European Symposium on Food Safety
20-22 April 2015
Cardiff City Hall, Cardiff, Wales

Programme

In Collaboration with ILSI Europe and with the Technical Cooperation of the Food and Agriculture Organization of the United Nations.

Hosted by the United Kingdom Association for Food Protection

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Exhibitors
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Organising Committee

Christina Harzman, Chair
BIOTECON Diagnostics — Germany

Patrice Arbault, Vice Chair
BioAdvantage Consulting — France

Alejandro Amezquita
Unilever — United Kingdom

Peter Ben Embarek
World Health Organization — Switzerland

Sarah Cahill
Food & Agriculture Organization of the United Nations — Italy

Allessandro Chiodini
ILSI Europe — Belgium

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ADRIA — France

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Netherlands Food and Consumer Product Safety Authority — The Netherlands

Akos Jozwiak
National Food Chain Safety — Hungary

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CEERAM — France

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INRA — France

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Lisa Hovey
International Association for Food Protection — United States

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Daniel Thomas
The Welsh Government

Carol Wallace
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Roy Betts
Campden BRI

John Holah
Holchem Laboratories Ltd.

Daniel Thomas
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Derrick Blunden
Whisper and Shout

Andrew Martin
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TU Delft – The Netherlands

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Maple Leaf Foods – Canada

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Aristotle University of Thessaloniki – Greece

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Sainsbury’s Supermarkets Ltd. – United Kingdom

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Ghent University – Belgium

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University of Milan – Italy

Pietermel Luning  
Wageningen University – The Netherlands

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University Medical Center Utrecht – The Netherlands

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NSF – United Kingdom

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National Center for Electron Beam Research – USA

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Agricultural University of Athens – Greece

Taran Skjerdal  
Norwegian Veterinary Institute – Norway

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ARDIA – France

Valérie Stahl  
Aerial Technical Institute for Food Industry – France

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Global Food Traceability Center – USA

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University of Aberdeen – United Kingdom

Helen Taylor  
Cardiff Metropolitan University – United Kingdom

Clément Trunet  
Université de Brest – France

Purnendu Vasavada  
University of Wisconsin-River Falls – USA

Maria Vitale  
Istituto Zooprofilattico Sperimentale della Sicilia – Italy

Jaap Wagenaar  
Utrecht University – The Netherlands

Steve Wearne  
Food Standards Agency – Wales

Marjon Wells-Bennik  
NIZO Food Research and Top Institute Food and Nutrition – The Netherlands

Anett Winkler  
Mondelez International – Germany

Ian Young  
Food and Agriculture Organization of the United Nations – Italy and Public Health Agency of Canada – Canada

Donald Zink  
U.S. Food and Drug Administration-CFSAN – USA

Marcel Zwietering  
Wageningen University – The Netherlands
Day 1 - Monday, 20 April

9.00 — 17.00 Registration Open

Posters will be on display from 10.00-18.00. Poster presentations will take place during coffee breaks.

PL1 Opening Session
Assembly Room
Chair: David Loyd

10.00 Introduction to IAFF and the IAFF European Symposium
DAVID THARP, International Association for Food Protection, Des Moines, IA, USA

10.15 The International Association for Food Protection
DONALD ZINK, U.S. Food and Drug Administration-CFSAN, College Park, MD, USA

10.30 A Welcome to Wales
VAUGHAN GETHING, Assembly Member for Cardiff South & Penarth Cardiff, Cardiff, Wales

11.00 Strategic Challenges to Food Control until 2020
STEVE WEARNE, Food Standards Agency, Cardiff, Wales

11.30 Effort: A European Project on the Ecology from Farm to Fork of Microbial Drug Resistance and Transmission
JAAP WAGENAAR, Utrecht University, Utrecht, The Netherlands

12.00 — 13.30 Networking Lunch in the Exhibit Hall

S1 Integration of Omic Data into Microbiological Risk Assessment
Assembly Room
Organizers: Luca Cocolin and Marcel Zwietering
Chair: Jeanne-Marie Membré

13.30 Seizing the Potential of “Oomics” to Explore the Behavior of Foodborne Pathogens
KALLOPI RANTSIOU, Valentina Alessandria, Luca Cocolin, DISAFA—Università di Torino, Torino, Italy

14.00 Metagenomics and QMRA: Is There a Match?
EELCO FRANZ, Romain Pielast, RIVM — Centre for Infectious Disease Control, Bilthoven, The Netherlands

14.30 Omics and QMRA: Challenges and Promises of the Future
NABILA HADDAD, Sandrine Guillou, Jeanne-Marie Membré, UMR INRA 1014 SECALIM, Nantes, France, LUNAM Université, Oniris, Nantes, France

S2 Current Issues in HACCP Training: Making Systems More Effective
Ferrier Hall
Organizers: Carol Wallace and Lone Jespersen
Chair: Carol Wallace

13.30 Embedding HACCP and Food Safety Culture through Organisational Training Approaches
SARA MORTIMORE, Land O’ Lakes, Inc., St. Paul, MN, USA

14.00 Using HACCP Knowledge Metrics to Revamp Manufacturing HACCP Systems — A Case Study
LONE JESPERSEN, Maple Leaf Foods, Mississauga, ON, Canada

14.30 Challenging All Trainers — Can You Successfully Train HACCP Concepts to SMEs and Micro Manufacturers? A Case Study on the Institute of Food Science & Technology/Safe and Local Supplier Approval—HACCP Training Initiative (UK)
HELEN TAYLOR, Cardiff Metropolitan University, Cardiff, United Kingdom

S3 Cryptosporidium and Giardia: Food Safety Issues of Important Concern
Syndicate Room C
Organizer: Fabienne Loisy
Chair: Roland Salmon

13.30 Importance of Food as a Transmission Vehicle for Cryptosporidium and Giardia
LUCY ROBERTSON, Norwegian University of Life Sciences, Oslo, Norway

14.00 Standardisation of Methods to Detect Cryptosporidium and Giardia in Berry Fruit and Leafy Green Vegetables
NIGEL COOK, The Food and Environment Research Agency (Fera), York, United Kingdom

14.30 Challenges of Identifying and Investigating Outbreaks of Foodborne Cryptosporidiosis
RACHEL CHALMERS, Public Health Wales Microbiology, Swansea, United Kingdom

T1 Technical Session 1 — Laboratory and Detection Methods
Syndicate Room D
Chair: Cian O’Mahoney

13.30 The Use of Microbial Flora Analytical Tools for the Discrimination of Organic Foods
CÉLINE BIGOT, Cirad, Qualisud, Montpellier, France

13.45 A Method to Validate Bacterial Thermal Destruction Taking into Account Recovery Conditions in Food Matrix
IVAN LEGUERINEL, Loic Chene, Veronique Huchet, Université de Brest, Quimper, France

14.00 Impact of Pooling Samples on the Detection of L. monocytogenes in Food
NATHALIE GNANOU BESSE, Jean Christophe Augustin, ANSES, Laboratoire de Sécurité des Aliments (PRES Paris Est), Maisons Alfort, France

14.15 Microplate Immunoassay (IMC): A New Method for the Isolation/Concentration of Escherichia coli O157:H7 in Food
PATRICE ARBAULT, Delphine Larose, Nicolas Desroche, Jean Guzzo, BioAdvantage Consulting, Orlienas, France

14.45 Discrimination of Saccharomyces cerevisiae and Non-Saccharomyces Yeasts Isolated from Spanish Grapes by Attenuated Total Reflectance Infrared Spectroscopy Combined with Multivariate Analysis
MIQUEL PUXEU, Imma Andorra, Anna Brull, and SILVIA DE LAMO, Universitat Rovira i Virgili, Tarragona, Spain

14.45 Pretreatments for Estimation of Viable Viruses through Reverse Transcription-qPCR in Shellfish: Pros and Cons
SILVIA MONTEIRO, Ricardo Santos, Laboratorio Analises, Instituto Superior Tecnico, Lisbon, Portugal
Day 1 - Monday, 20 April

15.00 — 15.30 Coffee Break in the Exhibit Hall

S4 Burden of Foodborne Diseases
Assembly Room
Organizer and Chair: Akos Jozwiak

15.30 Monetary and Adjusted Life — Year Estimates of Foodborne Disease Burden in the United States
MICHAEL BATZ, University of Florida, Gainesville, FL, USA

16.00 The Pathogen- and Incidence-based DALY Approach: A New Methodology for Estimating the Burden of Infectious Diseases in Europe
MARIE-JOSE MANGEN, University Medical Center Utrecht, Utrecht, The Netherlands

S5 Strain and Population Diversity: Implications for Food Safety and Food Spoilage
Ferrier Hall
Organizer and Chair: Heidy Den Besten

15.30 Strain Variability in Growth and Inactivation Parameters: Relevance for Pathogens and Spoilage Organisms
DIAH C. ARYANI, Wageningen University, Wageningen, The Netherlands

16.00 Single Cell Variability: Relevance for Fungal Spoilage Processes
KOSTAS KOUTSOUMANIS, Aristotle University of Thessaloniki, Thessaloniki, Greece

16.30 Causes and Consequences of Heterogeneity in Stressed Populations of Foodborne Pathogens
ABRAM AERTSEN, KU Leuven, Leuven, Belgium

S6 Virus Testing, Interpretation and What Do I Do with a Positive Result
Syndicate Room C
Organizers: Dan Li, Alvin Lee and Stephen Grove
Chairs: Dan Li and Alvin Lee

15.30 Recovery of Viruses from Food and Interpretation of Results
SOPHIE BUTOT, Nestle Research Centre, Lausanne, Switzerland

16.00 Batch Testing for Noroviruses in Frozen Raspberries
DAN LI, Ghent University, Ghent, Belgium

16.30 What If I Find a Positive and What Do I Do with It?
NIGEL COOK, The Food and Environment Research Agency York, United Kingdom

Technical Session 2 — Dairy and Beverages, Epidemiology, Food Defense and Non-microbial Food Safety
Syndicate Room D
Chair: Mickey Parish

15.30 Food Traceability Method Employing Tagged-DNA Based Barcodes
GEORGE FARQUAR, Antonios Zografos, KURT-PETER RAEZKE, Intertek, Bremen, Germany

15.45 Thinking Inside the Box: Using Cartoon Strips to Teach Food Defense at the Retail Level
MICHELE SAMARYA-TIMM, Somerset County Department of Health, Somerville, NJ, USA

16.00 Pig Herds Free from Campylobacter — Dream or Reality?
TRULS NESBAKKEN, Terje Iversen, Evan M Kolstoe, Øyvin Østensvik, Section for Food Safety, Faculty of Veterinary Medicine and Biosciences, Norwegian University of Life Sciences, Oslo, Norway

16.15 Characterisation of Staphylococcus aureus Isolated from Dairy Farms in Victoria, Australia
EDWARD FOX, Kate McMillan, Peter Chandry, Theo Alnutt, Narelle Fegan, CSIRO, Werribee, Australia

16.30 The Challenge of Histopathology in the Detection of Illicit Treatments with Hormones Associations
MARIO BOTTA, Guia Benedetta Richelmi, Marzia Pezzolato, Elisa Baioni, Danilo Pitardi, Serena Meistro, Elena Bozzetta, Istituto Zooprofilattico Sperimentale del Piemonte, Liguria e Valle d’Aosta, Turin, Italy

16.45 PRNP Analysis in Sicilian Goat Breeds for TSE Resistance and Full Eradication of Prion Strains
SERGIO MIGLIORE, Stefano Agnello, Sebastian Mignacco, Vincenzo Di Marco Lo Presti, Fabrizio Vitale, Maria Vitale, Istituto Zooprofilattico Sperimentale of Sicily, Palermo, Italy

17.00 — 18.30 Exhibit Hall Reception
Day 2 - Tuesday, 21 April

8.00 — 17.00 Registration Open

Posters will be on display from 10.00-18.00. Poster presentations will take place during coffee breaks

S7 The U.S. Food Safety Modernization Act: What Does It Mean for Europe?

Assembly Room
Organizers and Chairs: John Bassett and Purnendu Vasavada

8.30 FSMA Preventive Controls and FDA Expectations for Foreign Suppliers
PURNENDU VASAVADA, University of Wisconsin-River Falls, River Falls, WI, USA

9.00 Meeting FSMA Regulations — A European Perspective
PETE MARTIN, NSF Coventry, United Kingdom

9.30 FSMA and Microbiological Testing
ROY BETTS, Campden BRI, Gloucestershire, United Kingdom

S8 Stochastic Approaches for an Enhanced Control of Spore Germination and Development in Food Products

Ferrier Hall
Organizer: Daniele Sohier
Chairs: Panagiotis Skandamis and Florence Postollec

8.30 Stress Resistance and Mechanisms Governing Germination and Outgrowth Heterogeneity of Individual Bacillus subtilis Spores after a Given Preservation Treatment
STANLEY BRUL, Wishwas Abhyankar, Rachna Pandey, Linli Zheng, Alex Ter Beek, Jan P. P. M. Smelt, Sacha Stelder, Leo J. de Koning, Henk Dekker, Chris G. de Koster, Molecular Biology and Microbial Food (SILS), University of Amsterdam, Amsterdam, The Netherlands

9.00 Bacterial Spore Heterogeneity of Behavior Due to Sporulation and Recovery Conditions
CLEMENT TRUNET, Narjes Mirmet, Anne-Gabrielle Mathot, Florence Postollec, Ivan Leguerinel, Olivier Couvert, Frédéric Carlin, Louis Coroller, Université de Brest, Quimper, France, UMT14.01 SPORE RISK, Quimper, France, ADRIA Développement, Quimper, France

9.30 Stochasticity of Germination/Lag Time of Individual Spores of Clostridium botulinum and the Safety of Minimally Heated Refrigerated Food
MIKE PECK, Institute of Food Research, Norwich, United Kingdom

T3 Technical Session 3 — General Microbiology

Syndicate Room D
Chair: Nigel Cook

8.30 Prevention of Human Norovirus and Bacterial Pathogens at Public Access Watershed Sites in a California Central Coast Agricultural Region
PENG TIAN, David Yang, Michael Cooley, Lisa Gorski, Beatriz Quinoones, U.S. Department of Agriculture-PSMRU-WRC-ARS, Albany, CA, USA

8.45 Impact of Pulsed Light on Listeria innocua — A Viability Profile
BERND KRAMER, Peter Muranyi, Fraunhofer Institute for Process Engineering and Packaging, Freising, Germany

9.00 Quantification of Salmonella Transfer on Tomatoes and Tomato Bedding
JENNIFER TODD-SEARLE, Loretta Friedrich, Ruth Oni, Kenneth Shenge, Jeffrey LeJeune, Shirley Micallef, Michelle Danyluk, Donald Schaffner, Rutgers, The State University of New Jersey, New Brunswick, NJ, USA

9.15 Contribution of the Salmonella enterica sv Typhimurium Capsular Transcriptional Regulators rcsA and rcsB to the Persistence of Salmonella in Post-harvested Tomatoes
MASSIMILIANO MARVASI, Max Teplitzki, Marcelo Farias, Keith Jenkins, Middlesex University, London, London, United Kingdom

9.15 Biofilm-forming Ability of Salmonella Typhimurium on Polystyrene Surface in the Presence of N-acyl-homoserine Lactone Molecules Produced by Hafnia alvei
VASILIKI BLANA, Xenia Gkaripogki, GEORGE-JOHN NYCHAS, Agricultural University of Athens, Department of Food Science and Human Nutrition, Athens, Greece

9.45 Campylobacter jejuni — A Major Public Health Concern: Integration of Genomic Data to Characterize Virulence of an Atypical Strain
VICKY BRONNEC, Stéphane Cruveiller, Odile Tresse, Nabila Haddad, UMR INRA 1014 SECALIM, Nantes, France; LUNAM Université, Oniris, Nantes, France, Nantes, France

10.00 — 10.30 Coffee Break in the Exhibit Hall

S9 Risk Assessment of Unintentional Allergen Cross Contact

Syndicate Room C
Organizer and Chair: Sylvia Pfaff

8.30 Management of Allergen Cross Contact in Food Transport Containers Such as Tanks
HANS-DIETER PHILIPOWSKI, ENFIT e.V., Pinneberg, Germany

9.00 Management of Allergen Cross Contact in the Food Industry — Acknowledgment of Reference Dose
SYLVIA PFAFF, FIS Europe, Bad Bentheim, Germany

10.00 Management of Allergen Cross Contact at Household Level — Best Practice for Food Labeling
HAZEL GOWLAND, Allergy Action, St. Albans, United Kingdom

10.30 Method Validation-ensuring New Methods Meet the Requirements of European Legislation

Assembly Room
Organizer: Roy Betts
Chair: David Tomas Fornes

10.30 Microbiological Test Methods — The Requirements and Influences of European Legislation on the Way We Test Foods
ROY BETTS, Campden BRI, Gloucestershire, United Kingdom

11.00 Understanding If New Methods Work — Designing a Method Validation Procedure — ISO 16140
PAUL IN’T VELD, VWA Netherlands, Eindhoven, The Netherlands
11.30 How to Validate a Method — Practical Aspects and Problems Associated with Method Validation
DANIEL PECK, Institute of Food Research, Norwich, United Kingdom

11.45 A Large Scale Study to Test a Wide Variety of Additives in Broilers’ Feed to Decrease Campylobacter Shedding
MURIEL GUYARD-NICODEME, Ségolène Quesne, Typhaine Poozevara, Bernard Le Berre, Michel Amelot, Marianne Chemaly, ANSES, Laboratory of Ploufragan-Plouzané, Ploufragan, France

12.00 — 13.30 Networking Lunch in the Exhibit Hall

S13 The Importance of Microbiological Testing in Food Safety Management
Assembly Room
Organizers: Alessandro Chiodini and Lilou van Lieshout
Chair: Lilou van Lieshout

13.30 The Role of Validation, Verification and Microbiological Sampling in a Food Safety Management System
MARCEL ZWIETERING, Wageningen University, Wageningen, The Netherlands

14.00 The Relevance of End Product Testing: The Example of Canned Foods and Cooked Ham
JEANNE-MARIE MEMBRÉ, INRA, Nantes, France

14.30 Relevance of Microbial Testing for Verification in the Production of Chocolate
ANETT WINKLER, Mondelēz International, Munich, Germany

S14 Prediction of Shelf-life, and Product’s Microbial Quality Using Smart and Non-invasive Platforms
Ferrier Hall
Organizer and Chair: George-John Nychas

13.30 Use of Non-invasive Tools in Tandem with Bioinformatics for the Implementation of Process Analytical Technology in Food Industry
GEORGE-JOHN NYCHAS, Agricultural University of Athens, Department of Food Science and Human Nutrition, Athens, Greece

14.30 Hyperspectral Chemical Imaging in Microbiology: Methods, Recent Applications and Future Directions
AIOFE GOWEN, UCD, DUBLIN, Ireland

14.30 Metabolomics Serving Food Quality and Safety
EFSTATHIOS PANAGOU, AUA, Athens, Greece
Day 2 - Tuesday, 21 April

S15 Managing Foodborne Toxoplasmosis: Recent Advances and Future Directions
Syndicate Room C
Organizer: Edward Guy
Chairs: Edward Guy and Tom Humphrey
13.30 Human Toxoplasmosis: Infection, Clinical Spectrum and Burden of Disease
EDWARD GUY, Public Health Wales, Swansea, Wales
14.00 Molecular Epidemiology of Toxoplasma: Genotypes and Human Toxoplasmosis — A Foodborne Perspective
MARIE-LAURE DARDE, University of Limoges, Limoges, France
14.30 Toxoplasma Infection in Food Animals
ELISABETH INNES, Moredun Research Institute, Edinburgh, Scotland

T5 Technical Session 5 — Modeling and Risk Assessment and Communication, Outreach and Education
Syndicate Room D
Chair: Michael Batz
13.30 Regional Trends in GMP Failures
KALLIOPI ZERVA, AIB International, London, United Kingdom
13.45 Towards a Mechanistic Model for the Germination of Clostridium Species
ALINE METRIS, Jason Brunt, József Baranyi, Mike Peck, Institute of Food Research, Norwich, United Kingdom
14.00 Selecting the Most Appropriate Risk Ranking Methods
ESTHER VAN ASSELT, Ine Van der Fels-Klerx, Helle Korsgaard, Lea Bredsdorff, Maarten Nauta, Morten Poulsen, Martin D'Agostino, Marian Raley, David Coles, and Lynn Frewer, RIKILT — Wageningen UR, Wageningen, Netherlands
14.15 Designing a Data Base for Supporting Decisions on the Use of Predictive Microbiology Software
FERNANDO PEREZ RODRIGUEZ, Salvador Cubero, University of Cordoba, Cordoba, Spain
14.30 Towards Community Driven Food Safety Model Repositories
MATTHIAS FILTER, Carolina Plaza-Rodríguez, Christian Thoens, Annewarrie Kaesbohrer, Bernd Appel, Federal Institute for Risk Assessment, Berlin, Germany
14.45 Growth Potential of Listeria monocytogenes in Soft and Semi-hard Artisanal Cheeses
EVY LAHOU, Mieke Uyttendaele, Ghent University, Ghent, Belgium

15.00 — 15.30 Coffee Break in the Exhibit Hall

S16 Risk Perception and Risk Analysis — The Consumers’ View of Food Safety
Assembly Room
Organizer and Chair: Maria Vitale
15.30 Food Safety Risk Communication. What We Know and What We Need to Know
ANTHONY FLOOD, International Food Information Council, Washington, D.C., USA
16.00 Two Case Studies on Risk Perception: Trust in Institutions and Gender Differences in Risk Perceptions
SEDA ERDEM, Department of Economic and Behavioural Science Centre, University of Stirling, Stirling, United Kingdom
16.30 From Farm to Fork: Common Practices and Food Consumers Behaviours — BSE Versus Toxoplasmosis
MARIA VITALE, Istituto Zooprofilattico Sperimentale Della Sicilia, Palermo, Italy

S17 Practical Examples of Predictive Microbiology Used by the Food Industry
Ferrier Hall
Organizer and Chair: Cian O’Mahony
15.30 Practical Application of Deterministic and Stochastic Microbial Models for Risk-based Food Product and Process Design
ALEJANDRO AMEZQUITA, Unilever, Sharnbrook, United Kingdom
15.00 Hurdle Efficacy Predictions and Listeria Growth Inhibition in Complex Food Matrices
MICHAEL CALLANAN, Nestlé Research Center, Lausanne, Switzerland
16.30 Development of a Model and Software for Listeria monocytogenes Growth in Ready-to-Eat Meats with Different Antimicrobial Formulations
CIAN O’MAHONY, Creme Global, Dublin, Ireland

S18 Fresh Produce and Water: How Can Risk Assessments be Used in Ensuring Safety of Fresh Produce?
Syndicate Room C
Organizers: Alessandro Chiodini and Lilou van Lieshout
Chair: Lilou van Lieshout
15.30 Target Microorganisms Used in Risk Assessments Related to Water and/or Fresh Produce
ANA ALLENDE, TU Delft, Delft, Netherlands, CSIC, Murcia, Spain
15.30 Data Gaps to be Considered in Modelling Strategies for Fresh Produce
PETER MCCLURE, Mondelez International, Birmingham, United Kingdom
16.30 Usage of QMRA Studies in Risk Assessments Related to Use of Water and Fresh Produce
LIESBETH JACKSENS, Faculty of Bioscience Engineering, Department of Food Safety and Food Quality, Ghent University, Ghent, Belgium

T6 Technical Session 6 — Antimicrobials, Meat, Poultry and Eggs, and Seafood
Syndicate Room D
15.30 Strategies for Targeted Control of Listeria in Norwegian Food Processing Environments
EVEN HEIR, Solveig Langsrud, Trond Maretra, Nofima, Norwegian Institute of Food, Fishery and Aquaculture, As, Norway
### Tuesday Evening Social

Plan to join with your colleagues for a special night at the National Museum Cardiff. Harpist Hannah Stone, the Official Harpist to the Prince of Wales, will welcome guests in the gallery. Dinner, featuring Welsh culinary delights will be served in the Main Hall.

Musical entertainment from the Aber Valley Male Voice Choir will cap off an enjoyable evening.

A purchased ticket will be required.
Day 3 - Wednesday, 22 April

8.00 – 17.00 Registration Open

Posters will be on display from 10.00–13.00. Poster presentations will take place during coffee breaks

S19 Improving the Evidence Base and Transparency of Food Safety Decision Making
Assembly Room
Organizers and Chairs: Sarah Cahill and Andrijana Rajic

8.30 Knowledge Synthesis and Transfer in Support of Evidence-informed Food Safety Decision Making
ANDRIJANA RAJIC, Food and Agriculture Organization of the United Nations, Rome, Italy

IAN YOUNG, Food Safety and Quality Unit, Food and Agriculture Organization of the United Nations, Rome, Italy

9.10 Towards Transparent and Practical Risk-based Applications in Food Safety
MICHAEL BATZ, University of Florida, Gainesville, FL, USA

S20 Integrative Aspects in Food Safety Risk Assessment
8.30 – 10.00
Ferrier Hall
Organizer: Eelco Franz
Chair: Norval Strachan

8.30 Food Safety Management Systems and Safety Governance
PIETERNEL LUNING, Klementina Kirezieva, Wageningen University, Wageningen, The Netherlands

9.00 System Biology, Heterogeneity of Stress Response and Microbial Risk Assessment of Bacterial Spore Formers
STANLEY BRUL, Wishwas Abhyankar, Rachna Pandey, Linli Zheng, Alex ter Beek, Jan P. M. Smelt, Norbert O. Vischer, Chris G. de Koster, Erik M.M. Munders, Molecular Biology and Microbial Food (SILS), University of Amsterdam, Amsterdam, The Netherlands

9.30 Operationalising Factors That Explain the Emergence of Infectious Diseases
NORVAL STRACHAN, University of Aberdeen, Aberdeen, United Kingdom

S21 Recent Advances in Food Packaging to Ensure Quality and Safety of Foods
Syndicate Room C
Organizer and Chair: Suresh Pillai

8.30 Advanced Active Packaging Concepts
SARA LIMBO, Department of Food Science and Microbiology of University of Milan, Milan, Italy

9.00 Advanced Modified Atmosphere Packaging Concepts
ALAN CAMPBELL, Campden BRI, Chipping Campden, United Kingdom

9.30 Grafting and Crosslinking Polymers for Enhanced Packaging Purposes
SHIMA SHAYANFAR, Texas A&M University, College Station, TX, USA; National Center for Electron Beam Research, College Station, TX, USA

T7 Technical Session 7 – Produce and Sanitation
Syndicate Room D
Chair: Lone Jespersen

8.30 The Hygienic Design of Food Industry Brushware
DEBRA SMITH, Vikain, Swindon, United Kingdom

8.45 Potentiation of Disinfection Effect on Listeria monocytogenes Biofilms by Chlorine Dioxide and Hypochlorite in Rinsing Water
SOLVEIG LANGSRUD, Trond Møretrø, Even Heir, Nofima, Norwegian Institute of Food, Fishery and Aquaculture, Ås, Norway

9.00 The Seek and Destroy Process: Listeria monocytogenes Process Controls in the Ready-to-Eat (RTE) Meat and Poultry Industry
JOHN BUTTS, Martin Wiedmann, Thomas Malley, Food Safety By Design, LLC, Saint John, IN, USA

9.15 Growth, Colonisation and Internalisation of VTEC in Fresh Produce: The Potential Impact on Food Security
BERNHARD MERGET, Norval Strachan, Ken Forbes, Fiona Brennan, Nicola Holden, University of Aberdeen, Aberdeen, United Kingdom

9.30 Elucidating the Physiology of Attached and Internalized Salmonella Cells in Leafy Vegetables
NIKOLAOS GRIVOKOSTOPOULOS, Ifigeneia Makariti, Nikolin Hilaj, Zoi Apostolidou, Panagiotis Skandamis, Agricultural University of Athens, Athens, Greece

9.45 Microbiological Hazard Investigation and Evaluation of Dutch Fresh Produce Growers
JENNIFER BANACH, Joop van der Roest, H.J.(Ine) Fels-Klerx, RIKILT, Wageningen, The Netherlands

10.00 – 10.30 Coffee Break in the Exhibit Hall

S22 A Benchmarking Study of the Traceability Regulatory Environment in 21 OECD Countries
Assembly Room
Organizer: Tejas Bhatt
Chair: Brian Sterling

10.30 Global Food Traceability — Will It be Regulation or Collaboration?
BRIAN STERLING, Global Food Traceability Center, Washington, D.C., USA

11.00 Differences in Regulatory Environments around the World: Embrace or Eliminate?
BRIAN STERLING, Global Food Traceability Center, Washington, D.C., USA; COLINE DONON, GSI Global, Brussels, Belgium
11.30  Best Practices for Achieving Harmonization Using Standards
COLINE DONON, GS1 Global, Brussels, Belgium

S23  Non-destructive In Situ Techniques to Monitor Bacterial Colony Dynamics in Solid (Model) Foods
Ferrier Hall
Organizers: Jan Van Impe and Estefanía Noriega Fernández
Chair: Jan Van Impe

10.30  Exploring Bacterial Colonies in Solid (Model) Foods
ESTEFANÍA NORIEGA FERNÁNDEZ, KU Leuven, Leuven, Belgium

11.00  Use of Image Analysis as a Non-invasive Tool to Model Microbial Communities on Solid Surfaces
PANAGIOTIS SKANDAMIS, Agricultural University of Athens, Athens, Greece

11.30  Microscale Description of the Food Characteristics Surrounding Bacterial Cells: Relevance to Describe the Variability of the Individual Cell Behaviour
VALÉRIE STAHL, Bernard Hezard, Adrienne Lintz, Rachel Ferrier, Jean Christophe Augustin, Aérial Technical Institute for Food Industry, Illkirch, France

T8  Technical Session 8 – Laboratory and Detection Methods
Syndicate Room D
Chair: Daniele Sohier

10.30  Keeping up with Molecular Analysis — Smooth Transitions from Research to Risk Assessment
HELEN WITHERS, Ministry of Primary Industries, Wellington, New Zealand

CRISTINA PABLOS, Javier Marugán, Rafael van Grieken, Sandra Cristobal, Universidad Rey Juan Carlos, Móstoles, Spain

11.00  ISO/TS 13136 for STEC Detection: Method Performances Assessment to Go for Accreditation
DANIELE SOHIER, Maryse Rannou, Nadine Henaff, ADRIA, Quimper, France

11.15  Simultaneous Direct Detection of Shiga-toxin Producing Escherichia coli (STEC) Strains by Gold Nanoparticle Optical Sensing
IRWIN QUINTELA, Benildo de los Reyes, Chih-Sheng Lin, VIVIAN CHI-HUA WU, University of Maine, Orono, ME, USA

S24  Food Safety Decisions – Tools and Tips for Food Producers of Ready-to-Eat Foods
Syndicate Room C
Organizer: Taran Skjerdal
Chair: Catherine Halbert

10.30  The STARTEC Tool and Guidelines for Food Producers — Ingredient and Technology Choices for Food Safety, Quality and Nutrition
TARAN SKJERDAL, Norwegian Veterinary Institute, Oslo, Norway

11.00  Definition of Safety Criteria in RTE Products: A Whole Chain Approach from Ingredient Selection up to Consumption
ALESSANDRA DE CESARE, Alma Mater Studiorum, University of Bologna, Bologna, Italy

11.20  “A Day in the Life”— A Food Company Shares Its Wisdom
CECILIE FROM, Matbørsen, Stokke, Norway

11.35  Developing Tools for Food Safety Decision-making — Challenges and Recommendations, MATTHIAS FILTER, BfR, Berlin, Germany

12.00 – 13.00  Networking Lunch in the Exhibit Hall

PL2  Plenary Session
Assembly Room

13.00  Listeria monocytogenes: Recent U.S. Outbreaks and Implications for Control in No Growth Foods
DONALD ZINK, U.S. Food and Drug Administration—CFSAN, College Park, MD, USA

13.30  Campylobacter and Listeria — How Do We Control These Key Pathogens in Food Processing Establishments?
JOHN HOLAH, Holchem Laboratories Ltd., Bury, United Kingdom

14.00  BRC Food Issue 7 Changes in Response to Horsemeat
DAVID BRACKSTON, BRC Global Standards, London, United Kingdom

14.30  Problems of Future Data Interpretation for Food Microbiology: And the Answer is 42!
ROY BETTS, Campden BRI, Gloucestershire, United Kingdom

14.50  IAFP Poster and Technical Awards and Announcement on IAFP’s 12th European Symposium on Food Safety
DONALD ZINK, IAFP President

15.15  Adjourn

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PLENARY SPEAKERS

Abram Aertsen
KU Leuven, Belgium
Abram Aertsen obtained his Ph.D. at the Faculty of Bioscience Engineering of KU Leuven in 2004, after which he became a postdoctoral fellow. After postdoctoral stays in the groups of Prof. Laurence Van Melderen (ULB, Belgium) and Prof. François Taddei (INSERM U1001, France), he returned to the KU Leuven in 2009 to become a Principal Investigator in the Department of Microbial and Molecular Systems, where his research group focuses on the mechanistic dissection of stress-perception, -response and -adaptation phenomena and strategies in cells and populations of foodborne pathogens.

Ana Allende
TU Delft, The Netherlands
Dr. Ana Allende from CEBAS-CSIC (Spanish National Research Council) is a Senior Researcher with focus on safety of fresh produce. She obtained her Ph.D. in Food Science and Technology at the University of Cartagena, (Spain). During her scientific career she has conducted numerous postdoctoral positions in different International Research Centers including Ghent University (Belgium), USDA (USA) and PARC-Summerland (Canada). She has published multiple research papers in SCI journals about pre- and post-harvest risk factors affecting the microbial ecology and safety of fresh produce. She has built up more than fifteen years of scientific research and management experience by guiding research projects in the area of microbial safety of fresh produce. Currently, she is a member of working groups of the EFSA BIOHAZ panel.

Alejandro Amézquita
Unilever, United Kingdom
Dr. Alejandro Amézquita is a Science Leader in microbiological safety at the Safety & Environmental Assurance Centre of Unilever (based in the UK). He has a broad knowledge of risk assessment, predictive microbiology, and food safety management systems at an international level, gained from working for over 15 years in industry and academia. Dr. Amézquita has published numerous research papers in peer-reviewed scientific journals, and has presented and lectured extensively in his areas of expertise. In addition to his responsibilities at Unilever, he holds adjunct faculty appointments at the University of Nebraska-Lincoln and North Carolina State University, and actively contributes to professional societies and scientific organisations, and as a reviewer of key scientific journals. Dr. Amézquita received his BSc in food engineering from La Salle University in his home country (Colombia), and his MSc (food science & technology) and Ph.D. (biological systems engineering) from the University of Nebraska-Lincoln in the U.S.

Diah C. Aryani
WU, The Netherlands
Diah Chandra Aryani was born on 2 March 1980. She obtained her bachelor’s degree in food technology from Bogor Agricultural University. Later on she earned her MSc degree in food safety from Wageningen University, in which currently she is working towards a Ph.D. in food safety and preservation project.

John Bassett
John Bassett Consulting Ltd., United Kingdom
John Bassett has over 15 years’ experience of risk assessment and risk management, in both industry and government roles, and is a skilled communicator on risk and food safety. A veterinarian by training, he brings a “farm to fork” perspective on food safety challenges. He currently runs his own consultancy, John Bassett Consulting Ltd., which works with clients in the commercial food industry as well as governments and inter-governmental agencies. John is particularly passionate about using risk assessment approaches to drive business cost savings and create competitive advantage, through a better understanding of product and process risks, and optimal risk management. He is a past member of UK governmental advisory committees, the Advisory Committee for the Microbiological Safety of Food (ACMSF), and the Spongiform Encephalopathy Advisory Committee (SEAC); a member of the Royal College of Veterinary Surgeons; and a fellow of the Institute of Food Science and Technology (IFST).

Michael Batz
University of Florida, USA
Michael Batz is head of food safety programs for the Emerging Pathogens Institute (EPI) at the University of Florida. He has been conducting quantitative policy analysis for over 15 years. His research focuses on risk ranking and prioritization, foodborne illness source attribution, foodborne disease burden, and decision making under uncertainty. Mr. Batz leads EPI involvement in the Florida Integrated Food Safety Center of Excellence, one of five such Centers nationally, is executive director of the Food Safety Research Consortium, and consults for the U.S. Food and Drug Administration (FDA) and the U.N. Food and Agricultural Organization (FAO) on foodborne illness attribution and risk prioritization.
Roy Betts  
Campden BRI, United Kingdom

Roy Betts is Head of Microbiology at Campden BRI, and 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 CampdenBRI the 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 a number ILSI Europe Microbiological Risk Assessment Task Force, the UK Food and Drink Federation Food Hygiene Sub Committee and the UK Advisory Committee on the Microbiological Safety of Foods.

David Brackston  
BRC Global Standards, United Kingdom

David Brackston joined the British Retail Consortium (BRC) in October 2007 and has overall technical responsibility for the BRC schemes and BRC compliance activities. He has recently chaired the working groups developing the Agents and Brokers Standard and managed the rewrite of the BRC Global Standard for Food Safety issue 7. Prior to this he had worked for a Certification Body originally as an auditor before becoming the Quality and Processing Director with responsibility for all post farm gate certification schemes including BRC, IFS and ISO 22000. Having worked both in manufacturing with Associated British Foods and then for more than 10 years in retail as Technical Manager and Marketing Manager for ASDA (Walmart) he has a wealth of experience of the management of food safety throughout the supply chain.

Stanley Brul  
Molecular Biology and Microbial Food (SILS), University of Amsterdam, The Netherlands

Stanley Brul (1964) was trained as Biochemist and graduated “cum laude” in 1986. In 1991 he obtained a Ph.D. with a doctoral thesis entitled “Biochemical and Genetic Aspects of Peroxisome Biogenesis in Mammalian Cells.” He then started in 1990 as a post-doctoral fellow at Nijmegen University, did a short term fellowship supported stay at Rockefeller University and went in 1994 to work for Unilever Research and Development. From 1994–1999 he fulfilled at Unilever several scientific and managerial positions from project to program manager. In addition he received training in teaching, project and program management as well as general management and finance from the London Business School, the Lausanne Business School and INSEAD, France. In 1999 Stanley was appointed professor of Industrial Microbiology on an endowed chair at the Swammerdam Institute for Life Sciences of the University of Amsterdam while staying 4 days appointed at Unilever Research & Development as senior scientist novel (micro) biological technologies in the Science area Food Processing. As of 2003 on he acted as coordinator of the master’s program in Medical Biochemistry of the University. Since end 2007 Stanley Brul is fully employed by the University. Concomitantly he was appointed director of the bachelor’s program in Bio-medical Sciences at the University and received training in managing University professionals. Stanley teaches courses in molecular microbiology, biochemistry, nutrition and human molecular physiology.

Sophie Butot  
Nestlé Research Centre, Switzerland

Sophie Butot obtained her Master’s degree in food and environmental Microbiology at the University of Bourgogne and AgroSup in France. Since 2003, she works as a scientist at Nestlé Research Center in Lausanne, Switzerland. Her field of expertise comprises development and performance evaluation of molecular methods for detection of food- and waterborne viruses and assessment of the effect food processing technologies on virus survival. She is a member of the working group “Enteric viruses in food” of the European Committee for Standardization (CEN/TC 275/WG6/TAG4) and she actively contributes by being the bottled water matrix expert.

Michael Callanan  
Nestlé Research Center, Switzerland

Dr. Michael Callanan obtained his BSc and Ph.D. from the Microbiology Department at University College Cork. He spent the first 10 years of his professional career working on food fermentations and probiotic microorganisms in New Zealand, the U.S. and Ireland. He shifted focus 6 years ago to controlling microbial growth in food with natural antimicrobials at the R&D division of Irish food ingredients company, Glanbia, and has continued in this area since joining the Nestlé Research Center in Lausanne in 2010.

Alan Campbell  
Campden BRI, United Kingdom

Alan Campbell is Food/Packaging Technologist covering food manufacturing and packaging related areas within Campden BRI.

Alan trained in Food Technology at Grimsby College of Technology before working as a Development Technologist and QA Manager in the food industry. This was later followed by a technical role with a metal packaging company before joining Campden BRI in 1988.

During his time at Campden BRI Alan has been involved in a wide range of packaging and food processing related projects and consultancy. Additionally, Alan is involved in numerous Campden BRI training courses lecturing on food technology, packaging and the prevention of foreign matter in foods. During his time at Campden BRI Alan has written a number of articles and book chapters on food packaging related topics including modified atmosphere packaging and also acts as an expert witness when required.
Alan has been a member of the BRC/IoP Technical Advisory Committee for the BRC/IoP Packaging Standard since its beginnings in 1998. Alan has undertaken an extensive number of audits of food safety based systems such as BRC and HACCP as well as packaging sites (BRC/IoP). The audits have been undertaken in the UK, rest of Europe and internationally (Africa, SE Asia (including China) and South & North America). The majority of audits outside Europe have been in factories manufacturing canned foods.

**Frédéric Carlin**  
INRA, France

Frédéric Carlin is Research Director at INRA, the French National Institute for Agricultural Research, in the “Safety and Quality of Fresh and Processed Produce” laboratory in Avignon (Provence, South of France), in association with the University of Avignon. He is leading scientist in a research group of 13 permanent staff and 8 Ph.D. students and PostDocs dealing with Microbial food safety and the control of spore-forming bacteria in the food chain. The diversity within the foodborne pathogen Bacillus cereus is a major research topic in his group. Dr. Carlin is also associate editor of Food Microbiology and member of the editorial board of International Journal of Food Microbiology.

**Rachel Chalmers**  
Public Health Wales Microbiology, United Kingdom

Rachel Chalmers is Director of the national Cryptosporidium Reference Unit for England and Wales providing the national service for the investigation and management of Cryptosporidium and cryptosporidiosis. Her background includes food technology and the microbiology and transmission of zoonotic, foodborne and waterborne infectious diseases. Current research includes the molecular and sero-epidemiology of cryptosporidiosis, detection of gastrointestinal protozoa in clinical, food, water and environmental samples, assessing risks from different sources, and investigating the long-term health effects of infection. Rachel has co-authored over 100 scientific publications, and was made Honorary Professor in Swansea Medical School in 2013.

**Nigel Cook**  
The Food and Environment Research Agency, United Kingdom

Nigel Cook is a graduate of the University of Dundee. After postdoctoral research in the Universities of Aberdeen and Leicester he moved to the Central Science Laboratory (now the Food and Environment Research Agency [FERA]) at the Food Science Laboratory, Torry, Aberdeen in September 1994, before relocating to new facilities in York. At FERA, he studies the transmission of pathogens, particularly enteric viruses, through foods and the environment. He has a Visiting Professorship at the Katholieke Universiteit Leuven in Belgium. He is Councillor of the International Association for Food and Environmental Virology. He is a project leader within the standardisation working group ISO TC34 SC9 WG6, currently developing a standard for detection of Cryptosporidium and Giardia on berry fruits and leafy green vegetables. He was Coordinator of the European Framework 7 project “Integrated monitoring and control of foodborne viruses in European food supply chains (VITAL),” and Chair of COST Action 929 “A European Network for Environmental and Food Virology” from 2006 to 2010. Between 2009 and 2014, he was a member of various European Food Safety Authority’s Working Groups preparing opinions on the risk of foodborne viruses, and represented the European Communities on the Codex Committee on Food Hygiene Working Group developing Guidelines on the Application of General Principles of Food Hygiene to the Control of Viruses in Food. He was a member of the UK Advisory Committee on the Microbiological Safety of Food’s Viral Infections Subgroup. He was the founding Editor of the journal Food and Environmental Virology, published by Springer Publishing Company.

**Paul Cook**  
Food Standards Agency, UK, England

Following research in environmental and food microbiology Paul Cook joined the Department of Health in 1993 where he managed the Department’s research and surveillance programme on microbiological food safety. Since 2000 Paul has worked for the UK Food Standards Agency where he is Head of Microbiological Risk Assessment Branch. The branch is closely involved in managing research on foodborne disease, the assessment of microbiological food hazards and incidents, antimicrobial resistance and providing the secretariat for the Advisory Committee on the Microbiological Safety of Food (ACMSF). Paul has been an invited expert to various groups including EU Scientific Committees and WHO/FAO consultations.

**Marie-Laure Dardé**  
University of Limoges, France

Professor Marie-Laure Dardé qualified in medicine in 1982 and completed her Doctorate in 1990 at the University of Limoges. She was awarded her Chair at the University of Limoges in 1992 where she is currently Head of the Department of Parasitology-Mycology. Marie-Laure is a member of the French Agency for Food Safety ANSES Expert Panel on Toxoplasmosis and also of the EFSA (European Food Safety Agency) Expert Panel on Toxoplasma and food. She is also an Expert for the European Centre for Disease Control (ECDC) and was President of the French Society of Parasitology (2008–2011). She currently manages the international collection of Toxoplasma strains within the Biological Resource Centre in France and has been involved in a number of major international collaborations including participation in the EU-funded EUROTOXO prevention project and in the European Cost Action on Apicomplexa. Marie-Laure is currently conducting an international study of the association between atypical Toxoplasma strains and severe forms of toxoplasmosis in Africa and French Guiana including investigation of Toxoplasma strains in wild and domestic animals and water.
Alessandra De Cesare  
**Alma Mater Studiorum, University of Bologna, Italy**  
Alessandra De Cesare is Research Assistant in the Laboratory of Food Safety at the Department of Agricultural and Food Sciences, University of Bologna in Italy. She obtained her MD in Molecular Microbiology, her Ph.D. in Food Science and is currently enabled as Assistant Professor in Food Inspection and Hygiene. She participated in different EU research projects, like STARTEC, and has been deputy Coordinator in the BASELINE project on “Selection and improving of fit-for-purpose sampling procedures for specific foods and risks.” Her main current research topics are bacteria genotyping, metagenomic investigations of chicken guts, definition of food safety criteria for foodborne pathogens.

Coline Donon  
**GS1 Global, Belgium**  
Coline Donon is GS1’s Public Policy Manager for product safety and traceability. In her role, she coordinates globally the messages GS1 is providing to international and regional organisations. Before that, she worked at Havas Worldwide as Senior Public Affairs Consultant. Her missions included advising major healthcare and FMCG companies on consumer protection issues at EU level. Prior to this experience, she held the position of European Public Affairs Manager at Carrefour. She was in charge of representing the Group in Brussels on a number of issues including consumer protection and food. Coline studied European law and European relations in Paris.

Seda Erdem  
**Department of Economic and Behavioural Science Centre, University of Stirling, United Kingdom**  
Seda Erdem is a lecturer in the Department of Economics at the University of Stirling and a member of Stirling Behavioural Science Centre. Seda’s main areas of research broadly fall into public health economics, food safety and food choice, and behavioural economics. More specifically, she is interested in consumer choice behaviour, decision-making and eliciting preferences in the fields of food and health. Seda has involved in various research projects focusing on consumers’ food choice, perception of risks, trust in institutions, perceived responsibility for food safety, and in the development of decision-making tools for effective reduction of foodborne health risks. She is a member of a number of associations and part of the editorial board of a number of journals.

Matthias Filter  
**BfR, Germany**  
Dr. Matthias Filter has a diploma in biochemistry. He has experience in bioinformatics, cheminformatics, software development, data mining and QMRA modeling. He has more than 10 years of experience as a project manager in public and private sector organizations. Dr. Filter’s current position is senior research scientist, unit “Epidemiology and Zoonoses,” Federal Institute for Risk Assessment (BfR), Germany. His specific interest is development of community resources to increase food safety and security.

Anthony Flood  
**International Food Information Council, USA**  
As Director of Food Safety Communications at the International Food Information Council (IFIC), Tony Flood has worked for over 13 years developing a number of food safety education and outreach programs. Additionally, he directs the development and continuation of risk / crisis communication programs among academic, government and industry stakeholders on emerging food safety and defense topics. Tony is a graduate of James Madison University, Harrisonburg, VA, where he received a BS degree in Communications.

Tony is an active member of the International Association for Food Protection (IAFP), the Institute of Food Technologists (IFT), The Conference for Food Protection (CFP) and the National Center for Food Defense (NCFPD) risk communication core team.

Eelco Franz  
**RIVM – Centre for Infectious Disease Control, The Netherlands**  
Eelco Franz holds Master’s in Biology in the field of population biology at the University Utrecht, The Netherlands. His Ph.D. thesis was on “Ecology and risk assessment of *E. coli* O157 and *Salmonella* Typhimurium in the primary production of lettuce” at the Wageningen University, The Netherlands. After obtaining his Ph.D. he had a three year post-doc at the RIKILT – Institute for Food Safety (Wageningen, The Netherlands) University) where he did mainly modelling work on food safety risks regarding heavy metals, mycotoxins and pathogens. From 2010 he works at the Dutch Institute for Public Health and the Environment (RIVM), Centre for Infectious Disease Control. His main interest is he aims at integrating molecular/genomic, physiological, and epidemiological data for in order to get an enhanced understanding on the ecology and risks of foodborne microbial hazards.

Cecilie From  
**Matbørsen, Norway**  
Cecilie From is a veterinarian holding a Ph.D. degree in food safety from the Norwegian University of Life Sciences, Faculty of Veterinary Medicine and Biosciences. The focus area of her research has been toxin production in *Bacillus* spp. outside the *B. cereus* group and reduction/elimination of spore-forming bacteria in lightly heat treated foods. She also had the reference function of the Nordic Committee on Food Analysis, method no. 67: *Bacillus cereus* (2010). She has long experience working as a consultant for the Norwegian food industry in matters concerning food safety and quality issues. From 2012 Cecilie has been the quality manager of Matbørsen AS, a producer of ready-to-eat food to grocery stores and the HORECA sector in Norway owned by Norgesgruppen – Norway’s largest trading enterprise.
John Holah  
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/GHP’s 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 SME’s 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 on the hygienic design of food factories and food processing equipment; factory services and water systems; maintenance; cleaning and disinfection; personnel 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 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.

Paul in’t Veld
VWA Netherlands, 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. Obtained his Ph.D. in 1998 on the topic: The development and evaluation of microbiological reference material for food microbiology. Worked, from 1987 to 1999, at the National Institute of Public Health and the Environment (RIVM). Since 1999 he is 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 convener of ISO TC 24/SC9/WG3: method validation), to coordinate method development activities and support the organisation with microbiological advice on methods. Besides this he is technical assessor for various accreditation bodies and was until 2014 a member of AOAC RI Board of Directors.

Elisabeth Innes
Moredun Research Institute, Scotland

Elisabeth Innes has an honours degree in Immunology from the University of Glasgow and a Ph.D. in Tropical Animal Health from the University of Edinburgh. She has conducted research in the area of infectious diseases of livestock at several different research institutes in UK and Africa and currently leads a group at Moredun looking at developing solutions to control diseases caused by protozoan parasites. She has published over 170 scientific articles and has Honorary Professorships at Heriot Watt, Edinburgh and Glasgow Universities. She was recently awarded an MBE for scientific research and science communication. Elisabeth has a range of research interests including:

- Host-pathogen interactions in livestock species infected with Neospora caninum, Toxoplasma gondii and Cryptosporidium spp.
- Understanding protective immune responses against intracellular pathogens and developing novel vaccine targets
- Understanding disease risk posed by congenitally infected animals and the immune response to the endogenous infection
- Transmission routes of zoonotic pathogens, Toxoplasma gondii and Cryptosporidium spp.
- Risk to public health from environmental contamination with Zoonotic protozoa.

Liesbeth Jacxsens
Faculty of Bioscience Engineering, Department of Food Safety and Food Quality, Ghent University, Belgium

Liesbeth Jacxsens is Dr. assistant on quality assurance and risk analysis for the laboratory of food microbiology and food preservation (LFMFP) (prof Mieke Uyttendaele) and NutriFoodChem (prof Bruno De Meulenaer) of the Department of Food Safety and Food Quality of Ghent University. At this moment she is involved in the research of the EU 7th FP project Veg-i-Trade (www.vegitrade.org), where she is investigating the performance of food safety management systems and level of good agricultural practices along the horticultural chain in diverse regions in the world. Microbiological risk assessment studies are currently virusess in raspberry chain, enteric pathogens on strawberry, basil and leafy greens (www.foodscience.ugent.be/LFMFP). The expertise on chemical risk assessment research is concentrated on linking mould growth on agricultural raw materials and foodstuffs and mycotoxins production currently, Alternaria mycotoxins on tomato chain (www.nutrifoodchem.ugent.be). In the Veg-i-Trade project she is also training manager and responsible for trainings and capacity building in ICP countries. She was involved in the ITP food safety, quality assurance and risk analysis which is organized by UGent-VLIR (www.itpfoodsafety.ugent.be).

Lone Jespersen
Maple Leaf Foods, Canada

Lone Jespersen has been with Maple Leaf Foods since 2004 initially as Six Sigma Black Belt and later Director, Six Sigma. In 2009, Lone took responsible for the execution of the Maple Leaf Foods food safety strategy and recently took on leadership of all operations learning. Prior to that, Lone worked for Woodbridge Foam as Engineering and Operations manager responsible for the safety and quality of automobile safety products. Lone holds a Master’s in Mechanical Engineering from Syd Dansk University, Denmark, a Master’s of Food Science from the University of Guelph, and presently pursuing her Ph.D. with Dr. Mansel Griffiths and Dr. Carol Wallace at the University of Guelph. Lone’s passion for food safety culture has led to the development of her food safety culture measurement system “Cultivate Food Safety” which she is currently refining with North American and European food manufactures. Lone was the 2014 recipient of the Mary Edmunds Williams Scholarships in support of her food safety culture research.
Kostas Koutsoumanis
Aristotle University of Thessaloniki, Greece
Kostas Koutsoumanis is currently serving as an Associate Professor at the Department of Food Science and Technology, Faculty of Agriculture, Aristotle University of Thessaloniki, Greece. He received his B.S. degree in Agriculture Engineering from the Agricultural University of Athens, Greece, in 1997 and Ph.D. (Food Science) degree from the same University in 2000. After serving as a Research Associate in the Department of Animal Sciences at Colorado State University he took a lecturer position in the Department of Food Science and Technology at Aristotle University of Thessaloniki in 2002, and he was promoted to Assistant Professor in 2007 and Associate Professor in 2013. Currently, he teaches several graduate and MSc courses including General Microbiology, Food Quality and Safety Assurance, Predictive Microbiology and Risk Assessment and Applied Statistics in Food Science. From 2011 he is a member of Biohazard panel of the European Food Safety Authority (EFSA). He is member of the editorial boards of the Journal of Food Protection, International Journal of Food Microbiology, Food Microbiology and Current Opinion in Food Science. As a principal investigator or co-investigator, Kostas Koutsoumanis has received over 1.5 million euros in grants, contracts or donations for research in the field of microbiological quality and safety of foods. Recent research efforts have centered on the microbiological quality and safety of fresh and processed food products, predictive microbiology, microbial risk assessment, stochastic modeling approaches in food safety and quality, development and application of Time Temperature Indicators (TTI) for monitoring food quality and safety, etc. The research results have been presented and published at 65 refereed scientific journal articles, 7 book chapters, and more than 100 papers in conference proceedings with more than 2500 citations and h-index=30.

