lost crops, increasing contamination of cereals with moulds and resulting mycotoxin contamination. One of the most important mycotoxins found in crops is deoxynivalenol (DON).

Purpose: The aim of our study was to monitor the DON contamination of cereal and cereal product samples in Hungary and combine these results with official recordings of the Hungarian Meteorological Country Service in order to analyse the effect of weather data on DON contamination.

Methods: In this study, Hungarian wheat ($n=305$), maize ($n=108$), wheat flour ($n=179$), and pasta ($n=226$) samples (collected between 2008 and 2015) were analysed ($n=818$). Enzyme-linked immunosorbent assay and liquid-chromatography coupled with mass spectrometry were applied to determine DON toxin contamination.

Results: Among cereal samples, in 2011, wheat was contaminated with DON (overall average \pm standard deviation; 2,159 \pm 2,818 $\mu\text{g}/\text{kg}$ -1) that was above the maximum limit (ML). In wheat flour and pasta, no average values above the ML were found between 2008 and 2015, but higher DON contamination could be observed in 2011 as well (wheat flour: 537 \pm 573 $\mu\text{g}/\text{kg}$ -1; pasta: 511 \pm 175 $\mu\text{g}/\text{kg}$ -1).

Significance: Based on our survey, temperature and rainfall quantity/distribution are not the only weather conditions that affect the level of DON toxin contamination of cereals and cereal products. New risk factors, extremely dry weather and

very low annual average rainfall quantity, were determined for wheat samples and very high yearly average temperature for maize samples. This highlights that the consideration of other extreme weather conditions is also necessary when planning and monitoring, not just the previously determined weather conditions–matrix–DON toxin relationship.

P2-27 The Quality of Raw Milk (and Cheese Manufactured from It) in Ireland

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Introduction: Ireland has an international reputation for the quality and variety of its artisan cheese made from unpasteurised milk. It is important for the entire dairy industry that this reputation is not damaged.

Purpose: This work aimed to assess microbiological and residue (anthelmintic drug residues) risks associated with unpasteurised milk and the cheese made from it.

Methods: Raw milk, milk filters, curd, and cheese from ten raw milk artisan cheese producers in the south of Ireland were tested. Total Bacterial Count (TBC), presumptive *Bacillus cereus* group, *Escherichia coli*, *Salmonella* sp., and *Listeria monocytogenes* were determined. The determination of anthelmintic drug residues, including benzimidazoles, flukicides, macrocyclic lactone (ivermectin and milbemycins), levamisole, and morantel, was also performed.

Results: The TBC for milk samples ranged from 10^3 to 10^4 CFU/ml and milk filter samples were higher by about one log cycle. The *E. coli* results were similar between dairies with values of <10 CFU/ml for milk samples, between 10^1 and 10^2 CFU/g for curd samples, and 10^2 and 10^5 CFU/filter for milk filter samples. Presumptive *B. cereus* group were absent in most cases, although numbers were around 50 CFU/ml when they were present. Neither *L. monocytogenes*, nor *Salmonella* sp. were detected for any of the samples tested. One of the dairies stood out as having consistently higher numbers on all three types of samples for most of microbiological tests performed. None of the anthelmintic residue tests were found to be above the reporting limit of the method.

Significance: This survey has shown a good microbiological and residue quality of the raw milk used for raw milk cheese produced in Ireland. Moreover it has shown the importance of this kind of frequent assessment of raw milk cheese dairies, as it allows the identification of potentially problematic dairies, allowing them to address problems before causing any public health issues.

P2-28 The (FAO/WHO) International Food Safety Authorities Network: Infosan in Action to Control an Outbreak of Salmonellosis Linked to Infant Formula, 2017/2018

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Introduction: The International Food Safety Authorities Network (INFOSAN) is a global network of national food safety authorities, managed jointly by the Food and Agriculture Organization of the United Nations and the World Health Organization. In December 2017, a salmonellosis outbreak among infants in France triggered a series of product recalls with implications for dozens of countries around the world. INFOSAN was utilized during this event to facilitate rapid communication between affected countries.

Purpose: The purpose of this study was to examine how INFOSAN was utilized to allow members around the world to implement various risk management measures, including product recalls and public messaging.

Methods: All correspondence that took place between INFOSAN Emergency Contact Points (ECPs) in affected countries and the INFOSAN Secretariat was examined to determine the scope of impact and the actions taken

in various countries in response to receiving information through the network. This included a review of emails sent from and received by the INFOSAN Secretariat, as well as information shared on the INFOSAN Community Website.

Results: As of January 16, 2018, 43 INFOSAN ECPs were notified by the INFOSAN Secretariat (this excludes several European countries, informed exclusively by the European Commission Rapid Alert System for Food and Feed). Of the ECPs, 36 of 43 (84%) acknowledged receipt of the message and/or provided additional details of their investigations to the Secretariat. Ten ECPs shared information on the INFOSAN Community Website. Examples of actions taken included national multi-sector meetings, product recalls, and public alerts, including risk communication messages.

Significance: The rapid sharing of information through INFOSAN allowed members to take swift action to remove the offending products from retail markets, preventing illness. In addition, sharing information between INFOSAN members allowed for non-formal distribution channels to be investigated. Numerous members have shared the details of their subsequent investigations on the Community Website, helping to capture the truly global scope and impact of the event.

P2-29 Characterization of Binding Behaviors of Cd²⁺ to Rice Proteins

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Introduction: Heavy metal pollution is becoming a serious problem in China, threatening huge areas of cultivated lands, and therefore crop outcomes. The cadmium (Cd) pollution has long been a primary threat to the safety of common foods, especially rice.

Purpose: In this paper, for the first time, the binding behavior of Cd²⁺ to rice proteins (RPs) was studied.

Methods: By suspending Cd²⁺ into RPs containing aqueous solutions and characterizing the interplay between Cd²⁺ and RPs in the presence of binding inhibitors like coordination agents and metal ions, the binding mechanism was investigated.

Results: The results showed that the equilibrium of binding was reached within 30 min at 303 K with a maximum change in binding amount of 15.26 mg/g, and the pH was an important factor, positively influencing the change of binding amounts. At both 308 and 313 K, the binding of Cd²⁺ to RPs was spontaneous ($\Delta G^\circ < 0$ kJ/mol), endothermic, and with high-affinity interactions ($\Delta H^\circ > 80$ kJ/mol), which might be recognized as multidentate coordination. Except for acetate, all the investigated competing coordination agents, i.e., edetate, pyrophosphate, and citrate, showed inhibitory effects on RPs-Cd²⁺ binding, and edetate seemed to be the most effective one. At pH 6.5, Ca²⁺, Cu²⁺, and Zn²⁺ began to restrict RPs-Cd²⁺ binding when the metal ion concentration reached 500 mg/kg, and the decreasing of pH would strengthen the inhibitory effects of the investigated metal ions including Fe³⁺.

Significance: The study therefore provides us with new insights into removal of Cd²⁺ from RPs.

P2-30 Lead, Cadmium, and Arsenic in Human Milk and Their Socio-Demographic and Lifestyle Determinants in Lebanon

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Introduction: Exposure of newborns to toxic metals is of special interest due to their reported contamination in breast milk and potential harm.

Purpose: The aim of this study was to assess the occurrence and factors associated with lead, cadmium, and arsenic contamination in breast milk collected from lactating mothers in Lebanon.

Methods: A total of 74 breast milk samples were collected from primiparas according to guidelines set by the World Health Organization. A survey was administered to determine the demographic and anthropometric characteristics of participating lactating mothers. Dietary habits were assessed using a semi-quantitative food frequency questionnaire. The milk samples were analyzed for the presence of arsenic, cadmium, and lead using microwave-assisted digestion and atomic absorption spectrophotometry.

Results: Arsenic contamination was found in 63.51% of breast milk samples (mean $2.36 \pm 1.95 \mu\text{g/L}$), whereas cadmium and lead were detected in 40.54 and 67.61% of samples, respectively (means $0.87 \pm 1.18 \mu\text{g/L}$ and $18.18 \pm 13.31 \mu\text{g/L}$). Regression analysis indicated that arsenic contamination was associated with cereal and fish intake ($P=0.013$ and $P=0.042$, respectively). Residence near cultivation activities ($P=0.008$), smoking status before pregnancy ($P=0.046$), potato consumption ($P=0.046$), and education level ($P=0.041$) were associated with lead contamination. Cadmium contamination was significantly associated with random smoke exposure ($\beta:242$; $P=0.002$).

Significance: Our study is the first in Lebanon to report toxic metal contamination in breast milk. Although estimated weekly infant intake of these metals from breast milk was found to be lower than the limit set by international guidelines, our results highlight the need for developing strategies to protect infants from exposure to these hazardous substances.

P2-31 Determination of Acrylamide in Fried Potato Chips and Impact of Various Treatments on Acrylamide Formation during Frying

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Introduction: Acrylamide is a well-known industrial chemical, primarily used in the gel electrophoresis technique and in waste water treatment plants in polymer form. Acrylamide generates in heat-treated food products, mainly in carbohydrate-rich foods such as potato and cereal products via Maillard reaction.

Purpose: The current study was conducted to determine the acrylamide concentration in various potato chip samples from fast food chains and some commercially available brands. The objectives of this study were to determine the acrylamide content in fried potato chips available in the local market of Lahore, Pakistan and to minimize the concentration of acrylamide formation in fried potato chips by applying various treatments.

Methods: Twenty samples were analyzed from fast food chains, commercially available brands, local vendors, and homemade potato chips. Three treatments were applied to minimize the acrylamide concentration in potato chips. Twelve samples were treated in the laboratory, in oil (170°C) for two minutes. Extraction and high-performance liquid chromatography analysis were performed to determine acrylamide concentrations.

