EFSA Journal

SUPPLEMENT

Shaping the Future of Food Safety, Together

Proceedings of the 2nd EFSA Scientific Conference Milan, Italy, 14–16 October 2015

EXPO MILANO 2015
FEEDING THE PLANET
ENERGY FOR LIFE

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European Food Safety Authority

Shaping the Future of Food Safety, Together

Milan, Italy, 14-16 October 2015

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EFSA JOURNAL: SUPPLEMENT

Foreword | **EFSA**

Dear delegates,

It gives me great pleasure to welcome you to the second EFSA scientific conference, organised in collaboration with our colleagues in EXPO 2015 to whom we are very grateful for their invaluable cooperation in hosting this event. We have availed of the proximity of EXPO 2015 in Milan to reach out to the scientific community and stakeholders in this inspiring international setting so that we can keep abreast of the developments in risk assessment and ensure that we stand ready to address the challenges and opportunities that lie ahead.

The sheer innovation and diversity of exhibitions at EXPO 2015 bring home to us the complexity of the modern food chain and the need for ever closer international cooperation in ensuring that the food on the plates of our citizens is safe, nutritious and sustainably produced. Experience has taught us that many future threats to the food chain will have a transnational dimension and we are very pleased that we can bring together some of the finest scientific minds from across the globe over the three days of the conference. Together, they will help us to keep abreast of the very latest scientific thinking alongside the full range of views on food safety, from decision makers tasked with protecting consumers, to producers, civil society groups and interested citizens, among others.

Although EFSA's mandate is scientific, the social context of our work is very important as it impacts on our effectiveness as an organisation in building trust in the food chain. Many of you will already be aware that we are committed to a process of intensifying our engagement with stakeholders and opening our processes and data in order to support transparency and reproducibility in risk assessment. These themes are reflected in our conference programme alongside the state-of-the-art in the scientific fields we cover as part of our broad mandate. As we build our strategy to 2020, we will make effective use of the inputs received from this event to inform the future direction of travel of the organisation.

Our conference programme has been deliberately designed to address new and emerging issues and of course to develop the next generation of risk assessors. Our Young Researcher Initiative has enabled emerging scientists from across Europe and beyond to communicate their work and to develop their expertise and networks. The poster session, abstracts of which are available in this supplement of the EFSA Journal, greatly enhance the conference programme and I would encourage you to attend the poster presentations and enable these exciting young scientists to showcase their work.

Finally, I would like to thank you for contributing to this important event in the EFSA calendar and I trust you will find the conference engaging and professionally rewarding.

Bernhard Url
Executive Director

Conference programme

14 OCTOBER 2015

14.00 – 14.30 Opening ceremony

14.30 – 18.00 Plenary session

What does the future hold for assessment science?

18.00 – 19.00 Poster session

15 OCTOBER 2015

09.00 – 13.00 Parallel sessions

Open Risk Assessment: data

Weighing evidence and assessing uncertainty

Expertise for the future

Nutrition challenges ahead

14.30 – 18.00 Plenary session

Science, innovation & society

18.00 – 19.00 Poster session

16 OCTOBER 2015

09.00 – 13.00 Parallel sessions

Open Risk Assessment: methods and expertise

Novel chemical hazard characterisation approaches

Microbiological risk assessment

Drivers for emerging issues in animal and plant health

Advancing environmental risk assessment

14.00 – 15.00 Final plenary session

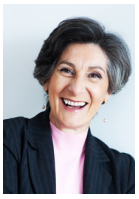
15.00 – 15.30 Concluding remarks

Plenary session: What does the future hold for assessment science?

The objective of the plenary session is to reflect on general developments that affect the conduct of assessment science work. These themes provide a basis for further discussion in the subsequent breakout sessions. One of them is the role of scientific advice for policy development in the context of societal decision-making. The 2014 Annual Report of the UK Government Chief Scientific Adviser (Walport and Craig, 2014) highlighted three generic issues in regulation: there is a danger that economic regulation will drive out innovation and reduce resilience; there are asymmetric incentives to many regulators, i.e. a regulator who stops something from happening that would have caused benefit will likely suffer no consequences; and societies are increasingly risk averse. The conference offers the opportunity for a deeper reflection on what the societal information needs are to reach decisions and how scientific advice can most usefully contribute to this. Around 2003, when EFSA became operational, several papers were already calling for a reflection on the relations between decision-makers, experts and citizens. Novotny et al. (2001) called for contextualisation as the key to producing science for public ends. They argued that science that draws strength from its socially-detached position is too frail to meet the pressures placed upon it. They imagined forms of knowledge that would gain robustness from their very embeddedness in society. Jasanoff (2003) argued that the credibility of regulatory science ultimately rests upon factors that have more to do with accountability in terms of democratic politics than with quality of science as assessed by scientific peers. More than 10 years on, there is merit in reflecting how much progress has been made in the world of assessment science to address these challenges and what lessons can be learned from the experience gained thus far.

CHAIRS

Angeles Rodríguez Peña — *Spanish National Research Council, Spain*



With a PhD on Molecular Endocrinology, Ángeles Rodríguez-Peña is senior researcher at the Spanish National Research Council. She has published more that 80 papers in peer-review journals, with 32 invited conferences. She has been Head of the Cell Signaling Department at the Biomedicine Institute of Madrid (1997–2002) and President of the Spanish Glial Network (1997–2002). She is a member of the Academia Europaea. From 2002 onwards she has been involved with different levels of responsibility in the European Research Policy and International Cooperation. She was appointed deputy head of the International Cooperation Unit of the CSIC responsible for implementing bilateral and multilateral relations of this organization (2005–2007). After that she was Deputy Director General for International Relations under the Ministry of Education and Science until 2008, when the Ministry of Science and Innovation was created and she became Deputy Director of European Affairs. She has been the Spanish delegate to the Strategic Forum of International Cooperation, SFIC (2009-2010), head of the Spanish delegation in the Scientific and Technical Research Committee of the Council of the European Union (CREST) and as a member of the newly named European Research Area Committee (ERAC). In 2010 she was elected President of the COST intergovernmental programme for European cooperation in Science and Technology (COST), responsibility that she makes compatible with her role as special Advisor to Deputy Director General of International Cooperation and Europe under the Secretary of State for Research and Innovation at the Spanish Ministry of Economy and Competitiveness. She combines her experience as a scientist with the design and implementation of R&I policies.

Bernhard Url — *European Food Safety Authority (EFSA), Italy*

Bernhard Url was appointed Executive Director of EFSA in June 2014, having served as Acting Executive Director for seven months. Dr Url joined EFSA in June 2012 as Head of the Risk Assessment and Scientific Assistance Department. A qualified veterinarian by training, he brings high-level management experience from food safety organisations to his role at EFSA. Prior to joining the Authority, Dr Url was Managing Director of the Austrian Agency for Health and Food Safety (AGES), which represents Austria on EFSA's Advisory Forum. From 2008 to March 2012, he also served as a member of EFSA's Management Board. During his 10 years at AGES, he was in charge of technical and scientific affairs with a remit that included the timely delivery of risk assessment and risk management services across a wide range of areas. This included ensuring effective risk communications during urgent food safety-related events. Prior to AGES, Dr Url spent five years as an Assistant Professor at the Institute of Milk Hygiene and Milk Technology at the University of Veterinary Medicine in Vienna before running a food quality control laboratory from 1993 to 2002. Dr Url graduated from the University of Veterinary Medicine in Vienna in 1987 and became a Doctor of Veterinary Medicine in 1990. He has published in the field of veterinary medicine with a particular focus on listeria and milk hygiene.

RAPPORTEURS

Hubert Deluyker — *European Food Safety Authority (EFSA), Italy*

Hubert Deluyker is EFSA's Scientific Adviser, providing the Executive Director with advice on issues related to EFSA's scientific activities and working in close cooperation with the Authority's scientific directors. Dr Deluyker joined EFSA in 2004. He established and was acting Head of EFSA's Assessment Methodology Unit prior to developing and becoming the Director of EFSA's former Scientific Co-operation and Assistance Directorate from 2007 to May 2011. He was Director of the Risk Assessment and Scientific

Assistance Directorate from May to October 2011, when he was appointed Director of Science Strategy and Coordination, a post he held until 16 May 2013. Before working for EFSA he was a clinical research scientist in the field of animal health from 1989 to 2004 for Pfizer Belgium, where he led a range of multidisciplinary and multinational research and development projects. He was also Associate Professor in Epidemiology from 1991 to 2000 at the School of Veterinary Medicine of the University of Ghent, Belgium, where he established the School's teaching and research programme on epidemiology. He previously worked as District Veterinary Officer for the Belgian Ministry of Agriculture. He holds a PhD in Epidemiology and a Masters in Preventive Veterinary Medicine from the School of Veterinary Medicine at the University of California, Davis. He also holds a Masters in Administration from the UC Davis School of Management and a BA in Business Administration from St Aloysius College, Brussels.

Jose Tarazona — *European Food Safety Authority (EFSA), Italy*

Jose V. Tarazona is the Head of the Pesticides Unit at EFSA. Doctor in Veterinary Medicine with a PhD in Toxicology, he started as Associated Professor of Toxicology and the University of Madrid, and from 1982 to 2009 worked at the Spanish National Institute for Agriculture and Food Research and Technology (INIA), serving as Head of the Division of Environmental Toxicology and Director of the Department of the Environment. His main field of expertise is ecotoxicology and environmental risk assessment He has participated in over 30 research projects in this area at national and European levels, and is co-author of over 250 scientific papers related with (eco)toxicology and risk assessment. Involved in the scientific advisory board of the European Union since 1992, he was a member of the CSTE, second vice-chair of the Scientific Committee on Toxicity, Ecotoxicity and the Environment (CSTEE), chair of the Working Group on Environmental Risks at the Task Force for the Harmonization of Risk Assessment Procedures of the Scientific Steering Committee, and vice-chair of the Scientific Committee on Health and Environmental Risks (SCHER). He has been external expert-consultant for different European bodies including the European Commission, EFSA, EMEA and EEA, as well as for the OECD, WHO and UN, chairing the OECD Expert Group on Chronic Aquatic Hazards and the OECD and UN Expert Groups on Terrestrial Hazards within the GHS strategy, and member of the UNEP POPs Review Committee. From 2009 to 2013, he worked at the European Chemicals Agency (ECHA) as Chair of the Committee for Risk Assessment and Scientific Chair of the Evaluation Directorate.

SPEAKERS

The science of assessment and the assessment of science: new frontiers in food safety evaluation

Sheila Jasanoff — *The John F. Kennedy School of Government, Harvard University, USA*

Sheila Jasanoff is Pforzheimer Professor of Science and Technology Studies at the Harvard Kennedy School. A pioneer in her field, she has authored more than 100 articles and chapters and is author or editor of a dozen books, including Controlling Chemicals, The Fifth Branch, Science at the Bar, and Designs on Nature. Her work explores the role of science and technology in the law, politics, and policy of modern democracies, with particular attention to the nature of public reason. She was founding chair of the STS Department at Cornell University and has held numerous distinguished visiting appointments in the US, Europe, and Japan. Jasanoff served on the Board of Directors of the American Association for the Advancement of Science and as President of the Society for Social Studies of Science. Her grants and awards include a 2010 Guggenheim Fellowship and an Ehrenkreuz from the Government of Austria. She holds AB, JD, and PhD degrees from Harvard, and an honorary doctorate from the University of Twente.

The safety of our food and drink is one of humankind's most ancient concerns. It is a branch of public health policy, a pillar of governance that some trace back 3500 years to the biblical book of Leviticus. Modern states began taking responsibility for food safety by the middle of the 19th century. The United States enacted its first federal food and drugs act in 1906, predating by more than 30 years the establishment, in 1938, of today's Food and Drug Administration. Early concerns, however, had more to do with intentional misleading of consumers, through the misbranding or adulteration of foods and drinks. While those concerns have not slipped off the radar screens of national and international food safety regulation, they have been augmented by more urgent concerns about the unintentional harms that humans, thickly settled on an increasingly interconnected and cultivated planet, may inflict upon themselves. Are our foods loaded with too many toxins, in the form of chemicals or traits engineered in through biological ingenuity? Does food carry undetected infectious agents? Are the supply chains that lead from field to table too long and convoluted to be controlled by existing regulatory authorities? Do we have the capacity to trace back to original causes when alerts are triggered by foodborne illnesses and, in unfortunate cases, the deaths of innocents?

Answers to all of these questions call for specialized knowledge and expertise, requiring new science and new modes of assessment. Yet, the problems of linking knowledge to policy have also grown more complicated in ways that render assessment more difficult and less credible. Fact-finding and decision-making authority is dispersed across countries and cultures. Time, particularly unforgiving in times of crisis, hardly ever allows for fully considered judgment. Media reports often heighten the public sense of urgency, while the very accumulation of data and its increasing accessibility undermine trust in formerly unassailable expert bodies. The stakes are high, as assessment failures can increase health risks for consumers and economic risks for producers and sellers.