Alec Kyriakides
Sainsbury’s Supermarkets Ltd., United Kingdom
Alec Kyriakides is Head of Product Quality, Safety & Supplier Performance at Sainsbury’s where he has worked for 22 years. Prior to Sainsbury’s, he worked in the manufacturing industry including the dairy and brewing sectors. His responsibilities include the management of specialist teams collectively responsible for the safety and quality management framework for Sainsbury’s. He is a member of a number of industry and government committees and sat on the government Advisory Committee on the Microbiological Safety of Food (ACMSF) for 10 years. Alec is the author of books on the microbiological safety of foods including Campylobacter, Salmonella, E. coli and Clostridium botulinum.

Dan Li
Ghent University, Belgium
Dan Li currently works as Postdoctoral Researcher at Ghent University, Belgium. Ph.D. obtained at Ghent University, Belgium in 2012. Research interests mainly on the application of RT-qPCR detection method for Noroviruses (NoVs) in fresh produce and sea foods, the prediction of NoV infectivity based on the viral integrity, the exploration of novel strategies to inactivate NoV in practical scenarios as well as the associated in-depth mechanisms, and the interactions of human NoVs with intestinal and external environment.

Sara Limbo
Department of Food, Environmental and Nutritional Sciences of the University of Milan, Italy
Sara Limbo is Assistant Professor at the Department of Food, Environmental and Nutritional Sciences of the University of Milan. The scientific activities deal with: a) the optimization of traditional and active packaging strategies to extend the shelf life of foods; b) the interactions between foodstuffs and packaging materials, in terms of migration of additives and contaminants; c) the modulation of physic-mechanical and barrier properties of polymers as function of fresh food requirements. Sara Limbo teaches Food Packaging Technology at the University of Milan. She is member of the Ph.D. School in Research and Innovation Technology for Agro-Food and Environmental Sciences at the University of Milan.

Pieterlun Luning
Wageningen University, The Netherlands
Dr. Pieterlun Luning, Wageningen University Dr. Pieterlun Luning graduated from Wageningen University in Food Technology, worked as post-MSc graduate, followed project manager function at Agrotechnological Research Institute where she did her Ph.D. in flavour research. She worked as post-doc for Unilever Vlaardingen, and worked several years at TNO Research and Nutrition Institute as product Manager “Innovative Packaging.” In 2000, she started as lecturer at Wageningen University, designed the 2-years interdisciplinary MSc Food Quality Management, and is since 2006 associate professor. She participated/is in national (PROFETAS on novel protein foods, Sensory Specific Satiation), and European projects (PathogenCombat, Veg-I-trade). Recently, she co-developed a fraud vulnerability assessment tool on behalf of SSAFE. Current research concentrates on QMS assessment tools, system dynamics modelling, risk-based auditing, food safety culture, food waste, product design dynamics, by applying a techno-managerial research approach. She supervises 12-15 Ph.D. students, teaches interdisciplinary courses, and supervises 15-20 MSc thesis students/year. She is author/editor of various books (Food Quality Management; Safety in agri-food chains), and published > 70 articles; h-index 20.

Marie-Josée Mangen
University Medical Center Utrecht, The Netherlands
Marie-Josée J. Mangen is an economist with special interest in public health, infectious diseases and food safety. She holds an MSc in animal science from the University of Bonn, Germany, and in Agricultural Economics and Marketing from Wageningen University, The Netherlands. She obtained her Ph.D. at Wageningen University, The Netherlands in 2002. She then moved for a short-term project to the Livestock Information and Policy Branch at the Food and Agricultural Organization in Rome, Italy. From 2003 to 2004 she held a post-doc at the Agricultural Economic Research Institute and at the National Institute for Public Health and the Environment (RIVM) in The Netherlands where she participated as an economist on the CARMA (Campylobacter Risk Management and Assessment) project. She then worked at RIVM on various economic evaluations in the field of public health and infectious diseases. Since June 2008 she has an appointment as assistant professor at the University Medical Centre Utrecht, Julius Center for Health Science and Primary Care within the Health Technology Assessment group.
Pete Martin  
**NSF, United Kingdom**  
Peter Martin has over 20 years of experience in the food law industry. Beginning in Trading Standards, he gained experience enforcing the whole range of consumer protection law and spending time in teams responsible for product safety, metrology and food safety and labelling. He has carried out routine inspections of retail and production premises, investigating consumer complaints and producing prosecution reports where required. In 1995, Peter joined a consultancy, Law Laboratories Ltd. Concentrating on food related law, he was involved in advising on food safety, labelling and auditing of food production sites in the UK and Europe. He has also spent time on non-food product related work, including product labelling and auditing of production sites. Throughout this time he has helped design and present courses on all aspects of consumer law and its' enforcement. Additionally, Peter worked independently as a trading standards consultant and more specifically in the areas of product labelling, trading standards challenges and provision of trading standards related courses. In 2009, Peter joined the consultancy NSF-CMi as Trading Law Manager looking after a team covering all aspects of Trading Law. In this role he has overseen the 5-a-Day scheme in its current format and managed a team of eight providing high quality trading law advice to clients, advising food retail and non-food retail clients on legal requirements, approving food/non-food specifications for retail sale, assessing advertising, marketing copy for legality, advising on a European basis and ensuring continuing high standards of advice and service.  

Peter is a qualified ISO Lead Assessor and also holds a Master's Degree in Food Law.

Peter McClure  
**Mondeléz International, United Kingdom**  
Peter McClure gained his BSc and Ph.D. from Cardiff University and then joined the Institute of Food Research in 1985, in the UK, to work in the areas predictive modelling and microbiological food safety. He worked for Unilever for over 20 years and recently joined Mondeléz International as the section manager for Microbiology and Food Safety for Europe, Middle East and Africa. 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, is a co-editor of *Foodborne Pathogens*, and is a visiting professor at Leeds University.

Jeanne-Marie Membré  
**INRA, 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 past (2003–2009), Dr. Membre worked at the Safety & Environmental Assurance Centre of Unilever, developing microbiological models in an industrial context. J.-M. Membre is a member of IAFP, of the scientific board of *Journal of Food Protection* and *International Journal of Food Microbiology*.

Sara Mortimore  
**Land O’ Lakes, Inc., USA**  
Sara Mortimore, is a Food Scientist with around 30 years of practical experience. She started her career with Glaxo SmithKline, working as a Research Technologist and then moved to a division of Croda International where she again worked in R&D before transitioning into Quality Assurance. In 1989 she joined Grand Metropolitan Foods which later became Pillsbury and subsequently was incorporated by General Mills Inc. She stayed there for close on 19 years moving through a series of global assignments all in Food Safety and Quality, working with brands such as Haagen Dasz, Green Giant, Old El Paso and many regional products. Sara joined Land O’Lakes in 2008 and is currently Vice President of Product Safety, Quality and Regulatory Affairs with enterprise wide responsibility.

Estefanía Noriega Fernández  
**KU Leuven, Belgium**  
Dr. Estefanía Noriega Fernández holds a Ph.D. on Chemical Engineering (Enhancing microbiological food safety and shelf-life through an innovative sustainable approach that integrates decision-supporting tools for the management of emerging processing technologies) from the University of Oviedo (Spain). She has been appointed as a (LOU) Assistant Lecturer at the Department of Chemical Engineering and Environmental Technology (University of Oviedo) and as a JAE-DOC Post-doctoral researcher at the Institute of Food Science Research CIAL (Spanish Council for Scientific Research CSIC, Spain). She is currently working as a Post-doctoral researcher at the division of Chemical and Biochemical Process Technology and Control (BioTeC - KU Leuven, Belgium). Throughout her scientific career, she has performed several research stays, as Ph.D. and Post-doctoral visiting fellow, at leading international research centres (Loughborough University, UK; Institute of Food Research, UK; KU Leuven, Belgium; University of Reading, UK), where she actively participated in some EU-funded projects. She has co-authored 23 journal publications, holding the first order of authorship in 12 of them (20 papers at SCI-indexed journals; 112 citations), and 45 scientific contributions at national/international conferences. Over the period 2009-2014, she has co-supervised 2 Ph.D. and 15 MSc/Postgraduate thesis and she is currently involved in 5 Ph.D. and 9 MSc/Postgraduate thesis in the domain of predictive microbiology at BioTeC (KU Leuven). She is a scientific (daily) co-coordinator of projects/collaboration agreements with industry and international research centres, and a member of the Flemish Cluster Predictive Microbiology in Foods CPMF2 (www.cpmf2.be) and the KU Leuven excellence centre OPTEC (www.set.kuleuven.be/optec/). She has been awarded, in regime of competitive concurrence, with several research fellowships (e.g., FEMS research fellowship), best paper/presentation awards and positive assessment as University Lecturer from the National Agency for Quality Assessment and Accreditation of Spain (ANECA). She is appointed as a reviewer at several SCI-indexed journals and she has been a member of several MSc/Ph.D. examination committees.
George-John Nychas  
Agricultural University of Athens, Greece

George Nychas is Professor of Food Microbiology at the Agricultural University of Athens. In the last 25 years he has coordinated 4 and participated in more than 30 EU projects (budget >6M€), while his research interests focus on: a) the assessment of food safety and spoilage through microbiological and physicochemical analysis (metabolomics) in combination with advanced statistical methods, b) responses of stress adapted pathogens grown either planktonically or as biofilms, and c) modelling the behaviour of pathogenic bacteria for the assessment of food safety. He has published 171 papers in SCI journals & 30 book chapters as invited author (ca. 5600 citations, h = 41).

Cian O’Mahony  
Creme Global, Ireland

Cian O’Mahony is head of Expert Modelling and Statistics at Creme Global, a data-science company specializing in predictive modelling and software. His background is in pure and applied mathematics, holding a first class honors degree from University College Cork followed by postgraduate studies in applied mathematics and pharmacy, focusing on physiologically-based pharmacokinetic models for local anesthetics used in post-operative care and cosmetic products. Many of the models developed by his team at Creme Global are now built into a range of applications used by regulators, industry and academia.

Efstathios Panagou  
Agricultural University of Athens, Department of Food Science and Human Nutrition, Greece

Efstathios Panagou is Assistant Professor of Food Microbiology at the Department of Food Science and Technology of the Agricultural University of Athens. His research interests focus on (a) the application of rapid analytical techniques on the assessment of food spoilage in tandem with multivariate statistical methods and exploratory data analysis, (b) modelling the microbial responses in various food ecosystems, and (c) microbial ecology of plant-derived fermented foods. He has participated in 15 research projects and he has published 68 original research papers in scientific journals and 6 book chapters (770 citations, h-index 15).

Mike Peck  
Institute of Food Research, United Kingdom

Professor Mike Peck was appointed a Research Leader at the Institute of Food Research in 1992. His research is focused on basic and strategic aspects of the physiology and molecular biology of Clostridium botulinum. He spends much time applying the findings of his research to deliver social and economic impact (for example, he works with industry and regulators to deliver safe minimally heated refrigerated foods). He holds Professorships in Applied Bacteriology at the University of Nottingham and Biological Sciences at the University of East Anglia, and has more than 150 refereed publications and book chapters.

Sylvia Pfaff  
FIS Europe, Germany

Dr. Sylvia Pfaff studied food chemistry in Hamburg. She has worked as a consultant for the food industry since 1996. Her main work experiences are food safety and quality management, allergen management, hygiene and packaging. Dr. Pfaff was in all research projects directly involved. She has intense experience with innovation projects on national and European levels; close networks to food companies, traders and science. At FIS Europe she works on several projects concerning integrated management systems for the food business where consumer issues are important. She organises seminars, workshops and focus groups regularly. She was involved in the EU research projects SPAS, Consumer Choice and EuroPrevall.

Hans-Dieter Philipowski  
ENFIT e.V., Germany

Hans-Dieter Philipowski graduated from HAW – University of Science, Hambourg with a Master’s Degree in Process Engineering. His career focus has been developing international standards for cleaning and safety, developing training for operators, food safety, food compliance and allergen traceability. He is the founder and owner of PROTEC-International, ACT-International and Philipowski-International. President and founder of the international Association ENFIT – International Federation for the promotion of innovative Technologies for Cleaning, Logistic Management and Service for Bulk, Transport and Storage tanks. Member of the Board University of Science Hambourg.

János G. Pitter  
Syreon Research Institute, Hungary

János G. Pitter graduated from the Semmelweis Medical University as a Medical Doctor (1999), and after 5 years in cellular physiology research he received his Ph.D. scientific degree in basic medicine. He completed a Pharmacoconomics and Pharmaceutical Policy postgraduate program at Eötvös Loránd University in 2012. He worked for 10 years in the pharmaceutical industry with experience in clinical development and regulatory correspondence, and was the head of a department responsible for project management and product differentiation for 3 years. From March 2014 he works at Syreon Research Institute, a public health and health economics oriented private research institute, as principal researcher.
Andrijana Rajic  
**Food and Agriculture Organization of the United Nations, Italy**

Andrijana Rajic works in Food Safety Programme of FAO-UN, Rome, Italy. She is also an Adjunct Professor at the University of Guelph and University of Saskatchewan, Canada. She has >20 years of professional experience on the interface of animal health, food safety and public health in Canada and Europe, and internationally. Andrijana co-advised two post-doctoral fellows, several MSc or Ph.D. students in epidemiology, and consulted many students / professionals with multi-disciplinary backgrounds. Her passions are journalism, music (live concerts) and history. To learn more about Andrijana’s presentation please visit: http://www.uoguelph.ca/omafra_partnership/ktt/en/worktogether/Handbook.asp

Kalliopi Rantsiou  
**DISA-University of Turin, Italy**

1993–1997: BS in Biology, University of Athens, Greece; 1997–2002: Ph.D. in Food Science, University of California, Davis (USA); 2002–2004: Research fellow at the Faculty of Agriculture, University of Udine, Italy; 2004–2005: Scientific expert on microbial risks in food, Hellenic Food Authority, Greece; 2006–2008: Research fellow and from 2008 to date: Assistant professor, at the Department of Agricultural, Forest and Food Sciences, University of Turin, Italy. Her research interests include molecular biology of foodborne pathogens and the use of culture independent methods to study the microbiota of foods. She is co-author of more than 70 papers. She is a member of the editorial board of the International Journal of Food Microbiology and ad hoc reviewer for major food microbiology scientific journals.

Lucy Robertson  
**Norwegian University of Life Sciences, Norway**

Lucy Robertson heads the Parasitology Laboratory at the Norwegian University of Life Sciences. She has been involved in research on intestinal parasites for over 25 years, and is particularly interested in zoonotic parasites, food and waterborne transmission, global and other drivers for transmission, and effects of parasitic infection on host nutrition and other physiological factors. She lectures to both medical and veterinary students, and is an advocate of the One Health concept.

Shima Shayanfar  
**National Center for Electron Beam Research, USA**

Shima Shayanfar is in the Nutrition and Food Science Department Texas A&M University. She has extensive expertise in the food industry and project management in USA, Europe and the Middle East in the areas of non-thermal processing and food packaging. She is currently pursuing her Ph.D. in Food Science and Technology on eBeam processing and molecular analysis of spoilage organism, pathogens and packaging polymers. She co-edited the book titled, *Electron Beam Pasteurization and Complementary Food Processing Technologies* that was recently published by Elsevier.

Panagiotis Skandamis  
**Agricultural University of Athens, Greece**

Dr. Panagiotis Skandamis holds a Ph.D. in Food Microbiology and has worked as a post-doctoral fellow in the Department of Animal Science of Colorado State University in USA. He is currently working as Associate Professor in the Agricultural University of Athens, teaching Food Microbiology, Predictive Modelling, Food Hygiene and Quality Control of Foods. Dr. Skandamis has authored 98 original research papers in journals of SCI and 26 book chapters. His research is funded by 5th, 6th and 7th EU Framework Program, National Grants and direct contracts with the Greek Food Industry in microbial food safety and predictive microbiology. He is Associate Editor in Food Research International and member of the Editorial Board of *Journal of Food Protection, Applied Environmental Microbiology* and *International Journal of Food Microbiology*. He is also the developer of GroPIN, a Predictive Modelling Software tool, which simulates over 400 kinetic and probabilistic models for pathogens and spoilage organisms under static and dynamic food processing and storage parameters.

Taran Skjerdal  
**Norwegian Veterinary Institute, Norway**

Researcher at the Norwegian Veterinary Institute. Coordinator of the EU project STARTEC, and involved in many other projects. Ph.D. from the Norwegian Technical University, Biotechnology in 1996. Has worked as researcher within microbiology and food in SINTEF, NOFIMA and in the company DNV.

Danièle Sohier  
**ADRIA, France**

Danièle Sohier is now heading the Food Safety & Quality unit in ADRIA (www.adria.fr), a Food Technology Institute qualified by the French Ministry of Agriculture. The team is involved in food microbiology R&D activities and expertise, and coordinates national and European innovation programs in close collaboration with food and diagnosis industrial partners. In relation with the oral communications planed at the IAFP-Eu meeting, ADRIA is one of the most important expert lab, which provides validation studies of alternative methods. Danièle participates to the AFNOR and MicroVal Certification committees. She is also involved in the ISO working group dealing with the ISO 16140 standard revision, as well as in the AOAC International Stakeholder Panel on Alternative Methods. ADRIA is as well managing the SPORE-RISK Joined Technology Unit, which deals with food safety and spoilage risks linked to spore-forming bacteria. Indeed, Danièle is the convener of the ISO working group on “Guidelines for conducting challenge-tests.” To learn more, please see her linkedin profile.
Valérie Stahl  
*Aerial Technical Institute for Food Industry, France*

Valérie Stahl is Project Manager in the field of microbiological quality and safety of food. She is working in Aerial (67 France), a Technology Resource Centre and a Technical Institute for Food Industry, member of ACTIA (F) www.aerial-crt.com. Its goal is to anticipate industrial needs and to propose innovative solutions to industry. Its research and development studies are conducted in partnership with companies, research centres and universities. Aerial has highly contributed to the development of new methodologies such as challenge tests, food micro-environment methods and is an active member of the Sym’Previus Groupment, performing predictive microbiology tools (www.symprevius.org). Aerial is the coordinator with Actalia of the national technological network RMT Actia QUALIMA.

Brian Sterling  
*Global Food Traceability Center, USA*

Brian Sterling is Managing Director for the Global Food Traceability Center (GFTC), which is a program within the Institute of Food Technologists (IFT) in Washington, D.C. In this capacity, he provides leadership for operations, business development activities, and projects. He is a partner and President of SCS Consulting, a management consulting firm near Toronto, Canada. He was previously the Chief Executive Officer for Ontario’s own agri-food traceability corporation (OnTrace), and since 2003 has been an outspoken advocate and successful implementer of food traceability solutions. Prior to OnTrace, he held senior roles in several consulting firms. He was Director of Business Development for RFID and Product traceability at IBM Canada during 2005 and 2006; and before joining IBM, he was Vice President/General Manager at RCM Technologies, responsible for their Canadian operations. His notable highlights in the area of food traceability include leading a seafood research program focusing on the impact of traceability on business performance, food waste, and consumer perceptions; a traceability gap analysis project for Ontario’s chicken producers and processors that provided recommendations for improving industry performance; providing leadership for development of a national livestock identification and registration system in Canada; as well as developed a business case decision support tool for Can-Trace which was used by smaller enterprises to understand the return on investment from traceability. He has also served as a member of Agriculture Canada/Department of Fisheries & Oceans industry-government committee for traceability with the Seafood Value Chain Roundtable. He was the founding co-chair of Canada’s Industry-Government Advisory Committee (IGAC) for the National Agriculture and Food Traceability System (NAFTS). Mr. Sterling has given numerous speeches and presentations concerning food traceability at international conferences and summits since 2004.

Norval Strachan  
*University of Aberdeen, United Kingdom*

Norval Strachan was trained as a Physicist and graduated with honours in Natural Philosophy 1986. In 1991 he obtained a Ph.D. in Engineering whilst working at the Ministry of Agriculture, Fisheries and Food. His research then moved into the area of rapid detection of foodborne pathogens and their toxins and this work carried on when he moved to the Robert Gordon University in 1996. In 1998 he joined the University of Aberdeen where his research focussed on risk assessment including dose response modelling, epidemiology, molecular epidemiology and mathematical modelling of infectious diseases (specifically Campylobacter, *E. coli* O157, *Salmonella* and *Listeria*). In 2012 he was appointed full professor and currently teaches in the School of Natural and Computing Sciences and carries out research in the Institute of Biological Sciences.

Helen Taylor  
*Cardiff Metropolitan University, United Kingdom*

Helen Taylor graduated with a BSc (Hons) Applied Biological Sciences. As an active member of the Institute of Food Science and Technology (IFST) she is registered as a Food Safety Manager, Professional Food Auditor & Mentor and HACCP Trainer. Helen is a qualified packaging Technologist (Dip. Pack.Tech) specialising in Food Packaging solutions for SME’s. She is the Operations & Technical Manager of the Food Industry Centre (FIC) at Cardiff Metropolitan University and has been an integral member of the team since 2006. Helen has responsibility for links with industry and the FIC including the management of specific technical projects for the food industry and the operational implementation of the Centre’s Knowledge Transfer programme (KITE).

Clément Trunet  
*Université de Brest, France*

Clément Trunet is Ph.D. student at the University Laboratory of Biodiversity and Microbial Ecology (LUBEM) of the University of Brest and at ADRIA Development, Quimper, France. He obtained an engineer diploma in microbiology and food safety (ESIAB) and a master’s degree in fundamental and applied microbiology of the University of Brest. The aim of his Ph.D. work is to describe and model the recovery behavior of heat treated *Bacillus* sp. spores regarding the conditions of sporulation, heat treatment and recovery.

Purnendu C. 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. 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) and the IAFP (2010).
Maria Vitale  
Istituto Zooprofiliattico Sperimentale della Sicilia, Italy  
Maria Vitale received her Molecular Biology Ph.D. degree at University of Palermo. Since the year 2000 Head of Laboratory of Genetics of Microorganisms at the Experimental Zooprofiliactic Institute in Palermo dealing in major foodborne bacterial and parasitic pathogens and in animal TSE. Involved in several educational training on molecular epidemiology and questionnaire administration on food safety aspects. Participating in scientific colloquia and several meeting at the European Food Safety Authority. Authors of several publications on molecular Biology applied to animal and zoonotic diseases, food microbiology and food safety. Involved in educational events for food microbiology and in questionnaire administrations for nutrition and food safety.

Jaap Wagenaar  
Utrecht University, The Netherlands  
Jaap Wagenaar is expert in the field of microbiological food safety and zoonoses. He is active for 20 years in microbiology research. He was trained as veterinarian and he performed his Ph.D. study at Utrecht University and at the USDA-National Animal Diseases Center, Ames, IA, U.S. In 1996 he started his research group at the Institute for Animal Science and Health in Lelystad, The Netherlands, on food safety and in particular on Campylobacter. Starting in 2000, Jaap became active in WHO-Global Foodborne Infections Network (WHO-GFN, formerly WHO-GSS), a WHO food safety program. Within that program he is member of the Steering Committee and he acts as trainer in international training courses. He is director of the WHO Collaborating Center for Campylobacter and the OIE-reference laboratory for Campylobacter. He worked with WHO (Headquarters, Geneva, Switzerland, and for the Tsunami-relief operations to WHO Indonesia), the Centers for Disease Control and Prevention (Atlanta, U.S.) and the USDA Western Regional Research Center (Albany, Ca, US).

From 2006, Jaap has been appointed as chair in Clinical Infectious Diseases at the Faculty of Veterinary Medicine, Utrecht University. His research group at the Vet School is focussing on Campylobacter and antimicrobial resistance. He is currently coordinator of a large EU-project on antimicrobial resistance (EFFORT). He is member of the WHO-AGISAR-group (Advisory Group on Integrated Surveillance of Antimicrobial Resistance). He is member of the scientific panel of The Netherlands Veterinary Medicines Authority (SDa).

Steve Wearne  
Food Standards Agency, Wales  
Steve Wearne was appointed Director of Food Safety in July 2013, the role becoming Director of Policy two months later. Previous to this he was Director of FSA Wales.

Steve is a scientist by training, with a degree in biochemistry and a spell of postgraduate research in molecular biology. Steve joined the Ministry of Agriculture, Fisheries and Food in 1990 and held a range of posts in food science and food policy development, before transferring to the Agency when it was launched in 2000. His first post in the Agency was head of the Private Office and Private Secretary to the first Chair of the FSA, Sir John Krebs (now Lord Krebs).

Marjon Wells-Bennik  
NIZO Food Research and Top Institute Food and Nutrition, The Netherlands  
Marjon Wells-Bennik is Principal Scientist Food Safety at NIZO food research, where she manages diverse projects relating to contaminants in foods. Furthermore, she is project leader of a large project on heat resistant bacterial spores at the Top Institute Food and Nutrition TIFN, a public-private centre of excellence in The Netherlands. She obtained her Ph.D. in Food Microbiology at the Wageningen University in The Netherlands and gained experience in molecular microbiology as a postdoc at Harvard University (Boston). Before starting at NIZO in 2004, she worked as a Scientist at TIFN, supervising Ph.D. projects on Listeria monocytogenes and Bacillus cereus and worked at the Institute of Food Research (UK) on Clostridium botulinum.

Anett Winkler  
Mondelēz International, Germany  
Anett Winkler joined Kraft Jacobs Suchard in December 1998 to head up the research microbiology laboratory in Munich. She developed the internal Microbiological Methods Manual before moving towards manufacturing and Research & Development microbiological support in all Kraft Foods categories (Cheese, Coffee, Chocolate, Beverages). Later on Anett concentrated on chocolate, biscuits and other low moisture foods including supplier developments and approvals. She also consolidated the scientific basis for microbiological process controls in low moisture foods by performing validation studies for nut and cocoa processing. Later on she took over responsibility for Microbiology in the Eastern European, Middle East & African Region. In her current role Anett is designing food safety programs, rolling out global training modules related to food safety and further supporting supplier development. Before moving to industry Anett had worked in Basic Research at the Max-Planck-Institute of Psychiatry in Munich, where she established the first Molecular Biology laboratory. Before that time she worked on different projects in Germany and USA looking at molecular and biochemical changes during drug withdrawal and studying relapse mechanisms. She has studied Microbiology in Germany, but completing her Master’s degree at the Lomonossov State University in Moscow about mutations in bacteriophages. After that she returned to Germany where she obtained her Ph.D. on work about stress responses in Bacillus subtilis.
Ian Young
Laboratory for Foodborne Zoonoses, Public Health Agency of Canada, Canada

Ian Young is currently a Food Safety Consultant with the Food Safety and Quality Unit of the Food and Agriculture Organization of the United Nations (FAO). He obtained a BASc in Public Health and Safety from Ryerson University in 2007 and a Ph.D. in Epidemiology from the University of Guelph in 2010. From 2011–2014, he worked as a Post-Doctoral Fellow and an Epidemiologist with Laboratory for Foodborne Zoonoses, Public Health Agency of Canada, on the application of knowledge synthesis, transfer and exchange methods to support evidence-informed decision-making and risk analysis in the food safety sector.

Donald Zink
U.S. Food and Drug Administration-CFSAN, USA

Dr. Donald L. Zink received a Bachelor of Science degree from Abilene Christian University. He earned an M.S. degree in Microbiology and a Ph.D. in Biochemistry and Biophysics from Texas A&M University. Between 1978 and 1983, he held faculty positions at Texas A&M University’s College of Veterinary Medicine and at the University of Arizona in the Department of Microbiology and the Department of Food Science. He joined Campbell Soup Company in 1983 as Manager of Process Microbiology where he worked in the area of refrigerated food safety and aseptic processing. In 1990, he joined Nestlé, where he held various positions in Quality Assurance for the Carnation Company and later served as Director of Food Safety for Nestlé USA. In 2000, he joined a new beef processing venture company, Future Beef Operations, as Vice President of Research and Development and Product Safety. In 2002, he joined the U.S. Food & Drug Administration’s Center for Food Safety and Applied Nutrition where he now serves as Senior Science Advisor for CFSAN in the Office of the Center Director.

Dr. Zink has served as a member of several advisory committees including the Committee on Program and Technical Review of the U.S. Army Natick RDEC for the National Research Council and the National Advisory Committee on Microbiological Criteria for Foods. He has been a member of various industry committees during his years with the food industry, including the ILSI North America Food Microbiology Committee and the Grocery Manufacturers of America Science and Regulatory Affairs Committee and Microbiological Safety Committee. Dr. Zink is the current President of the International Association for Food Protection.

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 professor in Food Microbiology at Wageningen University. Marcel is editor of the International Journal of Food Microbiology and member of the International Commission on Microbiological Specifications for Foods (ICMSF). Prof. Dr. Ir. M.H. Zwietering Professor in Food Microbiology Wageningen University personal page: http://www.wageningenur.nl/en/Persons/Marcel-Zwietering.htm laboratory: http://www.fhm.wur.nl.
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Symposia Abstracts
Integration of Omic Data into Microbiological Risk Assessment

The last decades in Food Safety Microbiology has been characterised by the development of new approaches in the domain of quantitative microbiological risk assessment (QMRA) and a revolution in the production of molecular data through the exploitation of high throughput omics. With the first we get more and more insight in needs and effectiveness of food safety control. With the second we get more much detailed insight in the behaviour and characteristics of microorganisms and in their ecology.

The next challenge scientists have to face is to find a way to integrate these domains. It is however not always easy and straightforward to reach this objective. This symposium will focus on examples of where it is obvious and where it is difficult to integrate results from omics and QMRA.

Seizing the Potential of “Omics” to Explore the Behavior of Foodborne Pathogens

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Understanding foodborne pathogen behavior and proposing biological models to describe the molecular and cellular mechanisms underlying virulence are the challenges that food microbiologists are currently tackling through the use of a set of analytical tools that are collectively called “omics” technologies. The type of information we obtain by using such tools is not different with respect to the past; nonetheless it is nowadays possible to achieve large sets of data that speed up our knowledge acquisition. In order to predict the physiological state and the response of pathogenic microorganisms under various food-related conditions it is necessary to integrate the “omics” results. The ultimate goal is a rational design of prevention and intervention measures to reduce the risk of foodborne illness.

Transcriptomics refers to the study of the RNA molecules synthesized by a specific microorganism, either grown in pure culture or within a mixed microbial consortium. Since transcription is one important point of control of gene expression, the transcriptome is a good predictor of the phenotype of a microorganism. The transcriptome can provide information at molecular level that could explain strain variability observed for different foodborne pathogens, especially for what concerns their ecology and virulence. Finally, by studying the transcriptome under different environmental conditions, it is possible to identify biomarkers related to specific characteristics of interest such as resistance, biofilm formation, virulence. Such biomarkers could be incorporated into quantitative microbial risk assessment.

Metagenomics and QMRA: Is There a Match?

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Next-generation-sequencing (NGS) has the potential of becoming the gold standard typing method in the near future. The potential of WGS as a technology that contributes to reducing the disease burden of foodborne infections depends on the success by which this type of data can be integrated in routine surveillance and applications such as microbial risk assessment and source attribution. The high-resolution nature of WGS enables us to develop a refined methodology for different steps in MRA including within species variation. However, using WGS data for microbial risk assessment requires a paradigm shift from applying the current MRA framework towards a methodology that is able to translate multidimensional genotypic data via reduced information on the phenotype to a single measure of risk (number of ill people). We have exploited the link between strain specific in vitro virulence with genotypic properties. This enables the identification of high-risk subgroups from a larger spectrum of strains. One step further is the recent increased interest in metagenomics, which enables simultaneous detection of all microorganisms in a clinical sample, without a priori knowledge of their identities, through the use of NGS. Where traditional microbial risk assessment relied on species/serotype abundance, qualitative risks in the context of metagenomics analysis should be determined on the basis of the level of evidence regarding the possibility that sequences identified in metagenomic analyses could be involved in adverse public health effects. There are a few fundamental issues with current metagenomics protocols that reduce their usefulness for accurate diagnostics and (quantitative) risk assessment, which include the genetic context (which gene in which bacterium) and quantification (concentration). The potential use of metagenomics to QMRA as well as the major hurdles and possible solutions will be addressed.

Omics and QMRA: Challenges and Promises of the Future

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In these last fifteen years, the significant progress in omics-based analyses has revealed new insights into responses of pathogens to their environment and their interaction with hosts. On the other hand, quantitative microbial risk assessment (QMRA) is an efficient approach that uses knowledge on microbial behavior to estimate, through predictive modelling of growth and inactivation and dose-response analysis, the potential risk that microbes represent to human health. Hence, the main challenge now is to integrate the massive amount of data generated by Omics technologies to improve QMRA. That seems possible as illustrated below.

Based on comparative genome analysis on a large number of strains, specific biomarkers of target pathogens could be identified to allow better hazard identification. This approach, with parallel development of low cost chip technology, could be used in diagnosis, but also for detection and/or identification of new emerging strains in foods. To deepen hazard characterization, transcriptomic and/or proteomic analyses coupled to in vitro co-culture of epithelial cells and bacteria, and the use of animal models for host-pathogen interactions studies, are valuable strategies for development of more accurate dose response models. In addition, transcriptomics, metabolomics and experimental investigations
using model systems could be profitably implemented to improve exposure assessment by linking phenotypes to cellular mechanisms underlying bacterial stress response and virulence. Ultimately, such ‘physiome’ approaches connecting omics and particularly metabolomics data, to physiological behavior of bacteria could enable construction of mechanistic models, integrating metabolic and regulatory networks to improve our understanding of the pathogen adaptation to its environment and its interaction with hosts.

Despite some limitations to integration of Omics data in QMRA, recent studies have offered promising perspectives in this field. Moreover, this challenging research topic will contribute to the proposal of efficient and innovative food safety strategies.

S2  Current Issues in HACCP Training: Making Systems More Effective

Food safety is as much a topic of debate now as when HACCP was first being developed, maybe even more so. HACCP remains a cornerstone of food safety management and in recent years there has been a surge in the demand for HACCP and food safety training. Unfortunately, although much training is being delivered by experienced trainers, there remains considerable variability in the standards of training available, given the lack of regulation or international standardization in this area. Many HACCP experts are skilled presenters yet few have a detailed education in learning theory and training skills whilst, on the other hand, training experts are rarely skilled HACCP practitioners. This situation can often leave HACCP trainees confused and with a superficial theoretical knowledge, which hampers development of effective HACCP systems. At the same time, the growth in HACCP and food safety management systems auditing and certification around the world has highlighted weaknesses in HACCP systems and often these ‘non-conformances’ relate to topics that should be covered in HACCP training. Clearly the messages are not getting through effectively.

This symposium aims to tackle these issues and provide real examples of effective HACCP education and training strategies in different food industry settings. Presenters are all both experienced HACCP practitioners and educators and will share their experiences and tips for successful HACCP training that will drive effective HACCP systems. With presenters representing the interests of HACCP training in industry sectors as diverse as farming, retail, and manufacturing large and small, as well as academic research and product sectors historically considered to be of low food safety risk, the breadth of application of effective HACCP training models will show that, whilst one size definitely does not fit all, ideas from different sectors can be used to strengthen HACCP training approaches throughout the food supply chain.

Embedding HACCP and Food Safety Culture through Organisational Training Approaches

SARA MORTIMORE, Land O’ Lakes, Inc., St. Paul, MN

HACCP is both widely used and widely debated. There is growing recognition that HACCP is perhaps not as effective as it could be and that this is related both to inadequate training and the lack of informed support at senior levels. This presentation will examine some of the approaches that have been used to more fully embed HACCP into a wider food safety culture. A North American company case study will be shared which shows how one company worked at improved awareness of food safety, and also leveraged that knowledge for continuous improvement of the HACCP and wider food safety program by making it a shared responsibility. Making food safety important at all levels in the organization and across all functions made a significant difference. HACCP was more effectively embraced as a tool by which to structure the food safety discussions and risk reducing decisions and strategies. It will be argued that there is complacency in some food producing organizations. Whilst the private food safety standards and scheme owners recognize that HACCP is one of the recurring gaps in compliance, the certification bodies continue to award certificates. Certified companies may ask themselves whether their HACCP systems were OK – the answer being yes. However, how many really take the time to ask whether they could be better? If they did, the answer is again almost certainly – yes.

Using HACCP Knowledge Metrics to Revamp Manufacturing HACCP Systems – A Case Study

LONE JESPERSEN, Maple Leaf Foods, Mississauga, ON, Canada

The multidisciplinary HACCP team is firmly embedded within the theory of HACCP and this collaborative approach is believed to provide a practical insight into how to institute this theory in practice. HACCP team members are normally selected for their operational skills and expertise rather than HACCP knowledge and, following selection, are usually trained in HACCP principle application. Often this training is done without any measurement that the required knowledge and skills have been acquired by the trainees. However, HACCP knowledge within manufacturing HACCP teams has been shown to be one of the factors linked to the effectiveness of HACCP plans developed by those HACCP teams.

Few organisations have used HACCP knowledge metrics in the continuous improvement of their HACCP systems. This presentation will outline how HACCP knowledge metrics have been used in one manufacturing organisation in the development and implementation of a major HACCP ‘refresh and revamp’ programme, both to understand training needs throughout the workforce and to measure whether training objectives have been met.

Challenging All Trainers – Can You Successfully Train HACCP Concepts to SMEs and Micro Manufacturers? A Case Study on the Institute of Food Science and Technology/ Safe and Local Supplier Approval – HACCP Training Initiative (UK)

HELEN TAYLOR, Cardiff Metropolitan University, Cardiff, United Kingdom

In 2010 it was clear to the IFST and SALSA operations that micro businesses developing their food quality and safety systems in the UK required further training and education in the application of HACCP. Many of the businesses had HACCP plans in place but they were wholly reliant on external support for the development of new HACCP plans and maintaining existing documentation. A team of experienced, like minded Food Safety practitioners set about developing practical interactive courses to address the knowledge deficit in the application of HACCP for the small manufacturer.
The informed development process created two separate HACCP training courses for UK businesses, at Level 1 HACCP Awareness and HACCP Understanding at Level 2. The learning outcomes for both courses would be achieved through practical application and feedback during interactive activities. The training ‘language’ was clearly defined to minimise jargon and technical acronyms. The training package was designed to be ‘trainer’ and ‘trainee’ friendly.

The personnel permitted to run these courses were carefully selected and independently assessed to deliver the ‘designer’ HACCP training packages. Thus, ensuring a consistent quality of delivery to ensure business and individuals expectations were met and optimising the impact of the courses.

The courses underwent a ‘consultative revision’ in 2014 working with the HACCP Trainer cohort in order to deliver an improved HACCP experience for all delegates in 2015. The success of the course has been monitored since its launch in 2012 with over 350 learners benefiting from the applied, practical workshops driving positive and practical application of HACCP into the UK micro business community.

S3 Cryptosporidium and Giardia: Food Safety Issues of Important Concern

Foodborne parasites cause a high disease burden in humans. In a recent WHO report (July 2014), Cryptosporidium and Giardia are named as parasites of major concern for food safety.

In this symposium proposal, three speakers with international expertise on foodborne parasites risks, and detection, in particular for these two protozoan parasites, will provide the audience with the latest information on Cryptosporidium and Giardia as a food safety issue.

Attendees will learn about the importance of food as a transmission vehicle for Cryptosporidium and Giardia. Approaches and challenges for investigating outbreaks of foodborne cryptosporidiosis will be addressed specifically. In addition, the forthcoming International Standard (ISO) methods for the detection of Cryptosporidium and Giardia in berry fruit and leafy green vegetables will be presented.

It is anticipated that attendees will be better equipped to address potential the risks posed by these parasites in food products after attending the symposium. They will have a greater understanding of the foods of greatest concern for the most important foodborne parasites, and how the parasites can be tested in their products.

Importance of Food as a Transmission Vehicle for Cryptosporidium and Giardia

LUCY ROBERTSON, Norwegian University of Life Sciences, Oslo, Norway

Water is well known as a transmission vehicle for both Cryptosporidium and Giardia, and many outbreaks have been reported in which water is considered to be the transmission vehicle. For food, however, a different picture seems to emerge – although there are many outbreaks of foodborne cryptosporidiosis there are relatively few reported outbreaks of foodborne giardiasis. In this presentation the relative importance of food as a transmission vehicle for both these parasites is discussed as well as drivers that may increase the importance of food in transmission of protozoan parasites, particularly Cryptosporidium and Giardia.

Standardisation of Methods to Detect Cryptosporidium and Giardia in Berry Fruit and Leafy Green Vegetables

NIGEL COOK, The Food and Environment Research Agency, York, United Kingdom

Fresh produce, as it is consumed with minimal preparation, is a potential vehicle of transmission of protozoan parasites, and Cryptosporidium oocysts and Giardia duodenalis cysts have been detected on produce in several countries. Practical and reliable standardized detection methods for monitoring foodstuffs will aid prevention of parasitic disease outbreaks associated with contaminated food. Methods to detect Cryptosporidium in lettuce and raspberries, and Giardia duodenalis in lettuce, were developed with a view to providing analytical tools suitable for routine adoption. They are based on four stages: 1) extraction of oocysts from the foodstuffs, 2) concentration of the extract and separation of the oocysts from food materials, 3) staining of the oocysts to allow their visualization, and 4) identification of oocysts by microscopy. Internal controls can be added to enhance the reliability of the interpretation of the results. The methods have been used in several studies in various countries to detect protozoan parasitic contamination of foodstuffs. The International Standards Organisation is leading the protocols towards formal standardisation. They are currently at the Draft International Standard (DIS) stage, with final publication expected later in 2015.

Challenges of Identifying and Investigating Outbreaks of Foodborne Cryptosporidiosis

RACHEL CHALMERS, Public Health Wales Microbiology, Swansea, United Kingdom

Cryptosporidium has been identified recently in a WHO report as a protozoan of major concern for food safety. Outbreaks demonstrate the importance of food as a transmission vehicle but there are challenges for their identification and investigation. Identifying an outbreak as being foodborne is important because this enables implementation of measures to restrict further spread or repetition.

Cryptosporidium has multiple sources (humans, farmed animals, wildlife) and is transmitted via the faecal-oral route either directly through contact with faeces or through the ingestion of the oocyst stage in contaminated water or food. Contamination of food may occur during production, processing or preparation; it may not be removed by subsequent washing nor be visible on inspection.

Although cryptosporidiosis outbreaks are often linked to drinking or recreational water, foodborne outbreaks are reported less frequently. The largest foodborne outbreak reported so far occurred in England and Scotland in May 2012, and has been linked epidemiologically to the consumption of fresh cut, pre-packaged salad leaves. Reporting and surveillance of microbiologically confirmed clinical cases, retrospective interviews for epidemiological descriptions and analyses, and environmental inspection of suspected food premises are important parts of the outbreak identification and investigation. The incubation period for Cryptosporidium is longer (days to weeks) than that of many other foodborne pathogens (hours to days), and microbiological investigation of implicated foods is difficult because material is often not available, having been either consumed or discarded by the time the outbreak is identified. Even if samples are available there is a lack of standard methods for testing for Cryptosporidium. Food traceability enabling timely trace-back of both domestic produce and imported food is important for successful and expedient outbreak investigations.

Food, and especially fresh produce, presents a risk for cryptosporidiosis and in outbreaks coordination between microbiological, epidemiological and food investigations are required for successful interventions.
Burden of Foodborne Diseases

The estimation of foodborne disease burden is subject to intensive research worldwide with the intention of ranking foodborne pathogens and food-pathogen pairs to guide foodborne disease related policy decisions. Instead, it also needs to carefully assess the feasibility, effectiveness, and cost of potential interventions, as well as their expected public health benefits. These considerations are also valid for broad resource allocation decisions and the planning of food safety programs.

Assessment of health burden, intervention feasibility, effectiveness, and costs allows the risk managers to conclude on risk management measures which reach their targets, are cost-effective, and are not over-restrictive. Ranking the pathogens according to their disease burden strongly depends on how the disease burden is measured. As an example, *Listeria monocytogenes* is responsible only for a negligible number of annual illness episodes as compared with other pathogens, but is ranked among the top three causes of foodborne disease related deaths.

The session aims to gather and discuss different approaches and practices in estimating the burden of foodborne diseases, and the consequences on food safety policy decisions.

Monetary and Adjusted Life-year Estimates of Foodborne Disease Burden in the United States

MICHAEL BATZ, University of Florida, Gainesville, FL

Foodborne diseases cause significant morbidity and mortality each year. In 2011, CDC estimated a mean of 48 million annual cases of foodborne disease in the United States, including 128,000 hospitalizations and 3,000 deaths. While disease incidence measures provide critical information for public health decision making, they do not allow for the direct comparison of diseases with different severities and incidence with a single measure, nor do they account for congenital illness or long-term health outcomes that may follow acute disease. A number of researchers have recently developed integrated measures of disease burden to quantify and directly compare the public health impacts of foodborne pathogens. These include estimates of the cost of illness in dollars, as well as in both Quality Adjusted Life Years (QALYs) and Disability Adjusted Life Years (DALYs). This presentation will provide an overview of available estimates and discuss both the methodological differences across studies and the implications for interpretation. The presentation will focus on disease burden estimates developed by the presenter and colleagues, namely the estimation and ranking of the cost-of-illness and QALY losses associated with 14 foodborne pathogens in the United States, and the attribution of these illnesses to food categories.

The Pathogen- and Incidence-based DALY Approach: A New Methodology for Estimating the Burden of Infectious Diseases in Europe

MARIE-JOSE MANGEN, University Medical Center Utrecht, Utrecht, Netherlands

In 2009, the European Centre for Disease Prevention and Control initiated the ‘Burden of Communicable Diseases in Europe (BCoDE)’ project to generate evidence-based and comparable burden-of-disease estimates of infectious diseases in Europe. The burden-of-disease metric used was the Disability-Adjusted Life Year (DALY), composed of years of life lost due to premature death (YLL) and due to disability (YLD). To better represent infectious diseases, a pathogen-based approach was used linking incident cases to sequelae through outcome trees. Health outcomes were included if an evidence-based causal relationship between infection and outcome was established. Life expectancy and disability weights were taken from the Global Burden of Disease Study and alternative studies. Disease progression parameters were based on literature. Country specific incidence was based on surveillance data corrected for underestimation. Non-typhoidal *Salmonella* spp. and *Campylobacter* spp. were used for illustration. Using the incidence- and pathogen-based DALY approach the total burden for *Salmonella* spp. and *Campylobacter* spp. was estimated at 730 DALY’s and at 1,780 DALY’s per year in the Netherlands (average of 2005–2007). Sequelae accounted for 56% and 82% of the total burden of *Salmonella* spp. and *Campylobacter* spp., respectively. The incidence- and pathogen-based DALY methodology allows in the case of infectious diseases a more comprehensive calculation of the disease burden as subsequent sequelae are fully taken into account. Not considering subsequent sequelae would strongly underestimate the burden of infectious diseases. Estimates can be used to support prioritisation and comparison of infectious diseases and other health conditions, both within a country and between countries.

Need for Reliable Disease Burden Estimates to Support Food Safety Decision Making: The Example of Human Campylobacteriosis in the European Union

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A cost-utility model on potential *Campylobacter* control measures in the EU-27 has recently been published. Model assumptions on disease burden and cost of illness were based on an EFSA scientific opinion, driven by data mostly from The Netherlands. The aim of our work was to gather country-specific cost of illness estimates for the EU-27, to generate a conservative European estimate of *Campylobacter* related disease burden per case, expressed in Quality Adjusted Life Years (QALY), and to assess the country-specific cost-effectiveness of control measure using the adapted model assumptions. RESULTS: 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. The adjusted country-specific cost of illness estimates ranged from 38 to 311 Euro/case (versus the assumed 267 Euro/case). Health burden due to acute gastroenteritis, Guillain-Barré syndrome, and reactive arthritis were estimated from published data as...
11.6, 5.3, and 2.3 QALY loss per 1000 human campylobacteriosis cases, respectively. Inflammatory bowel disease and irritable bowel syndrome were omitted, in line with a WHO opinion. The overall, discounted health burden per case was calculated as 0.0152 QALY loss (versus the assumed 0.0389 DALYs). The combined application of currently available control measures of low-medium consumer impact were found to generate both cost saving in the EU-27 and health benefits, even when using the adjusted, conservative model input parameters. Value based pricing of a control measure under development will also be presented in two EU countries.

**S5 Strain and Population Diversity; Implications for Food Safety and Food Spoilage**

It is a well-recognized problem within the field of food quality and safety assurance that microorganisms behave very diversely with respect to their robustness. Efficacy of control is challenged by diversity in robustness of strains belonging to same species, diversity within microbial populations, and the physiological state of micro-organisms. Food processing environments and food preservation measures can impose significant selection forces on foodborne microorganisms, and thereby possibly selecting for the most robust microorganisms. These robust survivors can cause food safety or food spoilage problems, or can become domestic flora in factories. This symposium will focus on robustness diversity of relevant food spoilage organisms (both bacteria and fungi) and foodborne pathogens. We will address these issues with leading experts in the field, discussing single-cell based approaches to investigate population heterogeneity, giving insight in mechanisms contributing to selection of persisters upon food processing, and the importance of quantification of microbial diversity to make prediction of microbial behaviour along the food chain more realistic.

**Strain Variability in Growth and Inactivation Parameters: Relevance for Pathogens and Spoilage Organisms**

DIAH C. ARYANI, WU, Wageningen, The Netherlands

The actual behaviour of microorganisms can differ from the prediction of microbial growth and inactivation models. Various factors might contribute to this difference, including strain variability and variability between experiments. Therefore, apart from quantifying the impact of strain variability on growth rate and thermal resistance, also biological variability was determined to prioritize their importance.

The impact of strain variability on the μmax was found to be comparable to biological variability as function of pH, a<sub>t</sub> temperature, and undissociated lactic acid concentration for *L. monocytogenes*. The integration of strain variability in the prediction of growth of *L. monocytogenes* caused easily a difference of about 2–3 log, depending on the conditions and type of food product, between the most and least robust strains.

In contrast to the results for growth rate, strain variability in thermal resistance of *L. monocytogenes* was found to be four times higher than biological variability. Strain variability in thermal resistance was also comparable to the effect of growth history, which was mainly determined by the effect of growth phase. Strain variability alone explained about ¾ of the variability in thermal resistance of *L. monocytogenes* found for a large variety of data in literature. Although with bias, the effect of both strain and growth history explained all the variability from literature. Strain variability in thermal resistance of a spoilage microorganism, *Lactobacillus plantarum*, was also found to be larger than biological variability. Thus, it may be suggested that strain variability is the most important aspect to be considered for designing an appropriate thermal process.

**Single Cell Variability: Relevance for Fungal Spoilage Processes**

KOSTAS KOUTSOUMANIS, Aristotle University of Thessaloniki, Thessaloniki, Greece

Most of the available studies on fungal behaviour in foods deal with monitoring growth of mycelia originated from a large number of spores. In practice however, the contamination of food with fungal spores is often associated with very low numbers. This presentation provides an overview of the recent studies on the variability of the germination time and mycelium growth kinetics of single fungal spores with examples for *Penicillium expansum* and *Aspergillus niger*. The methodology for taking into account single spore variability in modeling fungal behavior in foods is described. A large number of data on the germination and the lag time of mycelia originated from single spores at various storage temperatures is presented together with time-lapse microscopy videos on single spore germination and mycelium growth. In addition, the relation between germination and lag time of single spores is discussed. The importance of single spore variability is demonstrated through a quantitative risk assessment study of yogurt spoilage by *Aspergillus niger*. In this study a stochastic modelling approach was applied based on *Aspergillus niger* mycelium growth model taking into account the important sources of variability such as time-temperature conditions during the different stages of chill chain and individual spore lag time. By combining the output of the model with the mould prevalence, estimated by the industry using challenge tests, the probability distribution of the number of cups in which a visible mycelium of *A. niger* is formed at the time of consumption can be estimated. The risk assessment model which takes into account variability can lead to a more effective risk-based quality management of yogurt and support the decision making in yogurt production.

**Causes and Consequences of Heterogeneity in Stressed Populations of Foodborne Pathogens**

ABRAM AERTSEN, KU Leuven, Leuven, Belgium

Comprehensive insights into how foodborne pathogens exactly perceive, react, respond and adapt to the many stresses and selection pressures they encounter throughout the food production chain are elementary for a proper understanding of their resulting behavior and evolvability. In this context, the implementation of live cell biology approaches to study (stressed) bacterial populations with increased intra- and intercellular resolution is yielding an increasingly detailed view on the causes and consequences of population heterogeneity, which are likely to become indispensable in our efforts to mechanistically explain and better anticipate the mechanisms and strategies behind the inactivation, survival and resistance development of foodborne pathogens.

**S6 Virus Testing, Interpretation and What Do I Do with a Positive Result**

Enteric viruses, particularly human noroviruses (NoVs) are the most common cause of food borne disease, responsible for up to 50% of all outbreaks and cases per year worldwide. Viruses enter the food supply across the farm-to-fork chain by exposure to contaminated waters, surfaces, and/or human hands. A number of high profile outbreaks...
involved berries, including strawberries and raspberries have been reported. Recently, researchers from the EU have applied the ISO/CEN based-on method on batch testing of frozen raspberries. However, there is still large space for discussion and improvement for NoV detection methods especially in the virus recovery and the interpretation of the results.

The symposium will focus on batch testing of norovirus in berries and the impact of detection frequency, the removal of inhibitors from the food matrix and interpretation of real-time PCR result and Ct values. If a positive result is obtained, what would be the impact on the industry and what are the steps that would be taken.

**Recovery of Viruses from Food and Interpretation of Results**  
**SOPHIE BUTOT**, Nestlé Research Centre, Lausanne, Switzerland

Epidemiological evidence indicates that enteric viruses, in particular Norovirus (NoV) and hepatitis A virus (HAV), are the leading cause of foodborne illness in developed countries. NoV and HAV are mainly transmitted via the faecal-oral route, either by person-to-person transmission or by contaminated water or food.

Since NoV and HAV cannot be cultured, their detection in food can only be achieved through application of PCR-based techniques. The development of a reference detection method, which includes matrix specific sample preparation protocols, as proposed in ISO/TS 15216 is a milestone. However, these methods are relatively expensive and unable to discriminate infectious from non-infectious viruses which make the interpretation of positive results a challenge for the food industry.

Sampling for enteric human viruses in water and food should not necessarily follow bacterial sampling plans since important differences are evident, such as the low level of viral contamination, the inability to enrich viruses and the complexity and high cost of assays. As there is currently no specific mention of virus sampling in any of the available standards from international bodies, the uncertainty about the most relevant sampling plan to apply to big batches in the frame of virus testing is a concern for the food industry. These new challenges faced by the food industry to ensure the food safety will be discussed.