Results: Samples collected from local vendors contained the highest level of acrylamide (2,429 ppb), followed by homemade potato chips (1,460 ppb). Fast food chain B contained high levels of acrylamide (559 ppb) as compared to fast food chain A (255 ppb). Commercially available brands contained the lowest concentration of acrylamide (60 ppb). Three treatments were applied to reduce or minimize acrylamide in fried potato chips. Par fried potato chips contained 73 ppb; prepared frozen potato chips contained 144 ppb; and par frying prepared frozen potato chips resulted in the lowest acrylamide level, 44 ppb.

Significance: The present study illustrated that carbohydrates and amino acids are the main source of acrylamide in fried, heated, or processed food stuffs (starchy foods).

Acrylamide content can be reduced by decreasing precursor quantities (reducing sugar and amino acids) that are thought to be responsible for the Maillard reaction.

P2-32 Performance Assessment of the 3M™ Petrifilm™ Lactic Acid Bacteria Count Plate According to ISO 16140-2 (2016) Standard in Food Products and Environmental Samples

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Introduction: Lactic acid bacteria (LAB) are non-spore forming gram positive cocci or rods which produce lactic acid during carbohydrate fermentation. The 3M Petrifilm Lactic Acid Bacteria Count Plate is a self-contained, sample-ready culture medium system designed to enumerate LAB. Nutrients, selective agents, and oxygen scavenger compounds create an ideal environment for LAB recovery from food and beverage.

Purpose: An independent study was conducted to compare this new alternative method to ISO 15214 (1998) according to the ISO 16140-2 (2016) standard for NF Validation approval.

Methods: Different matrices were tested: meat, dairy and seafood products, composite foods, meal components, and environmental samples. The alternative plates were inoculated with 1 ml of successive dilutions in peptone salt and incubated 45 h at $30 \pm 1^\circ\text{C}$. Red colonies with gas (heterofermentative) or without gas (homofermentative) were enumerated. The possibility to store the plates for 1 week at -18°C after incubation was evaluated. The study investigated the relative trueness, accuracy profile, inclusivity, and exclusivity.

Results: Overall, 48 naturally contaminated samples and 49 artificially contaminated samples were analyzed by both methods. Depending on the tested food categories, the mean difference between the alternative method and the reference method counts ranged between -0.05 and $0.18 \log \text{CFU/g}$. After the storage at -18°C , these values ranged between -0.05 and $0.16 \log \text{CFU/g}$. For accuracy profile study, the lower and upper β -ETI were comprised within the acceptability limits. Among the 57 tested target-strains, 52 gave similar results with both methods, two were enumerated only with the alternative method, and three did not grow. No cross-reaction was observed with the 34 tested non-target strains.

Significance: The alternative method is reliable for the enumeration of LAB and offers more practicability to the user than the reference method.

P2-33 Performance Assessment of the 3M™ Molecular Detection Assay 2 – Cronobacter According to ISO 16140-2 (2016) Standard in Infant Formula, Infant Cereals, Raw Materials, and Environmental Samples

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Introduction: *Cronobacter* species form a group of gram negative bacteria which may cause lethal illness in infants. The 3M Molecular Detection Assay 2 - *Cronobacter* is designed to detect *Cronobacter* spp. in food by means of loop-mediated isothermal amplification of specific DNA target sequences and detection by bioluminescence.

Purpose: An independent study was conducted to compare this new alternative method to the ISO 22964 (2017) according to the ISO 16140-2 (2016) standard for NF Validation approval.

Methods: Different enrichment protocols were tested: food and environmental samples were 1:10 diluted and incubated at $37 \pm 1^\circ\text{C}$ for 18 to 24 h in Buffered Peptone Water (BPW, for 10-g samples) or pre-warmed BPW (for 300-g samples). 300-g samples containing probiotics were supplemented

with vancomycin (10 mg/L) and incubated for 22 to 24 h. After lysis (15±1 min at 100±1°C), DNA amplification was performed. The study compared the sensitivity, relative detection level (RLOD), inclusivity, and exclusivity.

Results: Overall, 314 samples were analyzed by both methods. Depending on the enrichment protocol, the sensitivity ranged between 80.0 and 96.0% for the alternative method and between 66.7 and 100% for the reference method. The relative trueness ranged between 77.4 and 97.9% and the false positive ratio for the alternative method ranged between 1.1 and 3.3%. Depending on the tested matrix, the RLOD ranged between 0.255 and 2.317, suggesting that both methods have a similar level of detection. The 50 tested target-strains were detected and no cross-reaction was observed with the 30 tested non-target strains.

Significance: The alternative method is reliable for the detection of *Cronobacter* spp. and the negative results are available two days earlier than the reference method.

Rapid methods are needed to assess the effectiveness of antimicrobials used in poultry chillers.

Purpose: Evaluate the new alternative TEMPO CAM automated method for the enumeration of *Campylobacter* to the United States Department of Agriculture Food Safety Inspection Service (USDA-FSIS) procedure using Campy-Cefex agar.

Methods: A total of more than 180 samples of chicken coming from different slaughter plants were analyzed by both methods. Cards and plate counts were obtained after 48 h incubation in microaerobic condition. Confirmation of the presumptive colonies on Campy-Cefex was performed by latex agglutination. There is no confirmation step required for the alternative method.

Results: The new method is a reliable alternative to the USDA-FSIS method with a slope close to 1 and a correlation coefficient of 0.96.

Significance: The alternative method offers equivalent performance to the USDA-FSIS method without the need to perform labor-intensive preparation and confirmation steps.

P2-34 Genetic Diversity of Staphylococcal Strains Isolated from Food and Enterotoxin Coding Genes

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Introduction: Identification and characterization of pathogens responsible for outbreaks of foodborne illness are necessary in public health and quality control in food industries. Some pathogenic strains belonging to the bacterial species *Staphylococcus aureus* can produce toxins in food, which could lead to staphylococcal food poisoning outbreaks (SFPO).

Purpose: Objectives of the study were to use whole genome sequencing to determine the genetic diversity of strains responsible for SFPO between 2005 and 2017 in Europe and the most frequent enterotoxin genes in these strains.

Methods: A collection of 143 genomes was sequenced using illumina Technology. This collection was comprised of strains responsible for SFPO and of reference strains isolated from food, environment, or humans. In order to study genetic diversity of *S. aureus* strains isolated from food within the known genetic diversity of *S. aureus*, 105 genomes available from public databases were included. Assembly and annotation were performed using an in-house workflow based on Spades and Prokka. The core genome was defined using Roary, and the phylogeny was performed using RAxML. Then, toxin profiles were established on the 23 enterotoxin genes available in the literature by using an in-house workflow based on the BLAST approach. Finally the genetic diversity of enterotoxin coding genes was studied using clustering approaches.

Results: Our results highlighted several divergent clones within *S. aureus* that were responsible for SFPO between 2005 and 2017. Furthermore, several enterotoxin coding genes were very frequent in these strains, including *seg*, *seh*, *sem*, *sen*, and *seo*. Finally, clustering of different alleles of enterotoxin coding genes showed a genetic proximity between *sea*, *sep*, and *see* genes.

Significance: These results are relevant for food safety as they allowed us to: (i) highlight the presence of enterotoxin coding genes not currently detected by PCR tools and (ii) determine new targets for the development of rapid detection methods.

P2-35 Assessment of an Alternative Method for *Campylobacter* Enumeration in Chicken Carcass Rinses

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Introduction: *Campylobacter* represents a leading cause of gastrointestinal infection worldwide. Chicken and poultry products are a major source of campylobacteriosis.

P2-36 Creating Synthetic Phages for Pathogen Detection

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Introduction: Bacteria play a major role in regulating human health and disease, but existing protocols for pathogen detection are limited. Bacteriophages can be used to infect specific bacteria, which make them good candidates for detecting and editing bacterial populations. However, creating phage-based detection assays is somewhat limited by the difficulties in engineering of phages. Traditionally, phages have been engineered with allele replacement methods, but this process is inefficient and time-consuming. In this work, we have used a synthetic biology strategy to engineer T7 bacteriophage in *Saccharomyces cerevisiae*.

Purpose: The aim of this study was to establish a yeast-based and an *in vitro* phage-engineering platform for T7 bacteriophage. Our platform will allow us, for example, to engineer T7 with a reporter enzyme or/and with extended host range for bacteria detection.

Methods: The entire viral genome was first amplified by PCR so that adjacent fragments had homology over 30 bp. The first and last fragments of the phage genome were amplified with primers containing homology towards yeast artificial chromosome (YAC) fragment. All viral genome fragments and the YAC were transformed into yeast, where gap repair joined fragments together in a template by the homology regions, yielding a replicative yeast plasmid. After extraction, YAC-phage DNA was transformed into competent *Escherichia coli* cells to restart the viral life cycle. An alternative *in vitro* method was developed using Gibson assembly of the PCR fragments.

Results: Purified YAC-T7 DNA could be transformed into a bacterial host and generate functional phages. The engineered T7 detected a single *E. coli* host from 100 ml of water within 8 hours of infection.

Significance: Bacteriophage have the potential to serve as rapid diagnostic tools for food and water safety. We have demonstrated two methods which efficiently allow the insertion of reporter probes into a phage genome. By improving genetic engineering methods we are better enabling their development into commercial kits.

P2-37* The Reporter Bacteriophage T7 Utilizes a Novel Fusion Reporter to Rapidly Detect *Escherichia coli* in Large Water Samples

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Introduction: Rapid detection of bacteria responsible for foodborne diseases is a growing necessity for public health. Reporter bacteriophages (phages) are robust biorecognition elements uniquely suited for the rapid and sensitive detection of bacterial species.