What can today's science-based approach to food safety assessment take away from historical experience? To what extent are local and national experiences germane to an era of transnational commerce and translocal decision-making? This paper sketches a framework for reflection, recognizing that there can be no definitive answers to questions such as these. Another way to put the point is that, in 'assessment science,' process may matter more than specific end results.

Drawing on experiences from decision-making under normal as well as crisis conditions, ranging from the 'mad cow' case to more recent events such as the 2011 European e-coli outbreak, it is possible to articulate four general propositions that should underpin the assessment of food safety. The talk will explore how these propositions might affect the design of assessment processes.

1. Framing matters. In policy advice, the answer to a technical question can be only as good and relevant as the question itself. Therefore, it is important to attend to the processes by which questions come before bodies such as EFSA. Opportunities for reframing are especially important.

2. 'Assessment science' is impure science. Facts, values, and judgments are always intermingled in assessing science for safety. What constitutes a 'fact' is the outcome of a decision-making process. A different process might result in a different determination of the facts.

3. Consensus is cultural. National decision-making systems have evolved distinct cultural approaches to producing credible knowledge. International assessment bodies should be aware of such cross-cultural variation, as well as the risk of producing their own closed and opaque cultures of consensus-building that do not respect national 'civic epistemologies.'

4. Trust is distributed. In today's complex food supply systems, centralized expertise is no longer sufficient to ensure public trust. States and suppliers (among others) share responsibility for generating expert knowledge, as well as for producing and maintaining public trust. Communication among these actors on such issues as the adequacy of evidence and the standards of precaution is essential to effective assessment.

From risk regulation to innovation democracy

Andrew Stirling — *The Sussex Energy Group, University of Sussex, UK*

Andrew Stirling is Professor of Science and Technology Policy and a former Research Director for SPRU at the University of Sussex. Among other projects, he co-directs the ESRC STEPS Centre, Sustainable Lifestyles Research Group and is on the leadership team of the ESRC Nexus Network. Among many other research projects, he's served on advisory bodies for the EU on Energy Policy, Science in Society, Collaborative Research, Sustainability and Science Governance and the UK government

on toxic substances, GM Crops, public engagement and science advice – as well as for the Royal Society, Nuffield Council, UN IHDP, European Environment Agency, Greenpeace International, Global Energy Observatory, Demos and the Green Alliance. He is a member of editorial boards for several academic journals and of the Research Committee of the ESRC.

Current debates over management of technological risks are often highly polarised. Nowhere is this more true than in the food and agriculture sector. Climate change, environmental pressures, rising populations, changing diets and globalising competition and concentration make the stakes high and growing. Few are more exposed to the implications, than the European Food Safety Authority (EFSA).

In these debates, many assert an apparent tension between science and innovation on the one hand and democracy and public values on the other. New forms of precautionary or participatory governance are held to compromise the rightful authority of objective expertise and rigour. Moves in these directions are often dismissed as capitulation to 'anti-technology' lobbyists or irrational public anxieties. Or they are interpreted more cynically as signs of special pleading or 'political correctness'.

This paper will argue that it is exactly these kinds of misleading assertion that actually drive the problematic polarisations they purport to contest. They suppress open political debate and so obstruct socially effective innovation. In the process, they undermine not only democracy but also science.

The reason is, that key issues in political choices in food and agriculture are not just about scientific questions of safety, but wider directions for innovation. Emerging technologies do not simply unfold a supposedly inevitable 'one track' 'race to the future', in which the only questions for decisions concern how to implement particular paths. In reality, both research and innovation are instead branching evolutionary processes. A radical variety of alternative possible trajectories typically exists, all equally engaged in science. Supposedly singular GM technologies, for instance, are highly diverse. And there are many very effective pathways beyond this for alternative social and technological innovations. Key questions are thus not just about 'how fast?', or 'what risk?', but 'which way?', 'who says?' and 'why?'

It is these wider questions that are systematically suppressed by polarising scientistic risk discourse in food and agriculture. Governance in this field is ill-equipped (and often ill-disposed) to address them. And uncertainties and ambiguities also lead supposedly 'sound scientific' risk analysis typically to be incapable of delivering single uniquely prescriptive recommendations concerning the most socially effective innovation trajectories. The highest quality evidence always remains absolutely necessary. But the only truly `evidence based' conclusion in food and agriculture, as elsewhere, is that the available evidence is only rarely sufficient definitively to justify a single policy. Instead of polarising 'science based', 'pro-innovation' rhetorics, then, precaution and participation more rigorously and accountably address ever-present uncertainties and ambiguities around public interests and values.

So why do such manifestly polarising misrepresentations persist? Realistic consideration of regulatory and innovation processes makes this easy to explain. For powerful but beleaguered commercial interests in the food and agriculture sector, stakes are high and competition intense. Those innovation trajectories that are most strongly favoured privately, are those offering narrow forms of comparative advantage (like intellectual property, appropriation of value or control of supply chains). In seeking to promote these aims, temptations are strong to invoke the more general authority of science. So it is understandable that such partisan arguments are made, obscuring the wider social and political choices presented by alternative innovation trajectories. What is less easy to justify, is where ostensibly neutral regulatory bodies find themselves perpetuating the same kinds of polarising error.

To remedy this, what is required are more robust ways to illuminate and interrogate these imprints of power in food and agriculture innovation and regulation. Based on a systematic analytical framework, this paper will highlight a series of concrete methods and processes to this end – offering practical ways to enable precaution and participation in order to resist polarising closure. By enhancing the depth, scope, transparency and accountability of policy, tensions disappear between scientific rigour and democratic legitimacy. It is by opening up a broader, deeper political space for this more mature form of 'evidence bound' (not evidence based) critical debate, that society as a whole faces the best hopes of realising the most effective directions for research and innovation in this challenging field.

Identification, prioritization and conduct of applied research and analyses impacting policy development: lessons learned from the US National Toxicology Program (NTP)

Nigel Walker — *National Institute for Environmental Health Sciences/NIH, USA*

Nigel Walker is the Deputy Director for Science for the Division of the National Toxicology Program at the National Institute of Environmental Health Sciences (NIEHS), one of National Institutes of Health (NIH). As Deputy Director of DNTP he is involved in the formulation, coordination and implementation of activities necessary to carry out the scientific goals of the National Toxicology Program (NTP), a US Federal government interagency program whose mission is to evaluate agents of public health concern by developing and applying tools of modern toxicology and molecular biology. He received his BSc in Biochemistry from the University of Bath, UK and his PhD in Biochemistry from the University of Liverpool, UK. Dr Walker's professional experience covers environmental molecular toxicology, quantitative dose response modelling and risk analysis, with particular emphasis on understanding the toxicity and carcinogenicity of mixtures, dioxins and other persistent organic pollutants, botanical dietary supplements and nanoscale materials. Dr Walker has over 120 peer-reviewed publications, including authorship on 37 NTP chronic toxicology and carcinogenicity study technical reports.

The US National Toxicology Program (NTP) is an interagency, government, research program, that was established in 1978 as a cooperative effort to coordinate toxicology testing programs within the federal government, strengthen the science base in toxicology, develop and validate improved testing methods, and provide information about potentially toxic chemicals to health regulatory and research agencies, scientific and medical communities, and the public. The need for a program like the NTP arose because of increasing scientific, regulatory, and Congressional concerns about the human health effects of chemical agents in our environment.

Over the past three decades the NTP has become a key partner in designing, conducting, and interpreting various types of assays for toxicity. Through its activities, the NTP provides, directly or indirectly, basic scientific data that other federal and state scientific and regulatory agencies, as well as private sector organizations, use in responding to issues relevant to the effects of chemical and physical agents on human health and the environment. The NTP seeks and accepts nominations of agents for studies that enhance the predictive ability of future studies, address mechanisms of toxicity, or fill significant gaps in the knowledge of the toxicity of chemicals or classes of chemicals. Selected substances are evaluated for a variety of health-related effects, among them general toxicity, reproductive and developmental toxicity, genotoxicity, immunotoxicity, neurotoxicity, metabolism, disposition, and carcinogenicity.

The NTP also plays a key role in the development and validation of alternative test methods for regulatory safety testing that will reduce, refine (by causing less pain and distress), or replace animal use – the 3Rs of alternatives. In support of this, ten years ago the NTP embarked on a new direction with the release of the NTP Vision for the 21st Century; to support the evolution of toxicology from a predominantly observational science at the level of disease-specific models to a predominantly predictive science focused upon a broad inclusion of target-specific, mechanism-based, biological observations. In the past decade NTPs involved in these activities and as part of US federal 'Tox 21' program has led to the quantitative high through in vitro screening of thousands of compounds, broadly characterizing and defining the chemical-biological space occupied by chemicals of toxicological concern. These data are being been utilized in the early prioritization of the toxicological assessment of classes of similar chemical agents, in providing biological basis of sufficient similarity of chemical mixtures, and in development of integrated pathways of biological response that can be used to replace certain traditional in vivo studies.

Over the past 5 years NTP has also been partnering with the USFDA to develop novel ways in which the expertise of both academic scientists and regulatory scientists can be more closely integrated to address an issue of public health concern. This proof-of-concept collaboration, known as the Consortium Linking Academic and Regulatory Insights on Toxicity of Bisphenol-A (CLARITY-BPA), involves the close integration of federally funded academic researchers with regulatory scientists at FDA conducting a traditional guideline compliant chronic perinatal toxicity and carcinogenicity study on the effects of BPA in rodents. As such it represents an unprecedented level of interaction among federal government scientists and NIEHS-funded university-based researchers for filling knowledge gaps, informing chemical risk assessment, and evaluating new methods or endpoints that may be used for regulatory hazard assessments.

In addition to its focus on identification of potentially hazardous substances through research and testing, the NTP carries out 'assessment activities' to identify substances that may cause cancer or non-cancer health effects through evaluations of the published literature. More specifically as part of this, the recent development and application of systematic review methodologies to environmental health, is starting to transform our strategic approaches to the objective integrated assessment of multiple data streams (human, mammalian, vertebrate, invertebrate, in vitro), to provide insights into the potential health risk posed by agents in our food and environment.

Scientific support for effective policy development: putting it in practice

Mark Walport — *Government Office for Science, UK*

Sir Mark Walport is the Chief Scientific Adviser to HM Government and Head of the Government Office for Science. Previously, Sir Mark was Director of the Wellcome Trust, which is a global charitable foundation dedicated to achieving extraordinary improvements in human and animal health by supporting the brightest minds. Before joining the Trust he was Professor of Medicine and Head of the Division of Medicine at Imperial College London. He is Co-Chair of the Prime Minister's Council for Science and Technology and has been a member of this since 2004. He has also been a member of the India UK CEO Forum, the UK India Round Table and the advisory board of Infrastructure UK and a non-executive member of the Office for Strategic Coordination of Health Research. He is a member of a number of international advisory bodies. He has undertaken independent reviews for the UK Government on the use and sharing of personal information in the public and private sectors: 'Data Sharing Review' (2009); and secondary education: 'Science and Mathematics: Secondary Education for the 21st Century' (2010). He received a knighthood in the 2009 New Year Honours List for services to medical research and was elected as Fellow of The Royal Society in 2011.

This presentation draws on key themes arising out of the 2014 Annual Report of the UK Government Chief Scientific Adviser, Sir Mark Walport, 'Innovation: Managing Risk, Not Avoiding It' (available at: https://www.gov.uk/government/publications/innovation-managing-risk-not-avoiding-it).The report distils leading edge EU scientific thinking to stimulate broader discussion on risk, hazard, uncertainty and vulnerability within the EU; and promotes a regulatory culture surrounding risk in which robust scientific evidence is openly considered alongside political and other non-scientific issues in shaping policy. The presentation also references work being done by the Government Office for Science into public attitudes on the future of food, and on the resilience of increasingly complex and interdependent food supply chains.

The EU economy is critically dependent on growth driven by science and innovation. Furthermore we badly need innovation in the UK, in Europe and around the world to deal with the increasingly complex challenges posed by growing and ageing populations, scarce resources, infectious diseases and the need to reduce carbon emissions. However, innovation is often held back by poorly framed discussions about risk. This has particular resonance for regulators responsible for protecting citizens and especially in the context of food, where discussions about risk are often laden with wider societal and cultural values and expectations.

Risk is a central theme to the work of the UK Government Chief Scientific Adviser; for every particular challenge the degree of risk and the level of certainty varies greatly and the more uncertain the risk the greater the challenge to scientists to provide the scientific context. Innovation in food production is essential to feed our growing global population in the face of climate change. Informed, independent scientific advice is a critical component to help to ensure effective policy development.

Innovation drives productivity and growth.
Innovations based on technology and science are essential to help us meet challenges such as ageing populations, scarce resources and infectious diseases.

The global population is forecast to be over 9 billion by 2050, leading to an increasing demand for food and placing further pressure on finite resources. We need to innovate so that we can produce more food with fewer resources.

Regulation needs to support innovation not hold it back.
Innovation inevitably entails risk as well as benefit. But if we fail to manage risk proportionately we can miss out on major potential benefits, or suffer needlessly.