**Batch Testing for Noroviruses in Frozen Raspberries**  
**DAN LI**, Ghent University, Ghent, Belgium

Berries, in particular raspberries, have been associated with a number of norovirus (NoV) outbreaks. This study demonstrated a defined approach for the interpretation of NoV-RT-qPCR signals obtained and puts this in a context of batch sampling.

A total of 130 samples of frozen raspberries were collected from 26 batches in four companies. In two companies the samples consisted of bulk frozen raspberries serving as raw material for the production of raspberry puree. In two other companies, the samples consisted of bulk individually quick frozen (IQF) raspberries serving as raw material for the production of frozen fruit mixes. RT-qPCR detection were performed for GI and GII NoV in 2 × 10 g. In cases where positive signals were obtained, an attempt to sequence the amplicons was undertaken.

Six of 70 samples from two of the 14 batches of frozen raspberries serving raspberry puree production provided a NoV RT-qPCR signal confirmed by sequencing. All six positive samples showed NoV RT-qPCR signals above the limit of quantification of the RT-qPCR assay. The mean NoV level in 20 g of these raspberry samples was 4.3 log genomic copies NoV GI/20 g. For the IQF raspberries, one batch out of 12 tested NoV positive, but only 1 out of the 5 samples analyzed in this batch showed a positive RT-qPCR GI NoV signal confirmed by sequencing. The RT-qPCR signal was below the limit of quantification of the assay used (3.7 log genomic copies/20 g).

In conclusion, the presence of quantifiable amounts of NoV and the clustering of several positive samples in one batch can indicate sanitary problems, although the impact to public health is unknown. The added value of additional confirmation of positive RT-qPCR signals were also demonstrated.

**What If I Find a Positive and What Do I Do with It?**  
**NIGEL COOK**, The Food and Environment Research Agency, York, United Kingdom

The increased awareness of the potential for foods such as berry fruit and leafy green vegetables to become vehicles for the transmission of pathogenic viruses, and the availability of methods for analysis of foodstuffs for hepatitis A and Norovirus, may prompt debate on whether routine monitoring of fresh produce can/should be performed. An important facet for consideration will be the interpretation of results which indicate that virus contamination is present in a sample, and what action to take subsequently. Several questions arise: what is a positive result using RT-PCR; can an indication of infectivity be obtained; why should action be taken if infectivity of the detected virus cannot be proven? This presentation will discuss these questions and others, with suggested answers, The advantages and limitations of current detection methods will be reviewed, and data presented from in-depth analyses of fresh produce food supply chains. Recommendations for actions which can be taken when virus-positive results are obtained will be made, using the philosophy of integrated monitoring and control.

**S7 The U.S. Food Safety Modernization Act: What Does It Mean for Europe?**

With their new FSMA legislation, the U.S. Food and Drug Administration (FDA) is revising expectations of food and feed producers in countries exporting to the U.S., as well as domestically. Does this mean a major change for how European companies exporting or operating in the U.S. manage food safety?

This session will focus on the implications of FSMA for European companies, specifically the requirements and the likely impacts. There will be a U.S. perspective on key aspects of FSMA relevant to European companies, followed by European company views to share what their companies are doing, or intend to do, to meet the requirements. The symposium will address several topics including food safety plans and preventative controls, testing and monitoring requirements and foreign supplier verification.

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With their new FSMA legislation, the U.S. Food and Drug Administration (FDA) is revising expectations of food and feed producers in countries exporting to the U.S., as well as domestically. Does this mean a major change for how European companies exporting or operating in the U.S. manage food safety?

This session will focus on the implications of FSMA for European companies, specifically the requirements and the likely impacts. There will be a U.S. perspective on key aspects of FSMA relevant to European companies, followed by European company views to share what their companies are doing, or intend to do, to meet the requirements. The symposium will address several topics including food safety plans and preventative controls, testing and monitoring requirements and foreign supplier verification.
The Food Safety Modernization Act (FSMA) represents biggest overhaul of the food safety system in a century. It stresses the U.S. Congress mandated paradigm shift from reactionary to prevention based food safety system that focuses on preventing food safety problems from occurring in the first place. The FSMA applies to domestic as well as imported foods and include two rules designed to help prevent unsafe food from reaching U.S. consumers by requiring importers to comply with the Foreign supply verification Program (FSVP). Under the proposed FSVP regulations, importers would be required to verify that food exported to the U.S. is produced under premises that provide the same level of protection as food produced in the U.S. Another proposed rule deals with establishing a program for accreditation of third-party auditors to conduct food safety audits and issue certifications of foreign facilities and the human and animal foods they produce for import to the U.S. The proposed rules on FSVP and Accreditation of Third-Party Auditors work in concert with the Preventive Controls rule for human and animal food. The FDA issued supplemental proposed rule based on FDA outreach and public comments and revised certain provisions regarding the proposed requirements concerning compliance status review of food and foreign suppliers, hazard analysis, and supplier verification activities. This presentation is designed to discuss the FDA expectations for foreign suppliers of food exported to the US, especially the proposed FSVP rule.

Meeting FSMA Regulations – A European Perspective
PETE MARTIN, NSF, Coventry, United Kingdom

FSMA and Microbiological Testing
ROY BETTS, Campden BRI, Gloucestershire, United Kingdom

There is no doubt that the Food Safety Modernisation Act has made a number of strategic changes to the way food safety is viewed and dealt with in the USA. We constantly see articles, seminars and conferences dealing with FSMA related issues advertised in journals and web articles. What is less clear and much less well discussed is the impact of the Act on businesses that export foods into the USA. Such companies will need to carefully consider and in most cases comply with FSMA requirements. Where this involves microbiological testing of foods, our interest soon turns to what to test and how to test. There is no hiding from the fact that testing needs can be very different on both sides of the Atlantic, with very different views on what to test and how to test. This paper will explore such differences and what may be required for organisation exporting foods into the USA.

S8 Stochastic Approaches for an Enhanced Control of Spore Germination and Development in Food Products

Gram positive sporeforming bacteria are ubiquitous in the environment and exhibit a wide range of phenotypic and genotypic features leading to their natural prevalence in foodstuff. These aerobic and anaerobic microorganisms further have the particularity to form endospores, i.e., metabolically dormant structures with extreme resistance, which enable them to survive process and sanitation procedures yielding their persistence in food industries. Spore contamination may hamper product stability required for ready-to-eat or canned products mostly due to spore germination, outgrowth and the exponential vegetative cell development. These conditions may alter the product yielding either food safety and/or quality issues. In processed ready-to-eat food, spores are usually present at low concentrations thus vegetative growth is likely to initiate from just a few spores, the ones, only ones but ones that resist, which can survive process conditions and develop in food during shelf-life.

Several studies report that germination of individual spores within a population is highly variable and is further increased when exposed to adverse conditions, possibly encountered during food processing and storage. This session proposes to illustrate three complementary approaches to better understand germination and lag time variability in order to refine risk assessment associated to Bacillus species and Clostridium botulinum food contaminations. Different technologies and approaches will also be discussed in relation to improved product stability.

Stress Resistance and Mechanisms Governing Germination and Outgrowth Heterogeneity of Individual Bacillus subtilis Spores after a Given Preservation Treatment
STANLEY BRUL, Wishwas Abhyankar, Rachna Pandey, Linli Zheng, Alex Ter Beek, Jan P.P.M. Smelt, Sacha Stelder, Leo J. de Koning, Henk Dekker and Chris G. de Koster
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Bacterial spores are the sturdiest structures known that are known in the living world. They are reinforced by various layers of peptidoglycan and proteinaceous material. Thus analysis of such protein layers is of major interest in spore biology. Here we report on Mass-spectrometry-driven qualitative and quantitative proteomics methods to expand knowledge about both the actual composition and the amount of proteins in the various spore macromolecular layers. In addition we show in the model organisms Bacillus subtilis that specific proteins are cross-linked as well as individual domains within specific spore wall proteins. Enhanced spore thermal; resistance observed upon spore maturation was correlated to enhanced cross-linking. Interestingly, B. weihenstephanensis, a spore forming food pathogen known for its ability to sporulate, germinate, grow and produce toxins at lower temperatures than its close relatives is known to produce the spore with different characteristics when sporulating at lower temperature resulting in lower wet heat resistance of the resulting spores. Analysis of the proteome of spores of B. weihenstephanensis strain WSBC 10204 produced at either 12°C or 30°C by mass spectrometry provided insight into variations in the protein content of the spore coat at different sporulation temperatures. Interestingly, only minor differences were observed, corroborating that wet heat resistance does not depend on the identity of the proteins making up the coat layers. Instead, low temperature might affect the rate of spore maturation and thus spore coat protein cross-linking. The presentation will explore also heterogeneity in spore germination and outgrowth and discuss a putative link with spore stress resistance. For this a new live-imaging tool will be presented.
**Bacterial Spore Heterogeneity of Behavior Due to Sporulation and Recovery Conditions**

**CLEMENT TRUNET**1,2,3, Narjes Mtimet4, Anne-Gabrielle Mathot5, Florence Postolle6, Ivan Leguerrinel7, Olivier Couver7, Frédéric Carlin2 and Louis Coroller2

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(4) ADRIA UMT14.01 SPORE RISK, Quimper, France
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Spore-forming bacteria in food are a major cause of food spoilage or food poisoning, leading to economic burden. Even after a heat treatment, surviving spores could be able to recover. The recovery consists of different physiological stages: germination, outgrowth and the first cell multiplication. The sporulation conditions, heat treatment intensity and recovery conditions have an effect on spore recovery ability and recovery kinetics. Nevertheless, the physiological stage which is the most impacted by the recovery conditions and the heat treatment intensity remains unclear. To quantify the impact of sporulation condition, heat treatment intensity and recovery conditions on the different recovery stages, recovery kinetics were performed using flow cytometry. The transfer from a physiological stage to the next one was monitored thanks to size and fluorescence intensity of cells which are labeled with Syto9 (permeability of spores) or 5-Cyano-2,3-ditolyl tetrazolium chloride (CTC, respiratory activity). Sporulation and recovery conditions could highly impact the yield and time of germination. The heat treatment impact is more complex as spores can be altered by various ways. It could be put in evidence by using cell criteria as size, Syto9 and CTC fluorescence intensity. Flow cytometry enables the quantification of the impact of sporulation, heat treatment and recovery conditions on the different stages during spore recovery. It allows the analysis of a large number of events over time and to discriminate what stage(s) is (are) the most impacted by each condition.

**Stochasticity of Germination/Lag Time of Individual Spores of Clostridium botulinum and the Safety of Minimally Heated Refrigerated Food**

**MIKE PECK,** Martin Webb and Sandra Stringer, Institute of Food Research, Norwich, United Kingdom

*Clostridium botulinum* is an important foodborne pathogen that is responsible for botulism a severe and often fatal disease. For *C. botulinum* to present a problem, spores need to germinate followed by cell multiplication and neurotoxin formation. Quantifying the stochasticity of germination and lag time of individual spores of *C. botulinum* is an important part of understanding the hazard, and can make a valuable contribution to improved food safety. Knowledge of the underlying distribution would allow greater refinement of risk assessments. Recently, we have quantified the variability of spores during the different stages of lag phase and examined the relationships between these stages. The effect of heat treatment, incubation temperature, and sodium chloride concentration on growth from individual spores of *C. botulinum* has been measured. These studies demonstrated that spores within a single population are very heterogeneous with large variability in all stages of lag phase. The duration and variability of times for germination, outgrowth and first doubling depended on both the historic treatment of the spores and the prevailing growth conditions, and the stage of lag most affected was treatment dependent.

**S9 Risk Assessment of Unintentional Allergen Cross Contact**

The Session will cover the risk assessment of unintentional allergen cross contact among The Food Supply Chain. The risk of Transport Units will be addressed

**Management of Allergen Cross Contact in Food Transport Containers Such as Tanks**

**HANS-DIETER PHILIPOWSKI,** ENFIT e.V., Pinneberg, Germany

Millions of tons of food in reusable containers by road rail and sea are transported daily. The risks of cross-contamination with allergens, microorganisms, dioxins, detergents and other contaminants are larger than expected by the industry. Cleaning equipment are cleaned without cleaning standards and no real result. Always applies when transporting the goal of all must be as cheap as possible. The quality of the cleaning and disinfection is frequently completely ignored. The logistics industry has many years ago created a certificate (ECD European Cleaning Document) that no quality guarantees, but only what activities a cleaning system has executed. Regardless of which quality has the cleaning and disinfection.

The presentation points out the risks of the industry and consumers provides an outlook on possible solutions. The solutions in addition to the standardization of cleaning procedures include the traceability of transport containers, allergens and their products was transported therein. ENFIT is because the current state of development.

**Management of Allergen Cross Contact in the Food Industry – Acknowledgment of Reference Dose**

**SYLVIA PFAFF,** FIS Europe, Bad Bentheim, Germany

The management of unintentional added traces of allergens due to cross contacts is an ongoing process in the food industry. The challenge is to gather all needed information concerning allergen traces in raw materials, impacts from production lines and equipment to the correct labelling. A help is the use of the VITAL concept launched by the Allergen Bureau Australia.

VITAL is a standardised allergen risk assessment framework for food producers – developed by the Allergen Bureau in response to an industry call for a risk-based approach to labelling for allergen cross contact in manufactured foods. The VITAL framework provides for ongoing monitoring and verification of risk assessment processes to ensure any changes to the level of risk are recognised and acted upon without delay.

A component of the VITAL framework is an Excel-based spreadsheet known as the VITAL Calculator. The VITAL Calculator allows the assessment of likely sources of allergen cross contact from raw materials and the processing environment, plus an evaluation of the amount present and a review of the ability to reduce the allergenic material from all contributing sources. The VITAL Calculator also specifies a particular precautionary allergen statement to be used according to the level of cross contact identified.
The presentation will introduce reference doses, action levels and portion sizes which are the key to determine if a risk still exists for the allergic consumer. Additionally, the VITAL calculator will be used for a food example of the audience.

Management of Allergen Cross-contamination at Household Level – Best Practice for Food Labelling
HAZEL GOWLAND, Allergy Action, St Albans, United Kingdom

The information needs and behaviour of consumers with food allergies, intolerances and coeliac disease vary. This depends on a wide range of factors including the particular food(s) they need to avoid, the level which they may consider their ‘threshold’ and the perceived potential severity of any reaction. In addition, third party allergen avoidance is practised more widely, by family members and friends, carers, nursery, school and university staff, restaurateurs, caterers, hospital and prison staff and many others. They all need skills and experience to read labels and interpret written and oral information effectively.

Food allergens of relevance vary between populations and geographical areas. Food suppliers need to ensure that they meet the relevant legal labelling requirements for the country in which the food is sold, and also recognise and identify other food allergens not on the labelling list but of relevance to the particular population.

In this symposium presenters will discuss the effect of current legislation on the implementation of food testing methods, the development of standards for method validation, and some of the issues and problems involved in method validation and choosing the right method to use in testing laboratory.

Microbiological Test Methods – The Requirements and Influences of European Legislation on the Way We Test Foods
ROY BETTS, Campden BRI, Gloucestershire, United Kingdom

In Europe testing can be drastically influenced by legislation, particularly the requirements of European Commission Regulation 2073/2005. This regulation precisely specifies the methods that have to be used in testing for its Criteria, these usually being based on the use of ISO test methodologies. Whilst ISO Methods are accepted reference procedures, they can tend to be complex and lengthy, driving more laboratories to use simpler and more rapid methods to get results more quickly. This can simplify the decision making process and will result in a safer, better quality food supply. Legislation allows the use of such rapid methods as long as they are correctly validated and give equivalent results to the ISO Reference methods.

In this symposium presenters will discuss the effect of current legislation on the implementation of food testing methods, the development of standards for method validation, and some of the issues and problems involved in method validation and choosing the right method to use in testing laboratory.

Understanding If New Methods Work – Designing a Method Validation Procedure – ISO 16140
PAUL IN’T VELD, VWA Netherlands, Eindhoven, The Netherlands

The development of the current ISO 16140, that was published in 2003, took just over 10 years. This long period might be explained by the fact that this was the first example of such a procedure. Not long after its publication it was decided in 2005 to start the revision of ISO 16140. The reasons for that were several, e.g. lack of clear criteria for acceptance of the alternative method.

This year the revised 16140, now called ISO 16140 part 2, will be published again after a long period of standardisation. This is mainly due to the big changes that are proposed in the new standard. So what are the ideas behind the new concepts of e.g. Accuracy profile study and Relative limit of detection. What are Acceptability Limits and how were these determined.

Another important aspect is what will be done with the methods that have been validated according to 16140 once 16140-2 is published.

How to Validate a Method – Practical Aspects and Problems Associated with Method Validation
DANIELE SOHIER, ADRIA, Quimper, France

A revised version of the ISO 16140-part 2 standard will be published in a few months, and is already used in ongoing validation studies. Indeed, let’s focus on these new study designs for the validation of qualitative and quantitative methods in food microbiology.

During a validation study of a qualitative method, the most challenging conditions are supposed to be tested in the various parts. How will the new rules impact on the selection of the qualitative method protocol? Where are the...
pitfalls?

The validation study of quantitative method gathers two main approaches. The first one, the trueness study, aims at testing naturally contaminated samples: indeed, the real life! The second one aims at defining the accuracy and bias of the alternative method in comparison to the reference method. How to deal with these complementary approaches in the final method approval?

At least, the revised ISO 16140-part 2 provides clear acceptance criteria all among the validation process. As final users, how can these criteria be used for routine method selection?

**S11 Bacterial Spores in Foods: Safety and Quality Aspects**

Bacterial endospores are dormant structures produced by bacteria. Such spores are ubiquitous and due to their intrinsic resistance properties endospores may survive treatments given to foods during manufacturing. Spores can subsequently germinate in the food, followed by growth, leading to considerable losses in the food chain, and potentially to foodborne outbreaks in the case of pathogenic sporeformers.

A number of endospore producing bacteria can cause foodborne illness including *Bacillus cereus*, *Clostridium perfringens* and *Clostridium botulinum*. While *B. cereus* and *C. perfringens* account for some of the most common foodborne infections, *C. botulinum* outbreaks are severe with a high personal and economic impact, but are rare.

Spores not only play a role in food safety, but can also determine the quality and shelf life of foods. A specific concern to the food industry are spores with high heat resistances, with some able to survive UHT or sterilization treatments, causing food spoilage upon germination and outgrowth.

This symposium will focus on recent developments and findings concerning bacterial endospores in relation to food safety and quality. The state-of-art will be described. Presentations will focus on clostridial spores relevant to foods, the foodborne pathogen *B. cereus* in relation to its diversity and adaptation to foods, and highly heat resistant spores relevant to food spoilage.

**Presence of a Particular Gene Cluster is Responsible for Dramatically Increased Heat Resistance of Bacillus Spores**

Erwin M. Berendt1,2,3, Jos Boekhorst 1,3, Oscar P. Kuipers1,2, Marjon Wells-Bennik 1,3

(1) Top Institute Food and Nutrition (TIFN), The Netherlands
(2) Laboratory of Molecular Genetics, University of Groningen, The Netherlands
(3) NIZO Food Research B.V., Ede, The Netherlands

Spores of *Bacillus subtilis* are able to survive various harsh environmental conditions, including different heat preservation treatments used in food processing. Eighteen food and environmental isolates of *B. subtilis* could be divided into two distinct groups with different spore heat resistances. To obtain the same level of spore inactivation, spores belonging to the group with high heat resistance required 100 times longer heating times at 120°C than those with low heat resistance. Whole genome sequencing was performed for these isolates of *B. subtilis*, followed by correlating the genome content to the corresponding spore heat resistance phenotype. A set of genes were uniquely present in the strains producing spores with high heat resistance. All these genes were located on one mobile genetic element. The integration site of the mobile genetic element was determined, and the insertion was found in the same gene for all strains that produced high heat resistant spores (these strains had diverse backgrounds). We demonstrated that the genetic element was directly responsible for the very high heat resistance of spores; transfer of the element to a strain with significantly lower heat resistance (i.e., *B. subtilis* 168) rendered a phenotype of high spore heat resistance. The roles of the genes on the mobile genetic element and the exact molecular mechanisms that mediate the increase of spore heat resistance are currently subject of investigation.

**Clostridial Spores in Food**

MIKE PECK, Institute of Food Research, Norwich, United Kingdom

*Clostridia* are an important cause of food poisoning and food spoilage. Important foodborne pathogens include *Clostridium botulinum* and *Clostridium perfringens*. *C. botulinum* is responsible for botulism, a severe and deadly disease associated with consumption of the highly potent botulinum neurotoxin. *C. perfringens* is responsible for a more common but milder illness. It has also been recently suggested that some cases of *Clostridium difficile* infection may be foodborne. A diverse range of clostridia are responsible for food spoilage. The ability to form highly resistant endospores is a primary reason why clostridia are an important cause of food poisoning and food spoilage. In particular, their resistance properties enable clostridial spores to survive the heat treatment applied to many foods.

**Bacillus cereus: Diversity and Adaptation to Conditions in the Food Chain**

FRÉDÉRIC CARLIN, INRA, Avignon, France

*Bacillus cereus* accounts for some of the most common foodborne poisonings. Reports of *Bacillus cereus* outbreaks in Europe (including very severe emetic outbreaks) markedly increased in recent years. Spores of *B. cereus* are widely dispersed in the environment and contaminate any sort of foods. With *Clostridium botulinum* and *C. perfringens*, *B. cereus* is a concern for mildly heat-processed foods because of spore resistance and many strains have the ability to multiply at refrigeration temperature. The phenotypic diversity of *Bacillus cereus* strains has been established long ago. Recently a robust description of the genetic structure of the whole group has been provided and defined seven genetic groups. With respect to food safety, these groups numbered I-VII have different growth temperature profiles, and also differ by their involvement in foodborne poisoning incidents, production of toxins causing poisonings, resistance to heat, adaptation to low pH or to low aw, etc... For instance most cereulide-producing strains are distributed in phylogegetic group III that also contains the most heat-resistant strains. The “diarrhoeic potential” is low for psychrotrophic group VI strains, while it is higher for mesophilic groups III and IV, which is consistent with the prevalence of foodborne disease strains in the groups. A procedure to identify a *B. cereus* strain in the seven phylogenetic groups has been proposed. The risk for consumers of *B. cereus* foodborne poisonings should be considered with respect to the type of strains present in foods.
S12  **Risk Assessment for Incident Management**

Faced with a potential safety incident associated with their products, food manufacturers need to be able to make a quick assessment of the likelihood and impact of adverse consumer outcomes that could result from products on the market, and follow up with robust risk management actions if necessary. This symposium examines the expectations that government and retailers have for risk assessment and suggests approaches that will help companies, small and large, meet those expectations.

**A Regulator’s View on the Challenges and Expectations for Microbiological Risk Assessment**


Dealing with food incidents is an important part of the Food Standards Agency’s work and a significant proportion of these are microbiological in nature. No two incidents are the same and whilst their assessment and management can often be straightforward, at other times they can be more complex to deal with. Time is a key consideration with all incidents and this can make risk assessment and risk management challenging for all concerned particularly when the information available is sparse, conflicting or uncertain. Often the challenge is trying to piece together where, what and how it happened by using differing but complimentary strands of evidence with the aim of preventing it happening again. Whilst well established hazards will continue to challenge us we also need to recognise the potential for new ones as exemplified by the E. coli O104 outbreak in 2011. The emergence of new microbiological hazards, methodology, more complex and diverse foods as well as consumer behaviour will continue to challenge our ability to assess and manage risk in this area.

**Risk Assessment for Retail**

ALEC KYRIAKIDES, Sainsbury’s Supermarkets Ltd, London, United Kingdom

The management of product risk in a retail environment across a large range of diverse products, each with differing and sometimes complex supply chains requires a practical and somewhat pragmatic approach. Balancing risk assessment approaches to deal with the challenges of actual and perceived risk in stakeholder communities is also a key consideration. This presentation will explore the challenges in delivering a strategy to manage risk in a retail environment.

**Rapid and Robust Risk Assessment in a Potential Incident Situation**

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

Risk assessment approaches are often used in the food industry to underpin the food safety management decisions made for products that are brought to market and for the processes required to produce them. The uses range from early product and process design through to “farm to fork” production aspects such as ingredient sourcing, safe manufacturing and distribution systems, to ensure consumers receive a safe and wholesome product.

But what happens when things go wrong, despite the careful work put in up-front? Even the most well-planned and implemented systems are prone to failures. When failures occur, and there is a potential for unsafe product in the marketplace, decisions on risk management actions need to be taken very quickly, ideally within 24 hours. Are the decisions that are made in the heat of the moment as robust as the ones that have been made in the planning stages? Is there time to do a proper risk assessment of the situation? This presentation will focus on the approaches and requirements to ensure a timely yet robust risk assessment that meets regulator expectations and ensures appropriate actions are taken to protect brands and consumers.

S13  **The Importance of Microbiological Testing in Food Safety Management**

It is being recognized that food safety of products can only be ensured by having adequate processes in place, i.e., food safety is manufactured, not tested. A food safety management system in a food processing company includes both (validated) control and verification activities. Control activities are aiming at prevention or reduction of a food safety hazard (e.g., heat treatments) and are typically related to product and process controls and covered in HACCP programs. In order to be effective such control activities have to be validated for the specific purposes they are used for, where microbial sampling and testing can be used as part of the validation. Pre-requisite programs such as infrastructure, pest control, cleaning and sanitation, zoning, air and water control programs, hygiene of the workers form a vital part of food safety management systems and are the basis on which HACCP is built. Their main aim is to limit / avoid contamination or further growth of contaminating microorganisms. Verification activities in a food safety management system have the objective to provide evidence that products are within the set specifications / values, as well as to provide evidence towards the effectiveness of prerequisite programs. There microbiological testing plays an important role. During the last decade food businesses focus on the design and implementation of food safety management to guarantee food safety, which was also driven by incorporating the requirement for applying HACCP principles in different legislations. Still there is much focus on end product criteria and testing of end products against set specifications. Differences between criteria for products coming from production lines with different levels of control do not really exist – or being evaluated and practically applied, although more confidence could be given to a product from a well-managed processing line than from a batch of products that complies only with specific microbiological criteria without any information on process control. Therefore, the relevance of end product testing will be looked at and discussed at different examples.

**The Role of Validation, Verification and Microbiological Sampling in a Food Safety Management System**

MARCEL ZWIETERING, Wageningen University, Wageningen, The Netherlands

There is a general understanding that control of safety is only to a very limited extent supported by end-product testing. Good management should be based on evidence that hazards are well under control and that the interplay between initial levels of organisms, reduction, recontamination and growth is supplying a final level or prevalence of the hazard that is appropriate. Whether these phenomena are well under control needs to be based on solid information (validation). For information on phenomena like reduction, survival, transfer and growth of microorganisms along the
production process or even the whole food chain, information from specific experiments, databases, scientific literature or predictive microbiology could be combined to determine proof of sufficient control. If in this manner, by validation, a process is shown to be under control, this can be verified by end-product testing at the food industry level and by epidemiology on governmental level. Neither absence of the microbial hazard in samples of an end-products, nor the lack of evidence for an epidemiological link, is a proof that a process, and consequently the safety of food products, is under control. On the other hand, if end-products are not complying or if there is a strong epidemiological link, this can be an indication that a process is not under control. Therefore sampling as a verification activity may be a useful tool. It can be stated that end-product sampling is a relevant part of the verification of a food safety management system, but that it is more the totality of information that provides the confidence, than the sampling only.

The Relevance of End Product Testing: The Example of Canned Foods and Cooked Ham

JEANNE-MARIE MEMBRÉ, INRA, Nantes, France

In this paper, the production processes of two different types of food products are presented as case studies. It was hypothesized that identification of the impact of process steps that may lead to reduction, recontamination or growth, can be used as tool to assess the importance of sampling and control in the process. This identification was done by analyzing the available data (e.g., scientific literature, RASFF portal, EFSA reports).

The first example deals with canned foods. Generally a minimal $F_{121\text{C}}$ value of 3 minutes is used to guarantee sufficient reduction of *Clostridium botulinum* spores (for non-acid products). With a $>12D$ processing, there is very low probability of survival spores. Likewise, in hermetically sealed cans, the recontamination is preventing. Consequently, the end-product contamination rate is expected to be so small that testing is not effective.

The second example is cooked ham product for which there is an effective thermal treatment but a relatively high probability of recontamination by *Listeria monocytogenes* at the slicing steps. This may lead to a non-negligible rate of contaminated end-products at retail as *L. monocytogenes* is able to grow under chilled conditions. Here, verification by end-point sampling might be useful.

In conclusion, to assure safety, an efficient food safety management system must be implemented e.g., based on the HACCP principles and with proper pre-requisite programmes. End-product testing can be then used for verification of the implemented food safety management system.

Relevance of Microbial Testing for Verification in the Production of Chocolate

ANETT WINKLER, Mondelez International, Munich, Germany

It is being recognized that food safety of products can only be ensured by having adequate processes in place, i.e., food safety is manufactured, not tested. A food safety management system in a food processing company includes both (validated) control and verification activities. Control activities are aiming at prevention or reduction of a food safety hazard (e.g., heat treatments) and are typically related to product and process controls. In order to be effective such control activities have to be validated for the specific purposes they are used for, where microbial sampling & testing can be used as part of the validation. During the last decade food businesses focused on the design and implementation of food safety management to ensure food safety, which was also driven by incorporating the requirement for applying HACCP principles in different legislations. Verification activities in a food safety management system have the objective to provide evidence that products are within the set specifications / values, as well as to provide evidence towards the effectiveness of prerequisite programs and / or process controls in place. There microbiological testing plays an important role.

There are several specifics to consider when defining meaningful microbial testing plans for chocolate, such as very low levels of pathogens being of concern, major part of last production steps not having a potential microbial reduction step, open processes & role of the environment. Those will be discussed in conjunction with potential controls in the course of the presentation.

Prediction of Shelf Life, and Product’s Microbial Quality Using Smart and Non-invasive Platforms

Highly perishable foods, which, unless correctly stored, processed, packaged and distributed, spoil quickly and may potentially become unsafe due to microbial growth of certain pathogenic bacteria such as *L. monocytogenes*, *E. coli O157:H7* and *Salmonella* spp., despite the introduction of HACCP (Hazard Analysis Critical Control Points), and the proliferation of food safety regulations. Indeed, the image of the European food industry has suffered major set-backs causing huge economic losses to the industry. These setbacks are key issue not only due to short-term economic losses but because they create an atmosphere of mistrust amongst food buyers and consumers in general and this may lead to long-term problem. That potential atmosphere stresses the need for more effective food quality and safety assurance systems, which are both effective and visible to the consumer. This quality (including safety) system should not only adapt the existing technological progress and evolution of consumer lifestyle but also foresee the potential of the latest information technology as well as the future contribution of converging technologies.

The current practice to meet these demands, address these issues and to assure the safety of food still relies heavily on regulatory inspection and sampling regimes. This approach, however, seems inadequate because it cannot sufficiently guarantee consumer protection since 100% inspection and sampling is technically, financially and logistically impossible.

It is evident that the food industry needs rapid, non-invasive and, if possible, hand-held analytical instruments/ methods that can be used on-line, in-line or at-line and can ensure that raw and in process materials are both of good quality and safety while food losses are minimized. These methods should be integrated in the design of systematic preventive approaches (e.g., within the HACCP hygiene assurance system, which is mandatory in Europe and is a globally recognized standard), and serve as elements of practical decision-making tools and an early warning system for critical quality attributes (CQAs) of (i) raw materials, (ii) materials under processing and (iii) final products before distribution and,
if possible, even further along the food chain, to facilitate the prediction of the remaining shelf life of these products; this is a significant tool in minimizing indirectly energy, water (used by the industry, e.g., for cleaning) and material expenditures.

In this symposium the potential use of Process Analytical Technology (PAT) will be introduced and will respond to above-mentioned requirements identified by the food industry e.g., to enforce research by integrating rapid and holistic analyses, including chemical, physical and microbiological analyses, by novel sensors, in tandem with converging mathematical technologies/tools/models, i.e., advanced statistical approaches (e.g., bioinformatics, chemometrics) and IT to provide a robust decision making tool.

**Use of Non-invasive Tools in Tandem with Bioinformatics for the Implementation of Process Analytical Technology in Food Industry**

GEORGE-JOHN NYCHAS, Agricultural University of Athens, Department of Food Science and Human Nutrition, Athens, Greece

The current practice of monitoring/evaluation of quality (including safety) in the entire food manufacturing as well as in the food chain, relies heavily on regulatory inspection and sampling regimes. For example, according to EU authorities the quality/safety/hygienic conditions of fresh meat is evaluated only by viable counts of bacteria able to grow on very generic media or on counts of the *Enterobacteriaceae* family. On the other hand counting colonies is certainly time-consuming, e.g., results can be available after 24–48h, and as a consequence it does not allow an online response, which would be needed to trigger appropriate corrective measures. The approach, described above, seems inadequate because it cannot sufficiently guarantee consumer protection since 100% inspection and sampling is technically, financially and logistically impossible.

Therefore, the food industry needs rapid, non-invasive, potentially hand-held analytical monitoring instruments/methods that can be used on-line and can ensure that raw and in process materials are both of good quality and safe. Furthermore, inspection authorities need reliable methods for control purposes, while the wholesale and retail sectors need these valid methods to ensure the freshness, safety and origin of their products and to resolve potential disputes between buyers and sellers. It is, therefore, crucial to have valid methods to monitor freshness and safety in order to be able to ensure quality. Thus, instruments based on vibrational spectroscopy or surface chemistry can be used as devices and robust model systems in assuring stakeholders that food product safety and quality are ensured.

**Hyperspectral Chemical Imaging in Microbiology: Methods, Recent Applications and Future Directions**

AIOFE GOWEN, UCD, DUBLIN, Ireland

Hyperspectral chemical imaging (HCI) is a broad term encompassing spatially resolved spectral data obtained through a variety of modalities (e.g., Raman scattering, Fourier transform infrared microscopy, fluorescence and near-infrared chemical imaging). It goes beyond the capabilities of conventional imaging and spectroscopy by obtaining spatially resolved spectra from objects at spatial resolutions varying from the level of single cells up to macroscopic objects (e.g., foods). In tandem with recent developments in instrumentation and sampling protocols, applications of HCI in microbiology have increased rapidly. This presentation gives a comprehensive overview of the fundamentals of HCI and recent applications in microbiology. Technical challenges and future perspectives for these techniques will also be discussed.

**Metabolomics Serving Food Quality and Safety**

EFSTATHIOS PANAGOU and George-John Nychas, Agricultural University of Athens, Department of Food Science and Human Nutrition, Athens, Greece

In food science, metabolomics has been proved a valuable tool to investigate quality changes during processing, distribution and storage of raw materials and final products. In the area of quality and safety of processed foods, metabolomics can contribute in (a) food classification, food adulteration and authenticity assessment; (b) quality control in food industry, (c) food spoilage, (d) association between sensory characteristics and chemical composition, and (e) impact of processing on food composition. Metabolomics have been proved very efficient in the detection of microbial metabolites that can be used as biomarkers in food spoilage under different packaging conditions (air, vacuum, modified atmosphere packaging) using HPLC, gas chromatography (GC) and/or mass spectrometry (GC-MS). In the context of the current presentation selected examples will be presented on the implementation of metabolomics in beef and pork meat spoilage stored under aerobic and modified atmosphere packaging, through the evolution of volatile compounds detected by headspace solid phase microextraction gas chromatography/mass spectrometry (Headspace SPME-GC/MS) and HPLC analysis. Moreover, pasteurized vanilla cream spoilage will be presented using SPME-GC/MS analysis. Finally, the contribution of metabolomics in food processing control will be discussed using green table olive processing as an example.

**Managing Foodborne Toxoplasmosis: Recent Advances and Future Directions**

S15

Approximately 2–3 billion humans are currently infected with *Toxoplasma gondii*, and toxoplasmosis was recently ranked 4th by FAO/WHO from an international food safety perspective. Although most infected individuals will remain unaware of the *Toxoplasma* parasites persisting as quiescent tissue cysts throughout their body for the remainder of life, approximately 10–20% will have experienced self-limiting “flu-like or glandular fever-like symptoms during acute infection. In pregnant women who become acutely infected, however, the parasite can also cause severe abnormality or death to the unborn child. In key vulnerable clinical groups such as the immunodeficient (HIV/AIDS) and the immunosuppressed (organ transplant recipients, patients receiving anti-cancer therapies, etc.), opportunistic reactivation of tissue cysts can cause severe or life-threatening disease.

Treatment of active toxoplasma infection is relatively effective but cannot clear quiescent tissue cysts from the host and there is no human vaccine. Thus, there has recently been renewed focus on prevention of food borne toxoplasma infection with international agencies such as EFSA, for example, developing guidelines for EU member states.
regarding surveillance capabilities for monitoring Toxoplasma infection in the food chain, as well as identifying key gaps in knowledge and technologies required for more accurate risk assessment and control.

The proposed session will review the current state of knowledge regarding the clinical spectrum and burden of toxoplasmosis and will describe and consider emerging molecular typing data generated through key international initiatives that have led to insights into the geographical origins and epidemiology of Toxoplasma gondii infecting the human and animal population. The session will also describe work being undertaken to assess relative risk of acquiring Toxoplasma infection through the consumption of specific tissues from different animal species entering the human food chain, including through major international initiatives and consortia, and will consider potential measures to further reduce the risk of foodborne infection.

Human Toxoplasmosis: Infection, Clinical Spectrum and Burden of Disease

EDWARD GUY, Public Health Wales, Swansea, Wales

Toxoplasma gondii is one of the most successful protozoan parasites in nature, being able to infect all warm-blooded animals including humans. It is an intracellular parasite with a predilection for brain and muscle tissues where, after infection has occurred, it persists in the form of latent tissue cysts, probably for the host’s lifetime. It is estimated that perhaps 2–3 billion people worldwide are infected. Infection in the immunocompetent is typically asymptomatic in 80–90% of individuals with the remainder usually experiencing a self-limiting, mild to moderate flu- or glandular fever-like illness. However, in pregnant women who acquire the infection after conception, the parasite may be transmitted to the unborn child and can result in mild to severe foetal abnormality or death. In the immunocompromised and immunodeficient, e.g., HIV/AIDS, cancer patients, organ transplant recipients, etc., significant or life-threatening sequelae may occur either to acute Toxoplasma infection or reactivation of latent tissue cysts associated with infection acquired previously.

While Toxoplasma can be transmitted between humans either vertically (congenital infection) or associated with transplantation of organs or tissues from an infected donor to an uninfected recipient, the two principal routes of infection are by ingestion either of oocysts in soil and water contaminated with the faeces of infected cats, or via the foodborne route through consumption of undercooked meat from infected animals. In a study in 2000 among pregnant women in 6 major European cities, over half of infections were attributable to consumption of meat. Further, from an international food safety perspective, Toxoplasma was ranked 4th among foodborne parasites with the greatest global impact in 2014, by FAO and WHO.

To serve as an introduction to the two following presentations, additional background information will be provided and some key questions highlighted, relevant to the investigation and control of Toxoplasma in the food chain.

Molecular Epidemiology of Toxoplasma: Genotypes and Human Toxoplasmosis – A Foodborne Perspective

MARIE-LAURE DARDE, University of Limoges, Limoges, France

The global population structure of Toxoplasma gondii is largely influenced by geographic distribution and by ecological aspects driving parasite transmission. In Europe and North America, two major clonal types, II and III, predominate. South America is a hotspot of diversity with many other haplogroups and highly divergent strains in its Amazonian part. In Africa and Asia, more recently explored, the diversity seems lower than in South America.

The pathogenicity due to a Toxoplasma infection depends on host species. It has been well defined in the mouse model for the three main genotypes described (types I, II, III). The mouse model permitted to find virulence markers, mainly proteins involved in Toxoplasma invasion or immune evasion. In human toxoplasmosis, the role of parasite genotype on the outcome of the infection is largely dependent on the immune status. However, in immunocompetent patients, some strains appeared to be more pathogenic, causing disseminated toxoplasmosis cases or more severe ocular disease.

In France, the parasitological network for Toxoplasma isolate collection permitted to genotype isolates or DNA from more than 1000 human cases. 84% of them belong to the clonal type II, and 4% to type III. This reflects the local epidemiology of strains in wild and domestic animals. Most of the human isolates different from these two genotypes were acquired directly or indirectly (imported meat from the Americas) outside Europe. They were often responsible for severe toxoplasmosis cases. Consequently, isolation of an atypical strain in human toxoplasmosis should prompt to an epidemiological investigation.

Toxoplasma Infection in Food Animals

ELISABETH A INNES, Alison Burrells, Julio Benavides, Paul Bartley, German Canton, Jackie Thomson, Francesca Chianini, Clare Hamilton, Joao Luis Garcia and Frank Katzer, Moredun Research Institute, Edinburgh, Scotland

Toxoplasma gondii is capable of infecting all warm blooded animals including humans. Congenital disease is a serious risk to women who contract T. gondii infection for the first time during pregnancy where the affected foetus may present with neurological and ocular disease. The disease burden of toxoplasmosis, as represented by disability adjusted life years, is one of the highest among all foodborne pathogens. Food animals such as pigs, chickens, sheep, goats and cattle may also pose a risk to public health if people consume undercooked meat products that harbour T. gondii tissue cysts, and recent studies have shown that this is a major transmission route to people. Outdoor reared animals are more likely to come into contact with the parasite and thus meat products from these animals are thought to be a risk. A recent survey of sheep flocks in Scotland showed that 56% of sheep were sero-positive for T. gondii and a further survey of UK pigs showed a prevalence of 7.4%. Work in The Netherlands has highlighted the fact that outdoor reared pigs represent a significantly higher risk of becoming infected with T. gondii compared to indoor reared animals. A current EFSA funded project is looking at the relationship between seroprevalence in the main livestock food species and presence and predilection sites of T. gondii in meat. Our current research has also focused on developing strategies to help mitigate this risk and has evaluated the use of vaccination to reduce the burden of tissue cysts in food animals resulting in safer meat for human consumption.
Whatever is our role in the society we all are food consumers. How we take care about food safety when we are at home handling food for us and family? Are we curious about typical cuisines when we travel around? What are our feelings toward “exotic food?” Do all scientists maintain their rationale behaviours in daily life? Do consumers trust in people and institutions involved in the food chain? The evaluation of risk perception is very important in food safety for the risk management and communication, as it drives consumer attitudes and behaviours. Risk communication, education for food safety intervention can be the same for all consumers? The scientific risk analysis process should always consider the consumer’s risk perception. Consumer decision-making is also determined by perceptions of benefit associated with food consumption, and risk-benefit communication may be needed to allow consumers to make informed food choices. Risk communication needs to take account of food choice habits and traditional food preparation practices of the population to whom the message is being targeted. The analysis of the traditional food dishes and common practices in different countries can help in tailoring risk communications and control risk perception. Good risk communication can ensure that consumers act to protect their health, and those of others. There are various factors, which can contribute to the development of effective risk communication with consumers. It is important to address consumer concerns, as well as technical risk assessments in communication. Risk uncertainty and actions to reduce these uncertainties. Consumers’ trust should also be explored especially when communicating information about the risks and benefits of mitigation strategies. Despite the existence of a considerable body of knowledge about effective food risk communication, some gaps in knowledge and future research needs can be identified, and these will be discussed.

**Food Safety Risk Communication. What We Know and What We Need to Know**

**ANTHONY FLOOD**, International Food Information Council, Washington, D.C.

Public understanding of chemical risks, and also of benefits associated with food production and chemicals, is one area where efforts to reach a mutually acceptable consensus have been largely unsuccessful. This study provides an initial step in bridging the communication divide between scientists and consumers. This research establishes a baseline understanding of how consumers view the potential risk of chemicals in the food supply.

**Research Approach:** This study assessed belief, attitudes and behaviors of mothers. The study employed a two-pronged approach to data collection. Phase I of the study implemented small group discussions (focus groups) to generate basic knowledge of how participants perceive food, food risks, and eventually chemicals. Focus groups consisted of 4 individuals in each group; 2 groups in 4 cities for a total of 32 participants in Phase I. Phase II used a quantitative, on-line survey to assess perceptions about specific chemicals in food. Survey participants totaled 1,000.

**Results/Findings:** Chemicals in food are not top of mind. When probed, results from phase one of the study confirm that, in general, attitudes about chemicals are negative especially for those mothers who are very sensitive to the issue. Our study found these mothers are typically young (18 – 34), have a higher household income, are knowledgeable about food topics, and are engaged with most aspects of food and shopping. They are also more likely to use social media as a source of information.

**Conclusions/Recommendations:** Initially, objectives for this project included the need to identify the key factors that influence those persons responsible for purchasing food for their families. Secondly, communication objectives for future work are identified in the survey. Participants indicate that the Q & A format for relaying chemical information was preferable to a more conventional narrative. Additionally, this study tested keywords, phrases, and concepts that will be implemented and tested again in the future. Finally, an analysis of how the Internet influences food purchasing decisions needs to be conducted.

**Two Case Studies on Risk Perception: Trust in Institutions and Gender Differences in Risk**

**SEDA ERDEM**, Department of Economic and Behavioural Science Centre, University of Stirling, Stirling, United Kingdom

This talk presents two case studies that utilise a relatively new approach, namely the best-worst scaling technique, to establish and analyse perceptions of risks and trust. The first case study investigates the levels of control respondents believe they have over twenty food and noon-food risks and determines the levels of concern they attach to each. The elicitation method is structured in a way to reduce the cognitive burden typically associated with ranking over large sets and permits the derivation of individual-level of perceived control and worry and how these vary by gender. The results show considerable heterogeneity in perceptions of control and worry, that the degree of heterogeneity varies across the risks, and that women systematically consider themselves to have less control over the risks than men. The second case study investigates the level of trust consumers have placed in sixteen different institutions who provide information about a new technology, nanotechnology, and its use in food packaging. The results show that consumers perceive information sources differently and present different levels of trust towards these institutions. By investigating the heterogeneity in perceived trust in various institutions, this research provides insights into the development of best practices in risk communication for novel foods produced by nanotechnologies. Overall, these two case studies provide information on the elicitation of perceptions of risks and trust in institutions and the analysis of heterogeneity in these perceptions with implications for risk communication.

**From Farm to Fork: Common Practices and Food Consumers Behaviours — BSE Versus Toxoplasmosis**

**MARIA VITALE**, Istituto Zooprofilattico Sperimentale della Sicilia, Palermo, Italy

Food safety is a complex area that encompasses the entire food chain from farm to fork. However food chains differ across several countries: for example, in the way farms are managed, traditions associated with farming practices and local cuisines. In recent decades consumers in developed countries have been increasingly concerned about the risk of disease transmission through food consumptions. Food is often associated with social events and celebration, and most people do not accept potential risks from food.
Food safety is highly relevant to both public health and economics. The BSE crisis in Europe has demonstrated the importance of consumer risk perceptions in relation to a food safety incident. The reduction of beef consumption resulted in huge economic losses for both producers and European governments, in particular in the UK.

In contrast to BSE (with low but fatal transmission from food to humans) toxoplasmosis is a neglected foodborne disease from the consumers’ point of view, mainly because the infection is usually asymptomatic. Due to the serious consequences of vertical transmission, seronegative pregnant women are conscious about safe behaviour. However quite often after pregnancy they relax and go back to their usual habits. Preventive educational campaigns are very important to diffuse good practices in food handling and consumption, but a question remains as to how efficacious they are. The analysis of common practices from farm to fork and the involvement of the target population could be very important for efficacy of educational campaigns.

**S17 Practical Examples of Predictive Microbiology Used by the Food Industry**

Over the last 3 decades, predictive microbiology has evolved into a mature area of science in the context of food safety and quality. Predictive models provide a powerful capability to: (i) support rapid and robust computer-aided design of microbiologically safe and stable foods, (ii) assess consumer safety risks, and (iii) inform risk management options and their implementation via operational food safety management systems (e.g., HACCP). In many cases, microbial models can support decision-making and inform a safe operational design space without the need for additional data generation (e.g. when a model has been appropriately validated in representative food-model systems and deemed to provide ‘fail-safe’ predictions). Even in cases where product-specific effects are expected to have a significant impact on microbial behaviour, predictions from lab-media based models are still very useful in guiding more focused and targeted challenge testing. In addition, there are currently several software tools available in the public domain which deploy microbial models in a user-friendly manner. Despite the readiness of predictive microbiology as a scientific capability and the obvious advantages it can bring to industry, its adoption and acceptance by the food industry remains variable.

This symposium aims at presenting examples of practical application of predictive microbiology in the food industry, striving to give a balanced view between advantages and opportunities vs. limitations and constraints. This session will be of relevance to food safety professionals in industry (e.g., food microbiologists, food scientists, food technologists, product developers), researchers in academia, and scientists working for government and international organisations interested the practical application of microbial modelling and risk assessment in the development of safe food products.

**Practical Application of Deterministic and Stochastic Microbial Models for Risk-based Food Product and Process Design**

ALEJANDRO AMEZQUITA, Unilever, Sharnbrook, United Kingdom

Designing food products and processes based on a risk assessment approach can bring a better understanding of the factors relevant to risk, thereby providing suitable information for risk management. Such proactive use of risk assessment by the food industry can facilitate safe innovation. In that context, predictive microbial models play a fundamental role in estimating levels of microorganisms at a given point in the food chain. Effective application of microbial models by industry must be fit-for-purpose. Both deterministic and stochastic approaches offer value in informing decision-making. The former approach has lower data demands than the latter; it also requires a lower level of expert knowledge for effective use and interpretation. Stochastic approaches, on the other hand, can be very valuable when there is high variability and/or uncertainty in the information needed to support decisions on safe product/process design, which would be otherwise constrained by the use of a deterministic model. This presentation describes the value added by deterministic and stochastic microbial modelling approaches in industry through several case studies. The deterministic case studies are focussed on non-thermal and thermal inactivation of infectious pathogens in acidic foods and dairy-based liquid foods, respectively. The stochastic case studies are focussed on growth of infectious pathogens at super-chilled temperatures and thermal inactivation of spore-formers during aseptic processing, both in neutral pH liquid foods. All case studies have been used as part of microbial risk assessments in support of a safe-by-design approach to establish the basis for consumer safety of such foods.

**Hurdle Efficacy Predictions and Growth Inhibition in Complex Food Matrices**

MICHAEL CALLANAN, Nestlé Research Center, Lausanne, Switzerland

The standard method of challenge testing to evaluate the efficacy of preservation systems is labour intensive and time consuming. One approach to overcome this problem is to use mathematical modelling based on pre-existing microbial growth data. It can also represent a more cost effective way to design preservation systems for food products. We looked to improve the accuracy of hurdle efficacy predictions made using microbial growth data by combining public data sources and internal challenge test data. Model predictions were compared to actual growth data generated using impedance technology in a real food matrix where the antimicrobial hurdles (pH, acids and water activity) were modified. The results indicate that predictions of hurdle efficacy were generally aligned with the experimental data. The alignment of the models and data could be further improved by adjusting interaction factors and lag time variables in the models.

**Development of a Model and Software for Listeria monocytogenes Growth in Ready-to-Eat Meats with Different Antimicrobial Formulations**

CIAN O’MAHONY, Creme Global, Dublin, Ireland

Antimicrobials are used to control the growth of microorganisms in foods under different product formulations, and predictive models derived from experimental data can be used to estimate microbial growth. The goal of this work was to develop a predictive model and software application that will estimate the growth of *Listeria monocytogenes* in ready-
Fresh Produce and Water: How Can Risk Assessments be Used in Ensuring Safety of Fresh Produce?

Risk assessment studies related to use of water and safety of fresh produce stem from both water and food microbiology oriented studies. Most of the studies from the water microbiology perspective focus on enteric virus risks, largely because of their anticipated high concentrations in untreated wastewater and their recalcitrance to common wastewater treatments. Risk assessment studies from the food perspective rather focus on bacterial pathogens such as *Salmonella* and pathogenic *E. coli*. A few risk assessments related to protozoa are available and some multi-target studies. Few site specific data points were available for most of these microbial risk assessments, meaning that many assumptions were necessary. Specific parameters lacking hard data included the rates of pathogen transfer from irrigation water, and other water sources (e.g., run-off water), to crops, pathogen penetration in food crops, and pathogen survival on or in food crops. Data on these factors have been accumulating over the last decade, and this should improve the reliability of future microbial risk estimates. QMRA studies are suitable in particular to evaluate different control scenarios but as the outcomes rely partly on assumptions, it should be regarded as an indication of the level of safety. Still, the outcome can be used to guide the risk management direction for effective pathogen control and to select the most appropriate control measures and it helps to focus research on the areas where important pieces of information are missing. The new approaches in QMRA are leading to more flexibility and more tailored guidelines on water treatment and levels of pathogens in irrigation or processing water for the fresh produce in certain regions.

Target Microorganisms Used in Risk Assessments Related to Water and/or Fresh Produce

Ana Allende, CSIC, Murcia, Spain; TU Delft, Delft, Netherlands

Contamination of leafy greens with foodborne pathogens may occur at any step in the farm to consumer chain from environmental, animal or human sources. In an EFSA opinion issued in January 2013, based upon the EU Zoonoses Monitoring data from 2007 to 2011, it was estimated that foods of non-animal origin were associated with 10% of the outbreaks, 26% of the cases, 35% of the hospitalizations, and 46% of the deaths. Recent publications highlighted several pre-harvest sources as the most probable origins of potential contamination including: contaminated water, soil amendments and fecal contamination from wildlife. In the fresh produce sector, water, is a usual suspect as the source of, or the vector for distribution of microbial hazards on the marketed produce. The evidence for water as a risk factor for introduction and transmission of pathogens on fresh produce will be discussed.

The most common etiologic agents associated with produce outbreaks are *Escherichia coli* O157:H7 and *Salmonella*. Recently, the EFSA highlighted *Salmonella* spp. and leafy greens eaten raw as salads as one of the five top raking food/pathogen combinations most often linked to human cases originating from Food of Non-Animal Origin (FoNAO) in the EU. This talk will review the main food/pathogens combinations. The potential use of indicator microorganisms to evaluate risk assessments related to water and/or fresh produce will be also discussed.

Data Gaps to be Considered in Modelling Strategies for Fresh Produce

Peter McClure, Mondelēz International, Birmingham, United Kingdom

Microbial Risk Assessment (MRA) is an integral component of food safety management which consists of the three components: risk assessment, risk management, and risk communication. A similar framework for safe (re)use of water has also been used this as a basis for water guidelines provided by the World Health Organisation, including the safe use of wastewater in agriculture. The principal aim is to support risk management by providing an objective, transparent, evidence-based assessment of the health risk of exposure pathways/scenarios. Fully quantitative MRAs are, however, data hungry and for MRAs applied to use of water for fresh produce, there are a number of data gaps that limit their development. Few site-specific data points are available for most of these microbiological risk assessments, meaning that many assumptions are necessary. Specific parameters lacking hard data include incidence and levels of pathogens in water and associated variabilities, rates of pathogen transfer from irrigation water to crops, pathogen penetration, survival in or on food crops and dose response. Data on these elements have been investigated over the last decade and this should improve the reliability of future microbial risk estimates. However, the large number of different foodstuffs and pathogens, combined with water sources and irrigation practices, means that developing risk models that can span the breadth of fresh produce safety is a considerable challenge. This presentation will describe these gaps in more detail.