Purpose: The advantages of phages include their host specificity, ability to distinguish viable and non-viable cells, low cost, and ease of genetic engineering. Upon infection with reporter phages, target bacteria express reporter enzymes encoded within the phage genome.

Methods: In this study, the T7 coliphage was genetically engineered to express the newly developed luciferase, NanoLuc (NLuc), as an indicator of bacterial contamination. While several genetic approaches were employed to optimize reporter enzyme expression, the novel achievement of this work was the successful fusion of the NanoLuc reporter to an affinity binding motif.

Results: This novel chimeric reporter bestows the specific and irreversible immobilization of NanoLuc onto a low-cost, widely available substrate. We have shown the possibility of detecting the immobilized fusion protein in a filter plate, which resulted from a single CFU of *Escherichia coli*. We then demonstrated that a similar substrate can be used to concentrate the fusion reporter from 100-ml water samples, allowing a limit of detection of <10 CFU/mL *E. coli* in 3 hours.

Significance: Therefore, we conclude that our phage-based detection assay displays significant aptitude as a proof-of-concept drinking water diagnostic assay for the low-cost, rapid, and sensitive detection of *E. coli*. Additional improvements in the capture efficiency of the phage-based fusion reporter should allow a limit of detection of 1 CFU/100 ml.

P2-38 Efficacy of Recovery and Detection of Sublethally Stressed *Listeria monocytogenes* Using Different Enrichment Media: From In Vitro to In Situ

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Introduction: As required by the majority of regulatory agencies, the current methods used for the detection of *Listeria monocytogenes* in ready-to-eat foods must be able to detect as low as one target bacterium per analytical portion. In spite of accelerating advances in the development and implementation of new rapid and highly sensitive methods for the detection of *Listeria* in foods, no existing technologies can approach the declared limit of detection without using efficacious enrichment broth.

Purpose: The study determined the effectiveness of selective enrichment broth for the recovery and detection of sublethally stressed *L. monocytogenes* in ready-to-eat foods.

Methods: Different microbial stress models were applied to evaluate the repair capacity and growth characteristics of sublethally injured *L. monocytogenes* using Actero *Listeria* Enrichment Media in comparison to other primary and single-step enrichment media. The detection of *L. monocytogenes* in deli meat and dairy products enriched with Actero *Listeria* was evaluated using Hygiena's BAX System Real-time PCR Assays for *L. monocytogenes* and genus *Listeria*, as well as by plating.

Results: *In vitro* culture studies, as well as matrix trials, demonstrated a strong potential for Actero *Listeria* to resuscitate sublethally stressed *L. monocytogenes* due to different stresses related to the food production chain. In co-culture studies, Actero *Listeria* showed an excellent capacity in controlling the growth of competing bacteria without affecting *L. monocytogenes*. Overall, 140 fractionally contaminated food samples tested by the alternative method using Actero *Listeria* yielded 40% more positive results as compared to the samples analysed by the United States Department of Agriculture Food Safety Inspection Service or U.S. Food and Drug Administration methods.

Significance: Using a real-time PCR assay for *L. monocytogenes* detection, Actero *Listeria* reduced the time-to-results to as short as 24 hours, enabling a rapid single-step enrichment to successfully reach the limit of detection of as low as one cell of *L. monocytogenes* in 25 g and 125 g of ready-to-eat food.

P2-39 Recovery and Rapid Detection of *Listeria monocytogenes* in Raw Dairy Products Using Actero™ *Listeria* Enrichment Media

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Introduction: In 2015, based on multiple outbreak data, dairy products were identified as one of the main sources of listeriosis. Despite this, the consumption of raw dairy products all over the world is greatly increasing. ISO 11290-1, a method currently adopted in Europe for the detection of *Listeria monocytogenes* in dairy products, is labour-intensive and time-consuming and often delays product release for sale. For this reason, the development of an alternative sensitive and rapid method for the detection of *L. monocytogenes* would be of great value for the dairy industry.

Purpose: The study evaluated the efficacy of application of Actero *Listeria* Enrichment Media for the recovery and rapid detection of *L. monocytogenes* in raw dairy products.

Methods: A total of 155 raw milk and semi-hard unpasteurized milk cheese samples (25 g each) were artificially contaminated with *L. monocytogenes* (0.2 to 16.4 MPN/sample) and stabilized at 4 to 8°C for 48 to 72 hours. The samples were homogenized with 100 to 150 ml of Actero *Listeria* and enriched for 22 to 24 hours at 35°C. *L. monocytogenes* was detected using Hygiena's BAX System Real-time PCR Assays for *L. monocytogenes* and genus *Listeria*, as well as by plating onto selective agars. The primary enrichments were incubated for an additional 24 hours at 35°C, followed by plating in order to confirm presumptive results.

Results: The one-step enrichment with Actero *Listeria* allowed for recovering *L. monocytogenes* in 94 of 155 (60.6%) raw dairy product samples containing high numbers of competing flora (~5 to 7 log₁₀ CFU/g). No false positive outcomes and only one false negative outcome (detected by the genus *Listeria* assay) were obtained.

Significance: The one-step enrichment with Actero *Listeria* enables a significant reduction in presumptive reporting time to as short as 22 hours without the loss of sensitivity and reliability of the methods used for the detection of *L. monocytogenes* in raw dairy products.

P2-40 No Growth of *Salmonella enterica* Could be Observed in Salad Juice at Refrigeration Temperatures

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Introduction: *Salmonella* have been widely recognized as mesophiles, and no growth of *Salmonella* has been observed previously on leafy greens at usual refrigeration temperatures (<8°C). However, contradictory results have been reported recently, which raises concerns as it detracts from the idea that the cold chain is the primary control in keeping food safe and preventing growth of *Salmonella*.

Purpose: To investigate if indeed *S. enterica* could grow in salad juice at refrigeration temperatures.

Methods: Two *S. enterica* strains (*S. Typhimurium* SL1344 and *S. Thompson* RM1987) and one *Listeria monocytogenes* strain (12MOB098LM) were tested, the latter serving as a positive control because it is cold-tolerant. Spinach and mixed leaf salad juice were prepared by homogenizing leaves with water (weight:volume=1:1), followed by centrifugation and filter sterilization. Selective media were used: Xylose-Lysine-Desoxycholate Agar (Oxoid) and Chromogenic Media (ALOA, bioMérieux) to monitor growth

of *S. enterica* and *L. monocytogenes*. Three temperatures (4, 7, and 12°C) were tested for up to 15 days.

Results: No growth of the two *Salmonella* strains was observed neither in nutrient broth, nor in the spinach or mixed leaf salad juice at 4 and 7°C within the 15 days of incubation (simulating the maximum shelf life of bagged salad leaves in Europe). At 12°C, both of the tested *Salmonella* strains grew from ~1 log CFU/ml to levels over 5 log CFU/ml within 5 days. In comparison, growth of *L. monocytogenes* was noted at all temperatures included, with faster growth in nutrient broth versus leafy green juices and faster growth with increasing temperatures.

Significance: We showed that keeping the cold chain (< 8°C) remains an effective control measure to inhibit the growth of *Salmonella*, including in fluids released by salad leaf damage.

P2-41 Survival of Murine Norovirus and Tulane Virus on Pre-Harvest Basil (*Ocimum basilicum*) Leaves

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Introduction: Fresh produce has been identified as an important vehicle for noroviruses (NoVs), and transmission and fresh culinary herbs have occasionally been associated with human pathogens and illness.

Purpose: To investigate the survival of murine norovirus 1 (MNV-1) and Tulane virus (TV), two common human NoV surrogates, on pre-harvest basil (*Ocimum basilicum*) leaves.

Methods: Three independent batches of basil plants were cultured under the same conditions. MNV-1 was tested in the first and second batches, and TV was tested in the second and the third batches. MNV-1 or TV was inoculated onto the adaxial leaf surfaces of the four-week-old plants in the grow box. For each leaf, 50 µl of virus suspension was distributed evenly in a 1-cm² area. A fake basil plant made with fabric was inoculated in the same way as a control. Three hours or three days after inoculation, the inoculated squares were collected for virus extraction. The viability of MNV-1 and TV were measured by plaque assay.

Results: On pre-harvest basil leaves, reductions above 1 log of virus viability were observed at 3 h after inoculation for both MNV-1 and TV. Three days after inoculation, the infectivity of MNV-1 decreased 4.4 and >5.5 log PFU/g and the infectivity of TV decreased >3.3 and >2.5 log PFU/g to non-detectable levels. Higher viral reductions were observed on pre-harvest basil leaves than on fabric leaves after three days in all the tested groups. No effect of the microbial background on the basil leaves was observed on virus survival.

Significance: An understanding of the behavior of enteric viruses on basil leaves can be used for relevant risk assessments. Virus inactivation was more rapid on basil leaves than on inert surfaces, suggesting a potential role of basil cells on viral inactivation.

P2-42 Influence of Seed Type and Seed Batch on the Long-term Survival of *Salmonella* and *Escherichia coli* O157 on Seeds Intended for Sprouting

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Introduction: Sprouts are consumed raw or minimally processed. Outbreaks occur and the bacterial pathogens most frequently associated are *Salmonella* and Shiga toxin-producing *Escherichia coli* (STEC). The seeds have been identified as the most likely source of contamination. They may be stored for several years before use. Although different studies have shown that *E. coli* O157 and *Salmonella* can survive for a prolonged period on seeds, little is known about the influence of seed type and variability of survival between batches of the same seed type.

Purpose: The aim of the study is to examine the long-term survival of two *E. coli* O157 and two *Salmonella* serovars artificially inoculated on three batches each of alfalfa and leek seeds.