Innovations such as precision agriculture, lab-grown meat and insects as animal-feed present significant opportunities to increase and develop more sustainable food production methods. Research shows that high-tech solutions to food sustainability need to be supported by trusted organisations such as regulators in order to find public acceptability.

We need to reframe the debate about risk.
In any public debate science is not the only lens being used, but scientific evidence and rigour are essential contributors to the debate. Discussions should be founded around specific possible uses of a technology, their respective alternatives, and the costs of inaction as well as action.

Public attitudes to food are often mediated through multiple lenses, of which cultural values, cost and safety are frequently foremost, with new scientific and technological techniques perceived to be 'unnatural' and viewed with suspicion.

Scientists have a role in reframing the debate to demonstrate the global costs of inaction and providing well-evidenced and impartial advice on the risk of specific technologies.

Key areas where we can build on existing approaches are:

- Investment: Aligning national priorities for investment on resilience, infrastructure and innovation with an evidence and risk-based approach;
- Coordination: Ensuring a more coherent and structured approach to assessing impact of risk in policy, regulation and crisis management;
- Regulators: Putting in place the right governance structures and incentives in relation to our regulators and regulated industries;
- Science-based EU: Rooting the approach to policy and decision-making in the EU in robust scientific evidence.

The scientific community should contribute to public engagement and increased public awareness of the issues; so that when policy is enacted there is a wider understanding of the science underpinning policy decisions.

Open Risk Assessment: data

Since its foundation, EFSA and Member States have made significant progress in the area of data collection for risk assessment and monitoring. In partnership with competent authorities and research organisations in Member States, EFSA has become a central hub for European data on food consumption and occurrence of food-borne hazards. Beyond EFSA's use of these data, they remain largely unexploited. In addition, for some of its risk assessments, EFSA also relies on published information as well as on scientific studies sponsored and submitted by industry. The environment in which the Authority operates has evolved significantly since its foundation. The growth of digital technology has granted scientists and consumers alike faster and more efficient access to data and information. The 'open data' movement, which has entered the sphere of EU institutions, is unleashing the potential for re-use of data. In parallel, the work of EFSA is increasingly subject to demands for more openness and transparency across its spectrum of stakeholders. EFSA is developing a data roadmap which outlines its future ambitions, focusing on open data, interoperability and data quality. This session will include a discussion on the opportunities and challenges associated with open data, data interoperability and data quality by sharing experiences in various sectors within and outside EFSA's remit.

CHAIRS

Ana Canals — *Spanish Agency for Consumer Affairs, Food Safety and Nutrition (AECOSAN), Spain*

Ana Canals holds a BSc in Biology and a PhD in Epidemiology by the University Complutense of Madrid. She has worked over 23 years as a scientists and has over 70 research publications in peer-reviewed journals in the fields of immunology, molecular biology and animal health. This research activity was performed at the National Institute for Food and Agricultural Research (INIA) in Spain and as a Research Associate in the Agriculture Research Service (ARS) from the United States Department of Agriculture in USA. During the last decade she has worked in management activities, as Research Technical Director at the Research Centre for Animal Health (CISA, INIA), and as a Deputy Director within the Ministry of Public Administrations working in the coordination, planning and management of a number of processes connected with human resources and budget management in the area of border control inspections. Since 2008 she works as a Technical advisor in the Spanish Agency of Consumer Affairs, Food safety and nutrition (AECOSAN), where she is responsible for the coordination of EFSA activities in Spain and international relations of the Agency.

Angelika Tritscher — *World Health Organization (WHO), Switzerland*

Food safety expert with strong scientific background and public health perspective. Over 20 years of international work experience, from basic training in food science, food technology and food law to practical experience in quality and safety assurance in food production; from cancer research to applied toxicology and human health risk assessment. Long-standing involvement in food safety risk assessment as basis for the development of international evidence-based food standards as developed by the Codex Alimentarius Commission. MSc in food science and a PhD in biochemical toxicology, certified toxicologist and member in several national and international professional societies (EUROTOX; SOT, IUTOX). Working at the WHO in the Department of Food Safety and Zoonoses and responsible for the unit on Risk Assessment and Management: providing strategic and programmatic leadership in developing the science-base for international (Codex) food safety standards, and providing risk management support in the negotiations. Responsibilities also include the support of capacity-building programs for WHO Member States in the prevention, early detection and management of food safety incidents through the International Food Safety Authority Network (INFOSAN).

RAPPORTEURS

Fabrizio Abbinante — *European Food Safety Authority (EFSA), Italy*

Fabrizio Abbinante is presently Program Manager of the Information Management Program at EFSA. He has been Data Manager and IT Business Analyst for both EFSA (from 2008 to 2014) and for the European Medicines Agency (from 2004 to 2008). He has managed several projects in the area of data warehousing, data collection, data harmonization, web reporting, interoperability, data standards, controlled terminology, and information management in general. From 2004 to 2007, he was member of the ICH M5 and ICH E2B(R) Expert Working Groups for the definition of an international standard for the exchange of medicinal product and safety report data between the three ICH regions (EU-JP-US) and Canada. In 2006, he became member of the ISO TC215/WG6. From 2002 to 2004, he worked for Bassilichi S.p.A, first as software developer, then as IT business analyst in the 'EudraVigilance project' at the European Medicines Agency (EMA), which included the design of a European central database for medicinal products. He graduated in 2002 as Software Engineer at the University of Pisa, in Italy.

Mary Gilsenan — *European Food Safety Authority (EFSA), Italy*

Mary Gilsenan is Head of the Evidence Management (DATA) unit at EFSA. The unit is responsible for the collection, collation, management and analysis of data on food consumption, and food-borne chemical and microbiological hazards, as well as dietary exposure assessments to underpin several of EFSA's risk assessments. The unit operates in close collaboration with a network of data providers in EU Member States. Prior to joining EFSA in 2012, Mary worked at Leatherhead Food Research in the UK where she managed its Global Regulatory Services department. She held previous nutrition research roles at Unilever global R&D in the Netherlands, Unilever UK, as well as Trinity College Dublin. Mary holds a BSc in Human Nutrition from the University of Ulster, Northern Ireland and a PhD on the application of probabilistic modelling to food chemical exposure assessments from Trinity College Dublin.

Eileen O'Dea — *Food Safety Authority, Ireland*

Eileen O'Dea has been working in data management for over 25 years, in the food safety sector for 13 years and previously in international data management for the pharmaceutical and medical sector. Eileen was chair of the EFSA working group who developed the EFSA data standards for laboratory analysis of food and feed samples and their electronic transmission (Standard Sample Description and Guidance on Data Exchange, both published in 2010) and the EFSA working group who revised these standards to cover additional data domains (SSD2 2013 and GDE2 2014). She has also contributed to many EFSA and EU working groups and colloquia including Data Warehousing and Web Reporting, Food Classification and Description, Zoonosis Reporting and Food Safety Statistics. She has managed many projects over her career focussed on capturing data to support the reporting needs of diverse customers and business needs, strategy definition, development of national & international data standards, data quality measurement and improvement, data standardisation and interoperability, master data management and controlled terminology, and information governance. Eileen graduated in Food Science with Management Studies in 1989 from King's College, University of London, and gained her Master of Business Studies (Information Systems) from Smurfit Graduate School of Business, University College Dublin in 2010, and her Postgraduate Diploma in Cloud Strategy in 2013 from University College Cork/Irish Management Institute.

KEYNOTE SPEAKER

European Commission's Open Science Initiative: co-creating added value with data

Jean-Claude Burgelman — *European Commission, Belgium*

Jean-Claude Burgelman is presently Head of Unit Science Policy, Foresight and Data in DG RTD. He joined the European Commission in 1999 as a Visiting Scientist in the Joint Research Centre (the Institute of Prospective Technological Studies – IPTS), where he became Head of the Information Society Unit in 2005. In January 2008, he moved to the Bureau of European Policy Advisers (attached to the president of the EC) as adviser for innovation policy. On 1/10/2008 he joined DG RTD as advisor and then Head of Unit in charge of top level advisory boards like the European Research and Innovation Area Board, the Innovation for Growth Group and the European Forum for Forward Looking Activities. Till 2000 he was full professor of communication technology policy at the Free University of Brussels, as well as director of the Centre for Studies on Media, Information and Telecommunication, and was involved in science and technology assessment. He has been a visiting professor at the University of Antwerp, the European College of Bruges and the University of South Africa and sits on editorial boards of several academic journals. He chaired the World Economic Forum's Global Agenda Council on Innovation and was a member of its Science Advisory Committee.

- *What is open science*
- *What do European stakeholders agree on*
- *The role of data*
- *Towards an open science policy agenda*

SPEAKERS

The European Commission's open data strategy and the EU Open Data Portal

Yvo Volman — *Publications Office of the European Union, Luxembourg*

Yvo Volman (1965) is head of the 'Common Portal and Open Data Portal' unit in the Publications Office of the European Union. Yvo studied at the Universities of Amsterdam and Strasbourg and holds a PhD in European law awarded by the European University Institute in Florence. He worked for the Dutch Ministry of Economic Affairs in the areas of industrial and technology policy, before joining the European Commission in 1998. In the Commission, he dealt with legislative and strategic issues as well as funding programmes related to the information market, digitisation and data. In 2013 he joined the Publications Office of the European Union.

In recent years, the rise of open government data has been facilitated by several major trends: IT-based government modernisation, the move towards 'big data', and the presence of more and more open content on the Web. It is now widely recognised that open data can lead to innovative products and services in a range of economic sectors. In addition, open data contributes to evidence-based policy making, and is of course an essential feature of any modern transparency policy. With the endorsement of G8 leaders in June 2013, and of the European Council in October 2013, open data has reached a stage of maturity and is explicitly pursued by many countries across the world. The European Commission has been promoting a policy on open data for many years, in view of its enormous economic and social potential. The Directive on the re-use of public sector information, adopted in 2003 and strengthened in 2013, is a cornerstone of this policy. It sets favourable conditions for the re-use of information produced by or for the public sector and ensures a level playing field across the EU, thus facilitating the creation of cross-border information products and services. The European Commission practises what it preaches and applies a very open policy to its own information resources. The directorates-general of the Commission have an obligation to make their datasets available to the public (for free and with a very limited set of conditions) and to bring them gradually into the EU Open Data Portal. This portal was launched at the end of 2012 and gives access to datasets from a range of EU institutions and agencies. Combining these datasets with other data financed by the public purse, for example datasets resulting from publicly funded research projects, can create a powerful tool at the service of the economy, research and society. Moreover a pan-European data portal is under development that will make it possible to find and combine the data from the EU institutions with data available at national and regional levels.

Data visualizations: drawing actionable insights from science and technology data

Katy Börner — *Indiana University, USA*

Katy Börner is the Victor H. Yngve Professor of Information Science in the Department of Information and Library Science, School of Informatics and Computing, Adjunct Professor at the Department of Statistics in the College of Arts and Sciences, Core Faculty of Cognitive Science, Member of the Leadership Team of the IU Network Science Institute, Leader of the Information Visualization Lab, and Founding Director of the Cyberinfrastructure for Network Science Center (http://cns.iu.edu) at Indiana University in Bloomington, IN and Visiting Professor at the Royal Netherlands Academy of Arts

and Sciences (KNAW) in The Netherlands. She is a curator of the international Places & Spaces: Mapping Science exhibit (http://scimaps.org). She holds a MS in Electrical Engineering from the University of Technology in Leipzig, 1991 and a PhD in Computer Science from the University of Kaiserslautern, 1997. She became an American Association for the Advancement of Science (AAAS) Fellow in 2012.

In an age of information overload, the ability to make sense of vast amounts of data and to render insightful visualizations is as important as the ability to read and write. This talk explains and exemplifies the power of visualizations not only to help locate us in physical space but also to help us understand the extent and structure of our collective knowledge, to identify bursts of activity, pathways of ideas, and borders that beg to be crossed. It introduces a theoretical visualization framework meant to empower anyone to systematically render data into insights together with tools that support temporal, geospatial, topical, and network analyses and visualizations. Materials from the Information Visualization MOOC (http://ivmooc.cns.iu.edu) and maps from the Places & Spaces: Mapping Science exhibit (http://scimaps.org) will be used to illustrate key concepts and to inspire participants to visualize their very own data.

Data interoperability and linked data technologies

Dave Weller — *Thomson Reuters, UK*

Dave Weller is the chief enterprise architect for Thomson Reuters, a position he has held since the beginning of 2012. He is responsible for defining the future state architecture and technology platforms needed to build Thomson Reuters products and support business operations. Additional responsibilities include building partnerships with technology companies to maximise investment and joint opportunities, identifying emerging technologies, and fostering internal innovation. He has worked for Thomson Reuters for more than 20 years in a variety of development and technical positions, including interim CIO and CTO of Factiva, a joint venture with Dow Jones. Dave is married with two children and in his spare time runs a small record label.