Usage of QMRA Studies in Risk Assessments Related to Use of Water and Fresh Produce

Liesbeth Jacksens, Faculty of Bioscience Engineering, Department of Food Safety and Food Quality, Ghent University, Ghent, Belgium

Risk assessments related to use of water and safety of fresh produce originate from both water and food microbiology studies. Although the set-up and methodology of risk assessment in these 2 disciplines may differ, analysis of the current literature reveals some common outcomes. Most of these studies from the water perspective focus on...
enteric virus risks, largely because of their anticipated high concentrations in untreated wastewater and their resistance to common wastewater treatments. Risk assessment studies from the food perspective, instead, focus mainly on bacterial pathogens such as *Salmonella* and pathogenic *E. coli*. Few site-specific data points were available for most of these microbial risk assessments, meaning that many assumptions were necessary which are repeated in many studies. Specific parameters lacking hard data included rates of pathogen transfer from irrigation water to crops, pathogen penetration, and survival in or on food crops. Data on these factors have been investigated over the last decade and this should improve the reliability of future microbial risk estimates. However, the sheer number of different foodstuffs and pathogens, combined with water sources and irrigation practices, means that developing risk models that can span the breadth of fresh produce safety will be a considerable challenge. The new approach using microbial risk assessment is objective and evidence-based and leads to more flexibility and enables more tailored risk management practices and guidelines. Drawbacks are, however, capacity and knowledge to perform the microbial risk assessment and the need for data and preferably data of the specific region.

**S19 Improving the Evidence Base and Transparency of Food Safety Decision Making**

Management of food safety issues provides numerous challenges from assessing the importance of food safety risks, through identifying, evaluating and selecting the most effective risk mitigation strategies, to communicating the basis of such decisions in the context of other competing (e.g., socio-economic) factors. In many sectors, systematic research (knowledge) synthesis and risk-based methodologies are recognized as the cornerstone of transparent, evidence-informed decision making. While relevant approaches are available and recently applied in the food safety arena, the extent of their use in support of transparent, evidence-informed food safety decision making is not optimal. In this symposium, we will first introduce and discuss some of the available approaches for identifying, appraising, summarizing and using data to inform food safety risk management either directly or through provision of transparent, structured inputs to risk-based approaches (e.g., risk assessment, risk ranking). The key principles and practices will be briefly described and coupled with practical, real-life examples illustrating specific contributions for transparent risk management and food safety decision making. Particular consideration will be given to principles and practices for optimizing use of limited data. The use of research (knowledge) synthesis and other methodologies in support of risk assessment and risk ranking will be specifically illustrated through recently completed global food safety activities. We will conclude the symposium with a group discussion with the symposium participants, focusing on the further improvement of these approaches to better support risk managers and address their specific needs.

**Knowledge Synthesis and Transfer in Support of Food Safety Decision Making**

**ANDRIJANA RAJIC**, Food and Agriculture Organization of the United Nations, Rome, Italy

Knowledge synthesis (KS) refers to reproducible and transparent methods to identify, appraise, characterize and synthesize the global body of knowledge about a topic. Systematic reviews (SR) and meta-analysis (MA) methods are the most frequently used KS methods in food safety. They can be used to investigate a variety of questions, including intervention efficacy, prevalence and concentration of outcomes, and diagnostic test accuracy. Scoping reviews answer broader research questions than SR-MA and can identify areas of research strength and gaps, which can guide prioritization of future research or the focusing of specific questions for SR-MA. Structured rapid reviews are streamlined SRs conducted within a rapid timeframe and are used to inform urgent food safety decision-making. Engagement of stakeholders early in the process is critical to ensure that the results will be appropriately transferred or exchanged and used to support risk management and decision-making. Key logistical requirements for application of KS in agri-food public health sector are experienced and multidisciplinary team, sufficient resources, and organizational commitment to use KS approaches. Various KS methods can be effectively used to support transparency, credibility and risk manager confidence in the process and results of scientific assessments (e.g., exposure assessment, quantitative risk assessment, MCDA). Throughout the presentation, unique and illustrative examples and insights will be provided within the agri-food public health context.

**Promising Principles, Practices and Tips for Using Data to Inform Risk Assessments/Tools**

**IAN YOUNG**, Food Safety and Quality Unit, Food and Agriculture Organization of the United Nations, Rome, Italy; Laboratory for Foodborne Zoonoses, Public Health Agency of Canada, Guelph, ON, Canada

Risk assessment and other risk-based tools (e.g., risk profiles, risk ranking) are frequently used to support risk management, communication, and decision-making about food safety hazards and threats. Structured and transparent procedures are necessary to ensure that data to inform these processes and tools are appropriately identified, appraised, and utilized, particularly in low- and middle-income countries (LMIC) where risk analysis capacities might be limited. In order to inform the development of new training modules for LMIC in this area, we conducted a rapid scoping review of the literature to summarize and describe promising principles and practices for identifying, appraising, and using existing data and generating new data to inform food safety risk assessment and other risk-based tools. We identified four key areas of promising principles and practices across a diverse range of situations and contexts relevant to food safety risk assessment and other risk-based tools: 1) identifying existing data sources; 2) overcoming data gaps; 3) selecting among data sources; and 4) ensuring data quality assurance and reliability. Detailed options and considerations will be presented and discussed for each of these categories, including their relevance and applicability for risk assessors, risk managers, and other stakeholders to ensure an evidence-informed approach to food safety risk analysis.

**Towards Transparent and Practical Risk-based Applications in Food Safety**

**MICHAEL BATZ**, University of Florida, Gainesville, FL

As we move towards ever greater pursuit of science- and risk-based food safety systems, the need for data, tools, and processes to provide objective but meaningful information to decision makers becomes more pronounced. The array of decision contexts is vast, ranging from broad resource allocation across an entire system to targeted risk management for a specific food-pathogen pair at a defined point in the food chain. Risk analysis provides a suitable
framework for incorporating data and analyses into the decision making process, but time and resources are often too limited, and available data too sparse, to allow for the development of quantitative models. Semi-quantitative and qualitative approaches can be utilized, though such approaches often rely upon subjective judgment or the elicitation of values from experts. Some decisions may be amenable to approaches that estimate risk in one dimension, but other situations require consideration of factors difficult to incorporate in traditional measures of risk. The challenge, for those providing input to decision makers, is to choose and design practical approaches that are best suited to task, which often involves the consideration of factors such as data quantity and quality, timeliness, available resources and analytic capacity, transparency, and repeatability. We will describe some recent experiences developing tools to prioritize food safety efforts, and discuss some key principles that might be considered by those attempting similar analyses in the future.

S20 Integrative Aspects in Food Safety Risk Assessment

There is increased awareness that food safety risk assessment should not be an isolated discipline. Quantitative Microbial Risk Assessment (QMRA) is a valuable tool for understanding, reducing, and preventing risks presented by hazardous microorganisms. However, QMRA should not be seen as a stand-alone bottom-up approach to assess microbial risks. Recent fast developments in systems biology have the potential to refine microbial risk assessment but have the pitfall to result in a to high level complexity and associated uncertainty. In addition, microbial risks are not only defined by microbial behaviour stricto sensu but by a larger context. Coupling of QMRA with social science and epidemiology may help in placing risks in the appropriate context. Advances made in social science and epidemiology can substantially aid microbial risk assessment by offering a look from different angles to microbial risks. This symposium will provide the opportunity to get up-to-date on the latest advances on the integration of molecular biology, social science and epidemiology in microbial risk assessment.

Food Safety Management Systems and Safety Governance

PIETERNEL LUNING and Klementina Kirezieva, Wageningen University, Wageningen, The Netherlands

Reoccurrence of foodborne illnesses concerns keeps the concerns on food safety management system (FSMS) performance. In the European Union, policies are following the principles of subsidiarity and multi-level governance, aiming at distributing policy responsibility among different governmental levels, the public and private sector. These principles are translated differently in the governance structure of each member state, leaving room for industrial self-regulation. Furthermore, market structures and climate conditions differ. Food safety requirements to companies differ substantially as their context characteristics, whereas all must comply with the same safety requirements. The study objective was to gain insight into current performance of FSMS in three European countries, with their specific governance and market context.

A framework was constructed to study the broad context including the food safety governance and diagnose the FSMS performance. The framework was applied in three case studies of leafy greens cultivation.

Results revealed that the companies operating in favourable broad context, including favourable climate, big companies in integrated market, and stringent standards as a result of market self-regulation, have demonstrated advanced FSMS, systematic information about the output and supporting organisational characteristics.

The FSMS of the companies that were operating in less favourable broad context, either in fragmented market with small companies or in less favourable climate, have demonstrated less mature FSMS. In the less favourable climate, companies were supported by public policy interventions and baseline food safety standards, they realised average FSMS. The small companies operating in fragmented market demonstrated basic FSMS. These companies have been dealing with both domestic and export market, private and public certifications. This mixture of standards and requirements has proved difficult for the implementation capabilities of small farmers.

System Biology, Heterogeneity of Stress Response and Microbial Risk Assessment of Bacterial Spore Formers

STANLEY BRUL, Wishwas Abhyankar, Rachna Pandey, Linli Zheng, Alex Ter Beek, Jan P.P.M. Smelt, Norbert O. Vischer, Chris G. de Koster and Erik M.M. Manders

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Microbial cells can be both beneficial and harmful to man. Their behavior in the food chain and upon ingestion in the intestinal tract of man should be highly predictable in order to facilitate the development of reliable fermented foods and preservation strategies for foods. Bacterial spores are a class of microbial structures of major concern to food manufacturing and distribution due to their extreme stress resistance. Spores can withstand elevated temperatures and the presence of adverse environmental conditions including high salinity, acidity and the action of hydrolytic enzymes. Spores are ubiquitous in nature and as such present in many food ingredients including prominently milk derived ingredients in the dairy chain and vegetable ingredients in the fresh produce and chilled chain. Predictability of the microbial cells’ viability and stability of predictive food microbiology models. Such predictability should be captured in spore germination, outgrowth and vegetative growth models. The conditions under which the data are gathered range here from optimal germination and outgrowth conditions to the use of commonly known preservation strategies including the presence of weak-organic acids.

In order to analyze the driving forces of spore germination and outgrowth at the population level as was done to date it is more and more evident that the underlying heterogeneity of individual spore germination and outgrowth needs to be captured. Such deconvolution of germination and outgrowth patterns is only possible if single spore analysis tools are available both at the ultrastructural and at the (molecular)physiological level. In this seminar we will present single cell live-imaging tools for Bacillus spore germination and outgrowth as well as illustrate how these can be used to generate data for microbiological risk assessment.

Operationalising Factors That Explain the Emergence of Infectious Diseases

NORVAL STRACHAN, University of Aberdeen, Aberdeen, United Kingdom

Most emerging infectious diseases are of zoonotic origin and spill over from animal reservoirs to humans, followed to a lesser or greater extent by cycling in the human population. These emerging diseases include the majority
of the major foodborne pathogens. Factors for disease emergence include: ecological changes (including those due
to economic development and agricultural land use); human demographics and behaviour; international travel and
commerce; technology and industry; microbial adaptation and change, and breakdown in public-health measures.

This presentation will provide examples of these factors for some of the key foodborne pathogens (e.g.,
Campylobacter, E. coli O157 and Listeria) and show how they can be made operational for understanding the changes
in incidence of these diseases. The methods involved will include: empirical epidemiology; case-case and case-control
studies; time series analysis; microbial sub-typing (source attribution, diversity, genetic distance) and quantitative microbial
risk assessment. In addition, examples will be given of the implementation and challenges to integration of these disease
emergence factors in quantitative risk assessment.

S21 Recent Advances in Food Packaging to Ensure Quality and Safety of Foods

Quality, safety, sustainability, and traceability are of high value to the food industry. Since packaging is the final
step in food processing before foods reach consumers, packaging integrity and functionality has significant impact on the
quality and safety of foods. Major technological advances have occurred over the last decade in materials sciences that
have a direct bearing on food packaging. Some of these advances include bio-polymers with customized barrier properties
and predictable environmental degradation patterns, branched 3D embedded networks, chemical-free sterilization of
packaging membranes, and nanomaterial impregnated packaging. The overall theme of this symposium is on the
technological advances taking place in food packaging. The purpose of this symposium is to provide food microbiologists
with an overview of advances taking place in food packaging that are designed to enhance the safety and quality of foods.
The symposium brings together speakers from academia and industry to highlight current research and commercialization
of enhanced active packaging concepts, chemical-free sterilization of packaging membranes, antioxidant immobilization
and enhanced modified atmosphere packaging concepts.

Advanced Active Packaging Concepts
SARA LIMBO, Department of Food, Environmental and Nutritional Sciences of the University of Milan, Milan, Italy

Active packaging is an area in which the most recent innovative ideas have been applied to satisfy the
requirements of safety, quality preservation and market globalization of foods. In this communication, the main principles,
mechanisms of action and technologies at the basis of the most common active packaging solutions will be presented and
discussed. Some specific considerations concerning advantages, limits and perspectives of active solutions integrated
in polymers will be also presented. The role of active packaging in food waste reduction along the supply chain will
be discussed, starting from some case studies and researches in this area. The presentation will be concluded with a
discussion on regulatory requirements in EU and some remarks concerning the future trends of active packaging.

Advanced Modified Atmosphere Packaging Concepts
ALAN CAMPBELL, Campden BRI, Chipping Campden, United Kingdom

Modified Atmosphere Packaging has been utilised for a large number of years. During that time we have seen
numerous changes in packaging formats covering a wide range of products. The presentation will include packaging
formats that fall under the MAP banner which includes those products that may be vacuum-packed.

The presentation will cover the changes in packaging materials and formats that are occurring within Europe.
This will include a background to current applications and gas mixtures followed by new developments. These new
developments cover all products and will relate to both safety issues as well as extension of shelf life. Where the use of
MAP can be enhanced by other technologies, such as Active/Intelligent packaging, these techniques will be highlighted.

Grafting and Crosslinking Polymers for Enhanced Packaging Purposes
SHIMA SHAYANFAR, National Center for Electron Beam Research, College Station, TX; Texas A&M University, College
Station, TX

Polymers play an essential role in delivering wholesome and safe food to consumers. However, one must
envision additional applications for polymers rather than just serving as a physical barrier. Such an approach opens up
novel applications that use packaging as a physical platform to incorporate additional, high value purposes. Polymers
can be strengthened by crosslinking using eBeam technology for different applications. Additionally, ionizing radiation
technologies have been used to graft materials onto polymers. Since the food industry is the largest user of packaging
materials, it is critical that food safety specialists consider the potential applications of packaging materials in addition to
just providing a physical/chemical barrier.

S22 A Benchmarking Study of the Traceability Regulatory Environment in 21 OECD Countries

This session will discuss a report published by the Global Food Traceability Center in 2014 on current
international food traceability standards and regulations for 21 major OECD countries. The goal of the session is to open a
dialogue concerning harmonization of food traceability requirements, so that stakeholders in the food system can mitigate
conflicting requirements and reduce unnecessary costs. The results of the benchmarking study will be presented to show
how countries in the EU stack up against those in North America, Asia and elsewhere. You will hear about the reasons
for the high levels of variability in regulations between these countries. Then the session will review possible solutions
from GS1 to overcoming the challenges, gaps and needs identified within the report. What are the lessons we can learn
from the development of the Hazard Analysis and Critical Control Points (HACCP) system for food safety? What industry
models are being considered for more harmonized traceability? What best practices are being used? Do we need a
model for food traceability similar to that of the Global Food Safety Initiative (GFSI)? How do we address the question
of harmonization of regulations? The session will attempt to answer some of these questions.
This presentation will review the results of investigation into the food traceability regulations of 21 separate countries, the efforts by governments to apply common requirements for food traceability, as well as a review of research into the utility and impact of traceability in the global seafood industry. Lastly, the presentation will conclude with a look into the near future and efforts underway to design and develop a harmonized and interoperable technology architecture for global food traceability.

### Differences in Regulatory Environments around the World: Embrace or Eliminate?

**ESTEFANÍA NORIEGA FERNÁNDEZ**

Exploring Bacterial Colonies in Solid (Model) Foods

Non-destructive *In Situ* Techniques to Monitor Bacterial Colony Dynamics in Solid (Model) Foods

Planktonic cells, typically found in liquid systems, are routinely used in predictive microbiology to, e.g., assess the effect of environmental/operational conditions on microbial growth or the efficacy of food preservation technologies. However, freely suspended cells often show different susceptibility to environmental hurdles than cell colonies developed in solid matrices. Limited oxygen, water and nutrient availability, metabolite accumulation and physical constraints derived from cell immobilization in the solid matrix, are main factors affecting microbial growth. Additionally, intra- and inter-colony interactions, as a consequence of the initial microbial load in solid systems, may also affect microbial physiology. Predictive food microbiology approaches are moving towards a more realistic resemblance to food products, which involves studies in structured solid systems instead of liquids. Since structured systems promote microbial cells to become immobilized and grow as colonies, it is essential to study the colony behavior not only for food safety assurance systems, but also for understanding cell physiology and optimizing food production processes in solid matrices. Traditionally, microbial dynamics in solid systems have been assessed with a macroscopic approach, by applying destructive analytical techniques, such as viable plate counting, which yield information about overall populations. In the last years, this approach is being substituted by more mechanistically-inspired ones at mesoscopic (colony) and microscopic (cell) levels. Therefore, non-destructive and *in-situ* monitoring is mandatory for a deeper insight into bacterial colony dynamics. Different methodologies that enable high-throughput data collection have been developed to characterise colony growth and the local environment, such as microscopy-based techniques coupled with image analysis, OD-based measurements in microplate readers and microelectrode-based techniques. This symposium will provide an overview of non-destructive *in-situ* techniques to monitor bacterial colony dynamics in solid (model) foods and will emphasize their advantages and inconveniences in terms of accuracy, performance and output information.

### Differences in Regulatory Environments around the World: Embrace or Eliminate?

**ESTEFANÍA NORIEGA FERNÁNDEZ** and COLINE DONON

(1) Global Food Traceability Center, Washington, D.C.
(2) GS1 Global, Brussels, Belgium

**Best Practices for Achieving Harmonization Using Standards**

**COLINE DONON**, GS1 Global, Brussels, Belgium

Is it possible for the business to leverage its traceability system towards control authorities? The use of standards is one answer. This presentation will examine how global standards can support a company’s needs, including complying with legal traceability requirements. How do you make sure that your business is prepared to face the worst case food traceability scenario? What other drivers of traceability can be supported by industry standards? The GS1 Global Traceability Standard is aimed at supporting companies in this effort by reducing resources and costs allocated to their traceability management system and practices. A review of case studies will illustrate how standards can support businesses in meeting actual and future traceability challenges.

**Global Food Traceability – Will It be Regulation or Collaboration?**

**BRIAN STERLING**, Global Food Traceability Center, Washington, D.C.

Differences in Regulatory Environments around the World: Embrace or Eliminate?

**ESTEFANÍA NORIEGA FERNÁNDEZ** and Jan Van Impe, KU Leuven, Leuven, Belgium

Exploring Bacterial Colonies in Solid (Model) Foods

Despite all EU efforts to tackle food poisoning and spoilage, about 5,400 outbreaks and 1.3 billion ton food waste are reported annually, with the significant healthcare and economic impact. Consumer demand for minimally processed foods and the shortcomings of existing preservation technologies have encouraged a constant seeking for alternative strategies that assist in improving food safety and quality assurance systems, e.g., “hurdle technology”-based approaches that integrate decision supporting tools for the management of mild processing technologies. However, the roles of strategies that assist in improving food safety and quality assurance systems, e.g., “hurdle technology”-based approaches in solid matrices. Limited oxygen, water and nutrient availability, metabolite accumulation, physical constraints and colony interactions may affect cell physiology, as a consequence of the solid environment. Moreover, a macroscopic approach has traditionally been used to assess microbial dynamics in solid systems, e.g., by means of viable plate counts, which yield information on overall populations. In the last decade, this approach has shifted towards more mechanistically-inspired ones at mesoscopic scale, e.g., subpopulation/colony level, and even at the microscopic or intracellular scale. Together with this trend, the need for robust methods that allow rapid data collection at the meso- and microscopic scale, has emerged. An overview of high-throughput techniques to monitor bacterial colonies is provided in this presentation, with emphasis on microscopy techniques coupled with image analysis and optical density measurements.
**Use of Image Analysis as a Non-invasive Tool to Model Microbial Communities on Solid Surfaces**

**PANAGIOTIS SKANDAMIS**, Agricultural University of Athens, Athens, Greece

Microbial growth in foods occurs in the aqueous phase. The structural characteristics (also called ’micro-architecture’) of this phase, in combination with the total concentration and dispersion of water compared to fat phase determine the form and rate of growth, i.e., the spatiotemporal microbial dynamics. The development of sophisticated image analysis systems for real-time monitoring of single cell division (or spore germination) under the microscope, during continuous exposure of attached cells to flowing liquid media, allowed further insight in the variability assessment of single cells. The variability in colonial growth associated with intra-colony cell-to-cell interactions cannot be quantified in liquid cultures, nor by direct imaging of cells, when the daughter cell is removed after division, as occurs in the presence of flowing media. Direct time-lapse imaging of microbial populations growing on agar surfaces of different intrinsic properties has enabled the characterization of population heterogeneity taking into account the interactions between adjacent cells. It may also depict the history of cells residing in different sites of a colony and their physiological adaptations, resulting from exposure to stresses, such as starvation or anoxia and affecting their subsequent resistance to inimical factors. Experimental protocols for direct imaging of surface-growing cells include the gel-cassette system, systems comprised of an agar layer on top of a microscope slide, covered by a cover slip, sealed with paraffin wax and placed under the microscope and more recently, the anopore strips. Finally, using multi-spectral imaging, it is possible to indirectly monitor colonial growth through the impact of bacterial metabolism on the chemistry, texture and colour of food. This is of particular significance for the assessment of heterogeneity in the onset of spoilage in different areas of food surfaces (e.g., containing different fat content, wounded tissues, etc.) such as in meat and fresh-cut salads.

**Microscale Description of the Food Characteristics Surrounding Bacterial Cells Relevance to Describe the Variability of the Individual Cell Behaviour**

**VALERIE STAHL**
(1) Aérial Technical Institute for Food Industry, Illkirch, France
(2) ENVA, Paris, France

Considering that pathogenic microorganisms generally contaminate foods with a few cells, several studies have investigated the individual cell behaviour to describe more accurately the variability of cell lag times and cell growth probability. These results highlighted the significance of the variability of the individual cell behaviour in the context of exposure assessment.

An individual-based modelling approach was developed to describe the behaviour of a few *L. monocytogenes* cells contaminating smear soft cheese surfaces and vacuum-packed cold-smoked salmon slides. As the microbial behaviour is also highly dependent on the variability of the food characteristics, the spatial and temporal variability of pH and water activity was described at a micro-scale level, with different batches of smear soft cheese and cold-smoked salmon.

The microscale pH of food surfaces was determined with a miniaturized 50 μm diameter pH electrode. The microscale *a*w of food surfaces was estimated using a cryoscopic micro-osmometer. A calibration curve linking the osmolality of cheese and salmon extracts with the *a*w measured with the dew-point analyser was established.

A specific technique to artificially contaminate food portions with a few cells was preferred to the usual protocol performed with a bacterial suspension to avoid altering the micro-local food environment. Irradiated food matrices artificially contaminated were used to validate model predictions.

The individual-based modelling approach combined with a description of the microenvironment surrounding contaminating cells was shown more effective than the traditional population/macroenvironmental one to describe the actual bacterial behaviour variability when food surfaces are contaminated with a few cells.

**Acknowledgment:** R. Ferrier was the recipient of a doctoral fellowship from the French ANRT (Association Nationale de la Recherche Technique). Studies on the microscale description of foods were supported by CNIEL (F), CITPPM (F) and ACTIA (F).

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**S24 Food Safety Decisions – Tools and Tips for Food Producers of Ready-to-Eat Foods**

Food providers make daily decisions regarding suppliers, ingredients, process conditions, distributors and customers. Even at “business as usual conditions” such decisions are complex and challenging. Several risk assessment tools and models have been developed during the last decades, but surprisingly, these are little used by the industry. In the STARTEC project, a mapping of decision and production processes was carried out. An outcome was that decision support is needed when tradeoffs between safety, quality and costs have to be done, both at the strategic level and in day-to-day decisions. We have developed a framework of a simpler but multidisciplinary tool which has the potential to support tradeoff decisions in the industry. The basis for the tool is the process flow chart of specific products and simulations where each parameter in the tool are categorised in a traffic light system. The workshop will present some of the studies in the project and present the prototype STARTEC IT tool: • Ingredient and technology choices for food safety • The STARTEC tool and guidelines for food producers • Developing tools for food safety decision-making - challenges and recommendations • ‘A day in the life’ – a food company shares its wisdom • The workshop is organized by the STARTEC project, which is a three-year EU-funded project which started in 2012, to develop a food safety decision support tool for SME manufacturers of ready-to-eat foods. STARTEC involves very close collaboration between food-producing SMEs, research organisations, and universities. In addition to the tool, STARTEC has developed guidelines for the food industry, based on the most relevant research findings from STARTEC and practical experience and inputs from industry. STARTEC Web site: www.startec-eu.info.
In the first part of the presentation, STARTEC presents an IT-based solution to help RTE meal business operators make decisions by estimating product safety, quality, nutritional value and shelf-life, as well as cost and benefits of production. The tool is developed to prototype level, and consists of database and prediction parts. The database allows business operators to build a portfolio of their products, including metadata and a uniform, formalized description of the product flowchart. The simulation part allows defining models. The system offers the possibility of calibrating these models using the data in the database.

As opposed to other readily available IT-based tools, STARTEC takes a multidisciplinary approach to decision-making, and it is there where its novelty lies. The IT tool is developed by the project partner IRIS, with support from all project partners. The second part of the presentation focuses on ingredient and technology choices for ensuring food safety, quality, and nutrition content of potato salad.

Experiments were carried out in the lab with simulated industry potato salads with more than 40 variations of ingredient composition, inoculated with 

Listeria monocytogenes. The results showed that the salad compositions, process and storage scenarios could be used to predict the level of pathogen levels within categories based on the content of lactic acid bacteria and protein rich ingredients in the salad. The data visualized how the salad composition could be adapted to improve the safety even at abuse temperature, and which formulations or deviations that lead to a change of food safety category and thereby a need for corrective action. Some guidelines for production of potato salads with less fat and for formulation of products suitable for hospitals will be given.

Definition of Safety Criteria in RTE Products: A Whole Chain Approach from Ingredient Selection up to Consumption

ALESSANDRA DE CESARE, Alma Mater Studiorum, University of Bologna, Bologna, Italy

The Performance Objective (PO) is a risk management concept we should become familiar with in the next future. The achievement of a PO for a target microbiological hazard in a specific food product should help food industries to put on the market lots compliant to the Food Safety Objective (FSO) defined by food authorities for that microbiological hazard at the time of consumption. Each PO must be calculated for specific ingredients according to the distribution of the microbiological hazard in those ingredients. Furthermore, the impact of each single production step and storage conditions on the hazard in the food up to consumption must be assessed. In this study the approach to derive PO for 

Bacillus cereus (BC) and 

Listeria monocytogenes (LM) in selected ingredients to be added in RTE mixed spelt salads, packaged under air or modified atmosphere, with a shelf life of 12 days, is presented. The PO values to meet a FSO for BC of 4 log CFU/g in spelt salads stored refrigerated under air or MAP for 12 days corresponded for spelt to -0.64 and 0.22 log CFU/g, respectively; for cheese, to -2.22 and -1.36 log CFU/g, respectively. The PO values to meet a FSO for LM of 2 log CFU/g in spelt salads stored refrigerated under air or MAP for 12 days corresponded for celery to -4.18 and -2.71 log CFU/g, respectively; for cheese, to -3.43 and -1.96 log CFU/g, respectively. The approach presented can be easily adapted to different FSOs and changing assumptions.

“A Day in the Life” – A Food Company Shares Its Wisdom

CECILIE FROM, Matbørsen, Stokke, Norway

It would not have been possible to develop the STARTEC IT tool without willingness from the industry partners to share experience about their real needs, production conditions and how decisions are made today. In this presentation Matbørsen will share their experience from everyday management of food safety and quality in a medium sized food production plant. Every day a quality manager has to deal with a wide variety of topics concerning both safety, quality and cost. Decision-making is fast and the consequences of making wrong decisions can be severe. Experience from being a partner in an EU-funded project and future work will also be discussed.

Developing Tools for Food Safety Decision-making – Challenges and Recommendations

MATTHIAS FILTER, BfR, Berlin, Germany

Software tools are getting more and more important for decision-support in food industry and governmental bodies. In the field of food safety a broad range of tools covering domains like epidemiological modelling, predictive microbiology and risk assessment have been developed in recent years. This talk will provide an overview on currently available food safety modelling software, summarize recent trends in tool development and point to still unresolved challenges for software developers and end users. On the basis of a risk assessment authority’s experience in several national and international software development projects recommendations will be given which could support ongoing and future software projects in the field. Examples of successful community driven software projects illustrate the benefits of the proposed strategy.
In Memory

Louise Fielding
B.SC., Ph.D., FIFST, FHEA
1968–2013

The 11th IAFP European Symposium on Food Safety is dedicated to the memory of Dr. Louise Fielding. Louise was an active member of IAFP, attending eleven Annual Meetings in North America and most of the European Symposia, many times as a presenter. Louise was a true champion of “Advancing Food Safety Worldwide.” She served as the President of the United Kingdom Association for Food Protection (UKAFP). Louise was the Director of Research at the Cardiff Metropolitan University School of Health Sciences. She had dreamed of the day that the IAFP European Symposium would be held in Cardiff.
Technical Abstracts
The Use of Microbial Flora Analytical Tools for the Discrimination of Organic Foods

CELINE BIGOT
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**Introduction:** The increasing interest for organic products made consumers exposed to inconsistencies. Consumers believe that organic products are safer and more nutritious than conventional products while there is no scientific evidence yet. For example, the fake Italian organic products sold around Europe in 2011, prompted questions about the credibility of the organic industry. The EU Regulation No 178/2002, applied since January 2005, imposed traceability of foodstuffs as a compulsory element of the consumer safety "farm to fork." This regulation is also applied to organic farming that differs from the others by a regulated use of chemical inputs as fertilizers and pesticides. But traceability of foods is mainly done at the administrative level, and the use of analytical tools is rare.

**Purpose:** Our study is based on the hypothesis that treatments associated to various farming types have a measurable effect on food microflora. That is why the main objective of this study was to use the microbial environment of foods to discriminate them according to their production mode.

**Methods:** The application of molecular microbial ecology techniques such as PCR-DGGE and real-time PCR/HRM showed that they could serve as discrimination tools using bacterial and fungal rDNA in a quick and cost effective way.

**Results:** The analysis of microbial genetic profiles of nectarines, peaches and apples showed that the observed differences between organic and conventional apples were significant enough to conclude that they exclusively originated from the applied treatments: It was possible to verify the robustness of our methodology by comparing results obtained on two successive harvest years for apple samples and we estimated the "intra-plot" variability and observed that organic apples could be discriminated from conventional ones independently on their position in the field (center or border).

**Significance:** We proposed this analytical tool as a first step to control and authenticate organic foods.

A Method to Validate Bacterial Thermal Destruction Taking into Account Recovery Conditions in Food Matrix

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**Introduction:** Observed heat resistance of microorganisms depends on environmental conditions (temperature, pH and $a_w$) encountered both during heat treatment (pre-treatment conditions) and recovery (post-treatment conditions). The effects of these factors have been described by mathematical models and quantified using a modular approach. The model parameter values were obtained for both bacterial heat treatment and recovery in laboratory media.

**Purpose:** The objective of this study is to present an original methodology to validate in food matrix models, determined parameter values quantifying thermal bacterial inactivation including both heat treatment and recovery conditions.

**Methods:** This method was applied to validate the effect of alkaline pH on the heat resistance of *Salmonella Enteritidis* MJG01 in egg whites. Heat treatments were performed at 56°C for different times using capillaries and then incubated 6 days at 37°C. The presence or absence of surviving bacteria was determined by bacterial count (inclusion in nutrient agar). Knowing the initial bacterial population as well as the treatment times that frame the first capillary where no growth is occurred was sufficient to estimate D-values in egg white.

**Results:** Using this method D$_{90°C}$ values were determined in white eggs for 2 different pHs to each a complete bacterial destruction. At pH 7, D$_{90°C}$ is determined between 6.03 and 7.04 min. While at pH 9.2, D-values are between 0.31 and 0.36 min. These experimental values determined in egg whites were compared to calculated D-values using the developed model and associated parameters values. Calculation yields a D$_{90°C}$ values between 0.69 and 0.80 min which is quite close to experimental values determined in egg whites at a pH 9.2.

**Significance:** This original method allows to validate model parameter values as well as determine the impact of the food matrix on bacterial heat resistance.

Acknowledgment: Study supported by the ADRO Ouest Association.

Impact of Pooling Samples on the Detection of *Listeria monocytogenes* in Food

NATHALIE GNANOU BESSE1 and Jean Christophe Augustini2
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(2) ENVA, Paris, France

**Introduction:** EN ISO 6887-1 Standard on samples preparation for microbiological analyses, allows to pool test portions or enrichment broths (wet pooling) before subsequent analysis, to reduce analytical costs, provided detection performances are not affected.

**Purpose:** We investigated the impact of wet pooling on the performance of the Standard EN ISO 11290-1 to detect *Listeria monocytogenes*. In particular, a model describing *L. monocytogenes* growth during pre-enrichment in half-Fraser broth was developed.

**Methods:** Three sources of variability were considered for simulations: initial contamination level, variability of $\mu$max and individual cell lag time distributions. To describe growth rates variability, we performed a uniform random drawing among 34 *L. monocytogenes* growth rates published or obtained previously. We deduced *Listeria* lag time distributions from DuPont and Augustin study (2009), allowing to assess the influence of stress on single-cell lag time for *L. monocytogenes* in half-Fraser broth. Uniform random drawing was performed among the 12 physiological states described. For validation purpose, *L. monocytogenes* concentrations obtained after pre-enrichment of 86 naturally contaminated products were compared to levels predicted. We obtained good agreement between observed and simulated microbial counts after pre-enrichment. Significant differences with observed values were found if lag time variability and initial physiological state were not considered, which confirmed their major impact on growth.
Introduction: Immuncapture (IMC) is used for the concentration of pathogenic bacteria from food product to facilitate their detection. Immunomagnetic separation has been traditionally the reference method for capturing foodborne pathogen bacteria.

Purpose: We have developed a new method for the immunocapture of pathogenic bacteria based on 96-well microplate coated with specific antibodies. The objective of this study was to demonstrate the efficiency of this method in capturing E. coli O157 in enriched food samples.

Methods: After adding 100 µL of enrichment broth (BPW) into the wells, antibodies are capturing present target bacteria. Then, after a washing step, a subculture is performed (3 to 5 h) in the wells with addition of fresh enrichment medium (100 µL mTSBn) for increasing the number of target bacteria. The efficiency of the system was assessed by performing immunocapture on food samples (ground beef / raw milk cheese, 25 g sample size), artificially contaminated with E. coli O157:H7. Naturally contaminated beef samples found positive by PCR screening were also confirmed by IMC plate. Tests were also done with magnetic beads.

Results: For ground beef samples, IMC with microplate allowed the capture of about 5.4 10⁵ CFU of E. coli O157 when magnetic beads recovered 2.7 x 10⁴ CFU. For both raw milk cheeses, IMC microplate captured about 3.2 x 10⁵ CFU (goat cheese) and 2.6 x 10⁵ CFU (Mont d’Or) of E. coli O157 when magnetic beads recovered about 6.2 x 10⁴ CFU (goat cheese) and 2.7 x 10⁴ CFU (Mont d’Or). IMC microplate allowed the confirmation of 33 out of 38 positive PCR screening for the naturally contaminated meat samples.

Significance: IMC microplate was shown effective for the capture isolation of E. coli O157 bacteria from enriched meat samples. The subculture step done into the wells, after immunocapture, contributes to a good confirmation rate.

T1-05 Discrimination of Saccharomyces cerevisiae and Non-Saccharomyces Yeasts Isolated from Spanish Grapes by Attenuated Total Reflectance Infrared Spectroscopy Combined with Multivariate Analysis
Miquel Puxeu¹, Imma Andorra³, Anna Brull¹ and SILVIA DE LAMO²
(1) Parc tecnològic del Vi (VITEC), Falset, Spain
(2) Universitat Rovira i Virgili, Tarragona, Spain

Introduction: Traditionally in the winemaking sector, Saccharomyces cerevisiae has been used for their oenological properties to successfully produce alcoholic fermentations. Lately, other indigenous Saccharomyces and non-Saccharomyces species have been also considered to make wine due to their impact on sensory attributes. Attenuated total reflectance infrared spectroscopy (ATR-FTIR) provides bands from all the cellular components of microorganisms, mainly from cell membrane and cell wall that permit the classification of microorganisms.

Purpose: The main objective of this research was to show the potential of attenuated total reflectance infrared spectroscopy (ATR-FTIR) combined with soft independent modeling of class analogy multivariate analysis to rapidly discriminate between S. cerevisiae and non-Saccharomyces species.

Methods: Forty indigenous S. cerevisiae and non-Saccharomyces strains isolated from five different grape varieties (Albariño, Tempranillo, Grenatxa, Xarel-lo and Carinyena) obtained from different Spanish wine regions were isolated after grape must spontaneous fermentation and grown in YPD agar and Lysine media at 28⁰C for 48 h. The identification procedure was done by 5 elements for Saccharomyces strains and by rDNA-RFLP for non-Saccharomyces strains. Thirty isolated strains were grown in grape must at 28⁰C for 24 h and were centrifuged and washed with saline solution to remove interfering compounds. Each pellet was placed onto ZnSe crystal and a minimum of eight spectra per sample were collected in ATR mode in mid-infrared region using a diamond crystal. Spectral data were analyzed by soft independent modelling of class analogy (SIMCA) and the models built up were validated with ten “unknown” samples.

Results: SIMCA models effectively discriminated between S. cerevisiae and non-Saccharomyces species and the major contribution to their discrimination was linked to IR bands related with components mainly in their cell wall, mannans and glucans.

Significance: ATR-FTIR could be used as a simple, fast, and reproducible technique to discriminate S. cerevisiae and non-Saccharomyces species.

T1-06 Pretreatments for Estimation of Viable Viruses through Reverse Transcription-qPCR in Shellfish: Pros and Cons
SILVIA MONTEIRO and Ricardo Santos
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Introduction: Enteric viruses are introduced in the environment through diverse routes including direct discharge of treated or untreated sewage effluents and urban and rural run-off. The wastewater treatments do not ensure complete removal of these microorganisms and therefore they are released into marine and estuarine waters. Shellfish filter pollutants from contaminated water, bioaccumulating them in their edible tissues. Shellfish contaminated by viruses inadequately cooked or eaten raw pose a major public health concern. As a result, the need for standardized, reliable and affordable methodologies for the detection of viruses such as noroviruses (NoV) and hepatitis A viruses (HAV) was created. In 2013, ISO methods were created for the aforementioned viruses (CEN/ISO TS 15216-1 and CEN/ISO TS
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Monday, 20 April – 15.30 – 17.00

T2-01 Food Traceability Method Employing Tagged DNA-based Barcodes
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Introduction: In this paper we discuss the development of a food traceability method employing tagged DNA-based barcodes. The barcodes are applied directly on the food surface or are mixed into the bulk, thus maintaining full traceability even if the product packaging is lost.

Purpose: The objectives of this project were to (a) design a simple barcoding scheme capable of encoding all the necessary traceability information (b) develop an application method that does not require significant incremental infrastructure (c) demonstrate that the barcodes can survive typical supply chain environmental conditions for a variety of commodities (d) demonstrate that traceability is maintained even when products of multiple origins are commingled (e) evaluate the feasibility of using the same method to detect adulteration of certain products such as olive oil, honey, wine, etc.

Methods: We employed an array of distinctly tagged DNA-based tracers called DNATrax, developed at Lawrence Livermore National Laboratory. The biological material can be mixed with existing food coating products (such as carnauba wax, silicone oil, etc.) and sprayed directly on the product or it can be encapsulated in maltodextrin, salt, starch, silica nanoparticles or other media and added directly to the bulk. Standard PCR methods allow a quick readout of the traceability information.

Results: We demonstrated that (1) that the barcodes outlasted the shelf life of several products, under typical supply chain environmental conditions; (2) we can recover the barcodes and identify the origin of commingled products such as apples and leafy greens and liquid products such as olive oil; (3) it is feasible to use digital PCR methods to detect adulteration.

Significance: Presently, traceability investigations frequently take several weeks to complete. The present method will allow rapid containment of food fraud and mitigation of risks to public health due to contaminated products and foodborne illnesses.

T2-02 Thinking Inside the Box: Using Cartoon Strips to Teach Food Defense at the Retail Level
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Introduction: Food supplies at the retail level, the final point of preparation and service to the consumer, are a point of vulnerability for malicious contamination. However, few user-friendly resources exist to engage and train frontline food workers in food defense. Available materials on the topic are lengthy, complex, full of jargon and not targeted to the average food handler. This project aimed to create targeted food defense resources that address areas of challenge to the retail food community: culturally and linguistically diverse workers, marginal foodhandler interest, and limited time to train.

Purpose: A series of printed multi-lingual training cartoons was developed under an FDA Innovative Food Defense Grant to depict a variety of plausible scenarios where a conscientious worker can take a small action to prevent potential injury or illness due to intentional contamination. Designed for use in a variety of approaches and settings by regulators and the foodservice industry alike, these materials are applicable for use during regulatory food inspections, for just-in-time trainings during emergencies or special events, for in-service trainings and/or for inclusion in food safety classroom settings.

Methods: With input from regulators and foodservice workers, a framework was developed to address the perceived susceptibility, severity, benefits and barriers, cues to action and self-efficacy aspects of food defense awareness. Storyboards of the issues were developed, and partnering with a cartoonist, four plausible scenarios were illustrated: unknown food sources; illness symptoms, employee contamination and suspicious persons.

Results: Introduced to regulators throughout 2013/2014, evaluations show that 83% would use these training materials in the course of their job. Reception from foodservice management was also positive and use of these materials in classroom settings increased engagement and interest of foodhandlers.
T2-03 Pig Herds Free from Campylobacter – Dream or Reality?
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Introduction: As Specific Pathogen-Free (SPF) pig herds are designed and managed to prevent specific pig diseases, it might be feasible to expand the list of microorganisms also including zoonotic pathogens such as Campylobacter coli since this agent has its origin in pigs.

Purpose: Three nucleus and seven multiplying SPF-herds were surveyed for Campylobacter to investigate whether the Norwegian SPF pig health and breeding pyramid might also be free from this agent.

Methods: In total, 625 fecal samples were collected from weaned piglets (n = 10), sows (n = 10), and slaughter-age pigs (n = 20) from each farm. Campylobacter was cultured and isolated following the Nordic Committee on Food Analysis.

Results: In conclusion the intervention of Campylobacter at the herd level might be possible as four of 10 SPF herds tested negative in two sets of samples. The four negative herds were all located in remote areas several kilometers away from conventional pig farming while the positive SPF farms were all situated in neighborhoods with conventional pig production.

Significance: According to the results in our study, even the high level of biosecurity in the SPF herds did not seem to be sufficient to keep Campylobacter out of 6 of the 10 herds we tested. It seems more difficult to control Campylobacter than some specific animal disease agents and another significant zoonotic agent, Yersinia enterocolitica, in pig herds. Anyway the fact that one nucleus herd was associated with three negative multiplying herds is a promising observation. But we did not observe obvious differences in the on-farm management practices that would be essential to practical management recommendations from this work. Further research is needed to better understand the efficiency of the on-farm practices and the significance of the outside environment for the control of Campylobacter in SPF herds. Might Campylobacter be used as an indicator for “biosecurity” in SPF-herds?

T2-04 Characterization of Staphylococcus aureus Isolated from Dairy Farms in Victoria, Australia
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Introduction: Staphylococcus aureus infection can range from gastroenteritis to more severe disease. Recent studies identified a population of S. aureus isolates associated with ruminants, having diverged from those associated with human infection.

Purpose: This study characterized S. aureus isolates from milk and milk filters from bovine, caprine and ovine farms, to understand population diversity and human public health relevance.

Methods: Whole-genome sequencing was performed on 13 isolates (6 bovine, 5 caprine, and 2 ovine), followed by comparative genomics of draft genomes. In silico analysis examined antimicrobial resistance and virulence markers. Population diversity was studied using MLST and PFGE analysis. Antimicrobial sensitivity against 7 antibiotics was examined.

Results: MLST identified caprine and ovine isolates as ST133, or a highly related, but previously undescribed ST differing in a single allele. Bovine isolates showed higher diversity including STs associated with human infection (ST1, ST5 and ST8) and bovine mastitis (ST705). A new ST, closely related to ST97 (implicated in human and animal disease), was identified among bovine isolates. PFGE identified 2 main groupings: one composed of bovine isolates; and a second group including caprine and ovine isolates. No isolates showed methicillin resistance, or harbored mecA. Resistance to penicillin and ceftriaxone was identified. Analysis of staphylococcal enterotoxin gene repertoires indicated lowest diversity among caprine and ovine isolates. The bovine grouping harbored 12 enterotoxins; with one bovine isolate coding 9 enterotoxins.

Significance: S. aureus isolates showed traits suggesting host adaptation to ruminants, and lacked many of the key virulence markers associated with human disease, or antibiotic resistance.

T2-05 The Challenge of Histopathology in the Detection of Illicit Treatments with Hormones Associations
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Introduction: The administration of health-risk related substances such as growth promoting agents is prohibited within the European Union. Despite the ban, the use of anabolic steroids in cattle is still practiced. Therefore, diagnostic methods are required that are specific and sensitive in detecting anabolic treatments. The use of the histological technique for the screening of the abuse of steroid hormones is performed in Italy within the framework of the National Residue Control Plan since 2008, showing high performances in detecting lesions in target organs (accessory sexual glands for sexual hormones, thymus for corticosteroids and thyroid for thyreostats), long after treatment. In recent years the diagnosis of illicit treatments in calves, has become challenging, because of the low dosages administered and the use of associations (especially androgens and estrogens).

Purpose: The aim of this study is to identify simple and reliable histological diagnostic criteria to reveal such treatment.

Methods: Eighty-six veal calves were raised under controlled conditions in two groups: 20 calves were given a weekly individual dose of 5 mg of estradiol and 50 mg of nandrolone, during four weeks; 66 calves were left untreated. Target tissues (accessory sexual glands) were collected at the slaughterhouse for histological analysis. A panel of microscopic lesions (hyperplasia, initial metaplasia, metaplasia, hypersecretion, cysts) was chosen. The accuracy of the histological examination was calculated taking into account the conditional dependency resulting from parallel interpretation of the lesions.
Results: Initial metaplasia and hypersecretion were found to be suggestive of an illicit treatment with estrogens and androgens in association. Results showed 95% (95% CI: 74% - 100%) sensitivity and 94% (95% CI: 85% - 98%) specificity.

Significance: The histological examination has proved to be a useful tool to monitor the abuse of sexual hormones and could be applied in the official control of residues of androgens and estrogens in calves.

T2-06 PRNP Analysis in Sicilian Goat Breeds for TSE Resistance and Full Eradication of Prion Strains
SERGIO MIGLIORE, Stefano Agnello, Sebastian Mignacca, Vincenzo Di Marco Lo Presti, Fabrizio Vitale and Maria Vitale
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Introduction: New concerns over risks related to Transmissible Spongiform Encephalopathies (TSEs) exposure through sheep and goats dairy products render important the full eradication of prion strains circulation. TSE control programs in sheep by breeding for genetic resistance are accomplished in several countries in Europe. In goats, recent studies provided a potential association of some PrP gene (PRNP) polymorphisms and resistance to TSEs, although further analysis is needed. Among the potential protective alleles, the Q/K polymorphism at codon 222 (Q222K) showed promising results.

Purpose: The PRNP polymorphism in Sicilian goat breeds is almost unknown and the study was aimed at the evaluation of genetic variability in autochthon breeds.

Methods: The sequence of caprine PRNP was determined in 568 goats of five breeds commonly reared in Sicily: Girgentana (n = 160), Red Mediterranean (n =153), Maltese (n = 143), Argentana dell’Etna (n = 40) and Messinese (n = 20). Genomic DNA isolated from blood was amplified by PCR and then sequenced in ABI 3130 genetic analyzer. Sequence alignment was carried out using the SeqScape software v2.5.

Results: 11 polymorphic sites were identified, G34V, V125I, M137I, I142T, H143R, R151H, R154H, P168Q, R211Q, Q222K e S240P. We found 222K variant in all breeds, among them the Girgentana and Red Mediterranean breeds showed a highest frequency: 18% and 15%, respectively. The results showed that the realization of breeding programs for TSE resistance in these breeds might be easier to accomplish compared to other European breeds in which K 222 is not present at such high level.

Significance: Breeding programs for TSEs resistance can reduce the risks related to TSE exposure with dairy products. The finding of significant differences among PRNP allele distributions in Sicilian goats, need to be considered for the feasibility of breeding program for TSEs resistance.

This study was supported by EMIDA project: “GOAT-TSE-FREE and RF-2010-2318525 from Italian Ministry of Health.

Technical Session 3– General Microbiology
Tuesday, 21 April – 8.30 – 10.00

T3-01 Prevalence of Human Norovirus and Bacterial Pathogens at Public Access Watershed Sites in a California Central Coast Agricultural Region
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Introduction: Human norovirus (HuNoV) is a major cause of foodborne illnesses in the United States. Public waterways can become contaminated and act as a reservoir for pathogens.

Purpose: We initiated a two-year survey of several public watersheds in a major leafy green production region of Central Coastal California to determine the prevalence of HuNoV as well as Shiga Toxin-producing Escherichia coli (STEC), Salmonella, and Listeria monocytogenes.

Methods: Moore swabs were deployed to sample environmental waters. Pathogens were eluted from swabs and measured by culture and/or molecular approaches.

Results: The overall prevalence of HuNoV, STEC O157, non-O157 STEC, Salmonella, and L. monocytogenes was 24%, 7.85%, 11.5%, 67.1%, and 50.5%, respectively. The prevalence of HuNoV and bacterial pathogens varied considerably between individual watersheds. However, there was no significant difference in HuNoV detection rates between samples testing positive and negative for bacterial pathogens. Among 626 samples tested by qRT-PCR, 107 samples tested positive for HuNoV of genogroup 1 (GI), and 104 samples tested positive for HuNoV of genogroup 2 (GII). Among the samples that tested positive for HuNoV, genomic copies averaged at 6.1×10⁶/l for GI, and 1.1×10⁷/l for GII HuNoV. Sequencing was able to confirm the identity of 62 samples as GI, and 20 samples as GII. Among the sequence-confirmed HuNoV samples, GI.1 is the predominant GI strain, and GII.4 is the only GII strain. The detection rate was highest in fall (35.3%), followed by winter (24.3%), spring (28.8%), then summer (2%). The predominant HuNoV genotype detected in 2012 was GII (18%), and in 2013 was GI (42.5%).

Significance: This is the first report toward measuring key bacterial and viral pathogens at the same sampling time in a major leafy green production area. High prevalences of bacteria and HuNoV were identified, which indicates potentials for contamination of produce fields if waterways flood after heavy rain events.

T3-02 Impact of Pulsed Light on Listeria innocua – A Viability Profile
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Introduction: Pulsed light is an upcoming non-thermal decontamination technology for surfaces of food and packaging material. Considerable research has been conducted in order to determine the inactivation efficacy against various microorganisms. Conventional plate count methods have usually been used to quantify the microbial reduction. Apart from the cultivability of microorganisms, the impact on the cellular level has not been investigated in detail so far. However, it is
known that stressed microorganisms may enter a “viable but not culturable” state, which could still pose some risk in case of pathogenic bacteria.

**Purpose:** This work aimed to investigate the effect of intense light pulses on selected structural and physiological properties of *Listeria innocua*, which is often used as a surrogate for the pathogen *Listeria monocytogenes*.

**Methods:** Additionally to the conventional plate count method, the effect of pulsed light on the cells respiratory activity, esterase activity, membrane potential, glucose uptake activity and membrane integrity was investigated. Furthermore, the integrity of the bacterial genome was studied by RAPD-PCR and qPCR. The impact of photo-reactivation was also assessed.

**Results:** The obtained results show that *Listeria innocua* exhibits substantial cellular activity even when the colony count was reduced by more than 4 log units after treatments with intense light pulses. The esterase activity and membrane integrity were almost unaffected, while the respiration activity, membrane potential and glucose uptake were disrupted only at high energy doses. Damages to the DNA molecule were already detectable at low energy doses. The occurrence of photo-reactivation has also been proven.

**Significance:** This study shows that distinct differences between the results of conventional plate count methods and direct viability markers of bacteria may arise after treatments with intense light pulses. In case of pathogenic bacteria, further research is needed to investigate if such germs could still pose a risk for human health.

**T3-03 Quantification of *Salmonella* Transfer on Tomatoes and Tomato Bedding**

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**Introduction:** *Salmonella* has been linked to foodborne outbreaks involving tomatoes. To reduce the risk of contamination, tomatoes that have fallen from the vine and have touched soil are typically not harvested. Tomatoes that have touched the plastic sheeting covering the soil (commonly called mulch) may be harvested.

**Purpose:** This study quantified the amount of *Salmonella* transfer between tomatoes and tomato bedding (soil, new plastic mulch, and used plastic mulch) to aid in *Salmonella* risk management decisions in the field.

**Methods:** Transfer studies were conducted in Florida, Maryland, and Ohio with soil and tomatoes common to each respective region. A five strain rifampicin resistant *Salmonella* cocktail was used to inoculate either the tomatoes or the tomato bedding. The contact of the tomato with the soil was initiated when the inoculum was wet, dried for 1 h, or dried for 24 h. The contact time was either a brief touch (1–5 s) or 24 h. *Salmonella* were enumerated on tryptic soy agar with rifampicin and by enrichment when below the direct plating detection limit.

**Results:** The transfer of *Salmonella* from both mulch materials was greater than the transfer from soil. The transfer from both mulches to tomato was between 0.5 and 2 log percent in all three states, for both wet and 1 h dried inoculum. The log percent transfer from Florida soil to tomato was between -1 and 1. The log percent transfer from Maryland and Ohio soils to tomato was below the enrichment detection limit.