Methods: Inoculated seeds were mixed 1:10 with uninoculated seeds to mimic spot contamination and stored at 15°C and 50% RH. The survival of the pathogens was monitored for more than two years, initially by direct counts on selective media; if numbers dropped below 2 log CFU/g, a mini MPN-technique was used.

Results: For *E. coli* O157 NCTC12900, only low numbers were recovered after inoculation and its survival was not further investigated. For the other *E. coli* O157 (BRMSID 188) and the two *Salmonella* strains (serovars Typhimurium and Thompson), initial levels ranged from 2.7 to 6.2 log CFU/g of seeds. Although gradually decreasing numbers were recovered, these strains were able to survive for more than two years. A faster decline was observed on alfalfa in comparison to leek seeds ($P=0.035$), but the decline in numbers was not significantly different between the three strains, nor between the different seed batches.

Significance: Our study confirms the long-term survival of enteric pathogens on seeds, but also shows that the survival depends upon the seed type.

P2-43* Germination of *Clostridium botulinum* Spores and the Implication for Food Safety

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Introduction: Spore germination in *Clostridium botulinum* is the key step in the transformation of dormant, highly heat-resistant spores into exponentially dividing vegetative cells that are capable of forming the deadly botulinum neurotoxin in food products.

Purpose: Our aim is to characterise the spore germination pathway in strains of *C. botulinum* Groups I and II by testing the effect of inputs of nutrient germinants, DDA (dodecylamine), CaDPA (calcium dipicolinic acid), and lysozyme.

Methods: The ability of these nutrient and non-nutrient germinants to induce germination of strains Af84 (*C. botulinum* Group I) and Eklund 17B (*C. botulinum* Group II) was established by measuring the change in optical density and by microscopic observation.

Results: Germination of strains Af84 and Eklund 17B was initiated by L-alanine and L-cysteine (nutrient germinants) and by DDA and lysozyme, but not by CaDPA (non-nutrient germinants). Spores of Af84 were more heat-resistant than Eklund 17B, and heating spores of Af84 for 4 hours at 95°C delivered a 3-log reduction in viability. Heat damaged spores of Af84 could not be recovered using lysozyme, L-alanine, or DDA. Thermal death of spores of Eklund 17B occurred within 2 minutes of heating at 85°C, and resulted in a 5-log reduction in viability. The presence of lysozyme (10 µg/ml) increased the recovery of heat damaged spores of Eklund 17B; however, the presence of L-alanine and DDA had no effect. The results indicate that part of the germination apparatus in Eklund 17B is damaged by heating, rather

than the spore DNA, as the spores could be recovered with lysozyme; whereas in Af84, DNA damage may occur, as the spores were not recoverable with lysozyme.

Significance: Understanding the mechanisms involved in spore germination can improve the control of botulinum neurotoxin-forming *Clostridia*, prevent foodborne botulism, and thereby contribute to the microbiological safety of foods.

P2-44 Effect of Lactic Acid on Pig-associated *Salmonella* and Human Pathogenic *Yersinia enterocolitica*

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Introduction: Currently, 60 and 43% of freshly slaughtered pig carcasses in Belgium are contaminated with *Salmonella* and *Yersinia enterocolitica* bioserotype 4/O:3, respectively. Lactic acid has been designated by the U.S. Food and Drug Administration as 'generally recognized as safe' for meat products, but is not allowed in Europe for pig carcass decontamination.

Purpose: This study evaluated the effect of lactic acid on different serotypes and strains of *Salmonella* and *Y. enterocolitica* isolated from pigs during slaughter.

Methods: Nineteen strains (nine *Salmonella* serotypes, two *Y. enterocolitica* bioserotypes) were grown at 25°C (environment) and 37°C (pig), exposed to 5% lactic acid (pH 4), and stored for 48 h at 2°C. Differences between treated and control cells were measured (0, 24, and 48 h) by plating on xylose lysine desoxycholate agar (*Salmonella*), cefsulodin irgasan novobiocin agar (*Y. enterocolitica*), and plate count agar to measure lethal and sub-lethal injury. Loss of virulence plasmid (pYV) in *Y. enterocolitica* cells was investigated by plating on Congo red magnesium oxalate agar.

Results: No lactic acid sensitivity differences were found between *Salmonella* serotypes or *Y. enterocolitica* bioserotypes ($P>0.05$). The exposure of *Salmonella* serotypes to lactic acid resulted in an immediate mean reduction of 1.1 (37°C) and 2.3 (25°C) log₁₀ CFU/ml. *Y. enterocolitica* showed an immediate mean reduction of 1.5 log₁₀ CFU/ml at both growth temperatures. Reductions remained stable during cooling ($P>0.05$). Sub-lethality of *Salmonella* was greater at 25°C (up to 99.1% after 48 h) than at 37°C (up to 47.1% after 48 h). The same trend was seen for *Y. enterocolitica*. The percentage of pYV positive *Y. enterocolitica* cells was lower when treated with lactic acid at all time points.

Significance: This study indicates that lactic acid treatment might be of great potential for pig carcass decontamination since it reduces the number of pig-associated pathogens in culture media and the virulence of *Y. enterocolitica* strains.

P2-45* Effect of Water Activity on the Heat Resistance of *Salmonella* Thompson

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Introduction: Thermal treatment is the most commonly used process to eliminate foodborne pathogens; however,

the heat resistance of *Salmonella* is known to increase at reduced water activity. Apart from the inactivation studies published on low-moisture food, little is known about the heat-resistance of *Salmonella* in broth (a_w 0.85 to 0.99) or on semi-dried product.

Purpose: To provide information on the heat resistance of *Salmonella* at different water activities and understand the effect of habituation on the heat tolerance of *Salmonella* strain.

Methods: Heat resistance of *Salmonella* Thompson (RM 1987) in peptone salt solution (PPS) with water activity of 0.85, 0.90, 0.95, and 0.99 was determined at different temperatures (55 to 65°C). Strain was washed and resuspended in PPS for immediate heat treatment or habituated at different water activities at 4 or 22°C for 24 h prior to heat treatment. Control and treated samples were plated on XLD and TSA media to examine the recovery rates. The same culture was inoculated on fresh and semi-dried basil (0.95), which were treated at 60°C to investigate the inactivation of *Salmonella* on basil.

Results: The recovery inhibition coefficient values increased substantially as the treatment temperature increased, which indicates that XLD media showed higher recovery capability at higher temperatures. Habituation at 22°C for 24 h in PPS with reduced water activity resulted in increased heat tolerance of *Salmonella* Thompson. Under the same habituation condition, Z_{TSA} values of *Salmonella* Thompson in PPS at a_w 0.99, 0.95, and 0.90 were 7.2, 4.7, and 5.2°C, respectively, which were higher than Z_{XLD} values. Maximal heat resistance of *Salmonella* was observed in broth at a_w 0.95 and on semi-dried basil.

Significance: Highlight the importance of media selection and cell habituation in heat inactivation studies. Thermal treatments are more effective in products with water activity higher than 0.95 to eliminate foodborne pathogens.

P2-46 Combined Effect of NaCl and Low Temperature on Anti-listerial Bacteriocin Production of *Lactobacillus plantarum* ST202Ch

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Introduction: Studies can be found about the characterization of antilisterial bacteriocin produced by *Lactobacillus plantarum*. Traditionally, the activity of the bacteriocin (arbitrary unit/ml) has been determined as an adaptation of the critical dilution method. Arbitrary unit/ml (AU/ml) is defined as the reciprocal of the highest dilution showing a clear zone of growth inhibition. With that methodology, the value of AU/ml is a discrete variable, which may cause information loss.

Purpose: The aim of our study was to examine the combined effect of NaCl and low temperature on antilisterial bacteriocin production of *L. plantarum*.

Methods: A redox-potential measurement method has been adapted for measurement of bacteriocin activity in *L. plantarum* ST202Ch. A bacteriocin sensitive *Listeria monocytogenes* strain was selected for this study.

Results: Evaluation of bacteriocin activity was described by new parameters: the difference between the detection times of the inhibited and the control (non-inhibited) *L. monocytogenes* suspensions (ΔTTD), and the elapsed fermentation time until the supernatant results in 2.5-log virtual decrease in *L. monocytogenes* (te).

Significance: On the basis of data obtained by redox-potential measurement methodology, a multiple regression model was established to describe the combined effect of temperature and NaCl concentration on the bacteriocin production of *L. plantarum*.

P2-47 Effect of Microbial Competition on Gene Expression of *Listeria monocytogenes* in Dry-cured Ham

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Introduction: Biopreservation is based on the competition exerted by commensal microorganisms towards foodborne pathogens or spoilage microorganisms and may involve production of antimicrobial compounds.

Purpose: The purpose of this study was to investigate the effect of microbial competition on gene expression of *Listeria monocytogenes* in dry-cured ham. We tested the effect of a purified enterocin as well as a strain of *Enterococcus faecalis* producing a bacteriocin.

Methods: Samples of dry-cured ham were artificially inoculated with *L. monocytogenes* and either co-inoculated with *E. faecalis* or supplemented with purified enterocin. The samples were vacuum packed and stored at refrigeration temperature for up to 28 days. During the storage period, viable counts of *L. monocytogenes* were determined while samples for RNA extraction were collected. RNA extracted was used in reverse transcription qPCR, targeting five genes involved in stress response and virulence. Comparative gene expression, using a housekeeping gene for normalization purposes, was then calculated.

Results: Both the purified enterocin and the *E. faecalis* strain had an effect on the viable count of *L. monocytogenes*. Population reduction was more evident in the case of the purified enterocin and in certain time points was close to 2 log₁₀ CFU/g. In the case of the use of *E. faecalis*, a small reduction (<1 log₁₀) or simply confinement of the growth was observed. Gene expression was influenced by the presence of both the enterocin and of *E. faecalis*. Gene expression was greatly influenced by the food matrix. In addition, important differences in the gene expression were observed for the two *L. monocytogenes* strains used.