Identifiers are fundamentally important in being able to form connections between data, which puts them at the heart of how we create value from data to make it meaningful. Simply put, identifiers are labels used to refer to an object being discussed or exchanged, such as products, ingredients, or people. The foundation of the web is formed by connections that hold pieces of information together. Identifiers are the anchors that facilitate those links. The lack of identifiers, or the poor use of them, stifles the power of information gained from linking multiple datasets together. Some of these shortcomings might be overcome using intelligent search and fuzzy matching, but the lower precision of these techniques means that the data never reaches its full potential and there is little incentive to drive improvement of precision over time. Identifiers are crucial to the process of sharing information. Managing identifiers is easier in a closed system. The web has many advantages, but it presents challenges for identifiers because of its vast scale and their ad hoc usage. Communicating identity — the understanding of what is being described — is essential in conveying the accurate meaning of shared information. This is especially true if the information is shared in machine-readable form, without human intervention. The scale and ad hoc use of the web means that those who produce and consume identifiers cannot easily coordinate an agreement on the representation and meaning of identity. Much of this coordination relies on the ability and inclination of consumers to look up the definition, usage, validity and equivalence of identifiers. There is no clearly established method for ensuring the communication of identity precisely and at an equitable cost to all. This paper draws lessons from illustrative examples and proposes some guiding principles, both for those creating and managing identifier schemes and those who are using them, and examines why and how information identity must evolve from being an inherent part of dataset design to being a distinct discipline in its own right.

Opening clinical trial data

Hans-Georg Eichler — *European Medicines Agency (EMA), UK*

Hans-Georg Eichler, M.D., M.Sc., is the Senior Medical Officer at the European Medicines Agency in London, United Kingdom, where he is responsible for coordinating activities between the Agency's scientific committees and giving advice on scientific and public health issues. Prior to joining the European Medicines Agency, Dr. Eichler was at the Medical University of Vienna in Austria for 15 years. He was vice-rector for Research and International Relations since 2003, and professor and chair of the Department of Clinical Pharmacology since 1992. His other previous positions include president of the Vienna School of Clinical Research and co-chair of the Committee on Reimbursement of Drugs of the Austrian Social Security Association. His industry experience includes time spent at Ciba-Geigy Research Labs, U.K., and Outcomes Research at Merck & Co., in New Jersey. In 2011, Dr. Eichler was the Robert E. Wilhelm fellow at the Massachusetts Institute of Technology's Center for International Studies, participating in a joint research project under the MIT's NEWDIGS initiative. Dr. Eichler graduated with an M.D. from Vienna University Medical School and a Master of Science degree in Toxicology from the University of Surrey in Guildford, U.K. He trained in internal medicine and clinical pharmacology at the Vienna University Hospital as well as at Stanford University.

Abstract not provided

Data collection by public bodies: joint EFSA–Member State activities

Leif Busk — *National Food Agency, Sweden*

Leif Busk obtained his basic training as a Toxicologist at Uppsala University and the Karolinska Institute in Stockholm. He has forty years of experience in regulatory toxicology, both from the pharmaceutical and food sector and has been the Swedish representative in the Advisory Forum of EFSA since the very beginning. He is presently working as a Scientific Adviser at the Swedish National Food Agency.

Stefano Cappe — *European Food Safety Authority (EFSA), Italy*

Currently responsible for the team 'Evidence and Data management' in the Evidence unit in EFSA. The team manages the data collection of chemical contaminants, additives, pesticide residues, veterinary drug residues, zoonoses, antimicrobial resistance, molecular typing data. The collected data provide the basis of many of EFSA's opinions and reports in the risk assessment area. Project manager of the EFSA scientific data warehouse, the tools that enable access on the web of the collected data according to rules agreed with Member States and the EC Commission. Employed since 2007 in EFSA, contributed to the creation of harmonised standards for the data reporting 'Standard Sample Description' and of the data transfer 'Guidance on data exchange'. From 2001 to 2007, employed in EMA (European Medicine Agency) in the development of the pharmacovigilance system 'EudraVigilance'. In the same period, still in the area of drug safety, worked in the implementation of the related data warehouse to provide access and analytical functionalities to the collected data.

Collecting data on occurrence of contaminants and nutrients and gathering information on food consumption is a main task for both Member States (MS) agencies and EFSA. Such data collection forms the basis for Community regulations of different types. Some data collections are mandatory and some are more or less voluntary. Example of the former is pesticide residue data while non-regulated contaminants is a representative of the latter. Implications for food safety of this, based on the willingness of MS to provide data will be discussed. EFSA has, over the years, developed schemes and platforms for data

collection to be used by MS. This will in the long run result in more efficient data collection on a European level. In addition these platforms will inspire MS to develop more efficient data collection systems that will facilitate also work with national risk. Storing national data in a Community reposit maintained by EFSA provides additional advantages, especially for small and medium seize MS. Of special interest could be the possibility, via the Micro Strategy, to use up to date systems for exposure and intake calculations developed by EFSA. There is commitment, both from EFSA and MS, to improve data collection and transfer. However, there are also always discussions in MS on how to balance priority between more strict national work and the support to EFSA, and in the long run, support to the European project. A broader engagement in these discussions, involving e.g. more risk managers and politicians, would facilitate the future development of even more efficient data collection systems to support risk assessment of foods on the European market.

Metro's Global Standard Traceability Solution

Britta Gallus — *Metro Group, Germany*

Britta Gallus has been working for METRO AG since July 1st, 2011 as Head of Regulatory Affairs. Amongst other things, she is responsible for current issues in the fields of Quality, Safety, Health and Environment as well as the representation of METRO GROUP in European and international committees. More over, in her role as intermediate between Metro AG and the Sales Divisions, she assures fulfillment of specifically legal but also company requirements. From 2002 until 2011, Britta Gallus chaired the HDE (Handelsverband Deutschland e.V.) office in Brussels as Managing Director of the HDE whilst, at the same time, being Managing Director of the HDE Food Law and Quality Assurance Committee. Her responsibilities included representing interests of retail in Brussels, European committee work and intensive monitoring of the European legal activities on the issues of food, environmental and consumer law. From 1997 until 2002 Mrs. Gallus worked as adviser at 'Bundesverband der Filialbetriebe und Selbstbedienungs-Warenhäuser (BFS)'.

Traceability – a cloud for oceans and more. It started with one product in a few stores. Now we are running a successful international traceability program – the Global Standard Traceability Solution (GSTS) – in several countries for several articles.

The aim is to establish an open, seamless traceability with regard to the careful use of our resources and the responsible procurement of products. High level of transparency in the value chain is not only vitally necessary and increasingly demanded by consumers – it is definitely also possible.

The GSTS approach covers costumer information demands, helps acting responsibly and strengthens our Food Safety initiatives. As one of the largest fish retailers in Europe, METRO GROUP is an important player in the global values chain. Every year we sell about 200,000 tonnes of fish in our stores worldwide. Protecting fish stocks is part of our social responsibility and we feel that it is our duty to find answers to global challenges. And fish is just the starting point.

The presentation will highlight the WHAT, WHY and HOW of the solution and invite the audience to be part of the solution!

Weighing evidence and assessing uncertainty

Methodologies for integrating (weighing) evidence and assessing uncertainties are of utmost importance to ensure that scientific assessments are transparent, robust and fit for purpose to support decision makers. One of the key challenges remains the development of harmonised methodologies for both the integration of scientific evidence and the assessment of uncertainties in food safety. Such challenges arise mainly because of the multi-disciplinary nature of the topics, which include microbiology, animal health and welfare, epidemiology, toxicology, ecology, plant health, bioinformatics and statistics. In addition, the beginning of the 21st century as the 'post genomic era' has seen the emergence of methods and tools such as omics, systems biology and computational tools (i.e. in silico tools). These new methods generate a vast amount of data and evidence (Big Data) which scientists are struggling to integrate into the current risk assessment paradigm. This break-out session aims to discuss the state of the art and future challenges in relation to weighing evidence and assessing uncertainties in critical areas of food safety, including chemical risk assessment, biological and environmental risk assessment and validation of animal-free risk assessment methods. Finally, presentation of international developments in uncertainty analysis for risk assessment and risk management will lead to a general discussion and conclusion of the session.

CHAIRS

Prabhat Agarwal — *European Commission, DG CONNECT, Belgium*

Prabhat Agarwal is currently Head of Sector for Evidence-based Policy-Making at DG CONNECT in the European Commission. His current work is focusing on the potential of digital technologies to transform the policy-making process. Prabhat studied Physics in Cambridge and Paris before working for seven years in industrial R&D in the area of exploratory semiconductor and nanotechnologies for a variety of applications. His other interests include Open Policy-Making, the relation between science, technology and society, and the history of science.

Derek Knight — *European Chemicals Agency (ECHA), Finland*

Derek J Knight, who is British, has worked at the European Chemicals Agency (ECHA) since September 2008. As the Senior Scientific Advisor, he is responsible for providing the Executive Director and the Director of Regulatory Affairs with expert scientific and technical advice on matters relating to chemical regulation, with the focus on the EU REACH, CLP and Biocides Regulations and the operations of ECHA. Previously he headed a team of regulatory affairs professionals at a UK contract research organisation for almost 18 years, covering a wide range of regulatory schemes worldwide. He has also registered medicinal products and worked as a Technical Support Chemist. He has a broad understanding of the regulation of chemicals and is especially interested in approaches to hazard and risk assessment using non-standard data. He is an external expert member of the Scientific Expert Panel of the SEURAT-1 research initiative 'Towards the replacement of in vitro repeated dose systemic toxicity testing'. He is a Fellow of the Royal Society of Chemistry and Chartered Chemist, a Chartered Scientist and a Fellow of the Organisation of Professionals in Regulatory Affairs. His doctoral studies at the University of Oxford in the UK were in organosulphur chemistry.

RAPPORTEURS

Bernard Bottex — *European Food Safety Authority (EFSA), Italy*

French agronomist specialised in plant protection products. Since 2007, Scientific officer in EFSA, coordinating the working groups of the Scientific Committee. Participation to the development of various methodological guidance documents for risk assessment, e.g. on biological relevance, or on how to characterise and communicate uncertainty or biological relevance. Coordinator of the EFSA Scientific Network on the harmonisation of risk assessment methodologies.

Jean Lou Dorne — *European Food Safety Authority (EFSA), Italy*

Jean Lou Dorne has been working as a Senior scientific officer at EFSA since 2006 to first support the panel on contaminants in the food chain in risk assessment of contaminants in the food and feed chain. In 2011, he moved to the Scientific Committee and Emerging Risks unit to develop EFSA's Chemical hazards database, summarising toxicological data used in all EFSA's chemical risk assessments, to support the identification of emerging chemical risks and to support EFSA's Scientific Committee and international scientific

advisory bodies in the development of horizontal methods and guidance documents for risk assessment. His current focus includes human and ecological risk assessment of multiple chemicals ('chemical mixtures'), use of modern methods in chemical risk assessment (omics, toxicokinetic models, in vitro and in silico methods, integrated testing strategies) and the use of the weight of evidence approach in scientific assessments. He holds an MSc in Toxicology from the University of Surrey (1995, UK), an Msc in molecular biology from the Natural History Museum in Paris (1997, Paris) and a PhD in toxicology and risk assessment from the University of Southampton (2001, UK) on the integration of human variability in toxicokinetics for chemical risk assessment (young scientist award (EUROTOX, 2002); young investigator's award (British toxicology society, 2009)). He has published over 60 peer reviewed publications, 5 book chapters and has contributed to more than 150 EFSA scientific outputs.

KEYNOTE SPEAKER

Weighing evidence and assessing uncertainties: where have we been, where are we going?

Lorenz Rhomberg — *Gradient, USA*

Rhomberg is an expert in quantitative risk assessment, including dose-response analysis, pharmacokinetic modeling, and probabilistic methods, with special experience in chlorinated solvents and endocrine-active agents. He is the author of books and of more than 50 articles on these topics. His practice includes work in support of environmental litigation as well as work relating to a variety of regulatory programs including CERCLA, FIFRA, TSCA, and REACH, among others. Before joining Gradient, Dr Rhomberg was on the faculty of the Harvard School of Public Health and was employed by US EPA. Dr Rhomberg is active in professional groups and environmental policy development, focusing on current issues in the interpretation of toxicological data in human health risk assessment through service on panels sponsored by government, industry, and such organizations as the National Academy of Sciences and the International Life Sciences Institute. Dr Rhomberg was recognized as the Outstanding Practitioner of the Year by the Society for Risk Analysis in 2009.

In this talk, I characterize the challenges in the process of weighing scientific evidence about toxicity, outline the needs of a regulatory process to do so by following a set of established procedures, review some of the processes that have been used and the questions about their adequacy that have been raised, and compare different strategies for structuring weigh-of-evidence inquiry. Finally, I propose some approaches that may achieve the twin aims of flexibility in the face of diverse scientific evidence and sufficient structure to ensure that consistency, rigor, and justification of conclusions can be documented.