**Significance:** Risk for cross-contamination of *Salmonella* from plastic mulch to the tomatoes is greater than the risk for cross-contamination from soil to the tomatoes. Policies regarding harvesting tomatoes that touch mulch or soil should be evaluated in light of this new information.

**T3-04 Contribution of the *Salmonella enterica* sv Typhimurium Capsular Transcriptional Regulators rcsA and rcsB to the Persistence of *Salmonella* in Post-harvested Tomatoes**

**MASSIMILIANO MARVASI**1, Max Teplitski2, Marcelo Farias2 and Keith Jenkins2

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**Introduction:** As outbreaks of produce-associated gastroenteritis continue to occur, we are coming to recognize that little is still known about the ecology of *Salmonella* outside of its animal hosts. It is now clear, that the ability of *Salmonella* to persist in plants requires specific changes in the pathogen’s gene expression.

**Purpose:** We aim to define the contribution of *rcsA* and *rcsB* genes to the colonization and proliferation of *Salmonella* inside tomatoes. The *rcsA* and *rcsB* genes are both transcriptional regulators involved in the capsular polysaccharide synthesis and biofilm formation in *Salmonella*. These genes were selected based on the previous observation that *Salmonella* surface structures contribute to the survival of the pathogen in tomato fruit.

**Methods:** An *in vivo* gene reporter technology was used to document the expression of the *rcsA* and *rcsB* genes within tomatoes. The *rcsA* and *rcsB* *Salmonella* Typhimurium reporter strains were inoculated into green and red tomatoes. Expression was measured after 3 d of incubation by tracking the activity of a sequence-specific resolvase. *rcsA* and *rcsB* *Salmonella* deletion mutants were also screened for their fitness and total proliferation in green and red tomato.

**Results:** *Salmonella* *rcsA* and *rcsB* genes were significantly expressed inside of both red and green tomatoes. Within tomato fruits, populations of *Salmonella* *rcsA* and *rcsB* mutants were up to 10^6 times lower than those of the wild type. Competitive fitness of the *rcsA* and *rcsB* mutants was also significantly reduced in tomatoes.

**Significance:** The identification of these transcriptional regulators is important for the persistence of *Salmonella* in tomatoes. We expect that this characterization will lead to the identification of targets or gene markers in tomato that specifically block *Salmonella* proliferation in post-harvested fruits.
T3-05  Biofilm-forming Ability of Salmonella Typhimurium on Polystyrene Surface in the Presence of N-acyl-homoserine Lactone Molecules Produced by Hafnia alvei
Vasiliki Blana, Xenia Gkaripogkli and GEORGE-JOHN NYCHAS
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Introduction: The formation of biofilms by Salmonella enterica is likely to be affected by the simultaneous presence of indigenous microflora on the food processing equipment. This microflora may also produce quorum-sensing molecules, e.g., N-acyl-homoserine lactones (AHLs) and autoinducer-2 (AI-2), which may influence both the formation and dispersal of biofilms of pathogenic bacteria.

Purpose: To evaluate the effect of microbial AHL molecules on the formation of biofilm by Salmonella Typhimurium (SeT) on an abiotic substratum.

Methods: SeT was left to form biofilms on polystyrene microplates at 20°C for a total period of 120 h under two growth conditions (TSB and diluted TSB, dTSB). Both growth media were supplemented by evaporated AHL extracts of Hafnia alvei culture re-diluted in the aforementioned media (50% AHLs). The biofilm-forming ability was measured at different time intervals using a crystal violet assay. The presence of AHLs and AI-2 activity in the wells was monitored by biosensor-based bioassays.

Results: SeT was found to form more biofilm under limited nutritional conditions (dTSB) during the whole storage period. The addition of AHLs under nutrient rich conditions (TSB) did not significantly influence the biofilm-forming ability of the strain under the whole tested period, while the addition of AHLs in the dTSB significantly reduced the biofilm-forming ability of SeT compared to the control treatment (0% AHLs). AHL content in the wells remained stable, whereas there was less AI-2 activity detected in dTSB compared to TSB, regardless of the AHL content, throughout storage.

Significance: Quorum-sensing molecules can affect biofilm-forming ability of SeT, while their effective control may improve food safety.

Acknowledgment: The action THALIS-BIOFILMS, has been co-financed by the European Union (European Social Fund-ESF) and Greek national funds through the Operational Program “Education and Lifelong Learning” of the National Strategic Reference Framework (NSRF).Research Funding Program: THALES. Investing in knowledge society through the ESF.

T3-06  Campylobacter jejuni — A Major Public Health Concern: Integration of Genomic Data to Characterize Virulence of an Atypical Strain
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Introduction: Despite being the leading cause of bacterial enteritis in the world, Campylobacter jejuni remains insufficiently controlled. C. jejuni species include several heterogeneous strains that are present in various environments and isolated from different hosts. The recent development of Next-Generation Sequencing technologies for genome analysis enable identification of the regions that differ between genomes. This genetic diversity could explain the persistence and adaptation of the pathogen to the food chain environment or expression of pathogenicity.

Purpose: The aim of this work was to analyze the genome of an atypical strain isolated in our laboratory, C. jejuni Bf. This clinical strain is characterized by an increased resistance to oxygen and an ability to growth under aerobic atmosphere, unlike other C. jejuni strains.

Methods: After the whole genome sequencing of C. jejuni Bf, a finishing and manual annotation, a genomic comparison was performed against referent strains of C. jejuni to highlight the genomic regions potentially associated with virulence or ability to manage environmental stresses.

Results: Results have showed at least 183 unique genes for the new sequenced strain. These included six prophage sequences and four restriction modification systems, involved in the genetic variability of bacterial strains. At least seven Campylobacter plasmidic genes integrated in the chromosome were found; they are known to carry virulence factors and antibiotic resistance. Seven unique genes encoding for surface structures related to virulence were also reported, like hcpC or vgrG corresponding to the type VI secretion system, newly described and mainly related to the adaptation of pathogens in hostile environment.

Significance: The genomic investigation of this atypical strain can contribute to understand virulence and adaptation abilities of C. jejuni Bf. More generally, this work can lead to the development of genetic markers for stress or virulence allowing rapid detection of C. jejuni strains particularly virulent or resistant in food industries.

Technical Session 4 – Low Water Activity and Microbial Food Spoilage
Tuesday, 21 April – 10.30 – 12.00

T4-01  Application of a Rapid Knowledge Synthesis and Transfer Approach to Assess the Microbial Safety of Low-moisture Foods
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Introduction: Low-moisture foods (LMF) are increasingly implicated in outbreaks of foodborne illness resulting in a significant public health burden. The Codex Alimentarius Committee on Food Hygiene initiated work to develop a Code of Hygienic Practice to set standardized and comprehensive international guidance on the microbial safety of LMF.
Purpose: To inform the development of the new Codex guidelines with rigorous and transparent scientific inputs, we applied a rapid knowledge synthesis and transfer approach to review global research on the burden of illness, prevalence, and interventions to control selected microbial hazards in LMF.

Methods: Knowledge synthesis methods included an integrated scoping review (search strategy, relevance screening and confirmation, and evidence mapping), systematic review (detailed data extraction), and meta-analysis of prevalence data. Knowledge transfer of the results was achieved through multiple reporting formats, including evidence "summary cards."

Results: From eight categories of LMF products and nine microbial hazards, 'cereals and grains' (n = 142) and Salmonella spp. (n = 278) were the most commonly investigated. Salmonella spp. were implicated in the most outbreaks (n = 96, 45%), resulting in the most hospitalizations (n = 895, 89%) and deaths (n = 14, 74%) attributed to LMF. Salmonella spp. had a consistently low prevalence across all LMF categories (0 – 3%), while other hazards (e.g., B. cereus) were found at highly variable levels. A variety of interventions were investigated in small challenge trials. Key knowledge gaps included under-reporting of LMF outbreaks, limited reporting of microbial concentration data from prevalence studies, and a lack of intervention-efficacy research under commercial conditions. Summary cards for each LMF category were a useful knowledge transfer format to inform complementary risk ranking activities.

Significance: This review builds upon previous work in this area by synthesizing a broad range of evidence using a structured, transparent, and integrated approach to provide timely evidence-informed inputs into the decision making process for developing international guidelines.

**T4-02**

**Microbiological Stability of Fermented Black Olives Using Osmotic Dehydration as a Pre-fermentation Treatment and Monosodium Glutamate as a Natural Flavor Enhancer**

Olga Hondrodimou1, Anastasios Stamatiou1, Vasia Oikonomopoulou2, Eleni Gogou3, F.J. Cui4, Petros Taoukis5, Magda Krokida6 and EFSTATHIOS PANAGOU1,5

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Introduction: Monosodium glutamate (MSG) is a food additive believed to impart a fifth taste called “Umami.” Osmotic dehydration (OD) is a mild dehydration process of food products resulting in intermediate moisture or minimally processed foodstuffs.

Purpose: To study the impact of partial substitution of NaCl with MSG on the fermentation of black olives subjected to OD prior to brining.

Methods: Three different mixtures of NaCl and MSG with/without osmotic dehydration were investigated, (i) 6.65% NaCl – 0.35% MSG (5% substitution), (ii) 6.30% NaCl – 0.70% MSG (10% substitution), (iii) 5.95% NaCl – 1.05% MSG (15% substitution). Changes in lactic acid bacteria, yeasts, Enterobacteriaceae, pH, titratable and combined acidity in the brine were analyzed for 140 d in parallel with sensory analysis.

Results: OD of the olives resulted in lactic acid processes based on the values of pH (3.7 – 4.1) and acidity (0.7 – 0.8%). In non-OD treatments the values of pH (4.5) and acidity (0.4 – 0.5%) could not ensure the microbiological stability of the product, especially in the higher levels of substitution. The use of MSG without OD resulted in higher buffering capacity in the brines (0.10 N) compared to non-OD olives (0.09 N) and hence to less microbiological stability. The flavor of the final product for all salt mixtures did not show any difference compared to control treatment.

Significance: The combination of OD pre-treatment with the use of MSG as flavor enhancer produces a safe and high added value product that provides new perspectives in the international market for table olives. The project “Development and adaptation of traditional Greek olive based products to Chinese dietary and culinary preferences” is co-financed by the European Union (European Regional Development Fund– ERDF) and Greek national funds through the Operational Program “Competitiveness and Entrepreneurship” of the National Strategic Reference Framework (NSRF) – Research Funding Program.

**T4-03**

**Will I Survive?**

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Introduction: Geobacillus stearothermophilus is a well-known flat sour spoilage organism, commonly used as biological indicator in the design of heat sterilization process. Several studies have investigated the heat resistance of the spores. However, little information is available regarding the behavior of the heat treated surviving spores during the can food storage.

Purpose: The purpose of this study was to model the spores’ behavior after a heat treatment when spores are exposed to conditions which do not allow growth (e.g., at low temperature or low pH). The model should take in account heat treatment conditions, medium pH and storage temperature.

Methods: Spores of G. stearothermophilus were heat treated at three different conditions to reach one decimal reduction: 115°C, pH 7 or pH 5.5 and 120°C, pH 7. Heat treated spores were stored in nutrient broth at conditions not allow growth. The spores’ behavior was evaluated by count plating for months.
T4-04 The Impact of Food Disinfection Methods Used in Fresh Ready-to-Eat Produce on Public Health
ANGELIKI BIRMPA and Apostolos Vantarakis
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Introduction: Fresh vegetables and fruits have come during the past decade to the forefront as important vehicles of foodborne illnesses, accounting for a large amount of reported outbreaks with an identified food source. Thus, important disinfection technologies are of paramount importance to reduce foodborne outbreaks.

Purpose: The purpose of the present study was to assess the effectiveness and exclude conclusions on the impact of food disinfection technologies on public health by taking into account the best microbial reductions that occurred with a series of disinfection technologies, and infectious doses of the tested microorganisms that have been reported in the literature. Thus, the infectivity of four bacteria (Escherichia coli, Staphylococcus aureus, and Salmonella Enteritidis and Listeria innocua) and one virus (HAdV 35) on three different Ready-to-Eat produce (romaine lettuce, strawberries and cherry tomatoes) was assessed.

Methods: Data on pathogen-specific relative infectivity of E. coli, S. aureus, S. Enteritidis, L. innocua and HAdV 35 were collected from the U.S. Food and Drug Administration’s (FDA), Public Health Agency of Canada (PHAC) and the European pathogen fact sheets. Selected disinfection technologies such as ultraviolet light (UV), ultrasound (US), sodium hypochlorite solutions (NaOCl) and combinations of the above for different treatment times were implemented.

Results: The combined technology of US followed by NaOCl was found to be the best disinfection technology for reducing bacteria (up to 4-log reduction) in different fresh produces. However, NaOCl seemed to be the only method that could reduce HAdV35. US seemed to be the most promising disinfection technology for reducing bacteria population in strawberries, followed by US+NaOCl technology. However, UV was recorded as the best option for reducing HAdV35 in strawberries.

Significance: This work provides a new perspective to deal with flat sour spoilage by taking in account the heat treatment efficiency and the impact of the physicochemical food properties during the storage.

T4-05 Modeling the Growth Rate of Clostridium perfringens as a Function of Residual Dioxygen Concentrations in Food Package
Marie-Laure Divanac’h1, Anne Lochardet1, FLORENCE POSTOLLEC1, Dominique Thuault1, Daniele Sohier1, Olivier Couvert2 and Veronique Huchet1
(1) ADRIA UMT14.01 SPORE RISK, Quimper, France
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Introduction: Packaging protects foodstuffs from contact with the external environment and from airborne contamination. Combined with a modified atmosphere, packaging also helps to prolong the shelf life of a food product and can also contribute to improving food safety. In industry, presence of residual oxygen concentrations exists in the packaging.

Purpose: The aim of this work is to study and model the impact of dioxygen concentrations on Clostridium perfringens growth and assess whether residual oxygen concentration is sufficient to inhibit anaerobic contaminant microflora.

Methods: Clostridium perfringens was cultured in RCM agar medium supplemented with 0.2% of glucose, 0.3% of yeast extract and reazurin. All Petri dishes were prepared in advance, stored in an hypoxia laminar flow hood, inoculated and then stored at 25°C in the same hood. To determine kinetics, 15 samplings were performed for given condition of O2-enriched atmosphere. For each sampling time, agar medium was collected, diluted and plated to determine bacterial population. Growth rate was estimated by fitting logistic primary model using delay and rupture.

Results: Growth rates were acquired for 9 concentrations of dioxygen between 0.1% to 6%. The addition of dioxygen resulted in a decrease in the growth rate until 6% O2 where bacteria stop growing. Based on Zwietering gamma concept (1995) and Rosso cardinal model (1993, 1995), this study integrates a new factor in predictive modeling which is the concentration of dioxygen.

Significance: This experimental protocol allowed performing data under statics residual dioxygen concentrations between 0.1% to 6% during several days. Taking into account the impact of residual dioxygen concentrations on bacterial growth in agar-based media, will further allow the transfer of these models to simulate the growth of foodborne contaminants in packaged food.

T4-06 A Large Scale Study to Test a Wide Variety of Additives in Broilers’ Feed to Decrease Campylobacter Shedding
MURIEL GUYARD-NICODÈME, Ségolène Queune, Typhaine Poezevara, Bernard LE Berre, Michel Amelot and Marianne Chemaly
ANSES, Laboratory of Ploufragan-Plouzané, Ploufragan, France

Introduction: Poultry meat is the major source of human Campylobacteriosis, the most frequently reported zoonosis in the EU and prevalence of Campylobacter colonization in European broiler flocks is 71%. However, there is still no effective strategy available to prevent or reduce Campylobacter colonization in broilers.

Purpose: This work is part of the 7th framework program “CAMPYBRO” and aims to determine the effect of 12 commercial feed additives to reduce Campylobacter shedding in primary poultry production.

Methods: Additives containing organic or fatty acids, monoglycerides, plant extracts, prebiotics, or probiotics were tested. For each additive, broilers contaminated with Campylobacter jejuni were fed with an additive free diet (control group)

Results: After a heat treatment, the surviving spores able to recover on agar medium decreased during the storage in no-growth conditions. Inactivation kinetics were biphasic, revealing the presence of two populations of spores after a heat treatment. The proportion of the most sensitive subpopulation was impacted by heat treatment conditions. Moreover, the resistance of the both subpopulations was affected by the pH and the temperature of storage. The higher resistance was observed at neutral pH (44 days ± 1.6 for one decimal reduction at 35°C) compared to pH 5.2 (18.5 days ± 2.7). Also, the surviving spores are inactivated faster for storage at high temperature (6.5 days for one decimal decrease at 55°C or 108 days at 25°C for pH 4.8).

Significance: This work provides a new perspective to deal with flat sour spoilage by taking in account the heat treatment efficiency and the impact of the physicochemical food properties during the storage.
or with supplemented diet (treated group). *Campylobacter* loads were assessed in cecal contents following the decimal dilution method. The comparison between control and treated groups was performed using statistical analysis based on mean and multiple comparison tests.

**Results:** No treatment was able to prevent broiler contamination with *Campylobacter* but a high individual variation was observed among birds regarding this contamination. At 14 days of age, eight treatments significantly decreased the colonization level compared to the control group by a maximum of 2 log CFU/g. At 35 days of age, three of these treatments still have a significant effect with a maximum reduction of 1.88 log CFU/g for a probiotic. At 42 days of age, only a short-chain fatty acid was still significantly efficient with a mean reduction over 2 log CFU/g. Moreover, a probiotic and a prebiotic-like significantly decreased the contamination by a maximum of 3 log CFU/g, only at this sampling period.

**Significance:** This work gives promising results about using feed additives to reduce *Campylobacter* infection in flocks. Nevertheless, a global approach, combining intervention measures at the different steps of the broiler meat production chain could have a greater impact on the public health risk reduction.

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**Technical Session 5 – Modeling and Risk Assessment and Communication, Outreach and Education**

**Tuesday, 21 April – 13.30 – 17.00**

**T5-01 Regional Trends in GMP Failures**

**KALLIOPI ZERVA**  
AIB International, London, United Kingdom

**Introduction:** This survey will focus on presenting results of food safety audits in a global extent. The survey will be based on audits conducted in more than 6000 different plants in 125 countries during 2014. Several industry sectors will be included such as bakery/snacks/confectionery, beverage, food contact packaging, non-food contact packaging and distribution centers.

**Purpose:** A review of efficiency and failure tendencies on prerequisite programs as being currently applied.

**Methods:** The audit is based on Good Manufacturing Practices and Food Safety Programs and consists of a detailed GMP inspection, followed by the corresponding necessary documentation review. A scoring system based on severity of the observation is applied as part of the audit with issues differentiated as “no risk for food safety,” potential risk, significant risk and imminent risk to food safety. Overall and specific scores and findings will be used in this essay to detect tendencies and review results per region and industry sector.

**Results:** The results will be presented as reviews and graphs taking into consideration the following aspects:

1. More solid GMP programs analyzed per country and/or region (Europe, Middle East, Africa, North and Latin America, Asia and Pacific)
2. GMP efficiency per industry sector
3. Specific section results, such as cleaning issues, maintenance and/or pest control, per country/region and industry sector.
4. Certain program reviews i.e., the number of facilities per country and per region with a well-developed Food Defense program (a Food Defense program includes a vulnerability assessment, a food defense team, annual program review, annual training and education).
5. Imminent food safety hazards (or program failures/departure from the Good Manufacturing Practices), analyzed as number of observations per country or per region.

**Significance:** A review of working efficiency of applied prerequisite programs in different food industry sectors and parts of the world.

**T5-02 Towards a Mechanistic Model for the Germination of Clostridium Species**

**ALINE METRIS,** Jason Brunt, József Baranyi and Mike Peck  
Institute of Food Research, Norwich, United Kingdom

**Introduction:** *Clostridia* form highly heat resistant endospores, enabling these bacteria to survive adverse conditions. Subsequently, spores may germinate, giving rise to vegetative cells that multiply and lead to toxin production and/or food spoilage. So far germination has been described with empirical models analogous to those used in thermal inactivation modelling. However the process is affected by sporulation conditions, therefore models should reflect this history-dependence.

**Purpose:** This study aimed to develop a quantitative model for germination based on a process suggested by recent advances in molecular microbiology and optical density (OD) measurements of *Clostridium sporogenes* in different conditions.

**Methods:** A calibration between OD and spore numbers was obtained by flow cytometry where the proportion of germinated spores was determined with SYTO16 and compared to percentages of phase-dark spores observed by phase-contrast microscopy. OD was measured in a Bioscreen in the presence of different concentrations of L-alanine and L-serine, after sporulation in TY or cooked meat media. Germination of both unheated and heat-activated spores was also compared. The modelling was carried out using Matlab software.

**Results:** A system of differential equations was proposed reflecting a signaling step where receptors are activated or inhibited by amino acids as suggested by the kinetics of mutants (Brunt et al.,PLoS Pathogens 10(9):e1004382). In a second step, it was assumed that germination is the result of activation of pre-formed enzymes in the spore cortex. We demonstrated that sporulation conditions have an effect on spore germinant receptors. Altogether, this model differs fundamentally from traditional predictive models.
**T5-03 Selecting the Most Appropriate Risk Ranking Methods**

ESTHER VAN ASSEL†, Ine Van der Fels-Klerx†, Helle Korsgaard‡, Lea Bredsdorf§, Maarten Nauta‖, Morten Poulsen‖, Martin D’Agostino‖, Marian Raley¶, David Coles¶ and Lynn Frewer¶

(1) RIKILT - Wageningen UR, Wageningen, Netherlands
(2) DTU, Copenhagen, Denmark
(3) The Food and Environment Research Agency (Fera), York, United Kingdom
(4) University of Newcastle, Newcastle upon Tyne, United Kingdom

**Introduction:** Both governmental organizations and food safety authorities would like to prioritize their activities using risk-based approaches. For this purpose, various risk ranking methods are available ranging from full risk assessment to the application of expert judgment.

**Purpose:** The aim of the current study was to perform a review and systematic evaluation of current risk ranking methodologies for prioritization of food and feed safety hazards with anticipated health impact.

**Methods:** A literature review was performed using a systematic approach with predefined keywords and selection criteria to identify available risk ranking methods and their characteristics. Apart from applications within the food safety domain, applications within socio-economics and environmental research were also evaluated for their usefulness in ranking food safety hazards.

**Results:** In total, around 14,000 papers were evaluated, of which 253 were relevant for this study. These relevant papers showed that the following methods are used for ranking hazards: (comparative) risk assessment, ratio methods, scoring methods, cost of illness, disease burden methods, stated preference methods, multi-criteria decision analysis, risk matrix, flow charts/decision trees and expert judgment. Characteristics of these methods were described. Based on this overview, a framework was developed consisting of questions that will help in the selection of the most appropriate risk ranking method depending on the case study. The selection is based on both the risk manager’s prerequisites and data availability. The framework has been tested in a number of case studies to determine the appropriateness of the method.

**Significance:** This study provided an overview of current methods available for ranking food safety risks. The developed framework will help in selecting risk ranking methods for specific case studies, which will facilitate risk assessors in performing a risk ranking exercise.

**Acknowledgment:** The contribution of EFSA to perform this study is highly appreciated.

**T5-04 Designing a Data Base for Supporting Decisions on the Use of Predictive Microbiology Software**

FERNANDO PEREZ RODRIGUEZ† and Salvador Cubero‡

(1) Food Science and Technology, University of Cordoba, Cordoba, Spain
(2) University of Cordoba, Cordoba, Spain

**Introduction:** The range of predictive microbiology software is wide and diverse, making difficult the selection of the most suitable application.

**Purpose:** To develop a Data Base on available predictive models containing structured and standardized information to support a suitable selection of predictive microbiology software and models.

**Methods:** The information on predictive models contained in 24 different predictive microbiology software (Combase predictor, Seafood Spoilage and Safety Predictor, MicroHibro, PMM-lab, Pathogen Modeling Program, etc.) was analyzed, and vocabulary and taxonomy were proposed so as to facilitate the description (i.e., attributes, features, properties, etc.) of models and software with respect to the use, model type, prediction type, and mathematical basis.

**Results:** This information was used to build a Data Base on Structured Query Language (SQL) containing more than 400 predictive models. This Data Base was first implemented in Microsoft Access Software (Microsoft®) to enable querying and reporting systems based on specific questions that users should respond regarding: application; predictive microbiology model type; food process, bacterium species, model matrix, and prediction factors.

**Significance:** This work is part of an ongoing project whose main objective is to develop an on-line expert system to guide users both in the selection of the most suitable model(s) from existing predictive software and how models should be applied for proper use.

**T5-05 Towards Community Driven Food Safety Model Repositories**

Matthias Filter, Carolina Plaza-Rodriguez, Christian Thoens, Annemarie Kaesbohrer and Berndt Appel; Federal Institute for Risk Assessment, Berlin, Germany

**Introduction:** Transferring predictive microbial models from research into real world food manufacturing or risk assessment applications is still a challenge for members of the food safety modelling community. Such knowledge transfer could be facilitated if publicly available food safety model repositories would exist.

**Purpose:** This research therefore aimed at identification of missing resources hampering the establishment of community driven food safety model repositories.

**Methods:** Existing solutions in related scientific discipline like Systems Biology and Data Mining were analyzed.

**Results:** On the basis of this analysis two factors could be identified which significantly promote the establishment of community driven model repositories: a standardized information exchange format for models and rules for model annotation. As a consequence the establishment of a Predictive Modelling in Food Markup Language (PMF-ML) is proposed and a prototypic implementation on the basis of SML is provided. In addition a domain-specific extension of the MIRIAM guidelines for model annotation has been developed. In order to demonstrate the practicability of the proposed strategy, existing predictive models previously published in the scientific literature were re-implemented using an open source software tool called PMM-Lab. The models are made publicly available in a sample Food Safety Model Repository hosted at the “OpenML for Predictive Modelling in Food” community project.
Significance: This work illustrates that a standardized information exchange format for predictive microbial models can be established by adoption of resources from Systems Biology. Harmonized description and annotation of predictive models would also contribute to increased transparency and quality of food safety models.

Acknowledgment: This work has been funded by BMBF (13N11202) research grant.

T5-06 Growth Potential of Listeria monocytogenes in Soft and Semi-hard Artisanal Cheeses

EVY LAHOU and Mieke Uyttendaele
(1) Ghent University, Ghent, Belgium

Introduction: Renewed interest in artisanal food products results in a wide range of artisanal farmhouse cheeses offered in farm and cheese shops. The production of these cheeses is more and more subjected to strict hygiene and cooling requirements in order to ensure food safety. However, these requirements put much pressure on the traditional methods used by artisanal cheesemakers.

Purpose: The study evaluated the growth potential of L. monocytogenes in different soft and semi-hard artisanal cheeses.

Methods: Challenge testing with cheese storage at 7°C and 14°C was performed on 12 soft cheeses of 3 different types and 8 semi-hard cheeses of 2 different types. The obtained results were also contrasted to expected results from predictive models (Combase and SSSP).

Results: The (median) growth potential of L. monocytogenes on soft cheeses stored at 7°C ranged from 3.3 - 3.6 log units and stored at 14°C ranged from 4.1 – 5.2 log units, whilst this of semi-hard cheeses at 7°C ranged from 0.4 – 0.6 log units and at 14°C from 0.7 – 2.1 log units. It was observed that the growth potential will be mainly influenced by the pH of the cheese, i.e., 6.3 – 6.9 (soft cheeses) and 5.4 – 5.5 (semi-hard cheeses). The pH depends on the amount of lactic acid bacteria, as such the growth potential of L. monocytogenes is indirectly influenced by the concentration of lactic acid bacteria. No difference in growth potential was observed between soft raw milk cheeses and soft pasteurized milk cheeses.

Significance: Strict hygiene and cooling requirements are needed for soft artisanal farmhouse cheeses in order to ensure food safety. Semi-hard artisanal farmhouse cheeses seem to pose a lower risk to food safety.

Technical Session 6 – Laboratory and Detection Methods
Monday, 20 April – 15.30 – 17.00

T6-01 Strategies for Targeted Control of Listeria in Norwegian Food Processing Environments

EVEN HEIR, Solveig Langsrud and Trond Møretrø; Nofima, Norwegian Institute of Food, Fishery and Aquaculture, Ås, Norway

Introduction: Listeria monocytogenes (Listeria) is regarded as the most serious food safety challenge for industries producing Ready-to-Eat foods. Listeria control challenges are associated with the complexity of contamination sources and the ability of Listeria to grow, survive and persist in food industry premises and processing equipment.

Purpose: To document the Listeria situation in the food industry through systematic review and identification of sources, contamination routes and factors that influence the Listeria situation in the salmon processing industry.

Methods: The standards of production facilities, operating practices and Listeria control strategies in the salmon processing industry were systematically mapped through a web-based survey. A two-year follow-up study including visits, processing routine reviews and microbial sampling of four processing plants documented the Listeria situation. Listeria was identified according to standard procedures based on ISO 11290-1. Strain typing was performed by MLVA.

Results: Microbial sampling (> 800 samples) in four processing plants showed processing environments to be an important source of Listeria product contamination. Listeria was more prevalent on humid (30% positives) and dirty (46%) surfaces compared to dry (14%) and visibly clean surfaces (18%). Routine sanitation programs were not effective in elimination of Listeria and 14% of product contact surface samples were Listeria positive after cleaning compared to 15% during processing. The results also showed that gutted salmon could be an important and continuous source of Listeria in filleting plants and smokehouses and that persistent Listeria are present in several plants.

Significance: The study documented the need for improved and specific measures for Listeria control in the salmon processing industry. Of particular need are enhanced strategies for prevention and elimination of Listeria in specific harborage sites/niches.

T6-02 Inhibition Mechanism of Listeria monocytogenes by Lactococcus piscium CNCM-I 4031: A Transcriptomic Approach

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Introduction: Lactococcus piscium CNCM I-4031, a psychrotrophic lactic acid bacterium recently isolated from raw salmon packed under modified atmosphere, is able to inhibit the growth of Listeria monocytogenes in peeled and cooked shrimp (Fall et al., 2010) and in chemically defined medium (MSMA) based on shrimp composition. According to our
were inoculated (10^7 or without the addition of 30% fat, and (ii) 0% or 2.5% NaCl, simulating commercially available products. The samples loaded halloysites were investigated for their loading efficiency, controlled release of carvacrol and their antibacterial activity. Polymer films containing loaded halloysites were also investigated for their potential as antibacterial food packaging materials.

**Results:** Halloysites were shown to be loaded with carvacrol molecules up to 17.40 ± 3.05 wt %. The controlled release rate of carvacrol molecules from halloysites were determined to be 0.053 mg/h within the first 40 h, and 0.011 mg/h for the following 130 h at 30°C. Agar diffusion assays demonstrated that carvacrol loaded halloysites can inhibit the growth of *Escherichia coli* and *Aeromonas hydrophila* cells through the release of carvacrol molecules. The minimum inhibitory concentration of carvacrol loaded halloysite was determined to be 1.25 mg/ml for a cell culture containing 10^9 *Aeromonas hydrophila* cells. Polymer nanocomposites of carvacrol loaded halloysites were also demonstrated to inhibit the growth of microorganisms as demonstrated by the killing zones around the polymeric films obtained from agar diffusion assays.

**Significance:** This study provides a novel approach for the development of safe antibacterial food packaging materials composed of natural components that can significantly contribute to mitigation of food contaminations and enhance food safety.

**T6-03 Antibacterial Halloysite/Polymer Nanocomposites for Safe Active Food Packaging Applications**

**SAMAN HENDESSI,** Emine Bilur Sevinis, Serkan Unal, Fevzi Cakmak Cebeci, Yusuf Menceloglu and Hayriye Unal Sabanci University, Istanbul, Turkey

**Introduction:** Active food packaging materials with antibacterial properties can allow safe storage of food by acting against food contaminant microorganisms. In order to have safe active food packaging, it is crucial to choose active packaging components that do not pose any risks to human health.

**Purpose:** The purpose of the present study was to develop novel nanocomposites-based active food packaging materials with antibacterial properties by entirely using natural and safe active components.

**Methods:** Halloysites that are naturally occurring, non-toxic clay nanoparticles with hollow tubular structures were utilized for the encapsulation and the controlled release of carvacrol, the active component of antibacterial thyme oil. Carvacrol loaded halloysites were investigated for their loading efficiency, controlled release of carvacrol and their antibacterial activity. Polymer films containing loaded halloysites were also investigated for their potential as antibacterial food packaging materials.

**Results:** Halloysites were shown to be loaded with carvacrol molecules up to 17.40 ± 3.05 wt %. The controlled release rate of carvacrol molecules from halloysites were determined to be 0.053 mg/h within the first 40 h, and 0.011 mg/h for the following 130 h at 30°C. Agar diffusion assays demonstrated that carvacrol loaded halloysites can inhibit the growth of *Escherichia coli* and *Aeromonas hydrophila* cells through the release of carvacrol molecules. The minimum inhibitory concentration of carvacrol loaded halloysite was determined to be 1.25 mg/ml for a cell culture containing 10^9 *Aeromonas hydrophila* cells. Polymer nanocomposites of carvacrol loaded halloysites were also demonstrated to inhibit the growth of microorganisms as demonstrated by the killing zones around the polymeric films obtained from agar diffusion assays.

**Significance:** This study provides a novel approach for the development of safe antibacterial food packaging materials composed of natural components that can significantly contribute to mitigation of food contaminations and enhance food safety.

**T6-04 Effect of Freezing Rate and Frozen Storage Duration on the Survival of Escherichia coli O157:H7 during Cooking of Beef Burgers of Different Formulation**

**STAVROS MANIOS,** Evangelia Kanakaki, Akateriní Bosioli and Panagiotis Skandamis Agricultural University of Athens, Athens, Greece

**Introduction:** The thermotolerance of *Escherichia coli* O157:H7 during cooking of beef burgers may be affected by their formulation as well as the storage conditions prior cooking.

**Purpose:** To evaluate the combined effect of freezing rate, frozen storage duration, fat and salt content of beef burgers on the thermotolerance of *E. coli* O157:H7.

**Methods:** Beef burgers of 1.5 or 3 cm thickness (70 or 120 g, respectively) were prepared from lean ground beef (i) with or without the addition of 30% fat, and (ii) 0% or 2.5% NaCl, simulating commercially available products. The samples were inoculated (10^7 CFU/g) with a 3-strain composite of *E. coli* O157:H7 and were stored at -16°C or -28°C, as to achieve low or high freezing rates. After 0, 1 and 20 d, the burgers (n = 3) were cooked in a preheated (200°C) oven broiler to internal temperature of 65 or 71°C.

**Results:** The addition of salt and the size of burgers did not affect significantly (P > 0.05) the survival of the pathogen. In contrast, 30% fat content enhanced the thermotolerance of *E. coli* O157:H7, especially when burgers were cooked at 65°C. The pathogen showed overall better survival after cooking of raw samples (day 0) compared with those stored under frozen conditions, suggesting potential injury during freezing. Long-term frozen storage (20 d), increased the recovery of the pathogen (1.1 to 4.4-log CFU/g reduction), compared with the short-term storage (2.2 to 5.2-log CFU/g reduction) after cooking to 65°C, especially when samples were previously stored at -28°C, suggesting potential influence of the freezing rate on the thermotolerance of the pathogen. The above observations were less evident when samples were cooked at 71°C, due to the severity of the treatment.

**Significance:** Higher freezing rate and long-term frozen storage may enhance the survival of *E. coli* O157:H7 in frozen beef burgers of high fat content.
T6-05 Purchase, Storage, and Preparation of Eggs and Poultry in the United States Compared to Selected European Countries

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(2) The Sensory Analysis Center, Kansas State University, Manhattan, KS
(3) Tennessee State University, Nashville, TN
(4) RTI International, Research Triangle Park, NC

Introduction: Consumer practices related to food safety with poultry and eggs have been studied in many countries, however little comparison has been made among different countries which can be helpful in understanding potential trends, similarities, and differences.

Purpose: The objective of this study was to characterize consumers’ purchase, storage, handling, and preparation of poultry products and eggs in the U.S. and four European countries: Estonia, Italy, Spain, and Russia.

Methods: Approximately 100 consumers in each European country (200+ in the U.S.), who were primary purchasers and preparers of food, completed a questionnaire about poultry products and eggs’ purchase locations and times, storage locations, and preparation such as use of cutting boards.

Results: In the U.S., most eggs are purchased refrigerated while in Russia and Estonia consumers purchased both refrigerated and room temperature eggs and in Italy (84%) and Spain (87%) eggs typically were purchased at room temperature. Interestingly, more than 20% of consumers in Russia and Italy also stored cooked eggs at room temperature, a potential food safety hazard. In all countries it was common to store poultry and meat on upper or middle shelves and in about half the households nothing was placed underneath the meat to control leaks or spills that could contaminate other foods. About one-fourth of U.S. and European consumers (slightly less in the U.S. and Estonia and more in Spain) reuse cutting boards from raw poultry and meat for other items such as fruits and vegetables without washing them.

Significance: Consumer shopping, storage, and preparation behaviors show potential unsafe practices in both the U.S. and Europe. A uniform approach to food safety research-based information might be used to define critical points of control for consumers and create educational messages based on the information gathered.

T6-06 Qualifying Gilthead Sea Bream Freshness with Microbiological Indicators

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(2) Agricultural University of Athens, Athens, Greece

Introduction: Fish is among the most perishable foods, where spoilage symptoms are due to the undesirable growth of microorganisms to unacceptable levels. In the case of sea bream, pseudomonads and Shewanella species seem to be the main spoilage organisms.

Purpose: To assess gilthead sea bream freshness based on quantitative and qualitative microbiological data, based on the hypothesis that presence of specific microorganisms can be used as a potential marker of freshness.

Methods: Freshly caught fish (max. 8 h after catching) or processed fish (eviscerated and cleaned 24 h after catch) from 4 different Greek sea areas were analyzed with classical microbiological methods before and during storage at 0°C for 16 days and at -18°C for 18 months. The presence of specific microorganisms in fish was monitored using PCR-DGGE.

Results: Regarding the classical microbiological analysis, results showed that pseudomonads, H2S producing and non H2S-producing were the dominant microorganisms of freshly caught fish, as well as during storage. Enterobacteriaceae community growth on plats after incubation at 7°C was found 1 – 3 log CFU/g higher than that incubated at 37°C. After 16 days of storage at 0°C, the dominant populations of freshly caught fish, reached the level of 6 – 7 log CFU,g, and 5.6 log CFU/g on processed fish. DGGE analysis revealed a great variety of microorganisms during storage of processed and non-processed sea bream. Sequencing the detected microbiota revealed the presence of the following genera: Pseudoalteromonas, Shewanella, Pseudomonas, Aeromonas, Psychrobacter, Yersinia, Xylosandrus, Oceanisphaera, Arthrobacter, Hafnia, Exiguobacterium. The genera Psychrobacter and Aeromonas were detected widely in all fishes. On the other hand, Oceanisphaera was most frequently detected at processed fish, while Xylosandrus germanus was mostly tracked in fish after 8 days of storage at 0°C.

Significance: Such a study could give a better understanding in fish microbiota and qualify possible microbial indicators of gilthead sea bream freshness.

T7-01 The Hygienic Design of Food Industry Brushware

DEBRA SMITH
Vikan, Swindon, United Kingdom

Introduction: Thanks to the European Hygienic Engineering Design Group (EHEDG) many food manufacturers appreciate the benefits of using hygienically designed production equipment as it is quicker and easier to clean, and minimizes the risk of product cross-contamination by microbes, allergens, foreign bodies etc. This, in turn, maximizes food safety and quality, reduces the risk of expensive product rejection or recall, and minimizes food waste. However, when it comes to the cleaning equipment used in food production, very few tools are developed with good hygienic design in mind. In January 2015 the British Retail Consortium (BRC) issued version 7 of their Global Standard for Food Safety. New to this version (in Section 4.11.6) is the requirement for cleaning equipment to be ‘hygienically designed,’ but what determines whether a piece of cleaning equipment is of good hygienic design and what can be done to ensure that hygienic design is incorporated into future food industry standard cleaning equipment?

Purpose: To investigate the hygienic design of different types of food industry cleaning brushware and propose improvements for cleaning equipment in the future.
T7-02 Potentiation of Disinfection Effect on Listeria monocytogenes Biofilms by Chlorine Dioxide and Hypochlorite in Rinsing Water

SOLVEIG LANGSRUD, Trond Meretø and Even Heir
Nofima, Norwegian Institute of Food, Fishery and Aquaculture, Ås, Norway

Introduction: Sanitation is regarded as an important measure to avoid establishment of house strains and eliminate Listeria monocytogenes from food processing machines. Some food producers omit rinsing the equipment after disinfection to prevent growth of bacteria in the period between sanitation and production. However, the rinsing stages after cleaning and disinfection are important steps to remove soil and bacteria from the machines and questions have also been raised about possible resistance build-up by this approach.

Purpose: In the present study, it was investigated if the use of chlorine dioxide or hypochlorite in concentrations commonly used for drinking water could potentiate the effect of disinfection.

Methods: Four salmon processing factories and four RTE-meat plants were sampled for L. monocytogenes both after cleaning and disinfection and during production. A cocktail of six strains from the production environment were grown as biofilms on stainless steel and exposed to disinfection (hypochlorite, peracetic acid or quaternary ammonium compound) followed by rinsing in water, 0.5 ppm chlorine dioxide or 0.7 ppm hypochlorite. The number of bacteria surviving this procedure was determined by neutralization of disinfectant and plating on nutrient agar.

Results: Sampling of eight food processing factories revealed that the ordinary sanitation program failed to eliminate persistent strains of L. monocytogenes. Rinsing biofilms of L. monocytogenes with water containing hypochlorite or chlorine dioxide led to 90–99% reduction of attached viable bacteria. Rinsing biofilms pre-exposed to disinfectants led to a similar effect. Thus, including these chlorine compounds in the rinsing water, even in concentrations allowed for drinking water, could potentially be used to reduce Listeria levels in the food industry.

Significance: The work shows that the current sanitation regime in food factories is not efficient for eliminating L. monocytogenes. Including chlorine compounds in rinsing water is a promising approach to enhance the sanitation process and should be tested in practice.

T7-03 The Seek and Destroy Process: Listeria monocytogenes Process Controls in the Ready-to-Eat (RTE) Meat and Poultry Industry

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Introduction: The majority of human listeriosis cases appear to be caused by consumption Ready-to-Eat (RTE) foods that are contaminated, at the time of consumption, with high levels of this pathogen.

Purpose: While strategies to prevent growth of Listeria monocytogenes in RTE products are critical for reducing the incidence of human listeriosis, control of post-processing environmental contamination of RTE meat and poultry is an essential component of a comprehensive L. monocytogenes intervention and control program. Complete elimination of post-processing contamination with L. monocytogenes is challenging as this pathogen is common in various environments outside processing plants and can persist in food processing environments over years.

Methods: L. monocytogenes persisting in processing plants have been identified as the main source of post-processing contamination of RTE foods and as contamination source responsible for multiple listeriosis outbreaks. Identification and elimination of L. monocytogenes strains persisting in processing plants is thus critical for (i) regulatory compliance with the so called “zero-tolerance” for L. monocytogenes in US RTE meat and poultry products and (ii) reducing the incidence of human listeriosis. The “Seek and Destroy” process is a systematic approach to finding sites of persistent growth (“niches”) in food processing plants, with the goal of either eradicating or mitigating effects of niches.

Results: The Seek and Destroy process has been used effectively to address persistent L. monocytogenes contamination in food processing plants, as supported by peer-reviewed evidence detailed here.

Significance: This is a science-based strategy for controlling L. monocytogenes in RTE foods.

T7-04 Growth, Colonization and Internalization of VTEC in Fresh Produce: The Potential Impact on Food Security

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Introduction: Fresh produce accounts for a significant proportion of foodborne outbreaks. Verocytotoxigenic Escherichia coli (VTEC) and non-typhoidal Salmonella enterica are responsible for the majority of bacterial cases and outbreaks, and these pathogens can be associated with a wide range of fruit, vegetables and nuts. Risk assessments (RA) aim to evaluate the production chain and identify significant risk factors within the HACCP framework. However, pre-harvest factors, such as internalization, growth in/on the plant and systemic spread have been underrepresented in current analyses. Such underestimation of pre-harvest factors inevitably impacts and hence skews the contribution of subsequent processing steps in RA.

Purpose: The aim of this work is to obtain information that can be used in risk analyses of contamination of fresh produce crops by VTEC. Here we focus on bacterial growth in plant extracts and in planta, using fresh produce plants commonly associated with foodborne outbreaks.
Methods: Colonization of two *stx*- VTEC strains Sakai and ZAP1589 on *Spinacia oleracea* and *Lactuca sativa* was assessed. Growth within plants was observed by viable counts and confocal microscopy, using gfp-tagged *E. coli*. The metabolite complement of plant extracts was also determined by HPLC and GC/MS.

Results: VTEC shows marked differences of log 2 CFU/g in colonization and internalization in plant tissues that is dependent on the plant species; tissue type and bacteria strain type. Biochemical analysis revealed major differences in metabolites between host species, e.g., almost twice as much monosaccharides in lettuce roots as in fenugreek sprouts. This can be used to relate growth rate variances to plant species-specific differentiation.

Significance: Robust risk assessment of bacterial contamination requires fundamental data, not least the potential for bacterial growth in and on the substrate. Our data will make a significant contribution to RA to show the basis for important differences in bacterial growth potential on plant hosts.

T7-05  
**Elucidating the Physiology of Attached and Internalized Salmonella Cells in Leafy Vegetables**

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**Introduction:** *Salmonella* colonizes fresh produce surface or even penetrates it, a phenomenon called internalization. Despite the well-established risk of internalized cells evading common disinfection practices, little is known about their physiology and the underpinning stress resistance mechanisms.

**Purpose:** To assess: (i) the attachment and internalization of *Salmonella enterica* regarding different leafy vegetables, (ii) the acid tolerance of internalized and attached cells and (iii) the transcriptional changes of stress and virulence-associated genes.

**Methods:** Spinach, lettuce, green amaranth, arugula and endive were inoculated, individually, with *Salmonella Enteritidis*, Typhimurium and Infantis by immersion (7 log CFU/ml) and stored at 5°C and 20°C for 2 h and 48 h (n = 6). Population of total, attached and internalized *Salmonella*, as well as their survival against pH 2.7 (HCl) were assessed. Transcriptional changes of virulence T3SS (*hilA*, *prgH*, *invA*, *avrA*, *ssrB*) and stress related genes (*cadB*, *proV*) were determined using RT-qPCR (n = 3).

**Results:** Attached and internalized *Salmonella*, 2 h post inoculation, reached 3.6 – 4.8 and 3.2 – 5.0 log CFU/g, respectively, with spinach and endive allowing the highest (*P < 0.05*) internalization. Maintaining inoculated vegetables for 48 h at 20°C increased the recovery of attached and internalized cells by 1 – 2 log CFU/g, though it is unknown whether this derives from growth or colonization induction. However, habituation of *Salmonella* at 20°C induced the transcription (10 – 100 folds) of T3SS-related genes.

**Significance:** Our findings reveal physiology aspects of *Salmonella* colonizing leafy vegetables and therefore could be supportive material in quantitative risk assessment of pathogens in fresh produce sector.

T7-06  
**Microbiological Hazard Investigation and Evaluation of Dutch Fresh Produce Growers**

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**Introduction:** Public health risks associated with pathogenic contaminations from food of non-animal origin (FoNAO) are a concern for the Dutch fruit and vegetable industry. Food Safety Management Systems, such as Good Agricultural Practices, are often recommended; however, such recommendations often lack practicality for produce growers.

**Purpose:** The aim was to investigate the status of currently applied measures, like Global G.A.P. by Dutch fresh produce growers, and the hygiene status at certain points at the grower. In addition, incentives to stimulate growers to adapt grower management for the microbiological safety of fresh produce were investigated.

**Methods:** A questionnaire alongside visits to fresh produce growers for microbiological sampling and analyses was used to identify farm management and hygiene status. Questionnaires were also used to investigate which measures could stimulate farmers to adopt certain management practices to reduce potential microbiological contaminations.

**Results:** Personal interviews (n = 19) and water, swab, and/or product sampling for total psychotropic or aerobic mesophilic bacteria, coliforms, and *E. coli* indicated that Global G.A.P. was recognized and followed by most growers to a certain degree; however, growers had limited familiarity to possible microbiological contaminations from water storage sources with minimal or infrequent use, staff personal hygiene, and harvesting equipment. Hence, each grower was given specific advice on potential practices or locations of microbiological relevance. Farmers were price sensitive towards water testing and water and hygiene management measures. Also, they were willing to adapt management when there was a sense of urgency.

**Significance:** These results can further assist Dutch national authorities and industries in developing practical sampling guidelines for microbiological hazards at primary production.

**Acknowledgment:** The Ministry of Economic Affairs, and the Dutch Fruit and Vegetable industry are acknowledged for their financial contributions and assistance in this study.

T8-01  
**Keeping up with Molecular Analysis – Smooth Transitions from Research to Risk Assessment**

HELEN WITHERS

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**Introduction:** Since the first genomic sequences were completed in the late 1990s, the development of molecular techniques for the detection of bacteria and viruses in complex matrices has occurred at a rapid pace. Certainly for research laboratories this has opened up whole new fields of research within microbiology. Transition from research tool to commercial applications has been equally as rapid; more often than not outpacing validation against traditional “gold standard” culture methodology; and technical competence.
**Purpose:** The question for food safety risk assessors is can we be sure about the validity of this data given the genetic diversity within bacterial communities; driven by the promiscuity of bacterial DNA exchange.

**Methods:** Real-time PCR has been used to detect the presence of *E. coli* O157:H7 and other STEC commercially in New Zealand since 2012. We have carried out an analysis of the data gathered by laboratories to confirm the sensitivity and specificity of these assays for use in foods.

**Results:** Recent studies of *E. coli* O157:H7 and other Shiga Toxin-producing *E. coli* in New Zealand have suggested a degree of genetic diversity, which could lead to a potential decrease in specificity and sensitivity for molecular detection assays, particularly those developed internationally. We have analyzed Real-time PCR data sourced from a number of commercial laboratories to review the efficacy of these tests compared to culture confirmation.

**Significance:** Analysis of data generated is essential, particularly for real time PCR; we have shown that examination of this data can reveal much about the efficacy of the process and has value in assessing risk even within commercial settings. The cost benefit of using molecular assays for food industries is clear, and they are here to stay but how do we ensure that these tests are appropriately validated to detect pathogens that are found in food?

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**T8-02 Impedance-based Microbiological Methods in Meat Processing Industry: Detection and Quantification of *Salmonella* spp.**

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**Introduction:** The presence of *Salmonella* in raw pork meat is becoming an increasing concern for the meat industry. Despite attempts to control foodborne pathogens, a significant number of pork products are contaminated, leading to economic losses and increasing the risk of foodborne disease outbreaks.

**Purpose:** Validation of an impedance-based method for rapid detection of *Salmonella* in raw pork meat and assessment of its application for quantification of *Salmonella* compared to traditional methods.

**Methods:** A µ-Trac 4200 (SY-LAB) system was used for recording impedance changes throughout time using modified selenite-cystine selective medium at 37°C. Twenty-five g pieces of raw pork meat were inoculated with known amounts of *Salmonella enterica* and *Salmonella Typhimurium*, being analyzed after homogenization in 225 ml of buffered peptone water. Validation was carried out against the standard method (ISO 6579:2002) for 13 samples. A biochemical test (Entero pluritest®) was also used as confirmation for all the tests. Different calibration curves were also prepared for the quantification of *Salmonella* and analyzed by µ-Trac and the standard plate count technique using DCA as selective agar.

**Results:** Both the impedance-based and the plating methods show similar number of positives (2/13) and false negatives (0/13), whereas the latter leads to a higher number of false positives (5/13 vs 2/13). Impedance-based method leads to a linear correlation between concentration of viable bacteria and detection time (*DT*), reaching a detection bacterial limit of 1 CFU. The *DT* is influenced by initial bacterial concentration and also by the bacterial strain, showing *S. Enteritidis* a lower *DT*.

**Significance:** The impedance-based method allows a rapid screening of samples, requiring less than 24 h for positives and ca. 30 h for negatives with reliability comparable to standard methods, reducing costs for the industry and risk of outbreaks.

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**T8-03 ISO/TS 13136 for STEC Detection: Method Performances Assessment to Go for Accreditation**

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**Introduction:** The ISO/TS 13136 (2012) standard is a horizontal method for the detection of Shiga Toxin-producing *E. coli* (STEC) and the determination of O157, O111, O26, O103 and O145 serogroups. The protocol uses real-time PCR for detection of the virulence and serogroup-associated genes. This standard is a Technical Specification, and many steps are fully opened. Indeed, while the enrichment procedures are clearly described, many possibilities are offered to run the DNA extraction step or to select the PCR Internal Control (IC). The confirmation procedure includes an Immuno-Magnetic Separation, but the combination of media to be used to select specifically the STEC isolates is not yet proposed.

**Purpose:** A study was run in order to select the most appropriate protocol, and characterize the selected method.

**Methods:** The performances assessment was based first on the PCR parameters selection by determining the PCR efficiencies and the PCR Limit of Detections (LOD) for all the target genes, as well as by verifying the inclusivity and exclusivity. A comparison between four principles was run according to the ISO 16140 standard and the AOAC Guidelines using the LOD determination; two relevant matrices artificially contaminated with a mix of target strains were tested, raw milk Brie with 25 g sample size and beef trim with 375 g sample size. PCR efficiencies in enriched matrices were determined as well.

**Results:** The selected method show PCR efficiencies comprised between 85% and 105%, on DNA extracts from pure cultures or from enriched matrices. The PCR LOD are comprised between 6.5 and 18.8 genome copies, and the entire method LOD are comprised between 2.2 and 10.6 CFU/25 g for the raw milk Brie, and 0.8 et 2.9 CFU/375 g for the beef trim. The confirmation protocol combines the use of TBX, R-MAC, ChromAgar STEC and CT-SMAC, as well as PCR and latex tests.

**Significance:** This study provides a comparison of the performances of the various principles proposed ISO/TS 13136 (2012), as well as a procedure to be used to ensure the quality assurance all among the development of this ISO standard within a lab.
Simultaneous Direct Detection of Shiga Toxin-producing *Escherichia coli* (STEC) Strains by Gold Nanoparticle Optical Sensing

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**Introduction:** Shiga Toxin-producing *Escherichia coli* (STEC) strains (“Big Six” – O26, O45, O103, O111, O121, O145, and O157) represent significant groups of pathogens responsible for foodborne diseases.

**Purpose:** The objective of this study was to develop a colorimetric optical sensing assay that can simultaneously detect various STEC strains.