Significance: The use of lactic acid bacteria as competitive cultures to control foodborne pathogens and spoilage microorganisms is gaining ground. This study suggests that there is a need to proceed beyond the determination of viable counts and further elucidate the behavior of the target microorganism.

P2-48 UV-C Inactivation of *Bacillus cereus* Spores

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Introduction: *Bacillus cereus* is a toxin-producing bacterium found ubiquitously in the soil; thus, it is widespread on different food groups such as vegetables, rice, and herbs. The strength of *B. cereus* in its survival is the ability to produce spores. As highly resistant spores can survive pasteurization, *B. cereus* is an important food contaminant. An important technology to disinfect food contact surfaces is short-wavelength ultraviolet (UV-C) light. However, data is lacking in literature on UV-C inactivation of *B. cereus* spores.

Purpose: The purpose of this study was to determine the inactivation of three different strains of *B. cereus* spores under UV-C light.

Methods: Spores of three strains of *B. cereus* (ATCC 14579, LMG 26718, and LMG 18989) were produced in maltose sporulation medium. After harvesting, spores were stored at 4°C and subjected in liquid to UV-C treatment from one (3.2×10⁻² J/cm²) to eight minutes (3.5×10⁻¹ J/cm²). Surviving spores were serially diluted and plated on brain heart infusion agar. Inactivation curves were plotted in Excel using GlnaFIT. Experiments were performed in triplicates.

Results: Initial inoculum concentration of LMG 18989, ATCC 14579, and LMG 26718 was 8±0.2 log CFU/ml, 8.7±0.0 log CFU/ml, and 8.3±0.2 log CFU/ml, respectively. All inactivation curves were fitted log-linear with tail. Tails started at 4.0±0.1 log CFU/ml for ATCC 1479, 3.1±1.0 log CFU/ml for LMG 18989, and 2.6±0.6 log CFU/ml for LMG 26718. Average inactivation rates (K_{max}) for LMG 18989, ATCC 14579, and LMG 26718 were, respectively, 2.3 min⁻¹, 2.3 min⁻¹, and 5.4 min⁻¹. The thermotolerant strain, LMG 26718, showed the least resistance against UV-C with a 2.8±0.5 log CFU/ml reduction within the first minute (3.2×10⁻² J/cm²).

Significance: This data suggests that different strains show different resistance against UV-C inactivation and may provide necessary information on UV-C inactivation on *B. cereus* spores for further projects.

P2-49* Comparison of Enrichment and Isolation Methods of *Campylobacter jejuni* in Chicken Fecal Samples

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Introduction: *Campylobacter* spp. are common foodborne pathogens that are a global cause of human enteritis, with *C. jejuni* accounting for 80 to 90% of human infections. Poultry products are a main cause of campylobacteriosis. It is therefore important to detect *C. jejuni* on poultry farms to control and manage foodborne illness. However, there is no standard method to isolate *C. jejuni* in chicken fecal samples.

Purpose: The current study was performed to optimize a *C. jejuni* isolation method in chicken fecal samples. Specifically, we compared isolation methods of *C. jejuni* according to combinations of enrichment media, selective agars, and the ratio of sample to enrichment media.

Methods: Thirty-five fecal samples from the GI tracts of chickens at a slaughterhouse were collected and analyzed to compare isolation methods of *C. jejuni* in different combinations of procedures. They were then enriched in Bolton broth and Preston broth different ratios of sample to broth (1:10, 1:10², and 1:10³, respectively), followed by plating on modified charcoal cefoperazone deoxycholate agar (mCCDA) and Preston agar and direct plating on mCCDA and Preston agar.

Results: In samples enriched in Bolton broth, *C. jejuni* was not isolated regardless of the ratio of sample to broth and the types of selective agars. In Preston broth, the isolation rate of *C. jejuni* was highest (34 of 35, 97.1%) at the ratio of 1:10³. Furthermore, *C. jejuni* was isolated significantly higher in mCCDA than in Preston agar (*P*<0.001).

Significance: Since the initial contamination levels and characteristics of chicken fecal samples are different from those of chicken products, it is necessary to apply an efficient isolation method for *C. jejuni* in chicken fecal samples. This optimized procedure would be useful in further studies of *Campylobacter* isolation and prevalence at the farm level.

P2-50 Preliminary Study of Kinetics and Gene Expression for *Staphylococcus aureus* Strains Producing Enterotoxins SEA and SED during In Vitro Growth

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Introduction: According to data from the European Food Safety Authority, bacterial toxins are one of the main agents of foodborne illness. *Staphylococcus aureus* can produce up to 21 enterotoxins (SEs) and most of them can cause the sudden onset of vomiting after food ingestion. Studies on the conditions promoting SE production by strains carrying *se* genes are currently very limited.

Purpose: This study aimed to determine the growth kinetics of *sea* and *sed* gene *S. aureus* strains, *sea* and *sed* gene expression, and production of SEs.

Methods: Two wild strains carrying *sea* and *sed* genes were cultured to obtain an initial cell density of 10⁴ CFU/ml. Tubes were incubated at 30°C and optical density at 600 nm (OD₆₀₀), and *S. aureus* enumeration was performed at different times (1, 9, and 24 h). SE production and gene expression were verified during the *in vitro* growth at early (7 h), mid (9 h), and late exponential phases (24 h). *Se* gene expression was analysed using a $\Delta\Delta C_t$ relative quantification model with reference gene normalization.

Results: Both strains showed similar growth kinetics: OD₆₀₀ values were 0.03, 0.39, and 3.84 for *sea*-strain and 0.04, 0.31, and 3.20 for *sed*-strain at early, mid, and late exponential phases, respectively. *S. aureus* enumeration for *sed*-strain was lower at early (7.04 versus 7.40 log CFU/ml) and late (8.34 versus 9.28) phases, whereas enumeration at the mid phase was comparable (7.89 versus 7.82). *sea* was only detected at mid phase and late phases; *sed* was also detected at the early growth phase. Similarly, the gene expression showed that the transcription of *sea* started at mid phase and reached the maximum rate at late phase; transcription of *sed* started at early phase and reached the maximum rate at late phase.

Significance: These results are useful to define critical phases during food preparation and storage processes to prevent SEs production.

P2-51 SRL Pathogenicity Island in *Shigella flexneri* 2a Affects Persistence and Fitness in Post-harvest Tomatoes

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Introduction: Some enteric pathogens can persist and multiply within plants and their fruits. Yet, little is known about *Shigella* spp. in these conditions, especially in tomatoes.

Purpose: This study identifies the *Shigella* resistance locus pathogenicity island (SRL PAI) of *Shigella flexneri* 2a YSH6000 as an element affecting persistence and fitness in post-harvest tomatoes.

Methods: By using a fitness test, we documented differences in the ability of two tomato varieties to support populations of *S. flexneri* 2a YSH6000 and its isogenic pair with a deletion of the SRL PAI, *S. flexneri* 2a 1363.

Results: In tomatoes of the Vine variety (regular size), persistence of both *S. flexneri* strains increased by 1.5 log within the first two days and decreased up to 0.5 log at day six of incubation. Alternately, in cherry-type tomatoes of the Mini Plum variety, the persistence of the multidrug resistant strain *S. flexneri* 2a YSH6000 was stable to 1.5 log until day six, while the persistence of the *S. flexneri* 2a 1363 strain significantly decreased. However, when the fitness was tested on cherry-type tomatoes, it was shown that the

S. flexneri 2a 1363 strain was outcompeting *S. flexneri* 2a YSH6000 strain carrying the SRL PAI.

Significance: These results argue for a dual function of SRL PAI in cherry-type tomatoes. It can confer stability in persistence but, at the same time, its deletion would support an increase in fitness possibly due to the elimination of the expensive machinery of the antibiotic resistance genes.

P2-52 New Bioluminescent Alkaline Phosphatase Test for Verification of Milk Pasteurization

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Introduction: This study introduces a new technology for the monitoring of alkaline phosphatase verification from pasteurised milk samples. The technology uses new bioluminescent substrates in a simple, self-contained device using a new luminometer to measure and interpret results from samples

Purpose: To demonstrate the performance of ZymoSnap ALP as a rapid and efficient method for pasteurization verification.

Methods: Pasteurized milk from ten supermarkets across the United States covering all fat contents, from 0.1 to 4%, were evaluated in two assays for various Hygiene luminometers. Both assays produced results equivalent to the gold standard Fluorophos system. Each milk sample was spiked with four levels of bovine alkaline phosphatase to the following levels: 0, 100, 350, and 1000 mU/L. Each milk was assayed using five replicates with means compared and charted for correlation.

Results: The assay demonstrated excellent linearity at all milk fat contents and demonstrated an inversely proportional relationship between fat content and light output. At 4, 2, 1, and 0.1%, the conversion of mU/L alkaline phosphatase to relative light unit (RLU) was as follows: at 4%, RLU=3 mU/L; at 2% RLU=1.75 mU/L; at 1%, RLU=1.5 mU/L; and at 0.1%, RLU=1.0 mU/L. The correlation coefficients for the ten milk types at *n*=5 was as follows: at 4%, R²=0.9561; at 2%, R²=0.9561; at 1%, R²=0.9561; and at 0.1%, R²=0.9561. Flavoured milks were also tested and followed a similar trend, with fat content of 4% having the largest influence on signal output and not color of milk. Strawberry milk had a conversion of 1.2 RLU per mU/L and chocolate milk had a conversion of 1.1 RLU per mU/L.

Significance: The easy-to-use assay gives dairy processors of all sizes the ability to run a quick and inexpensive alkaline phosphatase assay on milks of any fat content or flavour for rapid pasteurisation verification.