When bringing to bear scientific evidence to support decisions about potential health effects of chemical exposures in foods, in the environment, and in the workplace, there are inevitable limits to what the available data can demonstrate directly. Scientific judgments about the existence and nature of causal processes of toxicity need to be made while contending with all the data gaps, extrapolations, inconsistencies, and shortcomings among the available studies. There is a need to characterize not only what conclusions can reasonably be drawn, but also the degree of confidence in them, noting different interpretations that might also be considered. In pure science, an iterative process of hypothesizing general explanations and seeking critical tests of them in further experiments is pursued, with continued skepticism toward and testing of tentative conclusions being the 'scientific method'. In the regulatory context, decisions to take (or to forgo) actions must be made, and the judgments about whether the interpretation of evidence is sufficiently robust to support such decisions is delegated to a limited set of assessors who must make judgments and defend their legitimacy to interested stakeholders and the public. To ensure consistency in standards of evidence to support conclusions, and to communicate the judgment process and its justifications, a variety of risk assessment frameworks – procedures for gathering, interpreting, and drawing conclusions from available evidence – have been put in place and used by various governmental and international organizations.

In recent years, the sufficiency of some of these evidence-evaluation frameworks, and their ability to make sound, well justified, and well communicated judgments, has been questioned. This stems in part from deeper understanding of underlying modes of toxic action (and their diversity and differences among different experimental animal strains and humans, and at different exposure levels), exposing the limits of earlier assumptions about toxicity processes being parallel in test systems and in humans. In part, it is due to an increasing number of examples in which existing evaluation frameworks seem to miss important scientific considerations that have been revealed by deeper probing of underlying biology. New kinds of testing data, in particular, high-throughput in vitro testing and gene-expression arrays, have opened new avenues for characterizing toxicity pathways (and not just traditional apical endpoints) and pose challenges to traditional methods.

Critiques by high-level review panels of several key regulatory assessments have found insufficient explanation of the basis for weight-of-evidence judgments. The advent of evidence-based medicine as a means for evaluating clinical efficacy of alternative treatments has provided a model for how a more systematic and rigorous process might provide better and more objective justifications for judgments. In consequence, a great deal of recent attention has focused on how the weight-of-evidence process can and should be reformed, and I review some of the activity that has been undertaken by regulatory and scientific bodies at the national and international level.

Progress has been made on instituting systematic review processes for identifying relevant studies, objectively evaluating strengths and weaknesses, making inclusion/exclusion decisions, and tabulating results. I review some of these and, while noting the benefits, also argue that this by itself goes only so far in resolving the challenges, since the relevance of studies, how interpretations of them interact, and how they do or do not support overarching hypotheses about the basis for possible toxicity still need to be considered, and a process to do so systematically is challenging to define.

Insights into the challenges and means to address them can be gained by examining the differing strategies that have been employed in constructing evaluation frameworks. I summarize results from a recent review and workshop doing this. One strategy, a rules-based or algorithmic approach, aims to build a decision-tree process that embodies the interpretive wisdom of the field, such that each decision can be made objectively, and conclusions are justified by how the decision-tree process disposes of the data at hand. The advantage is objectivity, but the shortcoming is that the interpretive wisdom needs to be built into the algorithm, which may be faulty, become out of date, or be unable to accommodate novel kinds of evidence. An alternative strategy is to be more unstructured but to rely on expert judgment from a set of appropriately chosen scientists who then explain the basis for their judgments. The advantage is flexibility and, possibly, extra scientific insight, but the shortcoming is that the choice of experts becomes controversial, the justifications are articulated after the fact and are keyed to judgments already made, and the process can lack transparency. The conclusions are justified by asserting the expertise of the judges. A process analogous to evidence-based medicine can be rigorous and transparent, but it does not easily deal with evidence that is not direct observation of the question of interest itself – that is, it emphasizes consistency of repeated observations, but does not handle inference across datasets very well.

I end by presenting my own approach of Hypothesis-Based Weight of Evidence, which seeks to gain the advantages of others while avoiding the disadvantages. It stresses constructing competing sets of tentative explanations for all of the relevant study outcomes, where explanations invoking a common causal toxicity process can be compared for plausibility and dependence on assumptions to an alternative set of possible explanations that denies the tested agent's toxicity and explains outcomes by alternative means, such as chance, confounding, and variable operation of non-agent-related causes in different test systems.

SPEAKERS

Weighing evidence of biological relevance: from empirical testing in rats to 21st century mode of action analysis

Harvey Clewell — *The Hamner Institute for Health Sciences, USA*

Harvey Clewell is the Director of the Center for Human Health Assessment at the Hamner Institutes for Health Sciences. Before joining The Hamner, Dr Clewell served as a consultant in risk assessment, first as a Research Manager with ICF and then as a Principal with ENVIRON. Prior to this he served for 20 years as an officer in the US Air Force Biomedical Science Corps, performing research in environmental fate and transport, toxicology, and management of hazardous materials. He received his Masters in Chemistry from Washington University, St. Louis, and his PhD in Toxicology from Utrecht University, NL. Dr Clewell is a leading expert on the use of tissue dosimetry and mode-of-action information in chemical safety and risk assessment. He is currently a visiting scientist at Utrecht University and a member of the ECVAM Scientific Advisory Committee. He has co-edited a text on 'Physiologically Based Pharmacokinetic Modeling: Science and Applications', and has published more than 200 peer-reviewed journal articles. His research focuses on the development of methods for in vitro to in vivo extrapolation of cell-based assays, pharmacokinetic modeling of early life exposures, and characterization of human pharmacokinetic and pharmacodynamic variability.

In the early days of chemical risk assessment, the focus was essentially qualitative: describing pathological changes observed after the exposure of laboratory animals to relatively high doses of a chemical. Despite the accumulation of a large volume of animal data, there has been a growing skepticism regarding its usefulness, due to the perceived difficulties of interpreting the relevance of the animal results for humans. A concept that has proved useful for weighing evidence on the toxicity of a chemical is the 'mode of action'. The International Programme on Chemical Safety (IPCS) (Sonich-Mullin et al. 2001) guidelines provide a discussion of the desired elements of a mode of action and a description of the kinds of data that can inform its development, using a conceptual framework for mode-of-action evaluation.

The IPCS mode-of-action evaluation framework is an extension of the criteria of causation originally presented by Bradford Hill to aid in the interpretation of epidemiological data (Hill, 1965), to include the evaluation of experimental animal data. Weighing the evidence for the likely human relevance of an animal outcome is particularly problematic and has frequently been a source of controversy. In order to promote transparent, harmonized approaches for such evaluations the IPCS extended its mode-of-action framework to address consideration of human relevance for both cancer and noncancer effects observed in animal studies (Boobis et al., 2006, 2008).

In 2007, the U.S. National Research Council report on 'Toxicity Testing in the 21st Century: A Vision and a Strategy' argued for a transformative shift away from in vivo animal toxicity testing and towards the use of mechanistic in vitro assays, typically using human cells in a high-throughput context. The shift to in vitro tests for assessing risks of chemicals entails new questions about weighing evidence: (1) how will we define adversity from in vitro tests; (2) how will the in vitro test results be used to predict expected outcomes in animals and people; and (3) how will regulatory agencies set exposure standards for human populations based on in vitro test results. These questions squarely pose the challenges that need attention in order to develop a 21st century toxicology for both collecting toxicity testing information and weighing evidence for purposes of human health risk assessment.

Weighing evidence and assessing uncertainty in microbiological risk assessment: approaches for preparing appropriate scientific support for decision making in complex questions?

Matthias Greiner — *Federal Institute for Risk Assessment (BfR) and University of Veterinary Medicine Hannover, Germany*

Greiner graduated in Veterinary Medicine in 1986 and holds an MSc degree in Applied Statistics. He started his career as head of serological laboratories at the Department of International Animal Health (previously Tropical Veterinary Medicine and Epidemiology) and lecturer in veterinary epidemiology at the Freie Universität Berlin, Germany. He is Diplomat and served as President of the European College of Veterinary Public Health (ECVPH). From April 2002 to December 2005, he held a position as research professor and head of the Animal Health Section and the International EpiLab at the Danish Institute for Food and Veterinary Research in Copenhagen. During his leadership, the EpiLab has been accredited as OIE Collaborative Centre for Research and Training in Population Animal Health Diagnosis and Surveillance Systems and has gained international recognition for its research work related to this mandate. In January 2006, Dr Greiner joined the Federal Institute for Risk Assessment (BfR) in Berlin, Germany. His scientific interest is exposure and risk assessment, diagnostic test validation and epidemiology in relation to food safety and veterinary public health. From 2006 to 2009, Dr Greiner contributed to several working groups of the European Food Safety Authority (EFSA) and was a member of the Animal Health and Animal Welfare Panel (2006-2009). Since August 2011, Dr Greiner is Head of Department for Exposure at the BfR in Berlin, and Professor for Exposure Assessment and Quantitative Risk Assessment at the University of Veterinary Medicine Hannover, Foundation, Germany.

The complexity of questions in food safety is addressed in formal exposure or risk assessments by deconstructing the problem into elements at different levels, typically involving a risk question, a scenario, a model, model parameters and data to support parameter estimation. Using examples mainly from the microbiological area it will be illustrated that specific uncertainties at those mentioned levels need to be assessed in order to derive an understanding of the overall uncertainty of the science-based risk assessment. On the other hand, the concept of 'evidence' is mainly applicable to support the choice of a scenario and to characterise the empirical knowledge about key model parameters.

Formal statistical integration of parameter estimates from multiply primary studies is possible using meta-analysis. This approach is well established and provides tools for diagnosing of and accounting for heterogeneity, which is highly relevant in situations of primary studies with conflicting results. Meta-analyses require that the individual parameter estimates are comparable in their metric and relate to the same type of observation (e.g. estimates of a treatment effect in humans). In contrast, evidence synthesis and evidence integration aim at collating and combining, respectively, information from primary studies with different types of observations (e.g. in vivo, in vitro, in silico, epidemiological), different study organisms (e.g. human and animal), which are also referred to as lines or streams of evidence. The two methodologies weighing of evidence and uncertainty assessment are complementary and have a common overall goal, which is to provide the best possible basis for science-based decision making.

Uncertainty, variability and weight of evidence: how well do we know environmental risks?

Glenn Suter — *US Environmental Protection Agency, USA*

Glenn Suter is Science Advisor in the US Environmental Protection Agency's National Center for Environmental Assessment, and Chairman of the Risk Assessment Forum's Ecological Oversight Committee. He has produced more than 200 publications including 3 authored books and 4 edited books over his 37-year career. He has received the Society for Environmental Toxicology and Chemistry's Founder's Award and the Association for Environmental

Health and Science's Career Achievement Award, and he is an elected fellow of the American Association for the Advancement of Science. He is Associate Editor for Ecological Risk of Human and Ecological Risk Assessment, and a Senior Editor of Integrated Environmental Assessment and Management. His interests include ecological epidemiology, ecological risk assessment, and the conceptual bases for environmental science and decision making.

Risk assessment is technical support for decision making under uncertainty, so without uncertainty there is no risk. However, what decision makers need from assessors is not uncertainty but relevant risk information and an expression of confidence in that information. Confidence in an assessment result has two components. The first is the conventional statistical issues of uncertainty and variability in the results which are both expressed as scatter of the data—less scatter, more confidence. Estimating and expressing scatter can be difficult but it is a well-studied problem and the conceptual issues are relatively well recognized (Bayesian versus Frequentist, statistical testing versus estimation, etc.).

The second component of confidence is weight. The weight of evidence metaphor is borrowed from jurisprudence—evidence that has more influence on a decision is weightier. Weight of evidence is qualitative and, although data analysis contributes, it is ultimately a matter of judgment. Constituents of weight of evidence are relevance, strength and reliability. Relevance is the degree to which evidence represents the issue and situation being assessed. Strength expresses the degree to which the evidence is distinguished from random variance, commonly expressed as correlation. Reliability expresses factors such as study quality and transparency of presentation that make evidence more convincing. Qualitative assessment results such as causation (e.g. carbofuran caused bird kills) or status (e.g. the stream is impaired) have weight but not scatter. However, quantitative information, such as a lethal threshold concentration or a cancer slope factor, should be accompanied by expressions of both scatter and weight. We must learn to convey the overall confidence in our results to decision makers, stakeholders, and the public. [The views expressed in this abstract are those of the authors and do not necessarily represent the views or policies of the US EPA.]

Coming to grips with unfamiliar uncertainties of a new predictive toxicology paradigm

Maurice Whelan — *European Commission, Joint Research Center, Italy*

Maurice Whelan is head of the Systems Toxicology Unit of the Institute for Health and Consumer Protection (IHCP) of the European Commission's Joint Research Centre (JRC), based in Ispra, Italy. He is also head of the EU Reference Laboratory for Alternatives to Animal Testing (EURL ECVAM), an integral part of the IHCP. The focus of his Unit/ECVAM is on the development, evaluation and promotion of new integrated non-animal approaches to the safety assessment of chemicals and nanomaterials. Whelan is co-chair of the OECD Advisory Group on Molecular Screening and Toxicogenomics that is responsible for the OECD programme on Adverse Outcome Pathways and he is a member of the Steering Committee of the European Partnership for Alternative Approaches to Animal Testing (EPAA). He holds an external appointment of visiting Professor of Bioengineering at the University of Liverpool (UK).