**Methods:** Pairs of single-stranded thiol-modified oligonucleotides (30-mer) were designed and immobilized onto gold nanoparticle (AuNPs) to target *stx1* (119-bp) and/or *stx2* (104-bp) genes of STEC big six and O157 serogroups. Oligonucleotide-functionalized AuNps were used as probes based on the sequence-specific hybridization properties of DNA and stable colorimetric properties of AuNPs. Ground beef and blueberries (50 samples each) were randomly inoculated with STEC strains at < 1 log CFU/g. A sample pooling plan with brief enrichment procedures (modified Tryptic Soy Broth, at 42 ± 1°C for 6 h) was incorporated into the assay to ensure detection of viable STEC in food.

**Results:** When amplified target DNA from post-enriched food samples was hybridized with complementary AuNP-probes, reaction mixtures retained its initial red color following an increased salt concentration (2M). For non-target/non-STEC strains such as *Salmonella* Typhimurium, a change from red to gray-blue was induced due to aggregation of functionalized AuNPs, providing the basis of direct and simultaneous detection of targets. The detection limit is < 1 log CFU/g in food. Twenty out of 50 ground beef and blueberries were tested positive for STEC, which is 100% accuracy. It requires less than one hour to complete after DNA sample preparation. Gel electrophoresis and spectrophotometric data confirmed successful DNA sandwich hybridization and AuNPs aggregation.

**Significance:** The optical sensing assay provided a reliable means for direct and simultaneous detection of various STEC strains, utilizing AuNPs as inexpensive materials. The strategy was found to be superior over traditional approaches for detection of STEC in food samples even at low-level contamination.
Poster Abstracts
Facilitating Sustainability and Innovation in Food-sector Small and Medium-sized Enterprises (SMEs) in Wales, UK

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Introduction: The KITE (Knowledge, Innovation, Transfer, Exchange) project has facilitated academic/industrial partnerships in food sector SMEs in Wales, UK to help SMEs to meet technical demands required for business sustainability. Improved technical and food safety compliance with 3rd party accreditation standards is required to deliver business needs, improve innovation and enable sustainability.

Purpose: The KITE academic/industrial collaborative partnership aimed to increase food safety/technology knowledge, improve technical capabilities and compliance with 3rd party accreditation standards and increase processing and new product innovation in food-sector SMEs.

Methods: Assessment of the project impact (between 2008 – 2015) related to SME innovation and sustainability was undertaken by reviewing project management reports (n = 274) detailing outputs/outcomes, SME growth, new product development (NPD) and technical performance/accreditations. In addition, media documentation (n = 134) were analyzed and in-depth interviews (n = 34) were conducted with collaborative partners.

Results: Findings indicate that the KITE project has provided an innovative sector-specific approach to knowledge-transfer, targeting an under-resourced area in food technology expertise. Provision of technical expertise to affiliates has resulted in embedded changes of food safety culture and achievement of 25 BRC(5/6) accreditations and 28 retail-quality standards in 39 SMEs. More than 579 new food products have been developed/lunched predominately in processed (including specialist/allergen-free) foods in ready-meal/ready-to-eat and dairy sectors. Advancements in processing methods included implementation of validated processes to (e.g.) enable production of sterile baby foods in retort pouches for ambient storage and novel nut-free bakery. Improved potential for SME sustainability has resulted from a combination of outputs encompassed by improved technical/operational performance resulting in increased retailer confidence, broadening/strengthening of customer bases and increased sales > £56 million (€73 million). More than 1298 jobs have been safeguarded/created (including manufacturing/quality assurance roles) and improvements in production and environmental efficiency, and local supply chains are evident.

Significance: The KITE project has provided technical expertise to overcome critical technical barriers encountered in food-sector SMEs. Improved SME technical compliance, implementation of innovative processes and new-product development has facilitated SME growth, increased sales, increased employment and the potential for sustainability.

Sulfur Dioxide Decay in Bovine Flesh Subjected to Refrigeration Method (+4°C)

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Introduction: An Italian Ministerial Decree (209/1996) regulates food additives permitted in the preparation and preservation of food. Food additive is any substance, intentionally added to food products for technological aims during the production, transformation, preparation, treatment and stocking phases. Sulphites belong to this macro-category. These compounds are added to many food products, such as meat, for their antimicrobial and antioxidant activity. Furthermore, sulphites are used for the improvement of food appearance by original color maintenance that make it more suitable for buyers.

Purpose: This study aims to determine the existence of sulfur dioxide concentration decay in refrigerated (+4°C) meat products (sausages, hamburger, minced meat), after 7 days by ion chromatography with suppressed conductivity detection analysis.

Methods: The sulfites concentration assessment was conducted by ion chromatography. In the first step 25 g of samples were weighed and distilled. 5 ml of the purified sample solution were injected into the ion chromatograph after conditioning. After a conditioning time of 30 min we proceeded with the calibration. After calibration curve validation, we carried out with the chromatographic run of the samples. The concentration of sulfites (ppm) is calculated by interpolation on the calibration curve.

Results: Results showed a statistically significant difference in sulfites concentration between minced meat samples examined during the initial phase and after 7 days at +4°C (P < 0.05). No significant differences were found in hamburger and sausages samples after refrigeration storage.

Significance: The ingestion of foods containing high concentrations of sulfites can cause food allergies and asthmatic reactions in sensitive individuals. Furthermore, these chemicals may reduce the nutritional quality of food by interacting with some vitamins such as nicotinamide, folic acid, thiamine and pyridoxal. Results from this study are useful for the determination of the best method of meat preservation in order to prevent diseases associated with excessive consumption of sulfites.
P1-03  Interplay between Food Safety Climate, Food Safety Management System and Microbiological Output in Farm Butcheries and Affiliated Butcher Shops
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Introduction: Despite the efforts to develop and implement Food Safety Management Systems (FSMS), foodborne outbreaks are still reported. Therefore, in this study the focus is shifted to a more human dimension of food safety with the introduction of the concept “food safety climate.” It is expected to influence human behavior in a food processing company and affect the final delivered food safety/quality.

Purpose: The aim of this work was to set a definition for food safety climate and to develop and validate a tool to measure the food safety climate in food companies. These concepts are further demonstrated via a case study in which it is investigated whether the negative impact of a less elaborated/fit-for-purpose FSMS on the microbiological quality level of a company can be compensated by a favorable food safety climate.

Methods: A food safety climate questionnaire with twenty-eight indicators was developed based on scientific literature study and was validated by experts in the field. Four farm-based and four affiliated butcheries were screened on their food safety climate, level of implemented food safety management system and via product and environmental microbiological sampling objective data on the microbiological output of the butcheries were collected.

Results: Food safety climate was defined as employees’ (shared) perception of leadership, communication, commitment, resources and risk awareness concerning food safety and hygiene within their current work organization. The study revealed that despite a less elaborated/fit-for-purpose FSMS, some butcheries are able to achieve a good microbiological output, if a good food safety climate is present in their organization.

Significance: The case study showed some interesting relations between food safety climate and the delivered microbiological safety/hygiene of food products. With the help of our assessment tool companies are able to go beyond traditional food safety management and mirror the human dimension in food safety.

P1-04  Steam Pasteurization of Edible Co-products from Sheep
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Introduction: Edible co-products from sheep such as hearts and diaphragms are used in dry fermented sausages in Norway. In 2006, dry fermented mutton sausages were found to be the vehicle of infection in a fatal outbreak of E. coli O103:H25. After this outbreak, Norwegian meat industry introduced decontamination interventions of sheep co-products.

Purpose: To investigate the effect of steam pasteurization of hearts from sheep with subsequent chilling in cold water baths and freezing.

Methods: The study was performed in one Norwegian abattoir. The carcasses were hung by the forelegs during evisceration, no rodding of esophagus. Hearts were sampled (a) before any treatment (n = 66), (b) after steaming for 40 s ± 3 s (n = 40), (c) after cold water bath (n = 30), (d) after steaming and cold water bath (n = 29), (e) after cold water bath and freezing (n = 30) and (f) after steaming, cold water bath and freezing (n = 30). Samples were analyzed for APC, Enterobacteriaceae, coliforms and E. coli.

Results: All treatments showed significant log reduction of APC, Enterobacteriaceae, coliforms and E. coli compared to non-treated hearts (a). No significant difference in log reduction was found between hearts that were steamed (b) and the ones only placed in cold water (c). Freezing, either after cold water (e) or after steaming and cold water (f), showed significantly more log reduction in APC compared to the other treatments. However, there was no significant difference between freezing after cold water (e) and the combination of steaming, cold water and freezing (f).

Significance: This study demonstrates that steam pasteurization of sheep hearts gives a significant reduction of microbial contamination. The hurdle effect of cold water bath with subsequent freezing gave further reduction and implementation of these steps will improve safety of dry fermented mutton sausages. However, freezing seems to be the most efficient treatment. More research is needed to see if freezing alone is sufficient to maintain food safety.

P1-05  Consumer Behavior When Shopping for and Storing Poultry May Result in Cross-contamination
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Introduction: Understanding how consumers handle poultry can highlight gaps in consumer knowledge/practice of food safety. Although many studies have been conducted related preparation and cooking of poultry, little work has been done on shopping behaviors for poultry related to potential cross-contamination.

Purpose: This study determined what products poultry microbes could potentially be transferred to during shopping/purchasing and initial home storage of poultry products.

Methods: An observational study where two researchers shopped along with consumers (n = 97) and then went home with them was used to assess actual shopping, transport, and storage behavior with raw poultry products. Shop-a-longs were conducted in three locations in the United States. Observers tracked poultry products handled by consumers; use of wipes, meat bags, or sanitizers; the next three items consumers touched after handling poultry; whether poultry touched other items in the cart; how poultry was handled at the checkout counter; and how poultry was handled at home.

Results: In 71% of the situations there was no hand sanitizer or wipes in the meat section. Plastic bags were found in the meat section 85% of the time, but only 25% of shoppers used the bag for their poultry products. Immediately after touching poultry items 85% of the time, but only 25% of shoppers used the bag for their poultry products. In the cart, poultry packages most often came into contact with dairy products, produce and other packaged goods. During checkout poultry was bagged separately from other products in 71% of the observations. A majority of shoppers (59%) stored poultry in...
the refrigerator or freezer without a plastic bag or other container, allowing the poultry to touch other refrigerated or frozen products.

**Significance:** Clearly potential for cross-contamination during shopping for poultry products exists. An increase in food safety education on the proper handling of poultry during purchasing, transporting, and storage is needed.

### P1-06 Recommendations for Determining Doneness Found in Egg Dish Recipes

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**Introduction:** Research has shown that many consumers do not follow recommended food safety practices for cooking egg dishes, which can lead to foodborne illnesses such as salmonellosis. For egg mixtures, the USDA recommends cooking until the center of the mixture reaches 160°F. In previous studies consumers have stated that they obtain their recipes from the internet, cookbooks, and family.

**Purpose:** The objective of this study was to determine what endpoint temperature information consumers receive when using egg dish recipes.

**Methods:** Two hundred twenty-six egg recipes from 65 websites, 50 cookbooks, and 9 magazine titles (multiple issues of each) were analyzed. Recipe types included in decreasing order: pie, quiche, custard, casseroles, frittata, strata, soufflé, omelet, torte, pudding, egg taco, baked French toast, macaron, and shakshouka recipes.

**Results:** Time was the most frequently used indicator, given in 90% of the recipes, with 15% using only time. Seventy-eight recipes gave multiple indicators for determining the endpoint of the cooking process, 95 gave a single indicator in addition to time, 50 gave only a single indicator, and 3 gave neither a visual nor a time indicator. Other indicators were if the product was set (95), browned (75), had a utensil inserted that came out clean (60), puffed (20), jiggling (13), and bubbling (5). Thermometer temperatures were given in only two of the recipes.

**Significance:** This review shows that consumers are not receiving information on endpoint temperatures recommended by USDA in the recipes they likely use for cooking egg dishes. Further work is needed to determine if the endpoint recommendations that are suggested result in safe cooking.

### P1-07 Effect of High Pressure Processing on the Survival of Campylobacter jejuni and Shelf Life of Chicken Fillets

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**Introduction:** High-pressure processing (HPP) is a preservation technology alternative to heat treatment that effectively inactivates the spoilage microbiota and foodborne pathogens of several foods.

**Purpose:** To evaluate the effect of HPP on Campylobacter jejuni, indigenous chicken microbiota and shelf life of chicken fillets.

**Methods:** Chicken breast fillets were inoculated with Campylobacter jejuni (3 strains - 10^6 CFU/ml), packaged under vacuum, subjected to HPP (500 MPa, 10 min) and stored at 4 and 12°C. Total viable counts, C. jejuni, Pseudomonads, Brochothrix thermosphacta, lactic acid bacteria, Enterobacteriaceae and yeasts/molds populations were determined, while enrichment was followed to ensure the presence/absence of the pathogen. Sensory analysis of non-inoculated samples determined the shelf life of the product.

**Results:** After HPP and during storage at both temperatures, the pathogen could not be detected even after enrichment. All the spoilage microorganisms remained below the detection limit of the enumeration method until the end of the product’s shelf life, except from Br. Thermosphacta which was found to be the main spoilage microorganism in HPP samples. In control samples, the pathogen’s population remained stable at 4°C, while it reduced approximately 0.5 log CFU/g at 12°C until the end of storage. The shelf life of the HPP samples was increased by 5 and 3 days at 4 and 12°C, respectively, in comparison with the control.

**Significance:** HPP may increase the shelf life of chicken meat and enhance the safety of the product.

**Acknowledgment:** The action THALIS: “Development, mathematical modeling and optional design of non-thermal technologies for processing, packaging, distribution and storage of safe high quality food products,” has been co-financed by the European Union (European Social Fund - ESF) and Greek national funds through the Operational Program “Education and Lifelong Learning” of the National Strategic Reference Framework (NSRF) - Research Funding Program: THALES: Reinforcement of the interdisciplinary and/or inter-institutional research and innovation.

### P1-08 Survival of Listeria monocytogenes during Shelf Life of Traditional Greek Dairy Products with Probiotic Potential

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**Introduction:** Food fermentation improves safety and shelf life of foods. However fermented dairy products have been incriminated in several cases of listeriosis outbreaks.

**Purpose:** To monitor the survival of Listeria monocytogenes in traditional Greek products (sour milk and yogurt) with or without a potential probiotic strain.

**Methods:** UHT milk was inoculated with Streptococcus thermophilus and Lactobacillus delbrueckii subsp. Bulgaricus (control) or with the former strains plus Lb. plantarum T571 (probiotic) following the fermentation procedures to produce sour milk or yogurt. Samples were also inoculated with L. monocytogenes (3 strains) at different initial levels (10^4, 10^5 CFU/ml). After the fermentation, samples were stored at 4 and 12°C until the end of the product’s shelf life. Microbiological analysis (TVC, LAB, thermophillic cocci and Listeria spp.) was performed in parallel with pH and titratable acidity measurements and sensory analysis (non-inoculated samples). Enrichment was followed to ensure the presence/absence of the pathogen. The survival of the probiotic and Listeria strains was assessed by Pulsed Field Gel Electrophoresis (PFGE).

**Results:** During fermentation, the pathogen increased approximately 0.5 log CFU/ml for both inocula, while LAB exceeded 8 log CFU/ml in all cases. At both control and probiotic, the low pathogen inoculum was detected in sour milk only after enrichment at 4 and 12°C, while the high inoculum was detected by enrichment at 12°C and enumeration at 4°C until the end of shelf life. In yogurt, the pathogen survived (enumeration) in all cases until the end of shelf life. PFGE showed that the survival of the different Listeria strains depended on the inoculum and storage temperature.

**Significance:** Although further research is required, high-quality products with increased safety were produced.
P1-09 **Biodiversity, Enterocin-producing Ability and Antilisterial Activity in Milk of Enterococci Isolated from Traditional Greek Graviera Cheese**

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**Introduction:** Enterococci often constitute an important part of the microbiota of traditional cheeses. Several *Enterococcus* strains possess technological, antimicrobial and potent probiotic properties which may be beneficial for cheese safety and human health, whereas other strains which may act as opportunistic pathogens should be suppressed.

**Purpose:** This study evaluated biodiversity and enterocin-producing (Ent+) ability of 46 spontaneous Enterococcus isolates from two batches of commercial starter-free Graviera cheese.

**Methods:** Selected Ent+ strains were further evaluated for antilisterial activity in thermized milk, whereas their enterocin gene/s type was characterized by PCR. Based on key biochemical criteria and intergenic spacer (IGS) profile similarity.

**Results:** Two major groups of strains were identified as *E. faecium* (15 isolates; L-arabinose +, sorbitol -) and *E. durans* (23 isolates; L-arabinose -, sorbitol -, mannitol -, sucrose -), whereas the remaining isolates were five atypical (sucrose +) *E. durans* and three *E. faecalis* (sorbitol +) strains. Both *E. faecium* and *E. durans* IGS groups showed high intra-species diversity based on additional differences in sugar fermentation patterns and whole cell protein profiles of the strains. Four strong Ent+ strains, which differed in their biochemical and SDS-PAGE protein profiles, were detected in the *E. faecium* group only; all four produced enterocin A (Ent-A+) and caused a complete growth inhibition of *Listeria monocytogenes* in culture media in vitro. Supplementation of thermized milk with the Ent-A+ *E. faecium* KE82 resulted in complete inhibition or significant retardation of *L. monocytogenes* growth at 37°C for 6 h, which was extended up to 3 days at 18°C; the degree of inhibition was dependent on the different cocktails of *L. monocytogenes* target strains used for milk inoculation.

**Significance:** Thus, Ent-A+ *E. faecium* strains that may persist in Greek raw ewe/goat milks or milk processing plants show promise for use as antilisterial adjunct cultures in traditional cheeses.

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P1-10 **Biochemical and Molecular Characterization of Lactic Acid Bacteria Isolated from Two Different Types of Greek PDO Galotyri Cheese**

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**Introduction:** Galotyri is a Greek PDO acid-curd cheese traditionally produced from ewes’ or ewes’/goats’ milk in the regions of Epirus and Thessaly. To date, commercial, starter-inoculated, industrial-type Galotyri is a fresh (non-ripened) soft cheese stored under refrigeration, whereas similar artisan-type Galotyri cheeses are also kept refrigerated after they are ripened shortly.

**Purpose:** This study examined the diversity of lactic acid bacteria (LAB) in commercial cheese products as affected by their production scale.

**Methods:** One hundred random isolates from industrial-type (30 isolates) or artisan-type (70 isolates) Galotyri cheeses were characterized by biochemical methods, and 31 representative strains of the phenotypic groups formed were then tested by molecular identification methods.

**Results:** *Streptococcus thermophilus* occurred as a LAB monoculture in industrial-type cheeses. Conversely, artisan-type cheeses included mainly *Lactococcus lactis* (28), *Lactobacillus paracasei* (21), *Enterococcus* spp. (14) and sporadic *Leuconostoc mesenteroides* (3), *Leu. lactis* (2) and *S. thermophilus* (2) isolates. Based on key biochemical criteria and intergenic spacer (IGS) profiles, enterococci were divided into three distinct IGS subgroups identified as *E. faecium* (L-arabinose +, sorbitol -), *E. faecalis* (sorbitol +, L-arabinose variable) and *E. durans* (L-arabinose -, sorbitol -, mannitol -, sucrose -); the former two subgroups included several enterocin-producing strains. Based on 16S rRNA gene polymorphism analyses with the primer pairs LacF/LacR and CefF/CefR, all representative *L. lactis* isolates tested were identified with the subspecies *lactis*. *Enterococcus* spp. and *S. thermophilus* isolates showed intra-species diversity in their sugar fermentation patterns and SDS-PAGE cellular protein profiles.

**Significance:** Overall, the results of this study indicate that industrial processing of Galotyri cheese limits its LAB diversity toward a high predominance of commercial thermophilic starter strains, whereas traditional manufacturing practices of artisan-type Galotyri cheese, including short ripening processes, favor the evolution of a more diversified technological microbiota consisting mainly of mesophilic LAB strains.

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P1-11 **Assessing the Response of Listeria monocytogenes during Storage of Various Cheese Types and after Subsequent Simulated Digestion**

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**Introduction:** Variability of cheese characteristics may significantly impact the *Listeria monocytogenes* dynamics during storage and the subsequent exposure to gastric acidity.

**Purpose:** The purpose was i) to evaluate the ability of various types of cheese to support growth of *L. monocytogenes* and ii) to estimate the impact of cheese matrix on the resistance of the pathogen in gastric fluid.

**Methods:** Portions of commercial cream (Mascarpone, Cottage), soft (Mozzarella, Camembert, Mastelo, Anthotyros, Manouri, Ricotta), and semi-hard (Halloumi, Gouda, Edam) cheeses (pH 5.0 - 6.7) of different initial microbial load (i.e., low vs high population of endogenous microbiota) were inoculated with ca. 2.0 log CFU/cm² or g of a 4-strain composite
(4b and 1/2a) of *L. monocytogenes* (n = 4). Samples were stored under vacuum or aerobic conditions at 7°C. *L. monocytogenes*, lactic acid bacteria, enterobacteria, yeasts/molds, total viable counts, pH, and *a*<sub>v</sub> were monitored during storage. Halloumi, Gouda, and Mozzarella were exposed to simulated gastric fluid (pH 1.5; HCl; 37°C) for 5, 10, 15, 20, and 40 min during storage at 7°C. Survivors of the pathogen were enumerated after exposure to gastric fluid using thin agar layer method (n = 3).

**Results:** Halloumi (semi-hard), Mozzarella/Camembert/Ricotta (soft), and Mascarpone (cream) cheeses supported growth of *L. monocytogenes* at 7.0 – 8.0 log CFU/cm² or g after 10 – 15 days at 7°C, while the pathogen remained almost stable in the other cheeses. High initial population of endogenous microbiota significantly suppressed (*P* < 0.05) growth of pathogen by 3.0 - 4.0 log CFU/cm² or g on Halloumi and Mozzarella. Survival in gastric fluid was only observed after habituation on cheeses that supported growth (Halloumi and Mozzarella), while on Gouda survivors were below the enumeration limit (10 CFU/cm²), regardless of the exposure time and storage day.

**Significance:** Mapping of *L. monocytogenes* response on various cheeses during storage and tolerance during simulating digestion may provide significant information to cheese industry.

**P1-12 Inhibition of *E. coli* O157 and *Listeria* Species by Antagonistic Bacteria Isolated from Food Raw Materials**

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**Introduction:** Consumption of foods contaminated with pathogenic bacteria may cause severe diseases or can lead to fatal cases; therefore, inhibition of foodborne pathogens is a significant task for participants of food industry. Application of microorganisms that are able to inhibit or even kill the cells of bacterial pathogens can be used for this purpose. Antagonistic bacteria may overgrow the pathogens or inhibit their growth by production of different antimicrobial metabolites.

**Purpose:** The aim of this study was the selection of bacteria from different food raw materials with antimicrobial activity against *E. coli* O157 and different *Listeria* species. Characterization of the extracellular metabolites of the antagonistic strains has also been performed.

**Methods:** Antagonistic effect of the tested bacteria was determined by co-culturing, while the antibacterial substances were characterized by different analytical methods.

**Results:** Out of the tested bacteria, 24 showed antilisterial, while 16 had anti-*E. coli* O157 activity, however it was observed that direct application of these bacteria would not be possible for pathogen inhibition, as the required high cell density could cause spoilage of the food. Cell free supernatants of the antagonistic cultures contained extracellular substances that were presumed to have been contributed to pathogen inhibition. Siderophores were identified as the main inhibitory compounds but other protease- and heat-resistant metabolites could also have had impact on growth of the pathogens.

**Significance:** Application of antagonistic bacteria or their extracellular metabolites in food processing can increase the safety of foods and contribute to the protection of consumers’ health.

**Acknowledgment:** This research was supported by the European Union and the State of Hungary, co-financed by the European Social Fund in the framework of TÁMOP 4.2.4. A/-11-1-2012-0001 ‘National Excellence Program’, and by Hungarian project OTKA 101716.

**P1-13 Thermal Inactivation of Feline Calicivirus in Pet Food Processing**

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**Introduction:** Extrusion is the most common manufacturing process used to produce heat-treated dry pet food for domestic use and international trade. Due to reoccurring outbreaks of notifiable viral animal diseases and their impact on international trade, experiments were undertaken to demonstrate the effectiveness of heat-treated extruded pet food on virus inactivation.

**Purpose:** The objective of the present study was to design experimental equipment and methods for deriving key parameters for virus inactivation in three representative pet food raw materials used in the extrusion cooking of pet food and to obtain thermal inactivation data.

**Methods:** Pet food raw materials consisted of uncooked chicken meat obtained from chicken racks (CKN), mixed poultry meal (PoM) and an animal-derived palatant (PAL). Small scale heat inactivation studies used a customized heating block and a small-scale pressure reactor capable of operating at extrusion pressures. The feline calicivirus vaccine strain FCV F-9 was used as a surrogate model RNA virus pathogen and FCV F-9 inactivation was measured by plaque assays.

**Results:** Small-scale heat inactivation experiments showed that a > 4 log reduction in infectivity occurred at 70°C prior to reaching the minimum manufacturing operating temperature of approximately 100°C. The data from the small scale pressure reactor showed complete inactivation of all samples heated to 70°C, irrespective of either pressure or the original matrix moisture content. FCV F-9 was shown not to survive the acidic conditions used to produce pet food palatants of animal origin that are typically used after the coagulation effect of the extrusion process.

**Significance:** This study provides evidence showing that the extrusion manufacturing process for pet food results in significant inactivation of the surrogate model feline calicivirus. In the future, the experimental procedures described here could be used to test the inactivation of economically relevant animal virus pathogens that may cause unnecessary trade disruptions.

**P1-14 Microbial Interactions in Blue-veined Cheese: Effect on *Listeria monocytogenes* Behavior and Shelf-life Prediction**

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**Introduction:** Among dairy products, blue-veined cheeses are known to be the most frequently contaminated with *Listeria* spp. This can be due to process contamination (during the piercing at the beginning of the ripening or during the cut at the beginning of the shelf life). The development of a complex microbial community (lactic acid bacteria, yeasts and molds) which metabolize different compounds, modulate the pH values that can affect the pathogen behavior.
Purpose: The objectives of this study were to i) investigate the effect of the physicochemical changes on the *L. monocytogenes* behavior during the cheesemaking, ii) study the pathogen behavior during the shelf life assuming different time contamination: at the beginning of the ripening and at the beginning of the shelf life, and iii) predict the shelf life with different hypothetical scenarios.

Methods: Two challenge tests were carried out with *L. monocytogenes*: (i) by cow milk contamination to evaluated the pathogen behavior during the process, and (ii) by contamination of portioned cheese to evaluate the growth pathogen during the shelf life. For microbiological analysis, counts were estimated by plate count. The growth curves of *L. monocytogenes* were fitted using DMFit web edition, in order to predict the shelf life at different storage temperature.

Results: The pH value decreases by 6.65 ± 0.02 in milk to 4.93 ± 0.01 in cheese (1 d); during the ripening the molds growth (from 2.77 ± 0.01 to 6.45 ± 0.39 log CFU/g) causes an increase of the pH until 6.64 ± 0.14, thus aiding development of *L. monocytogenes*. During the shelf life, the pathogen was able to grow in cheese in both contamination time, but with difference growth rates.

Significance: The present data can be a useful tool for the quantitative risk assessment for *Listeria* in blue-veined cheese and to study alternative ways to reduce the *Listeria* contamination in this type cheese.

**P1-15** Americans’ Food Safety Practices for Raw Poultry and Shell Eggs: Areas for Improvement
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Introduction: A major source of consumers’ exposure to *Salmonella* and *Campylobacter* is from raw poultry and shell eggs. These products are frequently consumed in the United States; thus, exposure to these products is great. Improvements in consumers’ food safety practices when cooking at home may help to reduce foodborne illness attributed to these pathogens.

Purpose: To characterize U.S. consumers’ storage, handling, preparation, and cooking of raw poultry and shell eggs and to identify specific areas for improvement.

Methods: Conducted a nationally representative web-enabled panel survey of U.S. adult grocery shoppers (n = 1,504) to estimate the percentage of Americans who follow recommended food safety practices for raw poultry and shell eggs when preparing food at home.

Results: The survey results identified areas of low adherence to recommended food safety practices for raw poultry: not rinsing raw poultry before cooking (31.3% followed recommended practice), proper refrigerator storage of raw poultry (11.0%); using a food thermometer to check the doneness of small cuts of chicken (16.3%); and proper thawing of raw poultry in cold water (11.0%). Areas of low adherence to recommended food safety practices for shell eggs included the following: washing hands after cracking eggs (48.1%), cooking fried eggs until the yolks and whites are firm (46.6%), and using a food thermometer to check the doneness of baked egg dishes (3.2%).

Significance: Adherence to some recommended food safety practices is low among U.S. consumers. The survey findings, coupled with other research findings, will help to inform the development of science-based consumer education materials that can help reduce foodborne illness from *Salmonella* and *Campylobacter*. This research was funded in part through a grant from the Agriculture and Food Research Initiative Competitive Grants Program (grant no. 2012-68003-19606) from the U.S. Department of Agriculture, National Institute of Food and Agriculture.

**P1-16** An in vitro Model of Intestinal Cells to Study the *Yersinia enterocolitica* Ability to Colonize Pigs and to Infect Humans
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Introduction: *Y. enterocolitica* is the fourth bacterial cause of human enteritis in Europe. The species is divided into six biotypes (BT). BT1A regarded as non-pathogenic and pathogenic biotypes 1B, 2, 3, 4 and 5. Pigs, the principal reservoir for human pathogenic strains, do not develop clinical signs. The BT4 is the most frequently biotype isolated from pig and encountered in human yersiniosis.

Purpose: This study investigated the use of *in vitro* cultured cells to assess the ability of *Y. enterocolitica* to adhere and invade pig and human cells.

Methods: We tested *in vitro* the adhesion and invasion abilities of a collection of 23 *Y. enterocolitica* on intestinal pork cells IPEC-J2 and on human intestinal cells Caco-2. The analysis was done by performing a hierarchical cluster and a Newman-Keuls test.

Results: This study has established adhesion / invasion profiles for strains belonging to the five biotypes found in France. With the IPEC-J2 cellular model two profiles of colonization were identified. A profile characterized by strains possessing invasion capacities of less than 3% and another profile containing predominantly BT4 strains and corresponding to strains possessing invasion capacities greater than 3%. For the *in vitro* tests performed on Caco-2 cells, the early results suggested that the overall profile of adhesion / invasion was different. However the BT1A and the BT5 strains which are rarely isolated from pigs still show a low capacity to invade and to adhere Caco-2 cells.

Significance: These results reflect the ability of BT4 to colonize pigs and the low capacity to BT1A and BT5 to colonize pigs and humans.

Acknowledgment of financial support: Brittany Region.
P1-17  
**Listeria monocytogenes** Strains in Co-cultivation; Effect on Resistance to Simulated Gastric Fluid and Detection During Selective Enrichment by ISO Methods

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**Introduction:** Food samples can be contaminated with more than one strain of a pathogen. *Listeria monocytogenes* strains isolated from foods by selective enrichment might not always represent the strains that can better survive a patient immune system. 

**Purpose:** To investigate the effect of co-cultivation on the (i) acid response of *L. monocytogenes* strains to simulated gastric fluid (SGF), and (ii) isolation of strains during selective enrichment (SE). 

**Methods:** The recovery of four *L. monocytogenes* strains (C5, 6179, ScottA, PL25/different MLST types) after SE and their gastric-acid tolerance, were assessed during growth in Tryptone Soy Broth (TSB) (10°C/8 d) in single or mixed cultures (1:1 strain-ratio). Antibiotic-resistance was induced to strains for selective enumeration. Acid-stress response (ASR) of strains to SGF (pH 2.0, 37°C) was evaluated for 18, 48, 60 and 90 min on 2, 4, 6 and 8 d, respectively. On the same days, ISO 11290-1:1996/Amd 1:2004 enrichment protocol was used to determine the recovery of strains in mixed cultures from TSB. 

**Results:** Co-cultivation did not affect the gastric-acid tolerance of strains in mixed cultures compared to their single cultures. Significant ASR differences (*P < 0.05*) between strains in mixed cultures were observed on days 6 and 8. On day 6 after 36 min exposure to SGF, C5 and 6179 survived by 3.8 and 1 log CFU/ml (enumeration limit), respectively. On the same day, 6179 had lower recovery percentage after SE compared to C5 (1% and 99% of colonies on ALOA, respectively). However, SE did not always favor the strains with higher ASR. On day 8, ScottA (clinical isolate) displayed increased acid-resistance compared to C5 (dairy-farm environment isolate), but was outcompeted during SE and could not be isolated on ALOA. 

**Significance:** The results underline the necessity for improvement of selective enrichment methods and could aid investigations of *L. monocytogenes* infection sources and routes.

P1-18  
**Comparison of Methods for the Detection of Contamination in UHT None Milk Products**

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**Introduction:** The testing of UHT products is an essential process to ensure contaminated products are detected prior to shipment and storage. ATP Bioluminescence has been used to detect UHT contamination in milk samples for many years and the same technology can be used for a range of none milk products. Unlike milk samples the removal of the back ground ATP can be more difficult in UHT none dairy samples. This has reduced the widespread use of this rapid test in the none dairy industries. 

**Purpose:** ATP Bioluminescence technology can be used as a method to detect contamination in UHT products but none milk samples have proved to be difficult historically. This project looked at comparisons between different methods and used food samples not traditionally tested with ATP bioluminescence methods. 

**Methods:** A range of none milk UHT products were tested using traditional microbiology, pH testing and 3M MLS II Bioluminescence technologies to see if low levels of contamination could be detected. Store bought material was used and contaminated with low levels of a range of microorganisms to simulate the effects of failed UHT process. The range of organisms used were all found in UHT production sites and have caused sterility issues in the past. 

**Results:** The 3M MLS II test allowed for rapid detection of low level contamination with a range of microorganisms. Low level contamination was found in none dairy samples in less than 48 h compared to 5 days with traditional microbiology and 8 days with pH testing. 

**Significance:** This rapid detection of sterility failures means customers can be more responsive to issues seen in production and help prevent the failure to detect microbial spoilage as was seen in some of the other methods.

P1-19  
**Study of the prfA Virulence Gene Cluster in Listeria monocytogenes Strains of Different Origin**

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**Introduction:** *Listeria monocytogenes* is an important foodborne pathogen that can colonize food-processing environments. Its intracellular virulence potential is dependent on the *prfA* virulence gene cluster (*vgc*) and can vary among strains of different origin and serotypes. 

**Purpose:** To investigate the diversification of *prfA*-vgc genes among *L. monocytogenes* strains (clinical and food isolates) of different geographical origin and serotypes. 

**Methods:** *prfA, mpl, actA, plcA, plcB and hly* genes were sequenced for 24 strains; 12 from Greece and 12 from Ireland. For each country, 6 strains (3 of serotype 1/2a and 3 of serotype 4b) originated from food processing environments and similarly 6 strains were clinical isolates. Gene and protein sequences of the *prfA*-vgc were aligned using MUSCLE, and maximum likelihood phylogenetic trees were created using PhyML. A Pulsed Field Gel Electrophoresis (PFGE) analysis was also performed using the PulseNet protocol. 

**Results:** PFGE separated strains of serotypes 1/2a and 4b into two distinct clusters. The phylogenetic trees of *actA, plcA, plcB* and *mpl* showed lineage-depending divergence with two main clusters, of 1/2a and 4b serotypes strains. The highest sequencing divergence at the protein level among the 6 virulence genes was exhibited by *plcA, actA* and *plcB*. Most conserved gene sequence was recorded for the *prfA* gene. The mutations in *prfA* DNA sequences were all silent-mutations as no modifications in the protein sequences were observed for all strains. No significant correlations of the *prfA*-vgc to food or clinical isolation source was observed. Country-specific grouping of the strains was recorded for the *hly* gene and its protein sequence, where 20 out of 24 strains were clustered according to their Irish or Greek origin. 

**Significance:** Strain-specific virulence determination provides new insights into *L. monocytogenes* virulence and epidemiology and allows a better risk assessment of the pathogen.
**P1-20 Study of the Validation of Low A_w Food Roasting Processes Targeting Salmonella spp., Using Cocoa Bean Roasting as a Model System**

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**Introduction:** Due to the increased heat resistance of microorganisms in low A_w foods, there is growing concern for the potential presence and survival of some vegetative pathogens, in particular Salmonella spp., through the decontamination steps applied to these products. This could cause enormous losses to manufacturers and risks the health of consumers.

**Purpose:** This study aims to evaluate and validate potential decontamination technologies for low A_w foods, using the roasting of whole cocoa beans as an example process with the aim of improving established approaches to validation of low A_w products.

**Methods:** Lab-based D- and z-values were established for Salmonella spp. and Enterococcus faecium NRRL B-2354. These data was then used to compare with thermal inactivation studies carried out in a pilot scale roaster with inoculated product.

**Results:** D- and z-values determined for Salmonella spp. and E. faecium confirmed the suitability of the latter as a surrogate organism for Salmonella in this food type. z-values of 37.0 and 34.2°C were calculated for S. Typhimurium ATCC 14028 and E. faecium NRRL B-2354, respectively. D-values calculated in the laboratory oven and the pilot roaster were 8.6 minutes and 8.3 minutes, respectively, showing very good equivalence. Log reductions predicted based on calculation of cumulative hold time at 145°C using laboratory-determined z-values were lower than those observed by analysis of inoculated product processed in a pilot roaster, which is to be expected given that the exact combined lethal effects of roasting are demonstrated in pilot equipment.

**Significance:** The results of the study suggest that the use of laboratory inactivation data alongside thermal profiles can be used where processing of inoculated product in pilot or production equipment is not possible. However, where product composition may change during heating (i.e., protect microorganisms), a validation study in pilot or production equipment would be required.

**P1-21 Fate of Listeria monocytogenes in Vanilla Cream with or without Cinnamon Extract under Different Isothermal Conditions**

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**Introduction:** Vanilla cream is a Ready-To-Eat milk-based product which, although typically subjected to pasteurization, may be exposed to post-process contamination with pathogenic bacteria in the industry environment compromising the product’s safety.

**Purpose:** Evaluation of the growth behavior of Listeria monocytogenes in pasteurized vanilla cream formulated with/without cinnamon extract.

**Methods:** Commercially prepared vanilla cream, formulated with/without cinnamon extract, was inoculated with a five-strain mixture of L. monocytogenes (ca. 2 log CFU/g) and stored aerobically at 4, 8, 12 and 16°C. At appropriate time intervals, duplicate samples were analyzed and L. monocytogenes populations were determined. A primary model was fitted to the microbiological data for the estimation of the pathogen’s kinetic parameters.

**Results:** The kinetic behavior of L. monocytogenes was comparable in cream with and without cinnamon extract during storage at 12 and 16°C. Some level of growth inhibition was evident in cream with cinnamon extract during storage at 8°C, whereas significant (P < 0.05) were the differences observed between the two product types at 4°C. The estimated mean values of the lag phase duration (days) and the maximum specific growth rate (days^-1) of the pathogen in cream without cinnamon extract stored at 4°C were 2.28 and 0.59, respectively, while the corresponding values for the product formulated with cinnamon extract were 4.29 and 0.18.

**Significance:** The inclusion of cinnamon extract appears to have an important anti-listerial effect, provided that efficient temperature control is ensured. This work has been supported by the project “Efficacy of NOVEL analytical techniques to prEdict the quality and safetY of newly developed pErishable food products 11SYN_2_1528” co-financed by the European Union (European Social Fund – ESF) and Greek national funds through the O.P. “Competitiveness and Entrepreneurship (OPC II)” ROP Macedonia – Thrace, ROP Crete and Aegean Islands, ROP Thessaly – Mainland Greece – Epirus, ROP Attica, Framework NSRF 2007-2013, COOPERATION 2011.

**P1-22 Improving the Enrichment Procedure for the Detection of Low Contamination Levels of Shiga Toxin-producing E. coli in Sprouts**

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**Introduction:** In 2011, an outbreak of Shiga Toxin-producing E. coli (STEC) infections in Germany was caused by a strain belonging to serotype O104:H4. Epidemiological data linked the outbreak to the consumption of sprouts; even so the pathogen could never be detected in sprout samples nor seeds. For STEC detection in sprouts, ISO/TS 13136 prescribes an unselective enrichment step in BPW for 24 h at 37°C, followed by a PCR detection of stx genes.

**Purpose:** The purpose of this study was to assess and improve ISO enrichment procedure for the detection of low STEC levels in sprouts.

**Methods:** First, 10 g of soy sprouts were spiked with 10-100 CFU of stressed STEC and enriched (1:10) overnight in BPW (ISO procedure). The presence of stx1 and stx2 in the enriched samples was tested with a real time multiplex PCR system. In a second step, six enrichment broths used for the enrichment of Enterobacteriaceae were compared for their ability to promote growth (at 37°C and 42°C) of six stressed and unstressed STEC strains of different serotypes. Based on the results, some of these media were further tested with spiked sprouts.
Results: The experiments with sprouts spiked with low levels of stressed STEC, enriched in BPW, showed that contamination levels below 10<sup>3</sup> CFU per 10 g could not be reliably detected with PCR. The main reason for the detection failure is probably the very high levels of the Gram negative background flora (10<sup>6</sup> CFU/g) in sprouts. The evaluation of selected unstressed STEC strains showed no growth differences between the strains at 37°C and 42°C in EE broth, BGB broth and BPW. Stressed strains, however, showed a longer lag time for both 37°C and 42°C and a greater strain variation.

Significance: An improved enrichment procedure for the detection of STEC will significantly strengthen food safety of sprouts.

P1-23 Public Health Relevance of Cross-contamination in the Fresh-cut Vegetable Industry
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Introduction: While preharvest control strategies during growing and harvesting are generally recognized as critical to reduce the risk of contamination, there is still much reliance on produce decontaminations strategies applied by the processing industry in various countries. However, in some EU member states the use of antimicrobial agents in fresh-cut produce processing is prohibited. Although the phenomenon of cross-contamination is real and intuitively important, assessment of the true public health relevance is largely neglected.

Purpose: The goal of this was to assess this role of indirect contamination (i.e., contamination as a result of cross-contamination) relative to the contribution of the direct contamination pathway.

Methods: We constructed a transparent and intuitive model that may serve as a tool to make science-based decisions on the public health relevance of water disinfection in the fresh-cut industry. The model quantifies the number of disease cases resulting from a point-contaminated batch of lettuce that is processed under standard conditions.

Results: The model provided some useful insights in the role of indirect contamination of lettuce via the washing process. First, the contamination is diffusely distributed; the contamination is dyed to the bath, the less important role the role of cross-contamination. Second, the results indicate that direct contaminated portions are responsible for more than 90% of the resulting disease cases, putting emphasis on the need for preharvest avoidance of contamination. The indirect contamination via spread in the washing water only plays an important role when initial concentrations exceed 10<sup>3</sup> per batch in combination with a highly virulent pathogen (i.e., STEC O157 or O104).

Significance: We showed that cross-contamination with the washing water in the fresh-cut vegetable industry is of limited public health concern. Although a frequent refreshment of washing water is a fail-safe method to control cross-contamination, further analysis is required into the balance between safety and sustainability.

P1-24 Antimicrobial Resistance, Molecular and Phenotypic Diversity of Escherichia coli isolates from Fresh Vegetable Products in Korea
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Introduction: Escherichia coli is generally considered an indicator of antimicrobial resistance due to its genetic flexibility and adaptability. Fresh vegetable products have been recognized as an important reservoir of E. coli, and outbreaks of foodborne illnesses caused by the consumption of such foods have dramatically increased.

Purpose: This study is focused on following characteristics of the E. coli isolates obtained from fresh vegetable products in Korea: a) resistance to antimicrobial agents, b) genetic relationships based on repetitive sequence-based PCR (rep-PCR) technology, and c) ability to form biofilm.

Methods: E. coli isolates were obtained from 413 samples collected from retail markets in South Korea. The antimicrobial susceptibility was examined by the broth dilution test against 17 antimicrobials using the VITEK<sup>2</sup> compact system (bioMérieux, Marcy l’Étoile, France). The genetic relationships of E. coli were analyzed based on repetitive sequence-based PCR (rep-PCR) technology. Biofilm formation ability was assessed using an assay based on crystal violet staining.

Results: Among the 120 isolates, 22 isolates (18.3%) were resistant to one or more antimicrobials and 11 isolates were concurrently multidrug-resistant against up to 6 antimicrobial agents. The highest resistance rate was detected for ampicillin (14.2%), followed by piperacillin (11.7%) and cefalotin (10.0%). Strains carrying the same antimicrobial resistance profile had discrete rep-PCR patterns. Significantly larger amounts of biofilm were formed by an isolate which showed no antimicrobial resistance, but 60% of the five intermediated biofilm forming E. coli revealed multi-drug resistance against three or four antimicrobial agents.

Significance: Antimicrobial resistance and biofilm formation ability of E. coli isolated from fresh vegetable products showed these products such as fresh-cut salads and unpasteurized vegetable juices may contribute directly to human exposure to antimicrobial resistant bacteria. These results provide useful information to develop intervention technologies and in risk assessment to assure food safety and public health.

P1-25 The Analysis of Microbial Contamination of Soils and Farm Products Near Disposal Sites of Animals
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Introduction: As outbreaks by foot-and-mouth disease and avian influenza extended, many livestock were culled and buried to stamp out in South Korea in 2010. The farm lands could be contaminated by leachate flow from burial sites and agricultural products also could be infected with hazardous microorganism which rapidly proliferated in contaminated soils.

Purpose: The objectives of this study were to analyze and monitor the microbial contamination of soils and products near carcass burial sites from 2012 to 2013.

Methods: Samples of soils (n = 253; 2012 - 2013) and farm products (n = 32, 2012; n = 16, 2013) were collected from agricultural lands where have been cultivated near burial sites. The Clostridium perfringens as bacterial indicator of contamination were analyzed on soils. The agricultural products showing contamination of C. perfringens from soils were analyzed for Bacillus cereus, C. perfringens, Escherichia coli, Staphylococcus aureus, Salmonella spp. and E. coli O157:H7.
Results: In regard to detection of *C. perfringens* in soil near foot-and-mouth disease and AI burial sites, of 253 soil samples, the pathogen was detected in 46 cases (18.2%) in 2012 while 2013 findings showed just 20 cases (7.9%), resulting in 10.3% decrease. In 2012, for 36 agricultural products, 11 cases of *B. cereus*, 4 for *C. perfringens* and for *E. coli* were detected in rice, green chili and maize, whereas in 2013, for 16 products, 5 cases for *B. cereus* and 1 for *C. perfringens* were found in rice, pumpkin and spring onion. There was no detection of *E.coli O157:H7, Salmonella* spp. and *S. aureus* from 2012 to 2013. According to the result from 2012 and 2013, it was shown that degree of microbial pollution around livestock burial sites was decreased, but microbial safety management is still required to continue.

Significance: Outcomes of this study showed decreased case of soils and products contaminated. However, more monitoring of microorganism is needed to provide safe products to consumers.

P1-26 Analysis of Simultaneous Mycotoxins in Pearl Barley Using Simple Extraction and LC-MS/MS

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Introduction: Mycotoxins are toxic substances produced by fungi, causing serious health risks to humans and animals. Mycotoxins can contaminate cereals under their favorable temperature and humidity conditions in the level of cereals storage and production. Many countries have established maximum levels for mycotoxins occurring in cereals. To control mycotoxins contaminants, a rapid, simple and effective detection method is required.

Purpose: This study was performed to validate simple extraction combined with liquid tandem mass spectrometry (LC-MS/MS) for simultaneous analysis of 7 mycotoxins in pearl barley. This method was applied to the occurrence of mycotoxins in real samples.

Methods: A total of 73 processed pearl barley samples were collected from retail markets in Gyunggido, Korea. Sample was extracted with a mixture of acetonitrile/water (84:16). This raw extract was diluted (1:10) with water containing 0.1% formic acid/5mM ammonium acetate, filtered and injected to LC/MS/MS. For simultaneous mycotoxins analyze, aflatoxins (B1, B2, G1, G2), ochratoxin A, deoxyxynivalenol, zearalenone were used.

Results: The limits of detection (LOD) and limits of quantitation (LOQ) were determined 0.36 (zearalenone) ~10.45 (deoxynivalenol) µg/kg, respectively. Matrix matched calibration curves showed good linearity for 7 mycotoxins with determination coefficients higher than 0.996. Mean recovery values ranged from 80.1% (aflatoxin G2) ~ 100.1% (deoxynivalenol). Deoxynivalenol was observed in 4 (5.5%) samples and its contamination levels were from 11.0 to 34.6 µg/kg. 2 (2.7%) samples were contaminated by zearalenone and its levels were 9.2 and 20.0 µg/kg.

Significance: The levels of mycotoxins contamination on 73 samples were far below the European Union regulatory limits. This method could be a simple, rapid, efficient way to determine 7 different mycotoxins in pearl barley, simultaneously.

P1-27 Histology as a Valid Tool to Differentiate Fresh from Thawed Marinated Fish

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Introduction: According to Regulation (UE) 1276/2011, fish intended for raw consumption is expected to be frozen at -20°C for 24 h or -35°C for 15 h, in order to reclaim parasites. To fulfill EU requirements, since 2009 at IZSPLVA a histological method to identify fish as fresh/frozen has been validated and accredited with the following performance: sensitivity 100% (95% CI: 90.70% – 95.90%), specificity 92.59% (95% CI: 75.71% – 99.09%) and is now in place for official controls. The method was validated independently from the species analyzed. In Italy, especially in coastal areas, the consumption of raw marinated fish is rather common. The marinating process, however, is not effective in destroying nematode larvae, so these preparations have to be frozen before consumption.

Purpose: In this study we evaluated the performance of histology to distinguish between fresh and frozen-thawed raw marinated fish, in order to extend the field of application of the method.

Methods: Sixty raw anchovy fillets (*Engraulis encrasicolus*) caught in Sicily were immersed in a marinating bath prepared with water, wine vinegar salt and citric acid at refrigeration temperature for 24 h. Thirty samples were refrigerated, thirty were frozen for 24 hours at -20°C before marinating. All samples were fixed in 10% neutral buffered formalin and routinely processed for paraffin embedding, stained with haematoxylin and eosin (HE), microscopically observed and classified as positive (thawed) or negative (fresh) according to the standard operating procedure criteria in use.

Results: Results showed very high values of sensitivity 100% (95% CI: 89.4% - 100%) and specificity 100% (95% CI: 87.2% – 100%) of the method.

Significance: These results highlighted that histology is a valid and reliable tool to differentiate fresh from thawed fish, including marinated fish and that the method can be applied to guarantee food safety and avoid consumer exposition to the risk of infestation from nematodes.

P1-28 Electron Microscopic Study on Kudoa septempunctata Infesting Olive Flounder

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Introduction: *Kudoa septempunctata* is a myxosporean parasite of olive flounder that causes more than 50 cases of foodborne illness in Japan each year. This illness begins between 2 and 20 h after ingestion of raw olive flounder. The symptoms include transient but strong diarrhea and emesis. For quantitatively assessing the presence of *K. septempunctata* spores in the causative fish at food poisoning outbreaks, both direct observation method under a microscope and quantitative real-time PCR method (qRT-PCR) are officially accepted in Japan.

Purpose: The lower correlations have been often noticed between the number of spores counted by the direct observation method and the DNA amount determined using qRT-PCR method. To elucidate this discrepancy, we observed muscular tissues of infected olive flounders with *K. septempunctata* by an electron microscopy.
Methods: Olive flounder infected with K. septempunctata were purchased from a local fish farm in Japan. The samples for electron microscopy were prepared in conventional procedure and observed by transmission electron microscopy at an acceleration voltage of 80 kV.

Results: The images demonstrated unsynchronized development of K. septempunctata spores in plasmodia found within myofibers; in other words, the plasmodium contained not only developed spores but also developing spores (sporoblasts). Furthermore, the ratio between developed spores and sporoblasts varied at different parts of muscles.

Significance: A direct microscopic observation method could count developed spores, whereas qRT-PCR method could quantify the DNA amount of not only developed spores but also sporoblasts regardless of the cellular development and differentiation. Considering that the food toxicity caused by K. septempunctata is induced by developed spores passing through the gastric environment, the direct observation method counting only developed spores is better than qRT-PCR method for assessing the cause of foodborne illness at the outbreak as well as the risk of human illness in monitoring surveys of aquacultured or natural-water fish.

Poster Session 2 – Communication Outreach and Education, Epidemiology, Food Defense, Food Toxicology, Modeling and Risk Assessment, Retail and Food Service Safety, Sanitation

Tuesday, 21 April – 10.00–18.00

P2-01 Cabin Crew Food Safety and Hygiene Training — An Exploratory Study
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Introduction: The production and service of airlines meals is a “high-risk mass catering operation” with food safety implications, including temperature control during receiving/loading, storing and regeneration of meals, personnel hygiene, cross-contamination, food allergy and poisoning. Food service is a crucial part of cabin crew on-board duties, therefore and according to the regulations, cabin crew should be instructed/trained on food safety and hygiene. However, to date, there is no in-depth study on this topic.

Purpose: The aim of this study was to explore the extent, and current features of food safety training for airline cabin crew. Information obtained can be used to inform development of future training models and training methods and evaluation.

Methods: Using a snowballing technique, a sample of 26 cabin crew training managers/supervisors participated in in-depth, semi-structured interviews (n = 20). A content analysis of documents, e.g., cabin crew manuals, identified current airline food safety procedures.

Results: Twenty-six respondents from 20 international airlines participated in the study. All respondents agreed that cabin crew food safety/hygiene issues are important in relation to food-handling on board, for example “food safety is always an important issue,” “it is almost a major issue while handling food on-board.” Findings indicated that most airlines (18/20) used classroom methods to train cabin crew about food safety and two airlines delivered food safety training using a self-access e-learning module. The duration of training varied from “no specific time,” “30minutes,” “an hour,” to “two days.” Training did not cover HACCP; different levels of cabin crew members were trained to the same level of food safety. Most airlines (14/20) delivered food safety training as a part of the induction foodservice training and all airlines relied on internal training.

Significance: Cabin crew food safety training appears to be lacking in time of training and its content in relation to different cabin crew roles. HACCP training is required to minimize potential food safety risks on board.

P2-02 Listeriosis Risk Factors of Women during Pregnancy and the Potential Impact of Targeted Food Safety Information
ELLEN EVANS and Elizabeth Redmond
Cardiff Metropolitan University, Cardiff, United Kingdom

Introduction: Historical surveillance data (1980s-1990s) suggest listeriosis incidence was commonly associated with pregnant women. Recent international data (1999s-onwards) suggest decreased incidence of pregnancy-associated infection, thus suggesting successful implementation of targeted food safety information initiatives. Research to determine pregnant women’s current food safety behavioral influences is lacking.

Purpose: This study aimed to determine self-reported practices associated with listeriosis risk factors and evaluate potential impact of food safety education during pregnancy.

Methods: A self-complete questionnaire was completed by pregnant (20%) and post-partum (80%) women (n = 40). Risk perceptions, home food safety knowledge, attitudes, self-reported practices implemented during pregnancy related to specific Listeriosis risk factors were determined.