P2-53 BAX PCR Confirmation of Presumptive Positive Hygiene InSite Listeria Rapid Chromogenic Tests

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Introduction: Rapid and accurate confirmation of presumptive positive environmental *Listeria* samples is critical for guiding a manufacturer's response to positive results, and protecting both consumers and the company brand. However, PCR results are prone to interference by sample matrices and often require extensive preparation before PCR testing.

Purpose: This study aimed to determine the accuracy of BAX PCR results when samples were transferred directly from an InSite Listeria (IL) chromogenic test device with no sample pre-treatment. A crucial question is whether a positive *Listeria* sample gives a positive confirmation by PCR as soon as there is a colour change in the chromogenic test.

Methods: *Listeria* species (including *L. monocytogenes*) were spiked into IL and incubated at 37°C. Samples were transferred from IL directly into BAX PCR at 16, 18, 20, 22, 24, and 48 hours. Chromogenic and PCR results were compared at each time point.

Results: BAX PCR was able to detect *Listeria*-positive samples taken directly from IL devices. All presumptive

positive IL devices were correctly confirmed as positive by BAX PCR tests. BAX PCR tests were also able to detect *Listeria*-positive samples before a visible colour change had appeared in IL devices.

Significance: The results of this study show that the BAX PCR system can be used to run accurate confirmation tests from presumptive positive chromogenic environmental swab tests. The ability to detect *Listeria*-positive samples before a visible colour change in the ILMG devices ensures that the BAX PCR test will give accurate results when started as soon as a colour change is observed in the chromogenic method, reducing the time to result for manufacturers.

P2-54 Simultaneous Detection of *Listeria* Species and *Listeria monocytogenes* Using Combined Chromogenic and Fluorescence for Environmental Monitoring

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Introduction: This study introduces a new test kit, Hygiene InSite *L. mono* Glo, for the simultaneous detection of *Listeria* species and *Listeria monocytogenes* from surfaces using a combination of chromogenic and fluorometric detection biochemistries in a self-contained device.

Purpose: The test is designed to make detection on surfaces easier and simpler for food manufacturers and food service retailers by combining both species and *Listeria monocytogenes* confirmation in one easy-to-use and easy-to-interpret rapid device.

Methods: Stainless steel coupons were spiked with serial dilutions of 20 *Listeria* species which included 10 *L. monocytogenes* species. The coupons were allowed to dry overnight at room temperature and then swabbed with the device. The incubation period was run at 37°C for 24, 30, and 48 hours, and the color intensity from the chromogenic substrate was measured with concurrent indication of *L. monocytogenes* using a small inexpensive UV black light to indicate *L. monocytogenes*.

Results: At 24 hours incubation, dilutions -1 through -7 were detected chromogenically at serial decreasing CFU levels from 2.8 e8 CFU at -1 down to 222 CFU at -7. The fluorescent detection of *L. monocytogenes* at 24 hours was from -1 down to -6 dilution at a lower mean of 2,221 CFU. At 30 hours, the detection limit of the test for both chromogenic and fluorometric detection dropped to 25 CFU, and at 48 hours the detection level dropped to <10 CFU for both chromogenic and fluorometric detection. The probability of detection was also calculated at each dilution level, with higher dilution levels for both species and *L. monocytogenes* being easily detected at 24 hours and single or <10 CFU being detected at either 30 or 48 hours, depending on stress level.

Significance: The simultaneous detection of both *Listeria* species and *L. monocytogenes* in a single device within 48 hours offers strategic advantage to an environmental monitoring program.

P2-55 Effect of Food Pathogenic and Spoilage Bacteria on *Listeria monocytogenes* Biofilm Formation

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Introduction: *Listeria monocytogenes* has been associated with major foodborne outbreaks; however, a stabilization in human listeriosis cases has been observed in 2015,

according to the European Food Safety Authority (EFSA). The importance of *L. monocytogenes* for the food industry has been linked to its ability to form biofilms; however, the presence of other species and their effect on biofilm formation should be considered.

Purpose: To evaluate the influence of food spoilage and pathogenic bacteria on the ability of *L. monocytogenes* to form biofilms on a stainless steel (SS) surfaces.

Methods: *L. monocytogenes* left to form biofilm in mono-cultures or co-cultures with pathogenic or spoilage bacteria on SS immersed in TSB at 20°C for 6 days. Specifically, spoilage bacteria belong to the genera *Leuconostoc*, *Lactobacillus*, *Serratia*, *Citrobacter*, *Hafnia*, *Proteus*, *Pseudomonas*, and *Brochothrix* where selected. In addition, two *Salmonella enterica* serotype Enteritidis and *Escherichia coli* O157:H7 strains were included in this study. Biofilm population was enumerated by bead vortexing-plate counting method.

Results: All microorganisms tested were able to produce biofilm on SS coupons after 6 days incubation at 20°C, while biofilm formation seemed to be influenced by the bacterial species and/or strains. In particular, *L. monocytogenes* reached biofilm population of about 5.0 log CFU/cm². Higher levels of biofilm population was enumerated for the most of the rest of the bacteria tested, except one *Brochothrix thermosphacta* strain (less than 4 log CFU/cm²). Dual species conditions did not seem to affect *L. monocytogenes* biofilm formation, as similar populations were enumerated in most of the cases. In brief, in only two cases when *L. monocytogenes* was co-cultured with *Serratia proteamaculans* and *Proteus vulgaris* was a slight reduction in the number of *L. monocytogenes* sessile cells (approximately 0.5 log CFU/cm²) observed compared to monocultures.

Significance: This research will hopefully improve our knowledge on the physiology of multi-species biofilms formed by food-relevant microorganisms under food-related conditions.

P2-56 Introduction of Hygiene InSite *Salmonella* as Rapid Method for Surface Surveillance of Stressed *Salmonella*

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Introduction: The contamination of surfaces with *Salmonella* species is one of the major routes for cross-contamination. InSite *Salmonella* is a simple-to-use, rapid device for screening surfaces for *Salmonella* species.

Purpose: The InSite *Salmonella* device is designed to swab a large area for possible salmonellae contamination, pre-enrich, and then selectively enrich in the same device giving a presumptive positive 24 hours from sample collection. This allows better and more rapid surveillance of high risk surfaces.

Methods: *Salmonella* Typhimurium ATCC 14028 and *Salmonella* Arizonae ATCC 13314 were grown and diluted into diluent. Sterile stainless steel squares were inoculated with 100 µl from each dilution -1 through -9 and air-dried overnight under asepsis. Each square was swabbed with one InSite *Salmonella* device and incubated at six hours in the pre-enrichment phase, and a further 18 hours in the selective phase. Devices are positive if a vivid yellow color appears in the viewing window. All devices used in the study were analysed for confirmation of positivity using the U.S. Food and Drug Administration Bacteriological Analytical Manual (BAM) *Salmonella* confirmation steps with a final identification by biochemistry. Directly inoculated devices without a drying step were performed as controls.

Results: The drying of the *Salmonella* on stainless steel surfaces rendered the bacteria stressed. Dilutions -1,-2,-3, and -4 were all positive for both *Salmonella* Typhimurium and for *Salmonella* Arizonae. The -5 dilution dried was also detected using the InSite *Salmonella* device. All dilutions from the directly inoculated devices were positive. All devices were then confirmed for negativity and positivity using a

confirmation protocol from BAM; the confirmation was 100% for both positives for *Salmonella*, but also negative for *Salmonella* from those swabs that indicated a presumptive negative at 18 hours.

Significance: An easy-to-use enclosed device that will reliably indicate presumptive positive and negative *Salmonella* from surfaces has advantages to many food producers and manufacturers.

P2-57 Strain Variability: Kinetics and Genotypical Characterization of *Listeria* spp. Isolated from Food

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Introduction: *Listeria monocytogenes* is a foodborne pathogen that can resist disinfectants, form biofilms, and survive under extreme physicochemical situations, such as dry environments, different temperatures, a wide range of pH values, and high salt concentrations. All these conditions promote the proliferation of the pathogen in a large variety of food matrices.

Purpose: The aims of this study were: (i) to evaluate the kinetics properties (growth and death rates) of *Listeria* spp. isolated from food and (ii) to type *Listeria* spp. isolates by pulsed field gel electrophoresis (PFGE).

Methods: A total 22 strains (17 *L. monocytogenes* and six *L. innocua*) previously isolated from different food matrices (vegetables, dairy, meat, and fish) and environmental samples were cultivated in brain heart infusion broth at 25°C to calculate the growth rates and at 55°C to evaluate the death rates. All the experiments were performed in duplicate. PFGE fingerprints were obtained with the restriction enzyme *Apal* according to PulseNet protocol.

Results: A wide strain variability was found in relation to the kinetics properties: The growth rates ranged from 0.25 to 0.46 log CFU/ml, while the death rates ranged from -1.02 to -4.61 log CFU/ml, confirming that, in stress conditions, the strains could assume different stress-response behavior. Two strains of *L. monocytogenes* isolated from fillet trout and bacon showed both the highest and the lower values for the growth and the death rates, respectively. PFGE subtyping of all the isolates showed 19 different PFGE types, proving a wide genomic variability. No correlation between food matrices, kinetics properties, and genotype was found.

Significance: These results may improve knowledge about the strains' behavior in optimal or stressful condition and may lead to the creation of a strain collection to use for challenge tests in different food matrices and in different conditions.

P2-58 Impact of Biocides on *Listeria monocytogenes* Viability in Biofilm and in Seafood Industrial Environment

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Introduction: *Listeria monocytogenes* is a food pathogen frequently isolated in the seafood industry. This bacterium adheres to surfaces and forms biofilms composed of an extracellular matrix. This extracellular matrix protects bacterial cells from environmental stress factors such as cleaning and disinfection operations (NaD). It is therefore essential for professionals to have effective biocides to eliminate these biofilms and limit the transfer of *L. monocytogenes* cells from surfaces to food.