Key objectives underpinning future food safety policies will never be achievable without a clear paradigm-shift in the way we profile the toxicological properties of chemicals. Regulatory toxicity assessment relies for the most part on laboratory-animal tests developed many decades ago, the majority of which were never conceived to meet the demands of 21st century society. However toxicology science has advanced tremendously in recent years and now offers a new generation of biotech tools and computer models that can be used to predict toxicity rather than just observe it. Importantly too, predictive approaches reveal and exploit mechanistic understanding of why a chemical might be toxic to an organism under certain conditions, opening the door to more tailored, substance-specific approaches to hazard assessment. Embracing this new toxicity testing paradigm offers considerable benefits to society including; improved assessment of more chemicals, simplified cross-sector legislation based on harmonised assessment approaches, cheaper and faster testing for industry, higher levels of protection for sensitive populations, and incorporation of safety-by-design and green chemistry practice in product lifecycles. What exactly a new paradigm will look like and what implications it will have for both risk assessment and risk management are still unfolding but it is clear that the tipping point is behind us. Change will need to be managed at many levels, and understanding unfamiliar 'non-standard' sources of uncertainty will be an essential step in the process of responsibly yet definitively transitioning to new ways of describing and predicting toxicological hazard.

Assessing and communicating uncertainties for risk assessment and risk management: recent international developments

Andrew Hart — *Food and Environment Research Agency (Fera), UK*

Andrew Hart is at the UK Food and Environment Research Agency (Fera). His primary focus is on developing improved approaches for the analysis of evidence and uncertainty and applying them to different areas of health and environmental risk. He has served three terms as a member of the EFSA Pesticides Panel, and has served on EFSA Working Groups on a range of topics including uncertainty, transparency and probabilistic modelling. He provides training courses on uncertainty and expert elicitation for EFSA experts and staff and has helped several EFSA Working Groups to apply these approaches in specific assessments, e.g. for bisphenol A. He was also a member of the WHO working groups on uncertainty in exposure and hazard characterisation.

The need to address uncertainties in food safety risk assessment has long been recognised at international level. The Codex Working Principles for Risk Analysis, established in 2003, say uncertainties should be explicitly considered at each step in risk assessment, documented transparently and quantified to the extent that is scientifically achievable. EFSA's Guidance on Transparency, adopted in 2009, says the same. There has been gradual progress towards implementing these principles. Guidance on methods for addressing uncertainty in human exposure assessment was published by EFSA in 2006 and IPCS/WHO in 2008, while ECHA's 2008 guidance also included hazard, risk and environmental assessments. In 2014-15, IPCS/WHO published guidance on uncertainty in hazard characterisation, accompanied by a spreadsheet calculator, and EFSA published draft guidance on addressing uncertainty in all areas of its work.

The requirement to quantify is driven by the needs of decision-making and transparency. However, this applies to the overall uncertainty of the assessment outcome, and does not imply that every source of uncertainty must be quantified individually. Those that are not quantified individually must be quantified collectively when possible, generally by expert judgement. Qualitative methods have an important role to play in supporting these judgements. The draft EFSA guidance offers a framework that is scalable to the needs of each assessment, enabling the assessor to select from a menu of qualitative and quantitative methods while taking account of any limitations in time and resources, including emergency situations. It also recognises that progress is more feasible by evolution than revolution, focussing first on addressing uncertainties within current assessment paradigms. Revising those paradigms to address uncertainty more fully is challenging, as illustrated by the 2014 IPCS/WHO guidance on hazard characterisation, and will require concerted action over a longer period.

Implementation of new guidance will require training and support for risk assessors, including provision of user-friendly tools and specialist help with the more sophisticated methods. Risk managers will also need training and support in meeting their responsibility, emphasised by Codex, for resolving the impact of uncertainty on decision-making. In addition, risk assessors and managers will need to work together when communicating with stakeholders about how uncertainty has been addressed.

Expertise for the future

EFSA depends on a system of scientific panels, working groups and the expertise of its staff to perform its role in providing high-quality risk assessments on food. The centralisation of this work at EU level increases efficiency but may also bring challenges with regard to maintaining and developing expertise in the areas of food, feed, plant, animal and environmental health. Risk assessment of food requires a multi-disciplinary and inter-disciplinary approach: excellence in relevant fields of science is a prerequisite, but knowledge of the full risk analysis process, EU food law, consumer behaviour, international relations and skills in risk communication are also needed. To meet future challenges in an always changing environment, appropriate expertise needs to be identified and a model of specialised and continuous training is required. This session aims to discuss the state of the art and future of education in risk assessment. It will provide an overview of current developments in higher education and training on food safety risk assessment and regulatory science in the EU and worldwide, including general risk assessment, food safety risk assessment, and environmental risk assessment. It will also examine future training needs and developments in building capacity for risk assessment.

CHAIRS

Pier Sandro Cocconcelli — *Istituto di Microbiologia, Università Cattolica del Sacro Cuore, Italy*

Pier Sandro Cocconcelli is Chair Professor of Food Microbiology at the Università Cattolica del Sacro Cuore (UCSC). He is Rector's delegate for internationalization projects of the same university and Director of UCSC ExpoLAB, a multidisciplinary research centre of the UCSC, acting on the theme 'Feeding the Planet. Energy for Life' of EXPO Milan 2015. He is the president of CHEI, the Centre for Higher Education Internationalisation at UCSC, which promotes and conducts research, training, and policy analysis to strengthen the international dimensions of higher education. Currently he is member of the Scientific Committee of Expo 2015 – Comune di Milano. In 1987 he worked at the Institute of Food Research of Reading (UK) on the development of cloning vectors for lactic acid bacteria and on the expression of heterologous genes in Lactobacillus. In 1994 he held a visiting position as Assistant Professor at the BTPI of the University of Minnesota (USA) on the adhesion properties of gut bacteria, with particular attention to the virulence determinants of enterococci. Since 2003, he is a scientific expert at the European Authority of Food Safety (EFSA) as Panel and Working Group member working on the risk assessment of microorganism internationally introduced in the food chain. Since 1992 he has been continuously involved in national and EU-funded research projects on food bacteriology and microbial biotechnologies, and is currently coordinating FP7 EU research projects on microbiological safety and quality of traditional foods. His research activities are focused on food microbiology, bacterial molecular biology, bacterial genomics, risk analysis of food pathogenic bacteria, and on the gene exchange of antibiotic resistance and virulence determinants in the food chain. In particular, ongoing research activities are focused on: microbiological risk assessment in the food chain; antimicrobial resistance in the food chain: role of food bacterial communities in the spread of antibiotic resistance determinants; germination pathways of food associated pathogenic or spoilage clostridia (C. botulinum, C. tyrobutyricum and C. difficile)*; molecular biology and application of lactic acid bacteria; biofilm of pathogenic bacteria in the food environment; genomics of food starter bacteria; and environmental and soil microbiology.*

Dominique Gombert — *ANSES, France*

Following studies in engineering, Dominique Gombert, age 48, initially worked for the Ministry of the Environment in Paris on air quality management issues. He was then an associate expert on cross-border pollution and the repercussions of acid rain for the United Nations Economic Commission for Europe, before returning to the Ministry of the Environment to work on specific issues concerning technological risks. He then worked for seven years as deputy director of the AIRPARIF network for monitoring air quality in Ile-de-France. Since 2004, he has worked mainly in the field of health and safety and the risk assessment of major environmental issues in various national institutes and agencies, first as head of the Risk Assessment Department of the French Agency for Environmental and Occupational Health Safety, and then as the deputy director for chronic health risks at the French national institute for the industrial environment and risks (INERIS). Since 2010, he has been director of the Risk Assessment Department of the French Agency for Food, Environmental and Occupational Health & Safety.

RAPPORTEURS

Dimitra Kardassi — *European Food Safety Authority (EFSA), Italy*

Dimitra Kardassi is a Scientific Officer in EFSA's Pesticides Unit dealing with the coordination of peer review processes of individual active substances and MRL activities. She has more than 14 years of experience in the food safety area working in the Hellenic Food Authority (EFET) as scientific officer and head of unit dealing with monitoring programs on contaminants, food additives and other chemicals, last position as Director of Training and Communication Directorate. She was a member of the EFSA Advisory Forum Working Group on Communication (AFCWG), and of EFSA Working Group on Collection of Chemical Occurrence Data, contributing on the collection of Chemical Occurrence Data from the food control activities, special contribution to chemical contaminants data collection. She was also Chairperson of the Council working group for the Codex Committee on Food Additives (CCFA), and Chairperson of the Council of the EU at the CCFA in Hong-Kong, on behalf of Greek presidency, in the second half of 2014. She was member of several Commission and Council working groups, among others the European Commission Governmental Expert working groups on Food Additives, Flavourings and Enzymes and European Council expert committee on food improvement agents, headed the national delegation at the Codex Alimentarius Committee on Food Additives and Contaminants (CCFA and CCFC) (2006, 2008–2009). She represented Greece in the Standing Committee on plants, animals, food and feed, section Toxicological Safety. She has contributed to many European legislative acts in the area of food improvement agents. Dimitra was Training Coordinator and lecturer on several BTSF ('Better Training for Safer Food' initiative of DG SANCO and EAHC) and Project manager of several European-funded programmes in EFET.

KEYNOTE SPEAKER

Expertise for the future: harnessing the power of digital technologies

Gráinne Conole — *University of Leicester, Institute of Learning and Innovation, UK*

Gráinne Conole joined the University of Bath Spa on 1st February 2015 as Chair in Education. She was previously at University of Leicester, where she was professor of learning innovation and director of the Institute of Learning Innovation. Her research interests include: the use, integration and evaluation of Information and Communication Technologies and e-learning, research on Open Educational Resources (OER) and Massive Open Online Courses (MOOCs), new approaches to designing for learning, e-pedagogies, social media and the impact of technologies on organisational change. She regularly blogs on www.e4innovation.com and her Twitter ID is @gconole. She has successfully secured funding from the EU, HEFCE, ESRC, JISC and commercial sponsors). She was awarded an HEA National Teaching Fellowship in 2012. And is also a fellow of EDEN and ASCILITE. She has published and presented over 1000 conference proceedings, workshops and articles, including the use and evaluation of learning technologies. She has recently published a Springer book entitled 'Designing for learning in an open world' and is currently working on a Routledge book on practical Learning Design.

Today's digital technologies offer an unprecedented variety of ways in which to interact with rich multimedia, and to communicate and collaborate with others. Mobile technologies mean that learning anywhere, anytime is now a real possibility. New exciting technologies are on the horizon, such as augmented reality, seamless interfaces and Artificial Intelligence. Coupled with this today's working environment is increasing complex and changing; we are training people for a future in which they will do jobs that do not even exist today. Hence Continuing Professional Development is more important than ever, and its focus must be beyond knowledge recall to provide them with the necessary competences and digital literacies to enable them to develop critical thinking skills, problem solving and team work. The keynote speech will provide an overview of today's digital landscape and a taster of emergence technologies. It will focus on examples of good practice on the use of technologies to facilitate different pedagogical approaches, as well as considering how more open practices through the use of Open Educational Resources, Massive Open Online Courses and social media can ensure the sustainability of Continuing Professional Development.

SPEAKERS

Recent advances in food chemical risk assessment training and capacity building

Paul Brent — *World Bank Global Food Safety Partnership (GFSP), Developing training in Chemical Risk Assessment, Australia*

Paul Brent is currently a scientific consultant specialising in food and chemical risk assessment and risk management, and was until very recently Chief Scientist of Food Standards Australia New Zealand (FSANZ) where he had responsibility for the stewardship of the integrity and capability of FSANZ's scientific work. Dr Brent was also responsible for coordination of the international scientific work undertaken by FSANZ at Codex, OECD, FAO/WHO and APEC fora. He was the leader of the Genetically Modified Food Team and Section Head of the Product Safety Standards Section within FSANZ. He was Australian delegation leader to the UN/WHO Codex Committee on Food Additives and Contaminants for several years, most recently leading the Australian Delegation to the Codex Committee on Food Additives. Dr Brent was also a member of the Australian BSE Committee, Chair of the Australian Committee on Novel Foods, a member for six years (2009–2014) of the Expert Advisory Committee of the Hong Kong Centre for Food Safety, and is a current member of the WHO IHR Roster of Experts as an expert in food safety. Dr Brent is a member of the International Advisory Committee of the newly established China National Centre for Food Safety Risk Assessment (CFSA). He currently leads a project to develop an e-learning module on food chemical risk assessment for the World Bank Global Food Safety Partnership, and was recently appointed Chair of the Australian Advisory Committees on Medicine (ACMS) and Chemical Scheduling (ACCS). Dr Brent was also recently appointed as Principal Government Advisor to the Asia Pacific Economic Cooperation (APEC) Food Safety Cooperation Forum (FSCF) project on MRLs Harmonisation in the APEC region. Dr Brent obtained his Bachelor of Science at Newcastle University and doctorate in pharmacology at the University of Newcastle Medical School, Discipline of Clinical Pharmacology, prior to working in various roles at the Newcastle Medical School, including as a research scientist in basic and clinical pharmacology, neuroscience and neuropharmacology. Whilst at Newcastle Medical School, Dr Brent was the recipient of National Health and Medical Research Council research grants as Chief Investigator. Prior to his appointment with FSANZ, he worked as a senior toxicologist at the Therapeutic Goods Administration, Office of Chemical Safety, Australian Government Department of Health and Ageing.