Results: The majority (90%) believed that during pregnancy it is ‘more important’ to implement food safety practices and 63% reported that practices changed. Consequently, many reported to be more careful with cooking (73%), following ‘use-by’ dates (67%) and with consuming foods within two days of opening (68%). The majority (70-83%) recalled receiving information regarding healthy eating and keeping active during pregnancy. Conversely, only 45% recalled receiving food safety information, of which 75% reported the information to be useful and 80% reporting that the information did change
their practices. No significant differences ($P > 0.05$) were determined in self-reported food safety practices relating to listeriosis risk factors during pregnancy whether food safety information was received or not. Furthermore, perceptions of risk, control and responsibility were not correlated ($P > 0.05$), suggesting perceived food poisoning risk during pregnancy is perceived to be beyond individual control and responsibility.

**Significance:** Many indicated awareness of food safety risks associated with pregnancy, resulting in reported improvement of practices during pregnancy. Although food safety information was reported to have a positive impact on practices of individuals, findings suggested no significant differences existed in self-reported food safety practices according to recall of receiving food safety advice. There is a need for targeted food safety education to further raise awareness of the potential ability to control risks associated with listeriosis during pregnancy.

**P2-03 Cancer Patients and Caregivers Risk Perceptions, Awareness of Food Safety and Self-reported Receipt of Food Safety Information during Chemotherapy**

**ELLEN EVANS,** Adrian Peters, Simon Dawson and Elizabeth Redmond
Cardiff Metropolitan University, Cardiff, United Kingdom

**Introduction:** Chemotherapy patients have an increased risk of foodborne illnesses, particularly listeriosis, due to immunosuppression. It's important that cancer patients consume foods prepared/stored at home according to recommendations to ensure food safety and minimize risk of illness. Personal invulnerability and perception of current behaviors endangering health is reported to increase implementation of risk-reducing behaviors. However, research detailing cancer patients/caregiver perceptions and awareness of food safety is currently lacking.

**Purpose:** This study aims to establish patients/caregiver awareness of food safety risks during chemotherapy in conjunction with reported receipt of educational materials. This will increase understanding of current practices and aid development of future targeted food safety communication approaches.

**Methods:** A self-complete questionnaire (online/paper-based) was completed by chemotherapy patients ($n = 63$) and caregivers ($n = 39$) responsible for food preparation. Attitudes towards risks associated with food safety and related education were determined.

**Results:** Only 61% of participants (56% patients/70% caregivers) were aware of increased risks of foodborne illnesses during chemotherapy; only half (50%) reported food safety practice improvement during chemotherapy. Perceived importance of implementation of key food safety practices increased significantly ($P < 0.01$) post-cancer diagnosis, more so among caregivers than patients. During chemotherapy, the majority received information concerning keeping active (66%) and healthy eating (58%) but only 36% recalled receiving food safety information. Of this 36%, the majority stated the information was useful (96%) and resulted in food safety practices (80%). Furthermore, 58% believed insufficient food safety information was available specifically for cancer patients/caregivers. The majority reported receptivity to information during chemotherapy treatment, with 76% reading everything provided, 94–95% searching for additional information.

**Significance:** Despite increased awareness of the importance of food safety during chemotherapy, perceived risk of foodborne illness during chemotherapy treatment was underestimated, particularly among patients. Information regarding food safety for cancer patients/caregivers was considered to be insufficient and sought after. Data will inform targeted food safety risk communication to reduce the risks associated with listeriosis among chemotherapy patients.

**P2-04 Time-temperature Study of UK Consumers’ Domestic Refrigerators**

**ELLEN EVANS** and Elizabeth Redmond
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**Introduction:** Consumer demand for refrigerated Ready-to-Eat (RTE) foods has increased in recent years; however, such foods are commonly associated with listeriosis due to the ability of *Listeria monocytogenes* to survive and grow at refrigeration temperatures. Consequently, effective temperature control of RTE foods by consumers in the domestic kitchen is critical for food safety as inadequate refrigeration practices are believed to increase the risk of foodborne illness. UK recommendations for domestic refrigeration are ≤ 5.0°C.

**Purpose:** This study aimed to assess actual domestic refrigerator temperature profiles and self-reported food storage practices of UK consumers.

**Methods:** Time-temperature profiles of refrigerators ($n = 43$) in domestic kitchens were determined using three Signatrol SL52T self-contained button data loggers (Range: −40°C to +85°C; accuracy: ± 0.5°C; frequency: every minute) over 136 h placed in a central storage area, a door storage area and outside of the refrigerator. Households ($n = 43$) documented self-reported refrigerator usage during profiling.

**Results:** Age of refrigerators ranged from 4 months up to 30 years, domestic refrigeration temperatures ranged from -1.7°C to 16.9°C, no statistical differences ($P > 0.05$) in mean operating temperatures were determined according to refrigerator age. Time-temperature profiles determined 40% of refrigerators (door/central locations) operated at > 5.0°C for whole profiling duration (136 h). Although 9% operated at recommended temperatures for 75% of profiling, no refrigerator operated at ≤ 5.0°C for whole duration. A positive correlation ($r = 0.29$, $P < 0.05$) existed between self-reported door opening frequency and daily temperature fluctuations. Fluctuations in temperature were determined when households reported putting food shopping in the refrigerators (ranging from -2.0°C to 7.1°C). No significant differences ($P > 0.05$) were determined between refrigerator temperatures and participant demographic.

**Significance:** Temperature profiles indicate that majority of the sample may store RTE foods at unsafe temperatures which may increase risk of foodborne illness. Findings highlight the need for improvement of domestic kitchen refrigeration practices; data may be used to inform development of targeted food safety strategies.

**P2-05 Microbiological Contamination of Older Adults’ Domestic Kitchens Associated with Self-reported Food Safety Practices**

**ELLEN EVANS,** Adrian Peters and Elizabeth Redmond
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**Introduction:** The domestic kitchen is associated with sporadic foodborne illness. Older adults (≥ 60 years) are more susceptible to acquiring foodborne illness due to weakened immune function, data suggests foodborne illness among older adults has increased. Consequently, food safety practices in the home are essential to reduce foodborne illness among older adults.
**Purpose:** The study aimed to determine domestic kitchen microbiological contamination and self-reported food safety practices of older adults that may increase foodborne illness risks in the home.

**Methods:** Food contact surfaces/equipment (n = 1292) in older adults (≥ 60 years) domestic kitchens (n = 100) were microbiologically analyzed to determine aerobic colony count (ACC), Enterobacteriaceae, Staphylococcus aureus and Listeria spp.; self-reported food safety practices were recorded using verbal-questioning. Statistical analysis was used to determine associations/differences between microbial contamination and self-reported practices.

**Results:** Cumulatively, microbial contamination was significantly greater (P < 0.001) at wet sites than dry; with highest contamination determined on in-use cleaning equipment (dish/washes/dishcloths/sponges) ACC < 9.28 log CFU, < 8.81 log CFU Enterobacteriaceae and < 7.03 log CFU S. aureus. Reported length of dish-brush usage significantly correlated (r = 0.349, P < 0.05) with Enterobacteriaceae contamination. Chopping boards were contaminated with ACC < 4.98 log CFU; ACCs were significant greater (P < 0.05) on boards reportedly ‘wiped with cloth,’ than boards ‘washed with detergent in-sink/dishwasher.’ ACCs on dishcloth/sponges were significantly correlated (r = 0.658, P < 0.05) with ACCs on chopping boards. Refrigerator food storage areas had ACC < 6.2 log CFU, ACCs correlated (r = 0.26, P < 0.05) with when the refrigerator was reportedly last cleaned.

**Significance:** The novel approach facilitated comparison between domestic food safety practices (reported method/frequency) with actual microbial contamination, which determined significant associations/differences. Findings suggest that older adults fail to implement adequate and regular food safety practices. This may increase the risk of foodborne illness resulting from pathogen cross-contamination in older adults’ domestic kitchens. Data from this study has determined a need for older adults to improve food hygiene practices in the domestic kitchen; such data can be used to develop targeted food safety education for older adults.

**P2-06 Food Safety Communication among Teachers in Home and Consumer Studies**

**MARIE LANGE**, Ingela Marklinder and Helen Goranzon

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**Introduction:** Home- and Consumer Studies in the Swedish Compulsory School is an opportunity to educate students about food safety. Based on previous studies in this ongoing project, teaching has been indicated to be influenced by the teachers’ working years and formal education. Issues related to one of the four “C’s” in food safety; Cleaning was routinely taught. However other risk areas related to the other “C’s; Cooking (e.g., handling raw minced meat), Chilling (e.g., cold food storage) and Cross-contamination (e.g., handling of dishcloths) are all areas that need further highlighting. Boys in Sweden are indicated to be at a higher risk regarding cross-contamination and hand washing actions.

**Purpose:** To gain a deeper understanding of what, how and why food safety issues related to the four “C’s” are a part of Home- and Consumer Studies teaching.

**Methods:** Qualitative interviews with focus on food safety issues are performed individually with 10 teachers in Home- and Consumer Studies. A mind-map with four different themes of interest was used. A convenience selection was used and all interviews were recorded and transcribed verbatim.

**Results:** Food safety issues related to Chilling, i.e., cooling, refrigerator temperature and cold food storage, often become a smaller part of teaching and then in form of theoretical education while issues related to Cleaning become routines. Food handling issues related to the food handling process which is very essential in teaching but also very complex as it is affected from different angles. Teaching is affected by frame factors and especially the lack of time is a common cause for teachers to skip food safety.

**Significance:** Preliminary results indicate that food safety teaching needs to be strengthened to include all four “C”s in the teaching and frame factors to have an impact on the food safety education.

**P2-07 Molecular Diversity and Characterization of Listeria monocytogenes Isolates from Australia**

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**Introduction:** Listeria monocytogenes causes the severe disease – listeriosis. Current legislation globally on the occurrence of foodborne L. monocytogenes does not differentiate isolates based on health risk; however, as our knowledge increases, it has become apparent that certain isolates may be more likely to cause human disease. This association has been linked to genotypic traits of an isolate.

**Purpose:** This study examined L. monocytogenes from Australia, to provide insights into epidemiology and potential clinical significance.

**Methods:** Whole genome sequencing and in-silico analysis of 71 isolates were performed to determine the ST distribution of the population, along with an analysis of virulence factors associated with severe invasive disease. PFGE analysis was used to further subdivide the population.

**Results:** A total of 18 STs were identified. ST3 (30%) and ST1 (17%), commonly associated with clinical infection, were the two most common STs identified. Isolates among these groups were identified from a variety of food chains, including meat, dairy and vegetable sources. ST204 (15%) was also frequently identified, which has rarely been described outside Australia. ST12 isolates were exclusively isolated from Western Australia; ST10 and ST155 were only identified in Victoria. Isolates among ST121 and ST202 included those with amber mutations in the inIA gene (4% of isolates), which is associated with attenuated virulence. All isolates had uninterrupted inIB and actA genes, both associated with intracellular pathogenesis. The LIP1-3 pathogenicity island was present in 20% of isolates. Analysis of genetic lineages identified isolates as lineage I (52%), lineage II (47%), and lineage III (1%).

**Significance:** This study provides insight into the Australian L. monocytogenes population structure, its potential virulence, and provides data useful for epidemiological surveillance.

**P2-08 Molecular and Serology Screening for Toxoplasma gondii in a Small Sicilian Island (Italy), Pantelleria as a Study Model for a Comprehensive Analysis on Food Habits and Food Safety**

**SERGIO MIGLIORE**, Maria La Giglia, Antonino Altavilla, Salvatore D’Avola, Maria Errera and Maria Vitale

Istituto Zooprofilattico Sperimentale di Sicilia, Palermo, Italy

**Introduction:** Foodborne infections caused by virus, bacteria and parasites such as Toxoplasma gondii are diffused worldwide. Preventive educational intervention for food safety can be more effective when the target population can be involved and all environmental, social and cultural aspects are known.
P2-09 Epidemiologic Investigation of Foodborne Outbreaks in Pharmacies: A Pilot Study
Carla Torre1, Roberto Brazão2, José Guerreiro1, Marta Gomes1, Suzete Costa1, Luísa Oliveira2 and SILVIA VIEGAS2
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Introduction: The analysis of foodborne outbreaks (FBO) investigation data provides knowledge on food vehicles and contributive factors of human infections allowing their risk management. However, FBO are commonly underreported and alternative sources of information may prove useful.

Purpose: To test the adequacy of pharmacies as an information source to identify foods involved in FBO, their confection type and acquisition place as well as the main symptoms and medicines taken.

Methods: Application of a FBO epidemiologic investigation inquiry to individuals with FBO suggestive symptoms that went to 249 pharmacies all over Portugal looking for treatment, between August 18th and November 15th 2014. Descriptive statistical analysis of the results was performed and absolute and relative frequencies measures of location and dispersion were calculated in the program SAS version 9.1.

Results: From 270 validated inquiries collected online until October 29, 72.9% of the individuals went to the pharmacy as the first health resort (average age 44.1 years), 56.3% individuals reported moderate symptoms, namely diarrhea and 58.0% of individuals took anti-diarrheal. The most frequent place of consumption of the suspect food was home (51.5 %), restaurants (22.9%) and parties (13.4%) and the food was meat (21.6%) and vegetables (18.3%), mostly cooked. About 67% of the individuals reported that an average of other 4.6 individuals has consumed the same food and 29.6% of them showed the same symptoms.

Significance: The findings are consistent with the Portuguese FBO investigation data reported to EFSA and therefore indicate that pharmacies can be a valuable source of information about FBO and also support consumer food safety education in order to prevent FBO.

P2-10 Characterization of Bacillus cereus Detected from Italian Dairy Products
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Introduction: Bacillus cereus is a Gram positive spore-forming foodborne pathogen commonly found in wide range of food; it is closely related (B. cereus complex) to B. thuringiensis, an insect pathogen, B. anthracis, the causative agent of anthrax, B. mycoides and B. weihenstephanensis. B. cereus is known as a causative agent of two different types of food poisoning, which are characterized by either emesis or diarrhea.

Purpose: This study investigated the presence of genes encoding for diarrheal-enterotoxin in the B. cereus strains isolated from Italian dairy products.

Methods: One hundred ninety-four samples of cheese were analyzed for B. cereus detection according to ISO 7932:2005. Isolates were confirmed as B. cereus and then analyzed by means two multiplex PCR assays targeting the presence of cytotoxin K (cytK), enterotoxin FM (entFM) and for the toxigenic complexes hemolysin BL (HBL) and non-hemolytic enterotoxin (NHE).

Results: Sixty-two out the 194 samples (32%) resulted positive to the microbiological analysis. Gene targets of more frequent detection were the entFM (100%) and NHE complex (95%). According to Ngamwongsatil et al. (2008), the strains analyzed in this study were clustered into four principal groups. These groups included strains positive for all targets (group I – 13/62), then strains positives for all targets except for, HBL (group II – 14/62) or cytK (group III – 1/62) and, finally, strains positive for NHE complex and entFM (group IV – 27/62). Moreover, other less relevant patterns were detected.

Significance: Our study highlights the significant level of B. cereus contamination in Italian cheeses. Given that most of the B. cereus strains analyzed were virtually pathogens and that the dairy products are generally Ready-to-Eat food, a further investigation on the risk assessment of these products is appropriate.

P2-11 PCR-based Detection of Coxiella burnetii in Cheeses Produced in Southern Italy
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Introduction: Coxiella burnetii (Cb) is the causative agent of Q fever. Although oral transmission of Cb by contaminated raw milk or dairy products is a possible route of human infection, there have been few studies investigating the presence of these bacteria in dairy products.
Recently, there has been an interest in examining alternative fungicides such as natamycin. A. carbonarius using real-time PCR. The results of this study highlights that the presence of Cb in cheeses made in southern Italy is significant, although lower than reported previously in Italian dairy products. Further studies will be needed to assess the risk for human being.

**Purpose:** Aim of this study was the assessment of Cb contamination in cheese produced in southern Italy from milk of cow, buffalo, and small ruminants.

**Methods:** Two tests based on real-time PCR assay targeting Cb IS1111 element were performed. First an assay based on the use of Taq-Man probe was performed to screen the samples. The positive samples were confirmed using both a SyBr Green test and the evaluation of the melting temperature of the amplicons. In addition, all cheese samples were also tested to determine the milk species utilized (Regulation EC 273/2008). The samples of cheese produced with a milk mix from different species were not included into the study.

**Results:** A total of 170 cheese samples were tested, and the obtained results showed an overall prevalence of Cb of 20.83%. A positivity rate of 36%, 28% and 7% was observed in cheese from cow, small ruminants and buffalo, respectively. The statistical analysis of the Cb prevalence in the samples from cattle and small ruminants compared with those from buffalo shows an OR of 8.4 and 4.9, respectively. Comparing the results between artisanal to industrial products, the former showed a higher risk of infection (OR = 4.2 - 11.22).

**Significance:** The results of this study highlights that the presence of Cb in cheeses made in southern Italy is significant, although lower than reported previously in Italian dairy products. Further studies will be needed to assess the risk for human being.

**P2-12 Influence of Atmospheric Pressure Plasma (APP) on Biofilm Produced by Listeria monocytogenes**

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**Introduction:** Listeria monocytogenes have been isolated from many different environments. It is introduced into food processing plants via animals or plants material where it can establish endemic populations. A high number of factors are involved in the ability of a strain to cause foodborne outbreaks, including differential ability to persist in a specific environment. Many bacteria are able to attach and colonize environmental surfaces by producing biofilm that allows microbial organisms to persist in the environments and resist desiccation, UV light, and treatment with antimicrobial and sanitizing agents. The need for new sanitization techniques have become an important aspect in the food environment and the development of antimicrobial measures that are not subject to evolving resistance, represent a new challenge in the food control. In this context, the cold atmospheric pressure plasma (APP) is a relatively new antimicrobial technique that has been recently adopted also for applications in the food industry.

**Purpose:** The purpose of this study was to measure the effectiveness of APP treatments against bacteria in biofilms surfaces, evaluating also the individual susceptibility of L. monocytogenes strains.

**Methods:** In the first part of the research, the ability of different L. monocytogenes strains to generate biofilms on the stainless steel surfaces was studied in standard condition after 6, 24, 48 and 150 h. In the second stage, in vitro tests have been conducted submitting stainless steel coupons with L. monocytogenes biofilm to the Plasma treatment.

**Results:** Attachment and/or biofilm formation showed a strain dependent response. A decrease of CFU/cm² was observed after the APP action. SEM observation was also applied before and after the APP treatments.

**Significance:** The results provide information on the behavior of attached L. monocytogenes cells to food-contact surfaces, which may contribute in the development of sanitation programs with the use of APP for effective pathogen removal.

**P2-13 Effect of Natamycin and Environmental Factors on Growth and OTA Production by Two A. carbonarius Isolates Using a Central Composite Design Experiment**

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**Introduction:** Aspergillus carbonarius has been reported as the major fungal species responsible for the presence of Ochratoxin A (OTA) in wine and grape derivatives. Several studies have examined the effectiveness of different fungicides. Recently, there has been an interest in examining alternative fungicides such as natamycin.

**Purpose:** The objectives of this study were focused on investigating the efficacy of natamycin on fungal growth and OTA content by two A. carbonarius isolates in concentrations until 1 ppm. A central composite design and response surface methodology were applied in a range of temperatures (16.4 - 33.4°C) and water activities (0.90 - 0.97 a.).

**Methods:** The efficacy of natamycin was studied using a central composite design (2³ factors) and a response surface methodology. Growth estimation was obtained by plotting the colony’s radius increase against time and fitting the data with linear regression. OTA production was assessed after 5, 10, 15 days of incubation.

**Results:** Natamycin showed a slight inhibition on fungal growth when assessed under the optimum conditions. However, its efficacy was always dependent on environmental conditions presenting a higher inhibition at sub-optimum for growth conditions. OTA production showed a slight inhibition at sub-optimum for the toxin conditions, whereas no significant effect was observed at optimum for OTA conditions. Interestingly, at the latter conditions OTA was stimulated in some cases.

**Significance:** Natamycin is a natural substance which may affect A. carbonarius growth and OTA production in a way that depends on environmental factors and time.

This work has been supported by the project 'Design and development of innovative tools for the detection of ochratoxigenic fungi in wine and table grapes – FungalPrognosis_242' co-financed by the European Union (European Social Fund – ESF) and Greek national funds through the Operational Program "Education and Lifelong Learning" of the National Strategic Reference Framework (NSRF) - Research Funding Program: ARISTEIA-I.

**P2-14 Promoting Safety Control of Ochratoxigenic Fungus Aspergillus carbonarius in Grapes by Studying Gene Expression through Different Ecophysiological Factors**

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**Introduction:** There is a great food safety concern regarding the presence of ochratoxin A (OTA) in food. Aspergillus carbonarius significance in OTA contamination in grapes and grape products is widely known.

**Purpose:** In this work, one toxigenic A. carbonarius isolate from Greek vineyards was assayed for its in vitro OTA production potential and was also subjected to relative gene expression study for the pks, nrps, veA and laeA genes using real-time PCR.
**P2-15** Distribution of *Anisakis* in Area FAO 37 in the South of Sicily and around Malta Island

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**Introduction:** The detection of parasites of the genus *Anisakis* in fish and the increasing consumption of raw, undercooked or improperly processed fish, require the correct knowledge about the distribution and the prevalence of this zoonotic pathogen that allows us to make appropriate health decisions.

**Purpose:** The aim of this work is to complete the knowledge regarding *Anisakis* species distribution in FAO sub-area 37.2.2 and GSA 15-16 of Mediterranean Sea in a project led by Italian Centro di Referenza Nazionale for Anisakiasi.

**Methods:** *Lepidopus caudatus* fish samples were collected from Mediterranean Sea in the south of Sicily (GSA 16) and around Malta Island (GSA 15). *Anisakis* larvae were identified using morphological and genetic tools. The genes targets were a ribosomal genomic regions (ITS1, 5.8 SrRNA and ITS2) and cox2 gene. After amplification, PCR products were sequenced and the sequences were analyzed by phylogenetic analysis.

**Results:** *Anisakis pegreffii* is the only species present in *Lepidopus caudatus* fish samples. Phylogenetic analysis showed the same nucleotide sequence in *Anisakis pegreffii* from both GSA 15 and GSA 16 areas studied.

**Significance:** The present study has allowed us to extend knowledge on the distribution and the prevalence of zoonotic pathogens bellowing to the genus *Anisakis* in *Lepidopus caudatus* fish samples in the FAO sub-area 37.2.2 in the south of Sicily (GSA 16) and around Malta Island (GSA 15). Further studies are advantageous to expand knowledge about the distribution of larvae in other fish species and in other sub areas of Mediterranean Sea.

**P2-16** Inactivation of Bacteria in Spices and Herbs

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**Introduction:** Spices and herbs are natural dried components, or mixtures thereof, used in foods for flavoring, seasoning and imparting aroma. Currently in the EU the most consumed spices are pepper, paprika and pimento (allspice), whereas the most consumed herbs are thyme and oregano. Despite their low water activity, which inhibits microbiological growth, spices and dried herbs can be naturally contaminated with large numbers of microorganisms, among them several pathogenic species and toxigenic molds, for example, *Salmonella* spp., *Escherichia coli*, *Clostridium perfringens*, *Bacillus cereus* and aflatoxigenic *Aspergillus* spp. Spices and herbs are therefore treated for reduction of microbial load.

**Purpose:** The purpose of this study is to identify factors influencing inactivation of pathogenic bacteria by irradiation.

**Methods:** Meta-analysis on the published data available on irradiation was performed and trends were tested for their significance with statistical tests.

**Results:** Inactivation data collected showed high variability in reduction kinetics. Irradiation treatment (gamma or electron beam) and product physical state (whole, ground or powdered) were not found to significantly influence reduction. Gram positive bacteria were significantly more resistant than Gram negative; subsequently spores were significantly more resistant than vegetative cells.

**Significance:** Irradiation may not be able to significantly reduce spores and gram positive bacteria in spices and dried herbs.

**P2-17** Thermodynamic Evaluation of Sterilization Cycle of Aseptic Tank

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**Introduction:** Sterilization of aseptic tanks is critical to assure microbiological integrity of aseptic lines. The current protocols are limited to quantify the delivery of basic parameters – time and temperature. This work evaluated an aseptic tank sterilization cycle and by application of thermodynamic principles provides of the phenomena, resulting in new guidelines and specifications for equipment and process design and validation, with cost reduction and food safety assurance increment.

**Purpose:** Review current validation protocols applied in aseptic tanks

**Methods:** Twelve temperature sensors were installed inside an aseptic tank. The tank internal surface temperature and pressure were collected in four sterilization cycles. By the application of the pressure on Antoine equation, it was obtained...
the correspondent temperature of saturated steam. The amount of air was calculated by partial air pressure observed in Clausius-Clapeyron equation, followed by the molar mass estmative. With the deviation of saturated state, change on lethality was estimated by thermobacteriology models.

**Results:** It was observed deviation between the temperature calculated by Antoine and sensor temperatures that indicates presence of residual air from incomplete venting. The analysis showed venting cycles were insufficient, with remaining air in the order of 13% - 23% of total tank volume. Consequently, the subsequent sterilization process was not conducted at steam saturation condition. This different condition changes the kinetics for thermal destruction of microorganisms from a wet state to partial dry state – which thermal resistance of spores is higher, resulting potential reduction of destruction. This finding raises a question regarding the true efficacy of the sterilization process and validation protocols currently applied.

**Significance:** The apparent success of current sterilization processes may be explained by the usage of excessive temperature and time. New operational and validation criteria which assure saturated steam conditions during sterilization could allow reducing sterilization time in the order of 80% at the same temperature.

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**P2-18 Setting Time-temperature Criteria (TTC) for Microbiological Food Safety Using a Predictive Model for Growth and QMRA Simulation**

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**Introduction:** This study provides a method of setting time-temperature criteria (TTC) by using a predictive model for growth and quantitative microbial risk assessment (QMRA) simulation as a way to ensure the safety of refrigerated food.

**Purpose:** For this purpose, on the basis of the preceding study, a new modified Gompertz model chose as the most suitable predictive model for growth of *Listeria monocytogenes* in refrigerated cooked sausage after the packaging had been opened.

**Methods:** We developed a simple QMRA simulation model that considered *L. monocytogenes* contamination levels of cooked sausages at the distribution stage and environmental factors (storage temperature and time) related to refrigerated storage in Korean households, and selected the new modified Gompertz model; using these simulation results, ultimately a time-temperature plot and a TTC model as second-order polynomial model were developed that could estimate safe consumption times according to refrigerated storage temperatures in households.

**Results:** Using this model, as an example of TTC, it was shown that it is safe to consume uncooked cooked sausages which have been in a refrigerated state within 7 days after the packaging has been opened if *L. monocytogenes* consumption levels are within 0 log CFU/g (1 CFU/g) at 3.53°C, which is the average refrigerator temperature in Korean households.

**Significance:** This method can be helpful in setting a food safety objective (FSO), which is the allowed intake at the consumption stage.

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**P2-19 Survival of Cronobacter Strains in Powdered Infant Formula at Ambient Temperature**

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**Introduction:** *Cronobacter* is an emerging foodborne pathogen, identified as the causative agent of serious neonatal infections. In most cases, the source of infection was powdered infant food formula. *Cronobacter’s* high resistance to desiccation explains its presence and survival in powdered infant food formula factory products and environments.

**Purpose:** We assessed survival of 7 *Cronobacter* strains belonging to 4 different species (*C. sakazakii, C. malonaticus, C. mytjensii, C. turicensis*) in powdered infant food formula stored at ambient temperature for 3 months.

**Methods:** Evolution of bacterial population and of stress percentage were monitored by direct plating on non-selective and selective agar. Population cell densities were described as a function of time by biphasic with shoulder model parameterized using GinaFiT Add-in for Excel.

**Results:** Stress percentage remained quite moderate and constant for all strains over time though ability to grow decreases, indicating that different physiological phenomena are involved. Considering ΔΔ (time for 2 log reduction) and 4Δ (time for 4 log reduction) there seems to be a moderate effect of the species on resistance to desiccation, but no effect of strains origin (clinical/non clinical). Data revealed high persistence of culturable cells during extended periods.

**Significance:** This information is important to contribute to quantitative risk assessment.

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**P2-20 Analysis of Shipping Methods, Packaging Materials, and Product Temperatures of Perishable Meat and Seafood Products Ordered from Online Vendors in the United States**

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**Introduction:** Perishable food products such as meat, fish/seafood and poultry marketed directly to consumers by online vendors are delivered using the same methods as non-perishable products, making them vulnerable to temperature abuse.

**Purpose:** This study reports on shipping methods, packaging, coolant materials, and product temperatures of fresh and frozen meat, fish, and seafood products ordered online and shipped via common carriers such as FedEx or UPS.

**Methods:** Samples of 679 raw meat, poultry, fish, and shellfish products were purchased online from 160 US purveyors between January and October 2013, and shipped using common carriers.

**Results:** Including nine replacements, 169 shipments were received; 684 food items were tested. Of these, 375 were meat, 47 poultry, 231 seafood, and 30 “others.” Shipping was ‘overnight’ (32.4%), ‘2-day’ (29.4%), and ‘standard/ground’ (27.1%). Mean transit time was 32.35 h (SD = 14.83 h). Few (4.1%) had external damage, although 3.64% had leakage on the inside. Only 36.7% had food safety information on the outside of the box; 7.7% had no labels indicating their perishable contents. Food safety information was only found in 25% of the packages. Most of the shipments (92.9%)
including a polystyrene foam box; 76.9% also used an exterior cardboard box. The majority (50.6%) contained gel packs, 42.4% had dry ice, 4.1% included both; 2.4% contained conventional ice, and 4.1% of the packages contained no refrigerant. Upon opening the packages, the surface temperatures on the top of the products ranged from −23.5°F to 72°F (M = 36.8°F, SD = 13.3°F). The temperatures on the bottom ranged from −22.7°F to 75°F (M = 36.8°F, SD = 12.9°F). Nearly half (46.6%) had a surface temperature above 40°F.

Significance: Many of the perishable meat, poultry, and seafood products arrived outside of the temperature safety zone; thus, combined with a lack of food safety information accompanying the packages, places consumers at increased risk for foodborne illness.

P2-21 Consumption Habits and Storage Attitudes towards RTE Cooked Meat Products – Useful Information for Reliable Risk Assessments
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Introduction: Consumer behavior can either increase or substantially reduce the risk of microbial foodborne illness, thus this information is essential to evaluate the associated risks.

Purpose: To examine consumption habits and domestic storage practices of RTE cooked meat products.

Methods: A survey consisting of 33 questions was distributed to consumers from Catalonia (Spain).

Results: From the 396 respondents, over three quarters regularly buy sliced and packed products. A weekly-based frequency of consumption was recorded in 72% and 31% of the participants for lean and fatty RTE cooked meat products, respectively. For both types of products, serving size generally ranged from 20 – 80 g. Slightly higher frequency of consumption for lean (84%) and fatty (35%) products but smaller serving size (20 – 40 g) was recorded for children under five. Most of the respondents thought that the recommended storage temperature for RTE cooked meat products is ≤ 4°C (53%) or 4 – 6°C (34%). However, 4% indicated ≥ 9°C as recommended storage temperatures, 15% did not know/failed to respond. In domestic refrigerators, the thermostat was usually set to the midpoint when regulation was manual. In digital devices, temperature was mostly set at ≤ 4°C (49%) or 4 - 6°C (44%). However, a high proportion of the participants (92%) had never checked with an external thermometer if their fridge was operating at the set temperature. In general, the maximum storage period of the purchased product was 3 weeks for unopened packs and less than 1 week after the product was opened. Three quarters of the respondents declared to check “use by” date at the moment of purchase and 68% at the moment of consumption. In 45% of the cases respondents declared to consume expired products if no evidence of spoilage is perceived.

Significance: Quantitative data about consumption habits as well as domestic storage practices will contribute to perform more reliable risk assessments.

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P2-22 Determining the Source of Listeria monocytogenes in the Scottish Smoked Salmon Industry and the Risk of Contamination of the Final Product
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Introduction: Listeria monocytogenes is a foodborne pathogen causing listeriosis mainly in vulnerable groups (e.g., pregnant and elderly people). The disease is rare but severe, with mortality of up to 30%. Smoked salmon is a potential source of contamination but it is still being debated whether raw materials, environment or the final product is the main source of infection.

Purpose: Our aim is to evaluate the source of L. monocytogenes and the risk of contamination of salmon by modelling the prevalence of the pathogen along the different stages of the processing chain. Considering every step from farm to fork will allow possible interdependencies between different stages to be identified. The risk assessment will be complemented by DNA sequencing analysis of samples isolated from fish farms, smoking plants and clinical cases. Comparison of these data will help to understand the sources of Listeria in the salmon industry and the risk of contamination of the final product.

Methods: We have developed a preliminary model covering the whole process from farm to final product.

Results: Based on published literature and internal data, critical stages were identified, where the prevalence of Listeria significantly increased (e.g., early processing) or respectively decreased (washing). Statistical analysis suggests that the prevalence of Listeria in early processing of salmon (from harvest to processing plant) does not significantly depend on environmental factors (e.g., season, region and farm of origin). We noted however, significant difference in the prevalence of Listeria in early processing in different years.

Significance: Existing research on contamination with Listeria typically focuses on particular stages of the production chain, mainly the final product. In contrast, we apply a holistic interdisciplinary approach covering the whole processing chain which will lead to a reliable assessment of the most likely sources of contamination of salmon and ultimately reduce the risk of Listeriosis in consumers.

P2-23 A Modular Open Source Software Integration Platform Applicable for Food Safety Analysis and Modelling
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Introduction: The analysis of food quality and safety data as well as knowledge generation using mathematical models is a field of increasing relevance to food safety professionals and public authorities. Today, large quantities of experimental, quality control and product related data are available in comprehensive data collections. Moreover computational and mathematical foundations for data analysis and model generation are available.
P2-24 Quality Control of Dairy Products: Contribution to the Establishment of the HACCP System in an Ice Cream Lollypop Production Line in an Algerian Agri-food Enterprise
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Introduction: In order to guarantee permanently the supply of safe products which meet, on one hand, new regulatory requirements, and on the other hand, the requirements of the consumer in terms of quality and safety of products, all agri-food enterprises nationwide are under an obligation to apply the HACCP system.

Purpose: This work, done in an ice cream production enterprise, is a contribution to the establishment of the of HACCP system in an ice lollypop production line in an agri-food enterprise.

Methods: It relates primarily to the hazard analysis and the establishment of a control system, established after an upgrade of the existing through a hygiene audit of the enterprise, according to the requirements enacted by the Good Hygiene Practices and the Good Manufacturing Practices. Our study was conducted in two phases: The first one involves the realization of state of play of the Agri-food enterprise by a valuation audit. The second one includes four phases, the preliminary stages; the hazards analysis; the determination of the CCP and the implementation of HACCP plan.

Results: The study allowed identification of five critical control points (CCP), the writing of procedures to ensure smooth functioning of the HACCP system, as well as the establishment of a number of fact sheets and forms that facilitate the work and make it more organized.

Significance: At the national level the Executive Decree No. 10-90 of 10 March 2010 reinforces the obligation of results already present in the regulations and defined the "HACCP" system as hazards prevention tool, it becomes mandatory for all the Agri-food enterprises for the acquisition of sanitary approval.

P2-25 Heat Survival of Clostridium difficile Spores in Ground Meat during Cooking Process
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Introduction: Clostridium difficile is a spore-forming pathogen considered as a major cause of enteric disease in humans, with fecal-oral route as the primary mode of transmission. However, recent studies have reported the occurrence of C. difficile in ground meats at retail stores, indicating that foods could be an additional source of infection in the community.

Purpose: The objective of this study was to determine the resistance of C. difficile spores in contaminated ground meat during cooking process.

Methods: Prior to testing, to obtain spores and to enhance heterogeneity, spores of two different strains were produced in two nutritious broths. C. difficile spores were experimentally inoculated in 45 g of ground meat (beef and pork) in order to obtain a final contamination of 4,500 CFU g⁻¹. Six heating temperatures (70, 75, 80, 85, 90 and 95°C) were chosen. Samples were heating in a water bath with an integrated program for time-temperature. One sample without inoculum was used as control with a temperature probe placed inside. Once the desired temperature was reached in the core of the sample, the heat treatment was prolonged for 10 min. Subsequently, all the samples were placed on the chilling room (4°C) before analysis. These experiments were conducted in duplicate with a spore enumeration in triplicate.

Results: Heating contaminated ground meat at 70, 75 and 80°C for 10 min was not effective for C. difficile spores inhibition. However, 10 min of heat shock at 80°C was the only temperature that significantly reduced the number of countable colonies. Heat treatment at 85°C (or more) inhibits the germination of both of the strains tested.

Significance: Ensure that ground meat, like burgers or sausages, is heated to more than 85°C would be an important measure to reduce the risk of C. difficile food transmission.

P2-26 Use of Relative Humidity Fluctuations to Kill Foodborne Pathogen Listeria monocytogenes
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Introduction: Environmental hydration fluctuations influence microorganism viability and activity. The ambient air relative humidity (RH) is a potentially effective parameter to control development and persistence of pathogenic microorganisms. However, efficiency of this parameter remains not well characterized compared to other environmental factors such as pH, temperature, etc.
Purpose: The objective of this study is to characterize the impact of RH variations on L. monocytogenes survival. L. monocytogenes is a pathogenic bacterium responsible for listeriosis and has the ability to survive and to adapt to hostile environments (low temperature, high salinity, acidity). This resistance allows L. monocytogenes to persist on food processing plant surfaces.

Methods: To assess conditions leading the largest cell viability loss, four strains of L. monocytogenes (different serotypes and different origins) were exposed to different drying and wetting conditions (several RH levels, different drying and rehydration kinetics).

Results: Our results show that an intermediate RH level causes the greatest L. monocytogenes destruction. Moreover, we show that destruction depends on drying and rehydration kinetics. Under optimal conditions, we measure a loss of cultivability between 2 and 3 log according to strains. Application of two dehydration and rehydration cycles led to an additional loss of cultivability compared to application of one dehydration and rehydration cycle. Our results show also that rehydration step after drying is responsible for the majority loss of viability.

Significance: In conclusion, controlled variation of air RH is an effective tool to control pathogenic bacteria such as L. monocytogenes. This study reveals that ambient RH could be optimized, like temperature, to control L. monocytogenes survival and can offer a real potential for control foodborne pathogens in food processing environments and so insure food safety.

P2-27 Addition of Acetic Acid to Limit Growth of Pseudomonas aeruginosa in MPV Washing Bath
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Introduction: The processing environment and decontamination treatments used in minimally-processed vegetables (MPV) are limited. Pseudomonas species are common and persistent contaminants reported by industrial surface samplings which growth is supported in MPV washing bath.

Purpose: This study aims at quantifying the impact of acetic acid on P. aeruginosa planktonic cells growth to determine growth/no growth boundaries.

Methods: P. aeruginosa ATCC15442 cardinal values, i.e., minimal, optimal and maximal values of temperature and pH enabling bacterial growth were determined in broth for 8 levels of temperature (12 – 42°C) and pH (4.8 – 9) using Bioscreen turbidimetric growth monitoring according to already reported method. Similarly, growth rate in the presence of acetic acid was determined at 39°C at a pH higher than pKa (4.76 at 25°C) to favor the presence of active dissociated form. For a robust acetic acid minimal inhibitory concentration (MIC) determination, experiments were performed at 2 pH values, i.e., 11 concentrations at pH 5.2 (0 – 14 mM) and 11 concentration at pH 5.5 (0 – 24 g/l mM). Growth kinetics were fitted using Rosso logistic model while acetic acid MIC was determined using Le Marc model. The secondary model describing the influence of environmental factors (temperature, pH, acetic acid concentration) on bacterial growth was based on the gamma concept and its multiplicative effect.

Results: The acetic acid MIC was determined together with cardinal values. A MatLab-based app was developed to quantify the impact of pH, temperature and acetic acid concentration on Pseudomonas aeruginosa growth abilities.

Significance: Within the frame of SUSCLEAN European project on sustainable cleaning and disinfection in fresh-cut food industries, a MatLab-based app was developed to quantify and quickly identify the combination of conditions ensuring no growth of P. aeruginosa to enhance industrial outreach and further in silico test industrial relevant scenario in a few seconds.

P2-28 Population and Resistance Patterns of Salmonella Typhimurium and Staphylococcus aureus Biofilms Exposed to Sublethal Chemical Disinfection under Mono- and Dual-Species Multi-Strain Conditions
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Introduction: In the food industry and following inadequate sanitation, the remaining microflora may contain pathogenic species forming biofilms on the processing equipment. Cellular interactions within such communities may influence both cell physiology and population dynamics.

Purpose: To evaluate the possible influence of bacterial interactions encountered in mono- and dual-species multi-strain biofilms of Salmonella Typhimurium (ST) and Staphylococcus aureus (SA) on: (i) the ability of strains to develop biofilm, and (ii) their subsequent resistance to sublethal chemical disinfection.

Methods: Three strains per species were left to develop biofilms on stainless steel (SS) coupons, under either mono- or dual-species conditions. Biofilms were then exposed for 6 min to benzalkonium chloride (BC, 50 ppm), peracetic acid (PA, 10 ppm) or sodium hypochlorite (SH, 10 ppm). The dominance of each strain in the sessile communities was monitored using PFGE.

Results: Dual-species conditions led to a reduction in the number of SA sessile cells (1.1 log CFU/cm²), compared to mono-species ones, while ST sessile population remained rather unaffected. Regarding disinfection resistance and under both conditions, BC was found to be the most effective disinfectant, while SH was the least effective in all cases studied. The presence of ST strongly decreased the resistance of SA biofilm cells to PA. PFGE analysis interestingly revealed that the different strains here employed behaved differently with regard to both biofilm formation and antimicrobial resistance.

Significance: Such research expands our knowledge on the physiology of multi-species pathogenic bacterial biofilms and may facilitate the development of efficient control methods to be used in the food industry.

Acknowledgment: This work was funded by the action THALIS: "Biological Investigation of the Forces that Influence the Life of pathogens having as Mission to Survive in various Lifestyles; BIOFILMS," under the Operational Programme (OP) "Education and Lifelong Learning (EdLL)" which is co-financed by the European Social Fund (ESF) and National Resources.
P2-29 Evaluation of Time Dependency of Surface ATP Test Devices at Different Environmental Temperatures
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Introduction: The hygiene of food contact surfaces is critical to ensure the safety and quality of foods. The use of surface ATP hygiene monitoring systems is widespread and provides food business operators with rapid, measurable and cost-effective test devices. This enables them to take corrective actions in a timely manner and improve surface hygiene. Therefore, it is essential that the ATP hygiene monitoring system provides accurate and reliable results.

Purpose: To evaluate the time dependency, following device activation, of seven brands of surface ATP test devices and luminometers at different environmental temperatures.

Methods: The performance of the luminometers and ATP test devices was evaluated at 5°C, 10°C, 20°C and 35°C using an environmental chamber. All devices were tested by pipetting 10 µl of a 4 x 10^-8 M ATP solution onto the mid-section of the swab/sponge bud of each device. Testing was completed following the instructions provided by the device manufacturer. The time dependency of test devices was assessed by reading the result in the luminometer immediately after activation and at 20-s intervals over two min.

Results: Seventy results per manufacturer, at each temperature, were obtained to evaluate the decay rate and percentage change in signal over two min from activation. The results suggest that only one of the seven devices retained signal strength within 10% over two min at all temperatures. The signal of two of the devices reduced by more than 40% when read at one min following activation for all temperatures.

Significance: The selection of the appropriate surface ATP device for its intended environmental temperature and the time taken to read the result in the luminometer following activation of the device is critical to ensure accuracy of the results. Effective and accurate surface assessment can be used for optimization of sanitation procedures and continual improvement.

P2-30 Nitric Oxide (NO) Donors to Disperse Biofilms of Industrial Significance Formed by Human and Plant Pathogens
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Introduction: Biofilms formed on industrial surfaces in post-harvest production facilities can be recalcitrant reservoirs of pathogens and new biofilm dispersants are needed. Current research has been instrumental in identifying nitric oxide (NO) donors and hydrogels as biofilm dispersants.

Purpose: To test the effectiveness of the NO-donors molsidomine, MAHMA-NONOate and the association with the hydrogel cellulose-nanocrystals (CNC) on biofilm formed by the human pathogen Salmonella enterica on materials of industrial significance.

Methods: NO donors (molsidomine and MAHMA NONOate) were tested on 24-h old and well-structured (1 week old) Salmonella biofilms at room temperature and 4°C on polypropylene and polystyrene. Biofilm dispersion was measured by staining the biofilm with 1% crystal violet, and the absorbed crystal violet was dissolved in 33% acetic acid and measured with a spectrophotometer. NO donors were also tested in association with CNC.

Results: Our findings show that these compounds are able to disperse biofilms. In particular molsidomine was able to disperse Salmonella biofilm (24-h old) at concentrations as low as 10 pmol l^-1 within 6 h of contact time. Molsidomine was also able to disperse more structured 1-week old biofilms formed by both the 14028 Salmonella strain and a cocktail of six Salmonella outbreak strains. The association of CNC with the nitric oxide donors MAHMA NONOate and molsidomine has shown a synergistic effect in dispersing well-established biofilms: After 2 h of exposure, moderate but significant dispersion was measured. After 6 hours of exposure, the number of cells switching from the biofilm to the planktonic state was up to 0.6 log higher when compared with the not-treated.

Significance: The tested molecules are active at very low concentrations and appear to be promising as Salmonella biofilm dispersant. This, in turn, can implement new sustainable cleaning strategies by expanding the toolkit of pro-active practices for GAPs and HACCP protocols.

P2-31 The Effects of Different Hygiene Procedures in Reducing Bacterial Contamination in a Model Domestic Kitchen
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Introduction: A significant number of foodborne infections are acquired at home, and improvement of consumer knowledge and practice could potentially both reduce the number of sporadic cases and the number of persons affected during outbreaks. A main source of bacterial pathogens at home is raw food ingredients or infected persons preparing food.

Purpose: Relatively few studies have been performed to compare the hygiene effectiveness of cleaning under simulated use conditions. Although standard tests are used on disinfectants, there is insufficient data on their effectiveness in preventing cross-contamination via hands and surfaces, relative to that of traditional cleaning methods.

Methods: We report results from two surveys on Norwegian consumers' cleaning procedures. The hygiene efficacy of commonly used cleaning methods together with new technologies such as sprays, single use wipes, and chlorine-based disinfectants were compared using a model which involved cutting boards, tap handles and mobile phones contaminated by contact with ground beef inoculated with E. coli and S. aureus.

Results: The survey showed that many consumers use traditional cleaning methods for cleaning cutting boards and tap handles, and the most commonly used methods produced a mean log reduction (LR) in contamination of 1.5 – 2.5. The results showed that even small changes in the present practice may increase the efficacy, such as drying (mean LR 3.2 – 4.5), rinsing with higher temperatures or including a disinfection step. Cleaning of mobile phones was common and was improved by including humidity (1.5 – 1.9 mean LR).

Significance: In public and domestic situations, there is pressure to deliver hygiene which involves prudent usage of detergents, water, energy and antimicrobial agents. This study demonstrates that data from models which simulate use conditions are required to better understand how different practices involving hygienic cleaning can work synergistically to ensure that contamination from foodborne pathogens is reduced to acceptable levels to prevent infection transmission.
**P3-01** Production of Antilisterial Bacteriocin by *Lactobacillus plantarum* under Different Stress Conditions  
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**Introduction:** Nowadays, bacteriocin producer lactic acid bacteria cultures are commonly used as bio-preservatives in fermented foods and numerous scientific studies examine their bacteriocins against *Listeria* spp. *Lactobacillus plantarum* is commonly used in starter cultures for fermented meats.

**Purpose:** The aim of this study was to investigate how *Lactobacillus plantarum* ESB 202 (isolated from fermented meat) produce bacteriocins under different stress conditions: pH, NaCl and temperature.

**Methods:** The modified (pH 3.5 and 8.5, NaCl 7.5%) MRS broth was inoculated with *Lb. plantarum* and incubated at 30°C (except for stress conditions of temperature: 10°C, 42°C). Changes in pH and optical density were recorded in every hours for 48 h. Bacteriocin activity in the cell-free supernatant was recorded in every 3 h for 48 h. *Listeria monocytogenes* serogroup IIb (from cheese), *L. monocytogenes* serogroup IVb (from cheese), *L. monocytogenes* serogroup IIb (from hamburger) and *L. innocua* NCTC 11288 were used as target strains.

**Results:** *Lactobacillus plantarum* could not grow well under osmotic stress but it was able to produce low amount of bacteriocin. At pH 8.5, the alkaline adaptation was clearly observed and it took ~ 20 hours. At pH 3.5 and high temperature, *Lb. plantarum* was able to grow and produced bacteriocin. At low temperature LAB could grow but was not able to produce bacteriocin.

**Significance:** It was demonstrated that under stress conditions (except low temperature), *Lb. plantarum* could produce antilisterial bacteriocins. *L. monocytogenes* IIb was the most sensitive to the tested bacteriocins, while *L. innocua* showed to be more resistant.

**Acknowledgment:** This work was supported by National Funds from FCT – Fundação para a Ciência e a Tecnologia through projects PEst-OE/EEQB/LA0016/2013 and SFRH/BPD/47826/2008 Pos-doc grant to Helena Albano. The work of Tekla Engelhardt was supported by the Campus Hungary program in the frame of TAMOP4.2.4.B/1-11/1 and TAMOP4.2.4.B/2/11-1 project.

**P3-02** Antimicrobial Activity of the Extracts of Desert Truffle *Tirmania pinoyi* against Gram Positive and Gram Negative Bacteria  
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**Introduction:** *Listeria monocytogenes* and *Staphylococcus aureus* are of big concern in the food industry for human infections. *L. monocytogenes*, in particular, with its capability to multiply at refrigerator temperature and to survive at freezer temperatures is responsible for an increasing number of severe listeriosis. It is commonly isolated in the filling or packaging equipment of the dairy industry. *S. aureus* strains producing enterotoxins are responsible for several food poisoning cases worldwide.

**Purpose:** To discover new strategies to tackle pathogenic bacteria, we focused on a species of desert truffle *Tirmania pinoyi* as source of new anti-infective agents. Some studies demonstrated that organic extracts of truffles of genus *Tirmania* and *Terfezia* possess antimicrobial activity with broad-spectrum effects against Gram positive, Gram negative, aerobic and anaerobic bacteria as well as *Saccharomyces*.

**Methods:** In this study, acid-soluble protein extraction (aqueous extracts) of *Tirmania pinoyi* were obtained and tested for *in vitro* antimicrobial activity against planktonic forms of some reference strains of human and animal pathogens. The two Gram positive reference strains *Staphylococcus aureus* ATCC 29213 and *Listeria monocytogenes* ATCC 7644 and the two Gram negative *Pseudomonas aeruginosa* ATCC 15442 and *Escherichia coli* ATCC 10536 were used to evaluate the antimicrobial activity.

**Results:** The acid-soluble protein extract of *Tirmania pinoyi* had minimum inhibitory concentrations ranging between 57 and 114 µg/ml against all tested pathogens. Further study will be performed to test the extracts against the same bacteria strains organized in biofilms.

**Significance:** Several infections can be transmitted from animals to humans through contaminated meat or dairy products. New antimicrobial molecules against pathogenic bacteria such as *S. aureus* and *L. monocytogenes* can be very important for an effective control of the bacterial infections not only in humans but also in livestock farms for a major biosafety of food of animal origin.

**P3-03** Assessing Red Cabbage Seed Extract as a Potential “Clean-label” Antifungal Solution in Food  
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**Introduction:** There is a current demand from the consumer for healthier food products, particularly for product free of/ containing less chemical preservatives. Manufacturers are therefore looking for new “clean-label” preservative solutions.

**Purpose:** The objective of this study was to quantify the effect of a red cabbage seed extract on the growth of three spoilage mold species to determine if it can be a “clean-label” alternative to chemical preservatives.
**Methods:** The effect of the plant extract have been tested on the growth of Aspergillus niger, Penicillium corylophilum and Eurotium repens, three molds isolated from spoiled bakery products. Lag time before mycelium growth and mycelium radial growth rate have been extracted from mold kinetics, collected at different concentrations (range from 2.5 to 20 mg g⁻¹) of plant extract on agar-based medium under stable conditions (25°C, aᵣ 0.99, pH 5.2). A secondary mathematical model, based on the Minimum Inhibitory Concentration (MIC), has been developed for the lag time. The expected gain provided by addition of 1% (weight/weight) of red cabbage seed extract was calculated as the ratio between the time to visible growth with and without extract (visible diameter = 3 mm).

**Results:** The radial growth rates were constant whatever the plant extract concentration tested. The Minimum Inhibitory Concentrations of the extract for Aspergillus niger, Penicillium corylophilum and Eurotium repens lag time have been estimated: 24.3, 12.0, and 13.2 mg g⁻¹, respectively. The gain obtained with addition of 1% extract was estimated in a range from 5.2 (A. niger) to 15.2 (P. corylophilum). These results must be confirmed in real food matrix.

**Significance:** These findings will help in replacing chemical preservatives by natural solutions in food product. In combination with other inhibitory factors (e.g., aᵣ), plant extract could enable to extend the mold-free shelf life by 2 weeks. This work has been performed through the ACIPCEV project.

**P3-04** Ozone Use in Industrial Vegetable Washing: A Critical Review of In-factory Trials and a Shelf-life Prediction Study

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**Introduction:** Ozone (O₃) is efficient in reducing pathogens. Its application to produce at the post-harvest stage could be efficient in inactivating bacteria and viruses and can cause destruction of pesticides and chemical residues. On the other hand the use of ozone has disadvantages such as instability and reactivity with organic materials, thus the effective elimination of microorganisms may require high concentrations which may cause sensory flaws in fresh produce or corrosion damage in plants.

**Purpose:** The aims of this study were to investigate the effect of different O₃ washing treatments (at 0.5, 1.5 and 2.5 ppm for 10 min) on i) vegetables decontamination, ii) water decontamination iii) shelf life.

**Methods:** The salad (Lactuca sativa) used for the challenge test was contaminated with Listeria innocua and subjected separately to washing for 10 min with different O₃ concentrations. Each min the samples were collected to verify the bacterial inactivation. After 3, 5 and 10 min of each treatment others samples were packed in modified atmosphere and stored at 12°C for 10 days. The growth curves of L. innocua were fitted using DFMFit web edition, in order to calculate the growth rates. The effect of the same treatments on the washing water contaminated with L. innocua and feline calicivirus (FCV) was also evaluated.