Purpose: The objective of present study was to evaluate the impact of two biocides on the cell viability of *L. monocytogenes* in biofilm using two recovery methods recommended in ISO 18593:2004.

Methods: Stainless steel or PVC coupons were incubated for 1 h at 8°C in the presence of filtered smoked salmon juice and then for 48 h at 8°C in the presence of a bacterial suspension of *L. monocytogenes* mixed with *Carnobacterium maltaromaticum* and *Carnobacterium divergens* to allow the formation of a mixed biofilm. Treatments with biocides (two) or water (control) were applied to the biofilms, followed by application of recovery methods (contact plate or sponge stick). Bacteria were enumerated on agar medium for the cultivable population and by qPCR and PMA-qPCR, respectively, for the total and viable populations of *L. monocytogenes*.

Results: In all conditions, the treatment with the two biocides tested did not allow the removal of *L. monocytogenes* cells on the surface but led to a change in the viability of the population with mostly viable but non-culturable cells. Quantification data and epifluorescence microscopy observations showed that the efficiency of recovery methods varied according to the biocide treatment used and the surface on which these treatments were applied.

Significance: These results have allowed us to better understand and manage the health risk associated with this bacterium in industries.

P2-59* The Impact of Inter-strain Interactions and Matrix Adaptation on Growth and Acid Resistance of *L. monocytogenes* Strains on Different Types of Cheeses

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Introduction: Contamination of cheese with multiple *Listeria monocytogenes* strains has been previously demonstrated. However, scarce information exists on the survival and/or growth of the pathogen in contamination with multiple strains.

Purpose: To evaluate the effect of inter-strain interactions and matrix adaptation of *L. monocytogenes* strains on their growth on ricotta and Camembert cheeses and subsequent acid resistance to simulated gastric fluid (SGF).

Methods: Antibiotic-resistant (for selective enumeration), matrix-adapted (MA), and non-adapted (NA) *L. monocytogenes* strains (C5, ScottA [serotype 4b]; 6179 [1/2a]; PL25 [1/2b]), were inoculated in single- or two-strain cultures (1:1 strain ratio) at approximately 2.5 log CFU/g on ricotta and Camembert (10 g each) cheeses. Adaptation of cells was performed in cheese broth (1:1 cheese in maximum recovery diluent) for 48 h at 7°C. Growth and acid-resistance of *L. monocytogenes* were assessed during aerobic storage of cheese samples at 7°C. Survival of middle exponential and early stationary bacteria cells was evaluated after 10, 20, 40, 60, and 120 minutes in SGF (pH 2.0; 37°C) ($n=3 \times 2$).

Results: On Camembert, co-cultivation and matrix-adaptation did not affect the growth of strains in mixed compared to single cultures. On ricotta, significant ($P < 0.05$) growth inhibition of certain strains in mixed cultures was observed as manifested by the final population levels of the pathogen. NA ScottA (4.8±0.5 log CFU/g) and NA and MA 6179 (5.2±0.3 log CFU/g and 5.1±0.6 log CFU/g, respectively) were suppressed by the presence of C5, compared to the corresponding single cultures, which reached 7.9±0.2, 7.5±0.5, and 7.8±0.5 log CFU/g, respectively. Habituation of the pathogen on Camembert resulted in acid sensitization against subsequent exposure to SGF. Regarding ricotta, ScottA displayed increased acid resistance compared to C5 and PL25, even though it was outcompeted during storage.

Significance: The results reveal how cheese matrix may affect the outcome of inter-strain interactions and could help explain the dominance of certain serotypes in foods where *L. monocytogenes* is a safety concern.

P2-60 Analysis of Microbiological Hazards in Chinese Cabbage and Growing Environment

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Introduction: Pathogenic bacteria causing outbreaks of foodborne illness are frequently associated with fresh produce. Public health concerns about the microbiological safety of Chinese cabbage, the main ingredient in kimchi, have been increasing in Korea.

Purpose: The purpose of this study was to find the contamination sources of Chinese cabbage by evaluating the microbial ecology among cabbage, field soil, and irrigation water, both quantitatively and qualitatively.

Methods: A total of 216 samples of Chinese cabbages, soils, and irrigation water were collected from randomly selected fields, storage, and markets. The samples were analyzed for total aerobic bacteria, coliforms/*Escherichia coli*, yeast/mold, and six pathogens.

Results: The levels of total aerobic bacteria on average were 6.94, 4.98, and 2.60 log CFU/g in soil, cabbage, and water, respectively. The bacterial levels of cabbage from storage and markets (6.25 and 6.01 log CFU/g) were higher than from the fields, and the number of bacteria in irrigation water collected from the surface (2.98 log CFU/g) was higher than from the ground (2.11 log CFU/g). The *E. coli* was detected in three of 87 Chinese cabbage samples (3.45%) and seven of 42 irrigation water samples (16.67%), but not in soil, while the detection rate of *Bacillus cereus* and *Staphylococcus aureus* were 5.98 and 2.72%, mostly found in soil and water. *E. coli* was found in both cabbage and surface irrigation water collected at the same three locations.

Significance: These results suggest that the pathogens in cabbage are carried over from the contaminated irrigation water, especially surface water. Thus, the use of ground water or the change of irrigation methods from spray to drip should be considered to ensure the microbial safety of Chinese cabbage.

P2-61 The Impact of Cold Stress on the Survival of Shiga Toxin-producing *Escherichia coli* on Field-grown Romaine Lettuce

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Introduction: Leafy greens have been identified as a significant vector for the transmission of Shiga toxin-producing *Escherichia coli* (STEC), leading to multiple outbreaks worldwide.

Purpose: The objective of this study was to evaluate the effect of cold stress on STEC serotypes experimentally inoculated onto romaine lettuce plants in the field.

Methods: Romaine lettuce (*Lactuca sativa* cv Parris Island) plants grown to maturity (55 to 60 days) were inoculated with 10⁷ CFU/ml (18 h, 37°C, 150 rpm) of non-pathogenic serotypes O157:H7, O103:H2, O111:HNM, O26:H11, O145:HNM, and O44:H18 that were cold stressed (4°C for 5 days) or non-stressed. For each serotype, a row of plants was inoculated with the cell suspension using a watering can (~100 ml per plant). At each sampling (day 3, 7, 14, 21, 28, and 35), 225 g of leaves randomly selected from three lettuce heads was added to 450 ml of modified tryptic soy broth in a large stomacher bag and placed on a rotary shaker (100 rpm) for 30 min. Serial dilution plating ($n=3$) was performed on selective media (MACVC, MacConkey Agar + Vancomycin 10 µg/ml + Cefsulodin 3 µg/ml) incubated overnight at 42°C for the determination of cell populations. Independent trials were performed over two growing seasons and mean populations of bacteria were converted to log₁₀ values. Analysis of variance was performed using the linear model in SAS software and differences between serotypes were assessed by LSMEANS ($P<0.05$).

Results: There was no difference ($P>0.05$) in bacterial populations of cold stressed and non-stressed cells recovered from the romaine lettuce plants over the 35-day experimental period. However, there were daily variations among individual serotypes ($P<0.05$) and cells persisted up to 35 days.

Significance: Results provide an increased understanding of the cold temperature acclimatization exhibited by STEC, thereby providing knowledge which can be used to develop effective control strategies.

P2-62 Assessment of Food Safety Knowledge and Practices of Workers Handling Produce at the Wholesale Market in Doha, Qatar

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Introduction: Preventing food from becoming contaminated with bacteria is problematic in many developing countries, like Qatar, where the majority of food comes from countries with poor hygiene practices. Fresh produce is considered a healthy food choice; therefore, many Qataris have started consuming such foods in their diet. The contamination of fresh produce can occur during any stage of the farm-to-table process (e.g., handling of produce by workers who might have been infected with pathogens) which eventually can lead to outbreaks. In Qatar, most of the food workers come from Far East. The workers' hygiene in the wholesale market in Doha is considered an important factor that affects the transfer of microorganisms to produce.

Purpose: This study was carried out to evaluate the food safety knowledge and practices of workers who are in direct contact with produce at the wholesale market in Doha, Qatar.

Methods: A total of 120 workers were surveyed in the study using a 21-questions questionnaire. During the survey application, hand-swab samples were collected to determine the workers' hygiene levels.

Results: All respondents indicated that they did not receive any official training on food safety. The major age interval of the workers was 31 to 40 yrs old (36.7%), only 37.5% of the workers had high school degrees, and 67% of them have been working at the market for more than five years. Most participants (92%) generally do not use gloves and claimed to wash their hands 4–5 times/day. Different pathogens were identified as microbial hazards isolated from handlers' hand-swabs, such as *Klebsiella* and *Enterococcus faecium*, indicating inadequate hygiene practices.

Significance: These results demonstrate the urgent need to train the produce handlers on food safety and hygiene practices and adopt better control measures at the produce market to improve sanitary conditions.