There is an ongoing need to build capacity in food chemical risk assessment global training, especially amongst developing country food safety regulators. The specific challenge for any food chemical risk assessment training is to find a balance between basic training on principles and procedures, the application of those general issues and techniques to specific examples, and the need to enable the student to apply the general elements of the training to contemporary food safety issues. In the absence of any full and comprehensive food chemical risk assessment package, a project has been undertaken to respond to the documented capacity building priority needs for both the Asia-Pacific Economic Cooperation (APEC) Food Safety Cooperation Forum (FSCF) and the World Bank Global Food Safety Partnership (GFSP) in the area of risk analysis, and specifically, to develop a stand-alone capacity building module on food chemical risk assessment, with global applicability through the GFSP, via both face-to-face and e-learning mechanisms. The project proved quite challenging, however the working group, have developed an e-learning module consisting of 7 chapters each of which has as their core (backbone) a series of PowerPoint slides, ranging from 50 to 100 slides. The text and figures of each of the slides belonging to a certain chapter has been developed so that they are self-explanatory within the slide and from slide-to-slide, i.e. without any need of vocal explanation (simultaneous lecturing). The 7 chapters of slides (course core) are to different extents supplemented

with additional text notes (in Word), and some additional slides (branching off series) with case studies (further described real contemporary cases or constructed examples), and with self-explanatory student exercises. Although the module is designed to be stand-alone e-learning tool, the module or part thereof can be used in face-to-face activities (e.g. workshops) with colleagues, or other students studying the same module, and facilitate/strengthen programs that focus on relevant local food safety problems. The presentation will expand of details of the development and design of the module.

Food safety risk assessment capacity building: educational cooperation programme in Europe

Wolfgang Kneifel — *University of Natural Resources and Life Sciences, Austria*

Wolfgang Kneifel studied Food Science & Biotechnology (1980), Doctoral thesis (1983), Habilitation (1989), Assoc. Professor (1995), Full Professor for Food Quality Assurance (2004), Head of the Department of Food Science and Technology (since 2009), Head of Christian Doppler Research Laboratory for Bran Biorefinery (since 2012). Several memberships (e.g. Austrian Association of Food Scientists and Biotechnologists: President; GfM – Society of Milk Science, Kiel, Germany: past President; German Society of Mucosal Immunology and Microbiome, Stuttgart, Germany: foundation member; Austrian Society for Microbiology, Hygiene and Preventive Medicine: Board member; Scientific Board Member of H. Wilhelm Schaumann Foundation, Hamburg, Germany). Advisory board memberships (mainly dairy industry, feed industry). Teaching: coordinator of the international Master programme ´Safety in the Food Chain´ within the Euroleague of Life Science Universities, (2006–2014; www.safetyinthefoodchain.com); Quality Management (I, II), Applied Quality Management, Hygiene for Food Scientists, Food Safety and Risk Management (BOKU Vienna); General Microbiology and Hygiene for Pharmacists (University of Vienna, Faculty of Life Sciences, Dept. of Pharmaceutical Sciences), Food Safety and Risk Management (University of Hong Kong; School of Biological Sciences, visiting professorship since 2009). Expertise: Food safety, food hygiene, quality management of foods, valorisation of side products from food production. More than 200 publications, Section Editor (Food Microbiology) in FEMS Microbiology Letters.

Owing to a multitude of criteria and to ongoing regional as well as global developments, food safety has become a topic of high complexity and diversity. In this context, problems often are multi-faceted and factors such as outbreaks of food- and feed-borne diseases, local incidents, mass production, criminal fraud, but also changing trends in nutrition and consumer food habits, may play some important role. Importantly, several internationally linked control measures as well as surveillance and alert networks have been established based on official regulations aiming at the protection of markets from (potentially) contaminated, mislabelled or unhealthy food. Undoubtedly, there are several interfaces that need to be further cross-linked and harmonized. These targets can only be managed if experts possessing sound knowledge are involved. Hence, well-educated and trained food safety specialists (either employed by the food industry or by inter/national authorities) are needed to assess, to control and to monitor all relevant issues contributing to food (un)safety and quality. Ideally, such experts have to possess some high degree of interdisciplinary background as they usually have to deal with medical, toxicological, microbiological, chemical and technological issues and often need to assess the risks and to re-assemble fastidious puzzles encountered with outbreaks. The presentation will, on the one hand, illuminate the demands placed on the required experts and on the other hand introduce existing University curricula dealing with this subject. In addition, attractive training concepts are shown as contemporary examples for capacity building in this important area.

Training in epidemiology and microbiological risk assessment

Arnold Bosman — *European Centre for Disease Prevention and Control (ECDC), Sweden*

Public health specialist, focused on providing specialised training to public health professionals in the EU, related to disease prevention & control. Experience in field epidemiology, such as outbreak investigations at regional, national and international level, surveillance and field research. Goals are to establish an effective network of training in applied epidemiology, public health microbiology, outbreak management and cross-border health threats in order to contribute to capacity.

International Health Regulation IHR-2005 and Decision 1082/2013/EU require countries to be in control of the risk management cycle for serious cross-border health risks. This includes event detection, risk assessment, control and prevention measures, monitoring, risk communication and evaluation. The diversity in the public health systems in the EU provides a challenge to training programmes for such functions, especially when international collaboration is required.

Since 1995 the European Programme for Intervention Epidemiology Training (EPIET) provides training in intervention epidemiology. EPIET is a two-year fellowship to build the core competencies for public health epidemiologists. The curriculum focuses on surveillance, outbreak investigation, field epidemiology research and communication. Ten percent of the time is used for formal training courses. For the remainder of the two years, fellows are placed in a training site in order to perform public health functions under supervision of senior experts. The programme is hosted at the ECDC since 2006.

Since 2008, the ECDC also hosts the European Programme for Public Health Microbiology Training (EUPHEM). EUPHEM is aimed at microbiologists with a background in medicine, biology, veterinary or environmental microbiology. The primary objective is to provide good practice and training in public health microbiology enabling its fellows to apply microbiological methods to a wide range of public health problems in Europe.

Detection, assessment and control of communicable disease risks require a multidisciplinary and inter-disciplinary approach. Within the ECDC fellowships, EPIET and EUPHEM fellows train together within the same 'cohort' and work together to address public health risks.

At the 2014 European Scientific Conference for Applied Infectious Disease Epidemiology (ESCAIDE), one third of the abstracts by fellows were about food- and waterborne diseases. Most (90%) dealt with risk assessment (outbreak investigations and field research) or threat detection (surveillance). Monitoring and evaluation of control measures (public health action) were under-represented elements. This suggests that there is still room programmes to complete the circle 'from evidence to action' when choosing foodborne disease projects within the fellowship.

In this workshop examples will be provided of engaging European Fellowship Programmes in international outbreak investigations and the benefits this may bring to risk assessment.

Short courses in food risk assessment

Andreas Hensel — *Federal Institute for Risk Assessment (BfR), Germany*

Andreas Hensel is since 2003 the first and acting president of the then newly founded Federal Institute for Risk Assessment (Bundesinstitut für Risikobewertung, BfR) in Berlin, which is an institution of the German Federal Ministry of Food and Agriculture. Subsequent to tertiary qualification and specialized studies from 1979 to 1988, which culminated with the title Dr. med. vet from the University of Veterinary Medicine Hannover, Andreas Hensel went on to attain a

PhD title at the University of Utrecht. Further scientific qualifications include his habilitation as university lecturer for Microbiology in 1997 and his qualification as veterinary specialist ('Fachtierarzt') in Microbiology, Animal Hygiene, Clinical Laboratory Medicine, Epidemiology and Food Hygiene. He is also a Diplomate of the European College of Veterinary Public Health (ECVPH). Before accepting the office as president of the Federal Institute for Risk Assessment, Professor Dr. Dr. Hensel successfully performed in the position of senior scientist at the University of Vienna (1990–1997) as well as at the University of Veterinary Medicine in Vienna in the position of full professor for 'Animal Hygiene and Federal Animal Pest Control', at the same time he occupied the position of Director and Chair at the 'Institute for Animal Hygiene and Veterinary Public Health', University of Leipzig (1997–2003). In 2004 he continued his academic activities in the position of appointed honorary professor for consumer protection and risk assessment. Professor Dr. Dr. Andreas Hensel is Germany's representative in the scientific council (Advisory Forum) of the European Food Safety Authority (EFSA).

The Federal Institute for Risk Assessment (BfR) is responsible for evaluating, assessing, and, if necessary and possible, recommending measures for minimising and avoiding risks to human health from substances, microorganisms, products, and processes and to provide possible options for action. In some instances it is also necessary to assess the claimed benefit of substances, products, or processes (e. g. health claims). The resulting opinions serve as a scientific basis for other authorities, legal disputes, and the legislature. This requires transparency in the argumentation, the inclusion of external expertise, and, ultimately, communication of the findings to the public. Where reasonable, potentially diverging interpretations of the data ought to be stated explicitly. The final risk assessment shall answer questions as to (a) who will be affected primarily, (b) how high is the probability of harm, (c) how severe may the latter become, (d) whether it is reversible, (e) how well the assessment is founded, as well as (f) who can control the risk and how. In order to be reproducible, scientifically sound, verifiable, and internationally comparable, risk assessments should follow stringent rules: they usually comprise the four pillars of (a) hazard identification, (b) hazard characterisation, (c) exposure assessment, and (d) risk characterisation. The presentation outlines this procedure using the particular example of foodborne microbial pathogens and explains how the margin of exposure is derived. In addition, it takes a look back at how this process was implemented in specific previous cases, like melamine in infant formula and toffee candy, printing colours on packaging materials (ITX), benzene in juices, or nicotine in eggs.

Environmental risk assessment training and capacity building

Amadeu Soares — *University of Aveiro, Portugal*

Amadeu MVM Soares, full professor, Head of Department of Biology of University of Aveiro (2001–present), Adjoint-Director of CESAM – Center for the Environment and Marine Studies (2010–present) and Director of AIMARE – Aveiro Institute of Marine Science and Technology (2015–present). PhD in Zoology, University of Sheffield, UK, 1989. Coauthor of more than 350 papers from ISI Web of Science, h-index=38. In 2015 ranked within the top-30 European most cited scientists in Toxicology, according to the ranking of LabTimes, based on the ISI Web of Science citations. Some of the individual awards/prizes: 2013: SETAC-Europe Environmental Education Award, for the contributions to post-graduate training and education in the area of Environmental Toxicology; 2004: Excellence Award from the Ministry of Science and Higher Education, Portugal. Former SETAC-Europe president. Supervisor of 53 MSc students, 44 PhD students and 20 post-docs, all concluded. Participated in research projects funded by the EC (13 as coordinator), INTAS (as coordinator), and nationally funded projects (16 as coordinator/PI). Participated in more than 250 panels for academic degrees and professorship positions. Participated, some as panel coordinator, more than 50 evaluation panels for research programs, projects and grants, 25 international. 'Associate fellow' of the Canadian Rivers Institute, University of New Brunswick, Canada, from 2006 to present, and Special Visiting Professor at Federal University of Tocantins – Campus Gurupi, Brazil. Former member of the Strategic Development Group of the University of Lúrio, Mozambique. Member of the Scientific Advisory Council of the Institute of Environmental Assessment and Water Research – IDAEA, Spanish Council of Scientific Research (CSIC), Barcelona. Research interests: Ecotoxicology (soil, freshwater and marine), Functional Biodiversity, Aquaculture, Wildlife Management.

We are in a world where the public is demanding more stringent laws and regulations to protect the environment and human well-being, and these must be inspired in the advances of scientific knowledge. Education, training and capacity building of future European generations, in times where youngers are less and less attracted to scientific fields, are one of the main future challenges in Europe. In addition to a deeper knowledge of a particular field, due to normal specialization in each field of science, we are faced with the multidisciplinary nature of the work that needs to be done and that underlies the risk analysis process. All this must go together with the development of good communication skills. We will present some examples of good practices at European level and discuss and present future training needs, where universities and scientific societies are collaborating to attain and fulfill the challenges.