**Results:** Only during the salad washing with 2.5 ppm of O₃ a significant reduction of L. innocua was observed (from 5.79 ± 0.01 to 5.12 ± 0.02 log CFU/g) after 1 minute. No correlation was found between the growth rates calculated during the storage of salad after different treatment. In water, log inactivations (range from 2.22 to 6.56) of L. innocua and FCV were observed after 1 minute.

**Significance:** O₃ treatments could be a viable decontamination method for the food industry.

**P3-05** The Development of a Small-sized Electrolytic Assembly to Produce High Purity Aqueous Chlorine Dioxide for Fresh-cut Industries

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**Introduction:** Chlorine dioxide (ClO₂) is a strong oxidant which is widely applied to bactericidal disinfection in agriculture and livestock production. Applications also go to environmental wastewater treatment. The use of high purity aqueous chlorine dioxide for bactericidal purposes may reduce or eliminate the total trihalomethanes (THMs) content of the treated material. The THMs is mainly generated using residues like hypochlorite or chlorite with organic compounds. With optimal operation conditions, it was shown that 2 l of ClO₂, with 99.8% purity and a concentration 1700 ppm can be produced in 30 min with 40A and 110V.

**Purpose:** A small-sized electrolytic assembly was developed for installation at small scale factories for the washing and disinfection of fresh-cut fruits and vegetables.

**Methods:** The main parts of the assembly consist of a reaction tube (0.8 m in length), a vacuum pump, reverse osmosis elements and two diluted aqueous ClO₂ storage tubes (0.8 m in length). Aqueous ClO₂ is applied to cucumbers, red cabbage, baby corn, red and green bell peppers, followed by a microbial analysis.

**Results:** Results indicated that 50 ppm of chlorine dioxide can reduce the total plate count from 1.2 to 4.0 log CFU/g on cucumbers, baby corn, red cabbage and bell peppers, and from 1.2 to 4.3 log CFU/g for yeast and mold count. E. coli and coliform were ND for all samples.

**Significance:** Currently ClO₂ is created using a chemical production process; namely, strong or weak acid is mixed with sodium chlorite to produce the ClO₂. The reaction creates an acidic solution containing byproducts such as Cl₂ and ClO⁻ and may generate carcinogens, such as TTHMs, it thereby poses food safety problems. Future studies will focus on commercialized designs.

**P3-06** Safe-Pack – A New Class of Antimicrobial Packaging Material to Improve Meat Safety

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**Introduction:** Sustainable active microbialid (SAM) polymers provide a new class of functional biocides. These polymers are intrinsic antimicrobial thus the antimicrobial agents are immobilized in polymer and do not migrate into the environment. Therefore, SAM polymers show a promising potential for application as packaging material to improve food safety and quality. As part of a current research project, SAM polymers are being further developed into antimicrobial packaging.
Purpose: The aim of this study was to investigate the effect of a new class of antimicrobial packaging materials against selected pathogenic bacteria on meat and meat products.

Methods: The antimicrobial activity of the SAM polymer Poly-TBAMS was analyzed by adapting the test method JIS Z 2801:2000. The test method is based on a comparison of bacteria counts (S. aureus, E. coli) on sample and reference materials after 24 h at 7°C. The influence of bacterial strains, meat and meat products, and low temperature on the level of antimicrobial activity was investigated by varying these parameters. Therefore, meat and meat products were inoculated with selected pathogenic bacteria, stored at 7°C, and tested after 24 h.

Results: The initial bacterial counts of S. aureus and E. coli (> 5 log units) are reduced approximately by 4 log units on Poly-TBAMS after 24 h at 7°C. By adding inocula to meat juice, Poly-TBAMS shows high antimicrobial activity. Moreover, reduction levels > 1.2 log units are detected on inoculated chicken breast and > 2.9 log units on inoculated pork sausages even after 24 h.

Significance: The investigations show a high antimicrobial activity of the SAM polymer Poly-TBAMS against pathogenic bacteria on meat. Thus, Poly-TBAMS bears a high potential for applications as meat packaging materials to improve safety.

P3-07 Performances Assessment of the 3M™ Molecular Detection Assay Salmonella Kit According to the ISO 16140 Standard for Salmonella spp. Detection in Spices, Aromatic Herbs, Concentrates, Culinary Products, Cocoa and Milk Product

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Introduction: The 3M Molecular Detection Assay Salmonella kit uses isothermal amplification of specific DNA target sequences. The amplification is detected by bioluminescence.

Purpose: An independent study was conducted at ADRIA as part of the NF VALIDATION approval process, in order to extend the scope of the ISO 16140 validation study to spices and aromatic herbs, concentrates and culinary products, cocoa and cocoa-based products, and milk powders. The ISO 6579 standard was used as reference method.

Methods: The 3M Molecular Detection Assay Salmonella test protocol includes a single enrichment step in Buffered Peptone Water (ISO), and specific enrichment procedures are dedicated to tested matrices. Two incubation temperatures are validated depending on the tested food categories (37°C and 41.5°C). After lysis, DNA amplification is performed in the 3M Molecular Detection Instrument.

Results: Two hundred eighty-nine samples were analyzed for relative accuracy, sensitivity and specificity study. The results demonstrate equivalent performances between the 3M Molecular Detection Assay Salmonella method and the ISO 6579 methods. Depending on the tested (matrix/strain) pairs, the relative detection limits of the 3M Molecular Detection Assay Salmonella method vary from 0.3 to 3.6 CFU/25 g for the alternative method, those of the ISO standard vary from 0.4 to 2.8 CFU/25 g.

Significance: The alternative method is a reliable method for Salmonella spp. detection in spices and aromatic herbs, concentrates and culinary products, cocoa and cocoa-based products, and milk powders. The 3M Molecular Detection Assay Salmonella kit offers important economic savings by reducing time to result and handling time.

P3-08 Performances Assessment of the 3M™ Petrifilm Tests According to the ISO 16140 Standard for Total Viable Count, Enterobacteriaceae Count, Coliforms Count and E. coli Count in Pet Foods and Environmental Samples

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Introduction: The 3M Petrifilm Aerobic Plate Count, Enterobacteriaceae Plate Count, Coliforms Plate Count, E. coli Plate Count are ready culture medium systems which contain standard methods nutrients, a cold-water-soluble gelling agent, and a tetrazolium that facilitates colony enumerations.

Purpose: An independent study was conducted at ADRIA as part of the NF VALIDATION approval process, in order to extend the scope of the ISO 16140 validation studies to pet food and environmental samples. The ISO 4833, 21528-2, ISO 4832 and ISO 16659 methods were used as reference methods.

Methods: Each ISO 16140 method comparison study of these four Petrifilm tests gathered a linearity study done on 2 (matrix/strain) pairs, a relative accuracy study with 20 samples minimum tested in duplicate.

Results: The Petrifilm Total Viable Count Plate, Enterobacteriaceae Plate Count, Coliforms Plate Count and E. coli Plate Count show all satisfying linearity performances, with linear correlation coefficients superior to 0.98. The intercepts close to 0 and the slopes close to 1 were validated for the tested categories in the accuracy study. Biases between the Petrifilm tests and the related ISO methods are characterized by low values. The repeatability of the four Petrifilm tests is similar to the compared ISO standards.

Significance: The Petrifilm Total Viable Count Plate, Enterobacteriaceae Plate Count, Coliforms Plate Count and E. coli Plate Count are reliable alternative methods for Total Viable count, Enterobacteriaceae count, Coliforms count and E. coli count in pet foods and environmental samples. These four Petrifilm tests offer important economic savings by reducing handling time.

P3-09 Rapid Detection of Salmonella spp. in Dry Pet Food

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Introduction: Cases of human infections due to the contamination of dry dog food or dry cat food have been reported, especially with young children. Further to these outbreaks, pet food industries are looking for a rapid and reliable solution for the detection of Salmonella spp. Combining different technologies (PCR and easyMag™), bioMérieux has developed a dedicated method for the detection of Salmonella spp. in dry pet food in less than 12 h.
**Purpose:** The purpose was to compare the new bioMérieux method for the detection of *Salmonella* spp. in dry pet food to the ISO 6579 Reference method.

**Methods:** One hundred nine artificially contaminated samples of 125 g each were tested with both methods. The alternative method consists in a single enrichment in pre-warmed DPF broth and incubated at 41.5°C during 8 h. Then, DNA was extracted using the Nuclisens easyMag®: 1 ml of enrichment is placed in the easyMag® and we ran a dedicated protocol. DNA extracts are used directly with the *Salmonella* spp. PCR kit: 15 μl of detection buffer is added with 10 μl of DNA extract on Ready-to-use PCR microplate.

**Results:** Forty-four of one hundred nine (44/109) samples were detected positive and 50/109 were tested negative by both methods. Seven samples were detected positive with the PCR method and negative with the reference method. Eight samples were detected positive by the reference method and negative by the PCR method. These differences are due to the unpaired nature of enrichment between the two methods. The comparison showed no statistical difference between the 2 methods in term of sensitivity.

**Significance:** The bioMérieux method for dry pet food enables a reliable detection of *Salmonella* spp. in less than 12 h, allowing rapid decision and cost savings.

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**P3-10 Validation of EN ISO 11290-1&2 Standard Methods for Detection and Enumeration of *Listeria monocytogenes***

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**Introduction:** CEN/TC 275/WG 6, in charge of standardization in food microbiology at European level, has received a mandate from the European Commission to validate by inter-laboratory studies (ILS) a set of reference methods in food microbiology, including Standards EN ISO 11290-1 & 2, under revision for detection and enumeration of *Listeria monocytogenes* in food.

**Purpose:** Since the method is applicable to all food, feed and food processing environment, the ILS have been performed in 2013 on 5 matrices (smoked salmon, milk powder, vegetables, environment, and cheese), representative of categories cited in the EC Regulation 2073/2005 on microbiological criteria for foodstuffs.

**Methods:** About 16 laboratories participated in each trial. For quantitative studies, 4 levels of contamination including a blank and 2 blind replicates per level were used. For qualitative studies, 3 levels of contamination including a blank, and 8 blind replicates per level were used. In some cases, a competitive background microflora (in particular other *Listeria* species) was added. Enumeration results were transformed in log, and analyzed according to ISO 5725-2 Standard.

**Results:** Results as a whole were good, comparable if not better to former validation studies. In particular, values of repeatability and reproducibility standard deviations were considered correct for enumeration with a confirmation stage, since they were all <0.3 log, also for low levels. According to multi-way variance analysis, participating laboratories, diluents type, chromogenic selective agar manufacturers and presentation had no impact on results. Detection sensitivity ranged from 91.1 to 100%, whereas specificity ranged from 97.6 to 100%. Though isolation after half-Fraser enrichment allowed obtaining most positive results, a second selective enrichment in Fraser allowed to obtain more positive samples, thus illustrating the importance of the second enrichment.

**Significance:** New drafts for revision of EN ISO 11290-1&2, including the results of the ILS, have been submitted to the CEN/ISO parallel enquiry (30/10/2014-30/03/2015).

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**P3-11 ISO 16140 Comparative Validation Study to Demonstrate the Equivalence of a 10-hour Dual *Salmonella* and *E. coli O157:H7* Enrichment for Assurance GDS® to the Reference Culture Methods for the Detection of *Salmonella***

**David Kerr and VERENA PEGGION, BioControl Systems, Inc., Bellevue, WA**

**Introduction:** Many foods are commonly tested for both *Salmonella* and *E. coli* O157:H7. Typically, these foods are enriched separately increasing time and lab costs. This study proposed harmonizing these enrichments so food samples could be enriched in a single modified EHEC (mEHEC) broth previously validated for use with Assurance GDS for *E. coli* O157:H7 in as little as 8 h.

**Purpose:** To demonstrate the equivalence of Assurance GDS and a new mEHEC enrichment to ISO 6579 (2002) reference culture methods for the detection of *Salmonella* in selected foods and environmental surfaces.

**Methods:** According to ISO 16140 standard requirements, 4 categories (meats, dairy products, fruits and vegetables, and environmental samples) and a 376 g sample for the raw beef meat analysis were tested. For each category a minimum of 60 products belonging to a minimum of 3 food types were tested with approximately 50% being positive and 50% negative. Twenty-five gram samples from each category were enriched at a 1:10 sample to media ratio in mEHEC for 8 h at 42°C. For raw beef, additional 375 g samples were enriched at a 1:5 sample to media ratio for 8 h at 42°C. All samples were analyzed using Assurance GDS for *Salmonella* Tq and the reference culture methods, EN ISO 6579 (2002).

**Results:** In total, there were valid results from more than 300 samples across 4 categories. The data were analyzed using the acceptability limits defined for the unpaired data studies in the ISO 16140 (FDIS, 2014). Assurance GDS performed within the acceptable limits and the results were statistically not different than the reference culture method for the detection of *Salmonella*.

**Significance:** The newly harmonized mEHEC enrichment for both *Salmonella* and *E. coli* O157:H7 saves time and lowers lab testing costs for customers.

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**P3-12 ISO 16140 Comparative Validation Study of Assurance GDS, a Novel PCR Method Using Immunomagnetic Separation, for the Detection of *E. coli O157:H7* in Foods and Environmental Samples in 10 Hours***

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**Introduction:** *E. coli* O157:H7 poses a public safety risk typically associated with meat, dairy, and vegetable products which have a short shelf life. This study validates the use of Assurance GDS, a novel PCR-based method incorporating immunomagnetic separation (IMS) technology for the detection of *E. coli* O157:H7 in food and environmental samples in as little as 10 h.
**Purpose:** To demonstrate the equivalence of Assurance GDS to the ISO 16654 (2001) reference culture method for the detection of *E. coli* O157:H7 in foods and environmental surfaces in accordance with ISO 16140.

**Methods:** According to ISO 16140 standard requirements, 4 categories (raw beef meats, dairy products, fruits and vegetables, and environmental samples) and a 375 g sample for the raw beef meat analysis were tested. For each category a minimum of 60 products belonging to a minimum of 3 food types were tested with approximately 50% being positive and 50% negative. Twenty-five gram samples from each category were enriched at a 1:10 sample to media ratio in mEHEC for 8 h at 42°C. For raw beef, additional 375 g samples were enriched at a 1:5 ratio for 8 h at 42°C. All samples were analyzed using Assurance GDS for *E. coli* O157:H7 Tq and EN ISO 16654 (2001).

**Results:** In total, there were valid results from more than 300 samples across 4 categories. The data were analyzed using the acceptability limits defined for the unpaired data studies in the ISO 16140 (FDIS, 2014). Assurance GDS performed within the acceptable limits and the results were statistically not different than the reference culture method for the detection of *E. coli* O157:H7.

**Significance:** Assurance GDS can be used to accurately detect *E. coli* O157:H7 in as little as 10 hours, allowing producers to release products quickly, reducing holding costs and maximizing shelf life.

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**P3-13 Sarcocystis spp. in Human Population from Northwest Italy**

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**Introduction:** The genus *Sarcocystis* consists of cyst forming coccidian with an obligatory two-host life cycle. Humans may acquire the infection by ingesting raw or undercooked beef or pork containing sarcocysts belonging, respectively, to the two known zoonotic species: *Sarcocystis hominis* and *Sarcocystis suihominis*. Even if gastrointestinal symptoms subsequent to the infection by these organisms have been reported, data related to distribution and prevalence of *Sarcocystis* spp. in human population are scarce.

**Purpose:** The objective of this study was to evaluate the presence of *Sarcocystis* species in human population in Northwest Italy, a region characterized by large consumption of raw beef and a high prevalence (30–40%) of *S. hominis* in cattle population, but without official report of human sarcocystosis.

**Methods:** Thirty-four frozen stool samples, collected from patients suffering from gastrointestinal symptoms without etiological diagnosis, were conferred, by the Laboratory of Microbiology and Virology of City of Health and Science University Hospital of Turin, Italy, and analyzed by a semi-Nested PCR approach for the research of *Sarcocystis* spp. Amplicons were subsequently sequenced for confirmation and species identification.

**Results:** Of 34 samples, 18 (53%) resulted positive to the first PCR; 4 more samples resulted positive to the Nested assay, for a total of 22/34 (65%) positive samples for *Sarcocystis* spp. Sequencing confirmed the genera and identified all the isolates as *S. hominis*.

**Significance:** The study confirms the suspect of unreported cases of human sarcocystosis; oocysts belonging to *Sarcocystis* spp., in fact, may pass unnoticed during the coprological examination. Even if our data do not allow to support the pathogenic role of *S. hominis*, clearly demonstrate the need of a more wide epidemiological study in order to evaluate *S. hominis* relevance to the public health.

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**P3-14 Listeria monocytogenes and Salmonella enterica Intraspecies Biodiversity: Effect of Recovery on Microbiological Media during Fermented Sausage Production**

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**Introduction:** Microbiological testing, based on the use of selective media, is a basic tool for monitoring the safety of foodstuffs, both during production but also throughout all the stages of transportation, storage and commercialization.

**Purpose:** The purpose of this study was to investigate intraspecies biodiversity for two major foodborne pathogens, namely *Salmonella enterica* and *Listeria monocytogenes*, focusing on the capacity of different strains to (i) persist during sausage production when they are artificially inoculated in the meat batter, (ii) be recovered in various synthetic media used for their detection in foods.

**Methods:** A mix of 4 different strains of *L. monocytogenes* and 5 strains of *S. enterica* was artificially inoculated in the meat destined for production of two types of fermented sausages. Then, the microorganisms were monitored during the production process (fermentation and maturation) by microbiological analysis performed at regular intervals and subsequent isolation and molecular characterization of the isolates.

**Results:** The mixed populations of the two species showed different evolution patterns. While for *S. enterica* all the strains inoculated persisted for the whole production period, for *L. monocytogenes* an evident variation of persistence was observed. Among the strains inoculated, one was isolated with higher frequency. Strikingly, for *L. monocytogenes* it was possible to observe an effect of the enrichment broth in the recovery of the various strains inoculated. On the contrary, for *S. enterica* no differences were observed in the recovery with the two enrichment broths tested.

**Significance:** The differences observed underline that strains of *L. monocytogenes* possess varying capabilities to survive during fermented sausage production. Perhaps of even greater interest is the fact that the synthetic media used for the determination of *L. monocytogenes* in foods may give a biased picture of the real ecology, due to dissimilarities among strains, concerning their recovery in such media.
Introduction: Recent evidence of the European Food Safety Authority (EFSA 2010) have focused on the risk assessment of parasites in fishery products, such as Anisakidae larvae and the detection methods in fish fauna. Fish can be examined for the presence of parasites by a variety of methods including digestion. The digestion method involves the use of a pepsin/hydrochloric acid solution to free anisakid larvae from muscle or other tissues. The method is derived from the review of Reg EC 2075/2005, applied to the detection of Trichinella. 

Purpose: In this study we want to determine the digestive method efficiency in 22 fish species examined for the regional monitoring plan of Sicily for the detection of Anisakidae larvae in fish products.

Methods: Two hundred seventy fish samples were examined for the detection of Anisakidae larvae by digestion method. One hundred g of samples were weighed and homogenized. Subsequently, 16 ml of HCl (25%) and 10 ± 0.2 g of pepsin (1:10,000 NF) were added to samples and put into a becher with 2 l of water. The becher was placed in a magnetic stirrer at 44–46°C for 30 min. The digestion product was filtered in filters with 180 µm meshes and examined under the stereomicroscope.

Results: A number of 39 samples found negative at visual inspection gave a positive result for Anisakidae presence with digestion method (17% of samples). Results demonstrate a good efficiency of the digestive method for the detection of Anisakidae larvae in fish products.

Significance: This study confirms the effectiveness of the digestive method as an additional tool for the Anisakidae larvae detection in fish product. Results from this work revealed an efficacy prevalence of the digestive method in Scomber scombrus; this evidence can be related to the intramuscular infestation prevalence of Anisakidae larvae in mackerel.

P3-16 Automated Food DNA Purification for Authentication and Genetically Modified Organism (GMO) Testing

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Introduction: The ability to trace and authenticate a food product is of major concern to the food industry. It has implications in consumer protection and regulatory compliance as well as cultural and religious aspects. Advances in molecular technologies and genomic analyses of food sources enable screening at the nucleic acid level.

Purpose: Here we examine the utility of a small benchtop automated instrument, the Maxwell® 16, and novel cellulose-based magnetic particles for purification of amplifiable DNA from a variety of foods.

Methods: We choose 4 key areas of interest for food ingredient testing: genetically modified organisms (GMO) detection, seafood identification, seed screening (rice and coffee) and meat product testing. For each area, nucleic acid was extracted, quantitated, and tested for amplifiability in either end point or quantitative PCR.

Results: For GMO testing, extracted DNA from both dried corn kernels and processed corn foods (chips and pretzels) were tested for the presence of cauliflower mosaic virus (CMV) sequence. Results showed amplification of CMV sequences in GMO branded samples only. For seafood authentication, DNA was extracted from processed and frozen tuna and salmon and successfully amplified using endpoint PCR of cytochrome c, typically used for fish species identification. For seeds, DNA was purified and demonstrated to be amplifiable from rice (medium grain and basmati) and coffee beans (Arabica and Robusta). Meat testing focused on isolation of amplifiable nucleic acid from processed foods including ravioli, sausage and gelatins.

Significance: These studies together demonstrate the use of the Maxwell® 16 System for automated purification of food DNA upstream of amplification-based GMO and authentication testing.

P3-17 Assessment of Compliance of High-capacity Slaughterhouses in Finland

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Introduction: In the European Union, slaughterhouse operators are obligated to implement own-check systems (OCS) based on good hygiene practices and HACCP. The purpose of the OCS is to ensure that the premises and operations meet the requirements set in food safety regulations. Official veterinarians verify that food safety requirements are met and that OCSs are properly implemented. A well-functioning OCS is important in securing meat safety.

Purpose: The aims of our study were to investigate how well slaughterhouse operators’ OCS function and to examine the incidence and severity of non-compliances as assessed by the chief official veterinarians.

Methods: An electronic questionnaire examining non-compliance in all high-capacity red meat (n = 13) and poultry (n = 4) slaughterhouses of Finland was sent to chief official veterinarians (n = 17) in the spring of 2014. Respondents were asked to report in which part of the OCS they had observed non-compliances in the slaughterhouse during the previous year, how often and how severe non-compliances were in terms of meat safety. A Likert-scale from 1 (very seldom when inspected/not severe at all) to 4 (almost always or always when inspected/very severe) was used. The response rate was 76% (13/17).

Results: Common non-compliances concerned cleanliness of premises and equipment, hygienic working methods, and hygiene and functionality of sterilizers (non-compliance present in 100%, 92% and 77% of the slaughterhouses, respectively). These non-compliances were observed occasionally or often when inspected (means 2.2 – 2.5) and assessed somewhat severe to severe in terms of meat safety (means 3.1 – 3.2). However, full compliance was observed for instance in the control of Salmonella and surveillance of enterohemorrhagic Escherichia coli and Campylobacter.

Significance: Hygienic problems are common in slaughterhouses and can threaten meat safety. Hygiene management should be further addressed in the OCSs and the official control of slaughterhouses.

P3-18 Evaluation of Capsid Integrity Measurement for Predicting the Integrity of Human Norovirus Particles – An Inter-laboratory Comparison

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Introduction: Human Norovirus (hNoV) is the biggest cause of gastrointestinal disease in the industrialized world. The precise attribution of food to hNoV infections in the UK is unknown. Although the quantitative reverse transcription polymerase chain reaction (RT-qPCR) is widely used for hNoV detection, it cannot discriminate between infectious and non-infectious virus particles. hNoVs cannot be readily cultivated and alternative approaches for predicting hNoV infectivity are required. Combining the measurement of capsid integrity with RT-qPCR detection methods may provide a method of obtaining more informative data from studies of hNoV contamination of foods.
Purpose: The purpose of this study was to assess the reproducibility of a capsid integrity assay (CIA) by inter-laboratory comparison as a precursor study for the potential application of this approach to food samples.

Methods: A standard operating procedure was developed to examine the effect of RNase and heat treatment (80°C for 2 min) on six dilute (0.1% w/v) hNoV GI and GII stool samples using the hNoV RT-qPCR method detailed in the standard method for detection of viruses in foods (ISO/TS 15216). Differences in Cq values (ΔCq) resulting from the different treatments and samples were measured in a blind study using three laboratories.

Results: The results showed that the method was reproducible between laboratories. No RNase digestible RNA was detectable in any of the samples without heat treatment. Mean inter-laboratory ΔCq values following heat treatment and RNase digestion were 6.7 (SD 1.9, range 0.7–9.4) depending on the sample and laboratory. One sample showed increased heat resistance requiring heat treatment at 90°C for 2 min to maximize RNA exposure.

Significance: The results have shown that the assay shows good reproducibility between laboratories and may be applicable to food samples. Further studies are required to assess the application of this approach to food samples.

P3-19 Loop-mediated Isothermal Amplification (LAMP) for the Detection of Salmonella spp. Isolated from Different Food Types

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Introduction: Salmonellosis, a major foodborne infectious disease worldwide, is caused by 2500 serovars, and is most often attributed to the consumption of contaminated foods such as poultry, beef, pork, eggs, milk, seafood, nut products, and fresh produce. Loop-mediated isothermal amplification (LAMP) is a novel nucleic acid amplification assay that is rapid, sensitive, specific, and simple, and has provided a new alternative for molecular diagnosis.

Purpose: The objective of the study was the application and further evaluation of a diagnostic LAMP assay for the detection of Salmonella in foods, and its potential use in routine food analytical laboratories.

Methods: Fifty strains of 15 different serotypes of Salmonella subsp. enterica (isolated from raw poultry and pig meat samples, taboulen salad, food topping, sesame from India, and pie with greens), and 10 strains of 3 bacterial species other than Salmonella, 2 virus species, and 2 yeast species were used to further evaluate the specificity of the LAMP assay developed by Hara Kudo et al. (2005). The assay targets Salmonella enterica invasion protein (invA) gene, and was performed at 65°C for 60 min using a thermal cycler. LAMP products were visually detected under daylight or ultraviolet light, or after agarose gel electrophoresis.

Results: All Salmonella strains were shown to be positive using the LAMP assay; whereas, all other bacteria, virus and yeasts tested in this study were negative. Salmonella specific invA gene sequences were successfully amplified at 65°C in 60 min, by the LAMP assay.

Significance: The study evaluated the inclusivity of a developed LAMP method for the detection of Salmonella spp. strains isolated from different food matrices, as a sensitive, rapid and inexpensive detection method, which could be of interest for screening purposes in food analytical laboratories.

P3-20 Determination of Antimicrobial Effect of Essential Oils Alone or Combined with Non-thermal Disinfection Technologies in Fresh-cut Lettuce

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Introduction: Foodborne diseases are a serious threat to public health. Fresh-cut produces have been associated with a number of foodborne illnesses in recent years. Non-thermal disinfection methods have been applied to a wide variety of food products to destroy microorganisms. Essential oils (EOs) hold promise as a natural hurdle for microbial safety and can be used in conjuction with novel techniques for eliminating microorganisms from fresh produce.

Purpose: The purpose of the current study was to evaluate the antimicrobial effects of EOs alone or combined with non-thermal technologies such as ultrasound and UV on the survival of Escherichia coli, Staphylococcus aureus, Salmonella Enteritidis and Listeria innocua inoculated into fresh-cut lettuce.

Methods: Commercially available romaine lettuce (Lactuca sativa) was purchased from a local supermarket. Experiments were conducted using four bacterial strains: Escherichia coli NCTC 9001, Staphylococcus aureus NCTC 6571, Salmonella Enteritidis NCTC 6676 and Listeria innocua NCTC 11288. The cocktail of the above microorganisms was inoculated on fresh-cut lettuce. Different EOs (basil oil, lavender oil and mint oil) grown in Greece were selected and two concentrations were implemented on inoculated food products. Then the additive effect of ultrasound or UV technologies followed by EOs was tested. For the microbial analysis, ISO methods were used for the determination of the above microorganisms.

Results: The results of the present study showed that differences in disinfection efficiency among different essential oils were significant (P < 0.05). Moreover, the combinations of US/EOs or UV/EOs exhibited an additive effect (1–2 log reduction) than the EOs alone (0.5–1 log reduction) for reducing the cocktail of microorganisms in fresh-cut lettuce.

Significance: These data suggest that the combination of non-thermal with EOs could find potential applications for decontamination in the food industry.

P3-21 Survival of Salmonella in Paprika Powder and Rice Flour

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Introduction: Although low moisture foods are generally considered a low risk to human health it is now believed that they contribute significantly to the total number of foodborne infections; therefore, more attention should be paid to controlling the persistence and elimination of pathogens from these foods. Knowledge of the survival patterns, mechanisms and heat resistance of pathogens in low moisture foods are still not well understood and further studies are required.

Purpose: To investigate the survival patterns and heat resistance of Salmonella at various water activities of dry products (paprika powder at a_w = 0.45 and a_w = 0.55 and rice flour at a_w = 0.2 and a_w = 0.55) at different temperatures in comparison with surrogate bacteria for use in a pilot-scale pasteurization studies.

Methods: Rice flour and paprika powder were inoculated with several Salmonella strains and survival was monitored over a period of 12 weeks. Inoculated samples were stored at 15 and 25°C and survival monitored by plating out on Tryptone Soya Agar. Heat resistance was evaluated using glass vials at a range of temperatures (65–95°C). In parallel, comparative studies with surrogate Enterococcus faecium were performed.
Results: Results showed that survival and heat resistance of Salmonella in both products was greater when stored at low aw and at 15°C compared to 25°C. Survival patterns of Enterococcus faecium are very similar to Salmonella but heat resistance was significantly different. Z-values for Salmonella (12–20°C) were higher than z-values for Enterococcus faecium (11–13°C), showing that Salmonella is more heat resistant at higher temperatures than Enterococcus faecium.

Significance: Results have shown that use of Enterococcus faecium ATCC 8459 as a Salmonella surrogate has some limitations especially when used at higher temperatures (> 75°C).

P3-22 Survival of Pathogenic Microorganisms in Spices and Herbs
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Introduction: Spices and dried aromatic herbs can be cultured where hygiene conditions might be poorly controlled and products can have high levels of spoilage and pathogenic microorganisms. Since spices and dried herbs are commodities with low water activity, they are usually stored at room temperature under dry conditions. Hence they have a shelf life of 2–3 years.

Purpose: Although drying can inhibit microorganism growth, it may not completely inactivate pathogens. Thus the purpose of this study was to investigate survival of pathogens during storage of spices and dried herbs.

Methods: We investigated the survival capacity of different pathogenic microorganisms in powdered paprika and also performed a meta-analysis to identify the most critical factors that influence survival. We performed a meta-analysis on the available published data to identify the most critical factors that influence survival in spices and dried herbs. Additionally, survival of different pathogenic microorganisms was monitored experimentally in powdered paprika under controlled storage conditions.

Results: From the meta-analysis we concluded that storage temperature and water activity both play significant roles in survival. Experimental studies which simulated storage conditions showed that different pathogens do not survive to the same extent under the same storage conditions as Salmonella spp. had a fifteen times lower inactivation rate than Listeria monocytogenes.

Significance: Reduction of pathogens during storage of spices and herbs might be limited depending on the type of organism present. Control of the initial levels of microbial contaminants is therefore of importance.

P3-23 Impact of Drying Kinetics on Foodborne Pathogen Salmonella enterica Survival
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Introduction: Salmonella enterica is a foodborne pathogen, salmonellosis agent. It is highly represented in outbreak across the world, with nearly 100,000 cases every year in the European Union. Its ability to survive in several environments and its low infective dose made Salmonella enterica a key bacterium in food protection.

Purpose: This study aims to evaluate and understand the impact of drying process conditions (such as relative humidity, duration, kinetics or the food product nature) on S. enterica survival.

Methods: To study the survival of S. enterica as a function of drying different levels of relative humidity, we selected two S. enterica serovars: Salmonella Typhimurium, for its implication in outbreak and its thermal resistance in dried state, and Salmonella Senftenberg, for its thermal stress resistance in aqueous state. These strains were suspended in phosphate buffered saline solution or milk and then dried in thin covering layers on glass in controlled-relative humidity containers maintained to 11%, 25%, 44% and 58% thanks to saturated salt solution.

Results: The higher and lower drying rate, 11% and 58% of relative humidity, respectively, correspond to the lower destructions (1.5 log and 3 log for Salmonella Typhimurium, respectively, for 15 min in phosphate buffer saline) and the intermediate drying speed, 25% and 44% of relative humidity respectively, correspond to the higher destruction (3.5 log and 4 log for Salmonella Typhimurium, respectively, for 15 min phosphate buffer saline). Furthermore, drying in milk brings about less cell mortality than drying in phosphate buffer saline (2.5 log and 4 log for Salmonella Typhimurium, for 180 min at 58 % of relative humidity).

Significance: These results indicate that the drying parameters have a high impact on Salmonella survival and drying processes could be a tool for food decontamination and food safety.

P3-24 Monitoring Spoilage of Sterile Pork Meat Fillets Inoculated with Specific Spoilage Microorganisms (Lactobacillus sakei, Leuconostoc mesenteroides) Packaged under Modified Atmospheres in Tandem with GC/MS Analysis and Chemometrics
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Introduction: Lactobacillus, Leuconostoc and Carnobacterium are most frequently found on vacuum or modified atmosphere packaged meat playing an important role in the spoilage of refrigerated meat.

Purpose: The purpose of this work was to determine the type of end-products produced during the growth of two main specific spoilage microorganisms in contrast to sterile meat and to determine the metabolomic profile of the samples during storage.

Methods: Sterile pork meat was inoculated with 2 log CFU/cm² of Lb. sakei, L. mesenteroides and mix cultures and stored under MAP at 4 and 10°C until spoilage was pronounced. Microbiological analysis (TVC, LAB) was performed in parallel with HP/SME-GC/MS analysis. The spectral data collected from GC/MS were subjected to factorial discriminant analysis (FDA), One-way ANOVA and PLS-DA to distinguish the metabolic compounds produced by the different microorganisms used.

Results: Results showed qualitative and quantitative differences on the volatile compounds (alcohols, carboxylic compounds, esters) detected by GC/MS, between microbial species in mono and mix cultures as well as on sterile meat. Analysis with ANOVA and PLS-DA showed that 1-butanol, 2-butanol and 2-hexanol was related more with sterile samples, while 1-octen-3-ol, 2-heptenal, 2-octanone were related mostly with samples inoculated with Lb. sakeii. Finally, FDA
provided good discrimination between different inoculated meats since the classification was very accurate for sterile meat in contrast to mono and mix cultures.

**Significance:** The relationships between mono-cultures and volatile profile bring a better understanding of the metabolites produced and could be used as a fingerprint to monitor food spoilage.

**Acknowledgment:** This work has been supported by the project “Intelligent multi-sensor system for meat analysis-imMeatSense _550” co-financed by the European Union (European Social Fund–ESF) and Greek national funds through the Operational Program “Education and Lifelong Learning” of the National Strategic Reference Framework (NSRF) - Research Funding Program: ARISTEIA-I.

**P3-25 Effects of Climate Change on the Microbial Hazards in Cabbages and Young Radishes**

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**Introduction:** Climatic changes likely to affect fate and transport of pathogens and food safety risks could be very different from today. Microbiological contamination of green vegetables has shown varied by growing regions and weather conditions. These environmental conditions would affect the safety of fermented product, such as kimchi in Korea.

**Purpose:** Therefore, the purpose of this study was to evaluate the microbiological hazards pattern with climate change and suggest the ways to secure the safety of kimchi manufacturing processes.

**Methods:** The raw, pickled, washed and dewatered cabbages, fresh kimchi, and salted, washing and rinsing water were collected from kimchi manufacturer in every month. The soil, surface water and cabbage were collected in fields. The raw young radishes were purchased from market in the summer. Total aerobic bacteria, coliforms and pathogens (Escherichia coli, Salmonella spp., Staphylococcus aureus, Bacillus cereus, Clostridium perfringens and Listeria monocytogenes) were analyzed by following method of the Korea Food Code.

**Results:** Total aerobic bacteria in samples were ranged from 3.93 to 9.08 log CFU/g and coliforms were 2.13 to 5.22 log CFU/g throughout the year, showing the low in winter and higher number in summer. No pathogens were detected except *E. coli* of 2.18, 4.36 and 1.81 log CFU/g in salted and washing water, and dewatered cabbage, which cabbage were cultivated when the dry and hot summer for a month. *E. coli* were also found at 1.96, 0.4 and 0.74 log CFU/g from field cabbages at edible leaf part, soil and surface water, respectively, as well as 0.84 log CFU/g from raw young radish in a hot and drought period.

**Significance:** These results suggest that the water supply method should be changed to drip from sprinkler irrigation to avoid pathogen contamination at field and kimchi should be fermented to reach pH below 4.8 during summer to prevent foodborne disease.

**P3-26 Salad and Salmonella**

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**Introduction:** Leafy green and other salad vegetables are an important part of a healthy diet, providing vitamins, minerals, and dietary fiber. However, increasing numbers of cases of enteric pathogen contamination of salads such as lettuce, spinach, watercress, and bean sprouts indicate that there is a food safety problem of growing economic importance. However, little is known of what influences enteropathogen behavior when it colonizes fresh produce.

**Purpose:** Salad leaves become damaged during processing, releasing water. We therefore investigated if salad juices affected the growth and virulence of *Salmonella*, one of the mostly commonly isolated fresh produce pathogens.

**Methods:** Growth, proteomic and biofilm assays were used to study the effects of exposure to microliter volumes of juices from gently blended mixed lettuce leaves and alfalfa beansprouts.

**Results:** Salad extracts enhanced *Salmonella* in all the media tested; effects were dose dependent but as little as 5 µl could catalyze > 1000-fold increases in cell numbers compared with un-supplemented controls (*P* < 0.01). Attachment to surfaces is essential for salad leaf colonization, which makes important our finding that exposure to the salad extracts massively increased *Salmonella* biofilm formation. Proteomic studies also showed that expression of *Salmonella* proteins involved in host cell invasion and intracellular survival were increased following salad juice exposure.

**Significance:** Salad juices at microliter volumes can enhance *Salmonella* growth and virulence. The objective of our BBSRC funded research is therefore to develop ways of preventing enteric pathogen attachment to fresh produce.

**P3-27 Label-friendly Reduction of Mold Spoilage on Semi-hard Cheese**

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**Introduction:** Molds play a major role in spoilage of various types of cheeses and can lead to high economic losses. Local regulations and consumer preferences for clean label products limit the toolbox to control spoilage of cheese. Label-friendly alternatives are adjunct cultures with bio-protective functionalities which on top of flavor contribution act as an additional hurdle thanks to the production of antimicrobial metabolites and competitive exclusion.

**Purpose:** The objective of the study was to evaluate the inhibitory activity of an antifungal culture consisting of a *Lactobacillus plantarum* strain against mold spoilage on semi-hard cheese.

**Methods:** Semi-hard cheese samples were prepared with a commercially available cheese starter culture and test samples were additional inoculated with the antifungal culture. The milk was warm-ripened, coagulated and the curd cut in grains. The curd was filled in molds and after pressing the cheeses were salted and ripened for six weeks. After ripening, the cheeses were sliced and surface inoculated with different spoilage molds. The cheese slices were packaged under ambient atmosphere, stored at 6°C and inspected daily for visual mold growth. In addition, cheese samples were evaluated for sensory properties.

**Results:** The application of the antifungal culture delayed the outgrowth of molds compared to the samples without antifungal culture added. In respect of some mold strains, the period without visible mold growth was up to four times longer than that of cheeses made with the cheese starter culture alone. The results from the sensory evaluation indicate that addition of *Lactobacillus plantarum* contributes to increased flavor intensity.
**Significance:** The results demonstrate that antifungal cultures can limit the outgrowth of spoilage molds on semi-hard cheese and thus maintain the shelf life and reduce food waste.

**P3-28 Inhibition of Salmonella Typhimurium and Escherichia coli O157:H7 in Enhanced Pork Loin by Buffered Vinegar**

**ANJA VERBIEST**; Ms, Herentals, Belgium

**Introduction:** To meet the growing demand for label-friendly ingredients, Kemin developed a buffered vinegar-based food ingredient for controlling food pathogens in processed meat and poultry.

**Purpose:** Evaluate the inhibition of Salmonella Typhimurium (ST) and Escherichia coli O157:H7 (EC) in pork loin by injecting a brine solution containing water, salt, sodium phosphate and buffered vinegar and to determine the effect on quality and microbiological characteristics.

**Methods:** Treatments include 3% sodium lactate-diacetate, 1% Buffered Vinegar Liquid, 0.75% Buffered Vinegar Dry and an untreated control, conducted in three replications. Pork loin was surface inoculated with ST and EC to provide 5 log CFU per 100-g package. Duplicate inoculated samples were assayed for changes in ST and EC populations by plating and incubated at 37°C for 24 h. Sampling was discontinued if there was > 1 log CFU/kg for two or more consecutive sampling intervals or > 2-log increase. Duplicate, non-inoculated samples were assayed for Aerobic Plate Counts and pH at 0, 1, 2, 3, 4 and 5 weeks storage at 4°C. Lightness, redness and yellowness values were measured on duplicate non-inoculated samples for each treatment at weekly intervals. The microbiological data was reported as average values and standard deviations (log CFU/ml rinse) for duplicate samples and three separate trials (n = 3).

**Results:** Statistical analysis confirmed that treated samples significantly inhibited (P < 0.05) the growth of ST and EC for 5 weeks. Untreated control showed > 1-log increase after 2 weeks for ST and 4 weeks for EC. APC counts of untreated control showed > 7 log CFU/g after 3 weeks and the counts for the Buffered Vinegar and lactate treatments were in the range of 2 – 6 log CFU/g after 5 weeks.

**Significance:** Clean label ingredients like Buffered Vinegar can extend the shelf life of pork loin without impacting the quality characteristics and at much lower application levels compared to sodium lactate-diacetate.

**P3-29 Contamination Routes of Spoilage Bacteria in Salmon Processing Plants**

**TROND MØRETRØ** and Solveig Langsrud; Nofima, Norwegian Institute of Food, Fishery and Aquaculture, Ås, Norway

**Introduction:** Photobacterium, Shewanella and Pseudomonas are recognized as spoilers of salmon. Knowledge about the prevalence of these bacteria in salmon processing plants is scarce.

**Purpose:** To investigate contamination routes of important spoilage bacteria of salmon.

**Methods:** Two salmon processing plants were visited and sampled after cleaning, before production. Ninety-seven samples from equipment/ machines, 9 water samples and 18 samples from fish from different processing steps were plated on iron agar and the bacterial composition was identified by 16S rDNA sequencing.

**Results:** Pseudomonas spp. dominated in both plants, and were found in 61% and 51% of the samples in the two plants. Pseudomonas spp. were more frequently found on equipment than on raw unprocessed salmon. Pseudomonas also dominated on ice stored filets. The diversity of Pseudomonas in the processing environment was found to be high based on 16S rDNA analysis. Shewanella spp. were detected from both fish and equipment in the slaughter department. The prevalence of Shewanella spp. were lower from equipment in the filleting department compared to the slaughtering department. Shewanella spp. was also present on ice stored filets. Photobacterium spp. were detected from salmon early in the slaughtering process, but not from fish later in the process. Photobacterium spp. were not detected from any of the 97 samples from surfaces of equipment, machines etc. after cleaning.

**Significance:** Critical contamination sites along production lines for salmon were determined and identification of the microbiota showed that the routes of transmission to products differed between important salmon spoilage bacteria. The knowledge can be used for selecting strategies in order to improve the quality of salmon.

**P3-30 FoodChain-Lab: Tracing Software Supporting Foodborne Disease Outbreak Investigations**

**Armin A. Weiser, Christian Thoens, Bernd Appel, Annemarie Kaesbohrer and MATTHIAS FILTER**; Federal Institute for Risk Assessment, Berlin, Germany

**Introduction:** In case of foodborne disease outbreaks, rapid identification of the causative food product is essential, since the medical and economic damages grow with the duration of the outbreak. Experiences from several outbreaks in Europe demonstrate that there is a need for an expert software system capable of supporting investigation on supply chains as well as exposure assessments in crisis situations. Furthermore, the expert software system should be able to provide a comprehensive data management infrastructure necessary to assure high data quality and integrity and prompt analysis capabilities.

**Purpose:** The purpose of developing this software was to create a free and open-source software resource for public health experts applicable in feed- or foodborne disease outbreak investigations as well as in exposure assessment tasks related to feed or food supply chains.

**Methods:** FoodChain-Lab has been implemented as an extension to the open source data analytics platform Konstanz Information Miner (KNIME). KNIME enables visual assembly of data analysis workflows. The installation guide, the source code, example workflows and play data are available via http://foodrisklabs.bfr.bund.de.

**Results:** Since its initial application during the EHEC outbreak in Germany in 2011 FoodChain-Lab has been used and tested in several outbreak investigations, e.g., the norovirus outbreak in Germany in 2012 or the hepatitis A virus outbreak in Europe. On the basis of these experiences the software evolved from a data visualization and analyses tool into a comprehensive tool box for data management, enrichment, visualization, analysis and interactive reasoning.

**Significance:** FoodChainLab proved to be helpful during several investigations of foodborne disease outbreaks since 2011.
**P3-31**  
Isolation and Characterization of a New *Listeria monocytogenes* Molecular Serotype  
HEND ALI GAHMI, Ogueri Nwaiwu and Catherine Rees; Div of Food Sciences, University of Nottingham, Loughborough, United Kingdom

**Introduction:** *L. monocytogenes* is a foodborne bacterium, responsible for listeriosis in humans and animals. It is the causative agent of most deaths associated with bacterial foodborne infection in the UK and the FSA has targeted this organism as a priority for action. The “at risk” groups for listeriosis are usually pregnant women, infants, the elderly and other immunocompromised individuals. Incidence of listeriosis has increased in several European countries, but this has not been associated with any specific epidemic clone on change in virulence of the isolates associated with human infection.

**Purpose:** A common food source of associated with *Listeria* infections are RTE products, including freshly prepared salads and sandwiches, hence a research project was undertaken to investigate the source of *Listeria* found in commercially prepared green peppers.

**Methods:** International standard (ISO 112090-1) methods were used to isolate *Listeria* samples of the peppers taken at different points from the production line. These were further characterized using API™ *Listeria*, PCR multiplex serotyping and lineage PCR analysis.

**Results:** The seeds removed during preparation of cut product were the most common source of *L. monocytogenes*. One particular isolate from the seeds was further characterized and gave an unusual PCR serotyping result, producing a band pattern that has not been previously reported in the literature. The strain was fully hemolytic, gave characteristic colony morphology on Listeria Brilliance agar. Both the *prs* and ORF2819 primers produced bands of the normal size, and therefore the genome variation lies in the region targeted by the primers specific to either *lmo1118* or *lmo0737*. In addition, the lineage PCR results were also anomalous indicating a further change in the *actA-plcA* region.

**Significance:** This is the first report of this new *L. monocytogenes* variant, and adds to previous evidence that members of this species can be quite genetically diverse.

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**P3-32**  
Prediction of Shelf Life of Bakery Products: Illustrated by Packed Pineapple Cake  
Hsin-I Hsiao and KAI-YI FENG; National Taiwan Ocean University, Keelung, Taiwan

**Introduction:** Consumers demand safe and high-quality products with superior texture and longer shelf lives. Conducting a complete shelf life test for shelf stable products can be very time and resource consuming, thus multivariate accelerated shelf life tests are often employed. But it is still challenging for long shelf life products like bakery products with reported shelf-lives of between 30 to 60 days.

**Purpose:** This study aims to design a model whereby the shelf life of bakery products can be predicted in a short time-period. Pineapple cake was chosen for analysis because it is one of popular bakery products in Taiwan. The first part aims to determine the shelf-life of pineapple cake and second part aims to identify key predictors for the end of shelf-life.

**Methods:** Microbial, chemical analysis and sensory evaluation were used. Sensory attributes that show change over time was applied to data obtained from a trained panel that evaluated seven sensory attributes of pineapple cake stored at 25°C, 37°C, 50°C between a three to six month time period. Survival analysis based on consumer’s acceptance or rejection of samples stored for different times and at different temperatures.

**Results:** Preliminary results indicated pineapple cake stored at 25°C at 63 days and 37°C at 28 days had reached cut-off point that identifies the end of shelf life of packed pineapple cake. Furthermore, compared to other predictors, acid value would be a good predictor like sensory attributes for the prediction of shelf life for bakery products.

**Significance:** The findings these predictors for the end of shelf life may assist product managers to evaluate other types of bakery products.
Emily E. Jackson
Nottingham Trent University
School of Science and Technology
Nottingham, UK

Emily E. Jackson is a native of Indianapolis, Indiana. She received her Bachelor of Science degree in Genetic Biology and Microbiology from Purdue University and her Master of Science degree in Bacteriology from the University of Wisconsin – Madison. After completing her Master’s degree, Emily worked as an ORISE fellow with the U.S. Food and Drug Administration (FDA). While there, she researched bacterial pathogens in raw milk and quality indicating organisms in leafy green wash water. During this time, Emily was also a member of the Proficiency Testing and Method Validation group and prepared artificially-inoculated food samples for analysis in laboratories across the U.S. This work earned her an FDA Honor Award for Development and Multi-laboratory Validation of a Rapid Molecular Method for Detection of Viruses in Foods.

In 2013, Emily moved to Nottingham, UK and began her Ph.D. with Professor Steve Forsythe at Nottingham Trent University. She is currently working on the characterization of Cronobacter species, with a particular focus on DNA sequence-based methods. Her published works include a re-evaluation of cultural, biochemical and molecular detection methods and the identification of a new species, Siccibacter colletis.

Emily is extremely grateful to have received the 2015 IAFP European Symposium Student Travel Scholarship to attend the 2015 European Symposium on Food Safety in Cardiff, Wales. She is excited to network with other researchers and is looking forward to hearing about food safety research from around the world.
Exhibitors
IAFP’s European Symposium on Food Safety
20–22 April 2015
Cardiff, Wales

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Marble Foyer
First Floor

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2 BioControl Systems
3 Food Standards Agency
4/5 Welsh Government
6/7 Kemin
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9 ELISA Systems
10 R-Biopharm AG
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3M Food Safety, a leading supplier of innovative solutions for the food and beverage testing and hygiene monitoring markets, would like to take this opportunity to welcome you to stand No. 19 at IAFP. Recently introduced to the market is the 3M™ Petrifilm™ Rapid Aerobic Count Plate, the next generation of its trusted, accurate and easy test that can detect aerobic bacteria counts starting in just 24 hours. Our extensive product range also includes the market leading 3M™ Clean-Trace™ ATP System, 3M™ Petrifilm™ Count Plates range as well as the 3M™ Molecular Detection System.

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ELISA Systems offers high quality kits to the food industry and our food allergen test kits are used extensively both locally as well as internationally. We have a wide range of both traditional ELISA based kits, which are developed and produced in Australia, as well as our new rapid detection devices. Our new devices are designed to be very easy to use with fast results. Our team, are technical experts in the field of allergen testing, participating in both local and international technical special interests groups. We look forward to working with you for all of your allergen testing needs.

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The Food Safety Centre, Norwich encompasses the world-leading expertise available within the Institute of Food Research (IFR) and on the Norwich Research Park in the areas of Food Microbiology and Food Security. Expertise includes:
- Bacterial Foodborne Pathogens including *Clostridium botulinum*, *Campylobacter* and *Salmonella*
- Predictive Microbiology and QMRA
- Microbial Spoilage Investigations
- Food Authentication

The Food Safety Centre is led by Professor Mike Peck (Director) and Elizabeth Saggers (Deputy Director)
For more information please contact: info.foodsafety@ifr.ac.uk  Web site: http://foodsafety.ifr.ac.uk/

The Food Standards Agency is an independent government department responsible for food safety and hygiene. We work with businesses to help them produce safe food, and with local authorities to enforce food safety regulations. We use science and evidence to tackle the challenges of today and address emerging risks of the future. We aim to ensure that food produced or sold in the UK is safe and what it says it is, that consumers have the information needed to make informed choices about where and what they eat and that regulation and enforcement is risk-based and focused on improving public health.

The Global Food Safety Initiative (GFSI) is an industry-driven initiative providing thought leadership and guidance on food safety management systems necessary for safety and efficiency along the supply chain. This work is accomplished through collaboration between the world’s leading food safety experts from retail, manufacturing and food service companies, as well as international organisations, governments, academia and service providers to the global food industry. They meet together at technical working group and stakeholder meetings, conferences and regional events to share knowledge and promote a harmonized approach to managing food safety across the industry. GFSI is facilitated by the Consumer Goods Forum (CGF), a global, parity-based industry network, driven by its members. www.mygfsi.com.
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*International Food Hygiene* is the only magazine of its type and it addresses food safety issues for its global audience of technical and production managers in the foods and drinks sector. For those in the meat sector we produce *International Meat Topics* as a partnering title to *International Food Hygiene*.
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www.rapidmicrobiology.com is a well-established on-line resource for food microbiologists worldwide, we have been providing information on microbiology products and services for over ten years. Keep up-to-date with the latest microbiology news by signing up for our free weekly newsletter which includes updates on products, services, webinars, training courses and conferences.

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The largest international publisher of scientific books, Springer is co-publisher with IAFP of both the revised 6th edition of Procedures to Investigate Foodborne Illness and the Food Microbiology and Food Safety book series. Stop by booth 24 to meet the Food Science editor, Susan Safren, and discover an authoritative range of books and our journal program in food science. All IAFP members now receive a 25% discount on our books.
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ZERO2FIVE Food Industry Centre
Cardiff Metropolitan University
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Cardiff, CF52YB, Wales
Phone: 02920-416306
www.zero2five.org.uk

Created to support food businesses, the Food Industry Centre has both a technical and operational capacity. Access to our facilities, equipment and resources enables companies to undertake previously impossible R&D projects to help them achieve their goals and drive their businesses forward. The Centre draws on expertise within Cardiff Metropolitan University, which includes Food Science and Nutrition and Dietetics, as well as Environmental Health, Trading Standards and Biomedical Sciences. Our team of experts is available to assist in a variety of food disciplines including baking, confectionery, packaging design, technical management systems and new product development.

Llywodraeth Cymru / Welsh Government
Cathays Park
Cardiff, CF10 3NQ, Wales
Phone: 03000 6 03000
www.gov.wales/foodanddrinkwales

The Food and Drink industry in Wales is reported to account for at least 1 in 5 of the employment in Wales with in excess of 23,200 businesses and 165,000 jobs from farm to fork in 2013 and hence is a significant industry for the economy in Wales. In 2014 the Welsh Government launched and published ‘Towards Sustainable Growth: an Action Plan for the Food and Drink Industry 2014–2020.’ The document details an ambition to grow the Welsh food and drink sectors by 30% to an industry turnover of £7 billion by 2020.

The Action Plan aims to encourage integration on the priorities for action, such as tackling poverty, managing our natural resources and encouraging a sustainable, productive and profitable industry. Other priorities include delivering against the targeted areas of:

- Education, Training, Skills and Innovation;
- Business growth and market development; and
- Food safety and Food security.

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