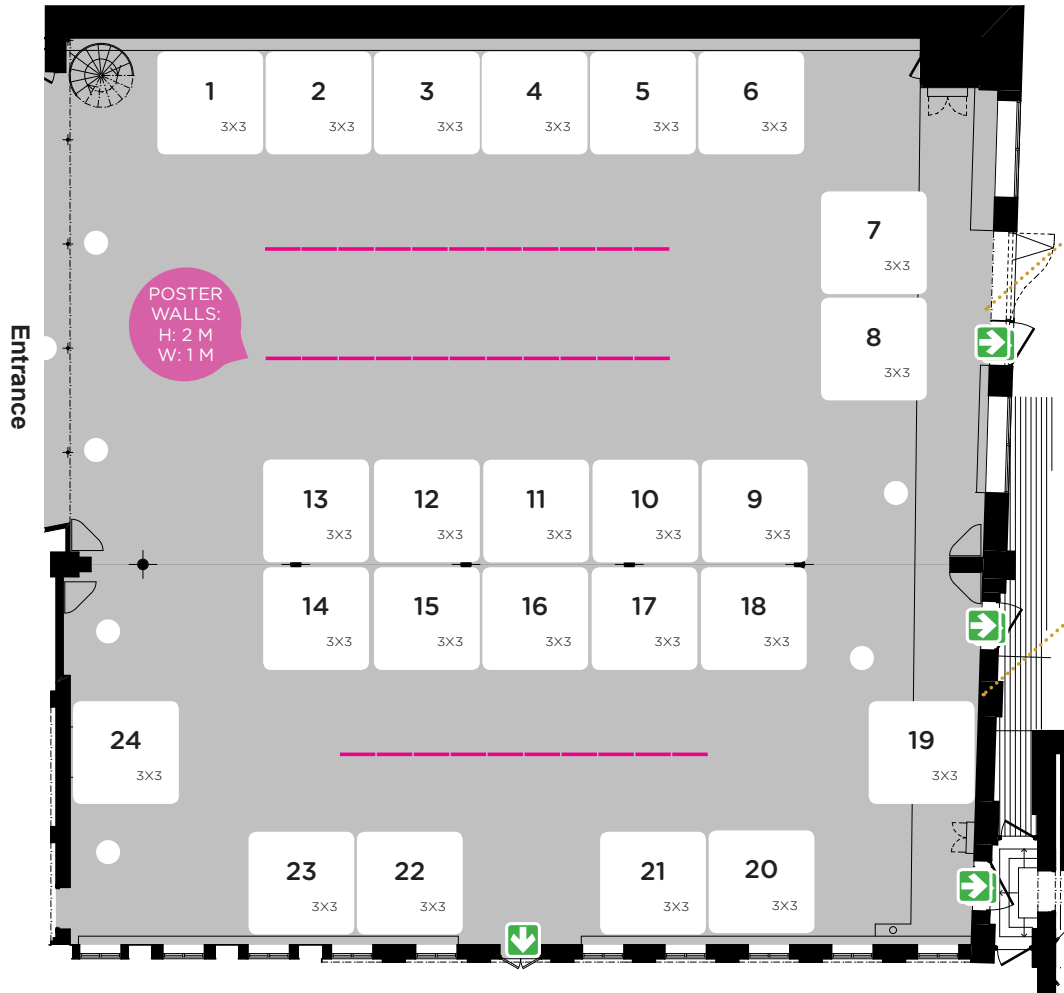


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7-8	Hygiene International Ltd.
20	ILSI Europe A.I.S.B.L.
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 **Stand 23**

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Stand 21

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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 (e.g., the *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 the food safety situations in developing countries.

The 26th International ICFMH Conference, FoodMicro 2018, will take place in Berlin (Germany) at Freie Universität Berlin, 3–6 September 2018, with the theme "Biodiversity of Foodborne Microbes" (<http://www.foodmicro2018.com/>). We shall be pleased to welcome you there!

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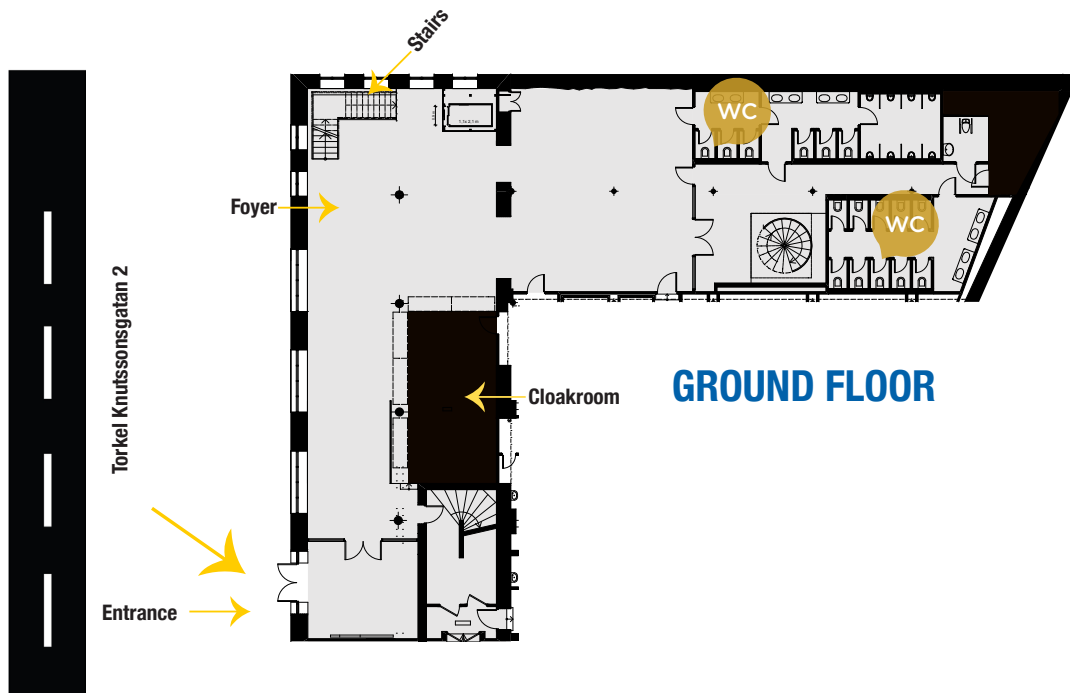
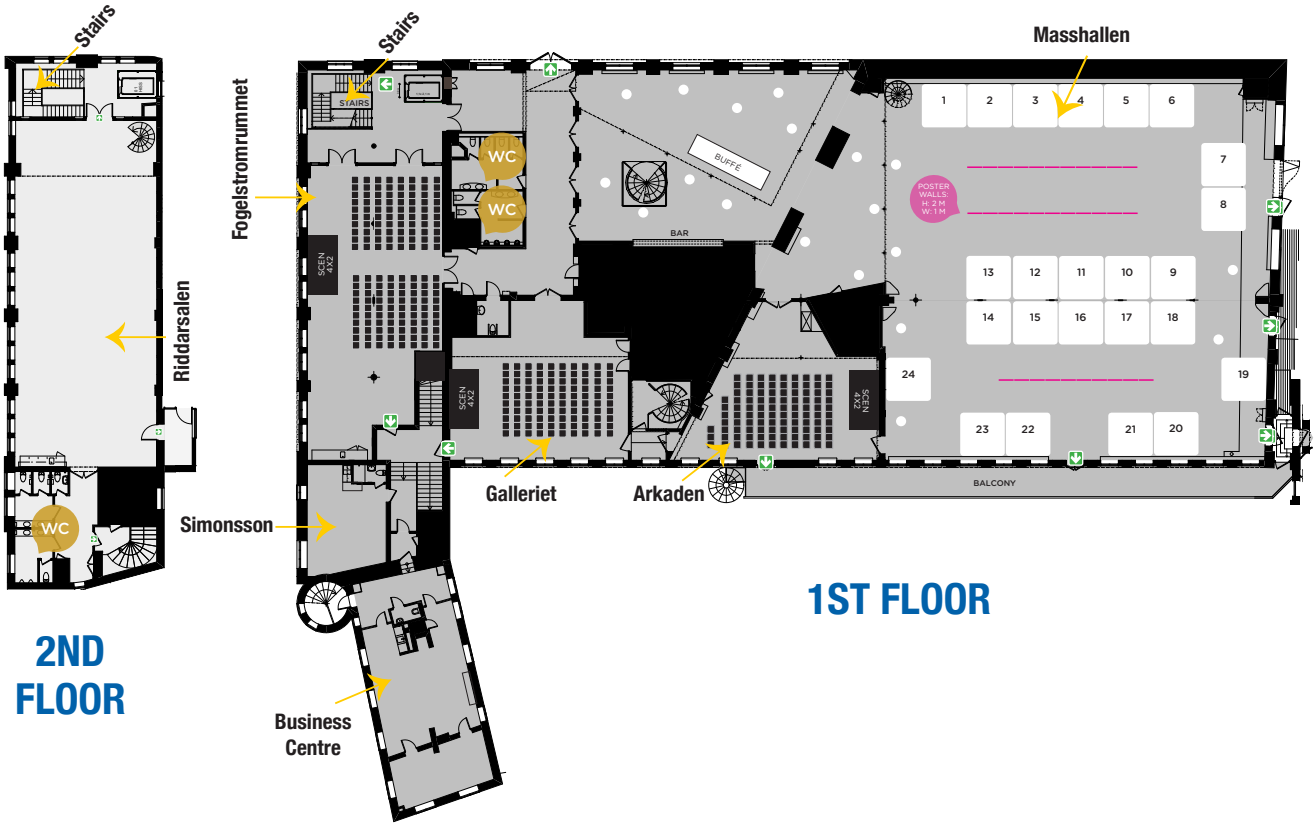
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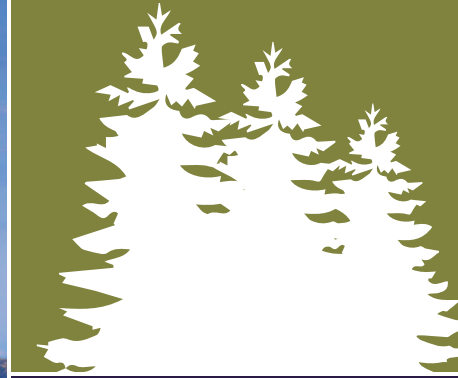
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