Beyond traditional learning — Ways for professionals to stay up-to-date on health risk assessment

Johanna Zilliacus — *Karolinska Institute, Sweden*

Johanna Zilliacus is Associate professor and Senior Lecturer at the Institute of Environmental Medicine (IMM), Karolinska Institutet, Stockholm, Sweden. Dr Zilliacus is an appointed member of the Karolinska Institutet Pedagogic Academy, for significant contributions to education at Karolinska Institutet. She has since more than 10 years been actively involved in developing international training in health risk assessment in Europe. Dr Zilliacus has within the EU-funded projects CACADE, RA-COURSES and TRISK developed and organised 20 different risk assessment courses as well as practical training projects in risk assessment for PhD students, post docs and professionals from authorities and industry. She was a partner in the EU-funded project TRACK_FAST that identified training and career requirements of future European food scientists. Dr Zilliacus is coordinator for the contract CT/EFSA/AFSCO/2012/01 that has organised 18 courses for EFSA Panel members and staff in risk assessment. She has also been actively involved in committees initiated by the European Commission aiming at identification of content and structure for training in health risk assessment in Europe. Currently Dr Zilliacus is a member of the EUROTOX subcommittee for education and is responsible for the Advanced International Training Programme in Health Risk Assessment at IMM, Karolinska Institute.

There is an increased requirement for trained health risk assessors in Europe for European authorities, national authorities, industry and others due to new regulatory framework and continuous development of health risk assessment methodologies. European universities provide training in food science and toxicology at Bachelor and Master's levels as well as doctoral education in these areas. However, there has been a lack of dedicated training in health risk assessment that is required in addition to the basic scientific training for individual who will be involved in performing health risk assessments for authorities and industry. During recent years a number of initiatives at European and global levels have been started to define content and structure of training in health risk assessment for professionals. This presentation will present the on-going activities to develop training for professionals and discuss the work that still needs to be performed to ensure that professionals have the right knowledge and skills for performing health risk assessments also in the future.

Nutrition challenges ahead

Rapid developments in science and technology mean that our understanding of the link between nutrition and health continues to grow. Genome sequencing has contributed to greater knowledge about the link between genetics and nutrition and, subsequently, about personalised nutrition. Such developments may help to prevent non-communicable diseases and conditions such as obesity, diabetes and cardiovascular diseases later in life.

At a global level, the sustainable provision of nutritious food to the growing population of the planet is a striking challenge. A significant section of the human population is suffering from nutrition-related problems, including under-nutrition and obesity. Finding solutions to this double burden are of paramount importance, given the multifactorial nature of malnutrition in all its forms and its evidence-based association with adverse health outcomes. The session will focus on developments and challenges in nutrition in the 21st century and options for addressing the growing need for food and key nutrients.

CHAIRS

Androniki Naska — *School of Medicine, National and Kapodistrian University of Athens, Greece*

Androniki Naska is a graduate of the School of Sciences of the University of Athens (BSc in Chemistry); the School of Health and Life Sciences, King's College, University of London (MSc in Human Nutrition); and the University of Athens Medical School (PhD in Nutritional Epidemiology, with distinction). Currently, she is Associate Professor of Hygiene and Epidemiology at the University of Athens Medical School. She has been core investigator in 10 and senior member of the coordinating team in 12 EU-supported research projects on nutrition monitoring; eating out; the development and testing of dietary assessment methods; the collection and analysis of dietary and related data; and the evaluation of diet-disease associations. Prof Naska has published 115 original research papers and reviews, and six chapters in nutrition-related books (3,482 citations and h-index = 32, January 2015).

Junshi Chen — *China National Center for Food Safety Risk Assessment, China*

Junshi Chen was graduated from the Beijing Medical College in 1956 and engaged in nutrition and food safety research for more than 50 years at the Institute of Nutrition and Food Safety, Chinese Center for Disease Control and Prevention (the former Chinese Academy of Preventive Medicine), Beijing. Since 2011, he took the position of Senior Research Professor at the China National Center for Food Safety Risk Assessment. He has conducted large epidemiologic studies on diet, nutrition and chronic diseases, in collaboration with Dr T. Colin Campbell, Cornell University, and Prof Richard Peto, University of Oxford, since 1983. From late 1980's, he conducted a series of studies on the protective effects of tea on cancer, including laboratory study and human intervention trials. He is the member of the expert panel who wrote the WCRF/AICR report 'Food, Nutrition and the Prevention of Cancer: a Global Perspective' (1997). Recently, he was appointed as the Chair of the Chinese National Expert Committee for Food Safety Risk Assessment and the Vice-Chair of the National Food Safety Standard Reviewing Committee. Internationally, he serves as the chairperson of the Codex Committee on Food Additives (CCFA), member of the WHO Food Safety Expert Panel and Director of ILSI (International Life Sciences Institute) Focal Point in China. Dr. Chen's research interests focus on nutrition epidemiology as well as food safety surveillance and risk assessment in the following areas: (1) Relationship between diet, nutrition and non-communicable diseases in different geographical areas and population groups in China; (2) Food fortification; (3) Studies on the protective effect of edible plant (tea, vegetables, fruits, etc.) components on cancer formation with special emphasis on biomarkers and human intervention trial; (4) Total Diet Study in China.

RAPPORTEUR

Silvia Valtueña Martínez — *European Food Safety Authority (EFSA), Italy*

Silvia Valtueña Martínez is Senior Scientific Officer at the (human) Nutrition Unit of the European Food Safety Authority (EFSA). The Nutrition Unit deals with the scientific evaluation of health claims made on foods, novel foods, infant formulae/dietetic foods, dietary reference values and upper tolerable intake levels of nutrients, and food allergens for labelling purposes. Silvia obtained a degree in human medicine (MD) from the University of Barcelona (Spain) and

earned a PhD degree in human medicine (branch of nutrition) from the University Rovira i Virgili (Spain). After 2.5 years of post-doctoral training at the Harvard Medical School (Boston, MA, USA) and two years of post-doc Marie Curie fellowship at the National Institute of Nutrition in Rome (Italy), she underwent a 5-year training in Internal Medicine at the University of Parma (Italy). During all these years, she conducted independent research in several branches of human nutrition, including the relationship between diet and the development of chronic diseases, namely obesity, osteoporosis, diabetes and cardiovascular diseases. She works at EFSA since September 2006.

KEYNOTE SPEAKER

Nutrition in the twenty-first century

Thomas Sanders — *King's College London, UK*

Tom Sanders graduated in Nutrition from the University of London and holds a PhD and DSc from the University of London. He was appointed Professor of Nutrition and Dietetics at King's College London in 1994 and was Head of the Diabetes and Nutritional Sciences Division in the School of Medicine until his retirement in September 2014. He is currently an emeritus Professor of Nutrition & Dietetics at King's College, a scientific governor of the British Nutrition Foundation and Honorary Nutritional Director of the charity HEART UK. He participated in expert consultations with the United Nations FAO/WHO on the role of fats and fatty acids in human nutrition; European projects (e.g. the EU Benefits and Risks Associated with Food project). Over the past decade he has conducted large randomised controlled trials of dietary intervention on cardiometabolic risk. These include the OPTILIP, RISCK, MARINA and CRESSIDA trials which have been designed to provide an evidence base for dietary guidelines for the prevention of cardiovascular disease.

An ageing population and an epidemic of obesity in younger people are the major nutritional challenges in Europe. There is also a need to reduce greenhouse gas emissions, which have implications for food production (e.g. fertilizer use and animal production). Dietary guidelines are concerned with meeting nutrient requirements as well as preventing diet-related diseases such as cardiovascular disease, type 2 diabetes and some types of cancer. Eating less meat would benefit health as well as contributing to a reduction in greenhouse gas emissions. Research indicates health benefits of a shift towards a Mediterranean type of dietary pattern, which is characterized by a lower intake of meat (especially red meat), more fruit and vegetables (including legumes) and moderate amounts of fish, dairy products. However, the diet in some Southern European countries is high in salt, which is linked to high blood pressure. There is good evidence that reducing the salt content of processed foods helps lower blood pressure, which is the leading risk factor for cardiovascular disease.

Europe is well placed to eliminate trans fatty acids from the diet and has an abundant supply of vegetable oils high in oleic acid but low in saturated fatty acids, which help maintain healthy blood cholesterol levels. The causes of the obesity epidemic are complex: while decreasing levels of physical activity contribute to weight gain obesity is primarily caused by excess energy intake from food and drink. The most pragmatic approach is to reduce the intake of 'empty calories' i.e. from sugar sweetened beverages, confectionary, alcoholic drinks and added fat because this would improve the nutrient density of the diet while reducing the energy content.

The priority for the ageing population is to keep people well and out of hospital. While controlling body weight is important for the prevention of and management of type 2 diabetes, weight loss leading to underweight weight is a serious problem for older people, especially those living alone. Attention also needs to be paid to ensure that adequate nutrient status with regard to calcium, vitamin D and vitamin B12 is maintained. As there appears to be strong synergy between diet and moderate physical inactivity (brisk walking, cycling, swimming) in the prevention of obesity, diabetes, osteoporosis and sarcopenia, advice should focus on adopting a healthy life-style appropriate for age rather than targeting individual food components.

SPEAKERS

Metabolic programming: Implications for feeding infants and children

Mary Fewtrell — *UCL Institute for Child Health, UK*

Mary Fewtrell trained in Paediatrics and has worked in Infant & Child Nutrition Research for the past 20 years. She is currently Professor of Paediatric Nutrition and Honorary Consultant Paediatrician at UCL Institute of Child Health, London, UK; and Chair of the ESPGHAN Nutrition Committee. Her research interests include the programming of health outcomes by early nutrition and growth, investigated in randomised nutritional intervention trials in both term and preterm infants, with long-term follow-up; and practical aspects of infant nutrition, with studies on breastfeeding, breast milk expression and complementary feeding.

A large number of animal studies, starting in the first half of the 20th century, suggest that nutrition in early life can 'programme' a wide range of health outcomes and even longevity in a variety of species, including non-human primates. Over the past 30 years, evidence from both experimental and observational studies has accumulated to support the concept that this same phenomenon – termed 'nutritional' or 'metabolic programming' – occurs in humans. Data have accumulated more slowly in humans largely due to the practical difficulties in conducting experiments to demonstrate causal associations; and the difficulty of obtaining information on long-term health outcomes or identifying suitable proxy measures or biomarkers for these outcomes.

Nevertheless, there is now convincing evidence that early nutrition and early growth patterns influence subsequent cardiovascular and metabolic outcomes as well as cognitive development; and that the observed effect sizes are likely to be important in public health terms. Hence, there is increasing appreciation that nutritional recommendations for infants should take into account the potential for effects on long-term as well as short-term health outcomes. Ongoing research is required to address a number of important issues: defining trade-offs between the effects of interventions on different outcomes; identifying differences in programming effects in different populations or groups of infants; pinpointing 'critical windows' for programming effects; and investigating nutrient-gene interactions. Furthermore, the mechanisms responsible for programming effects are a topic of intense research; in particular the role of epigenetic modifications and the possibility that these might eventually be amenable to intervention.

Notwithstanding these unresolved issues, there is already sufficient data to incorporate the concept of nutritional programming into feeding recommendations; notably supporting the role of breast milk for improving later metabolic health and reducing obesity risk and, related to this, the avoidance of encouraging or promoting rapid infant growth, with implications for the design of breast milk substitutes and the choice of complementary foods.

Personalised nutrition for the gut microbiome: feed it, change it, swap it?

Kieran Michael Tuohy — *Fondazione Edmund Mach, Italy*

Kieran Tuohy received his PhD from the University of Surrey (UK) in 2000. Between 2000 and 2006 he worked as a post doctoral researcher in the Food Microbial Sciences Unit of Professor Glenn Gibson, University of Reading, and in 2006, was appointed lecturer in the Department of Food Science and Nutrition, University of Reading. He now leads the Nutrition and Nutrigenomics Group at Fondazione Edmund Mach, Trento, Italy. His research interests are focused on dietary modulation of the human gut microbiota for improved host health. Current projects include work examining the impact

of whole plant foods, probiotics and prebiotics on; the gut microbiota, the host immune system, mucosal cell turnover and apoptosis, host systemic metabolite profiles, cancer biomarkers, body weight, blood lipid profiles and insulin resistance. Since January 2012 he has been associate editor of the International Journal of Food Science and Nutrition, and for the last year, he has been on the editorial board of Frontiers in Molecular Biosciences: Metabolomics. He is a member of the ILSI Europe expert group on 'Prebiotics: Microbial fermentation and metabolism' and sits on the scientific organising committee for the NutrEvent series of conferences, in Lille, France. He has published over 80 peer reviewed articles (http://www.researcherid.com/rid/G-9142-2011) and is co-editor of the book 'Diet-Microbe Interactions in the Gut', Elsevier.

The gut microbiome has emerged as an important contributor to human health and disease. Distinct microbiota profiles are associated with diet and life-style related chronic diseases such as obesity, colon cancer, and autoimmune diseases like inflammatory bowel disease and type 1 diabetes. Additionally, changes in human biofluid metabolite profiles derived from the gut microbiota also characterise these diseases and at least in animal models, microbial metabolic activities related to short chain fatty acid production, the enterohepatic circulation of bile acids, plant polyphenol metabolism and amino acid metabolism, have been shown to play an important regulatory role in both metabolic and inflammatory processes linked to chronic human diseases.

This presentation will discuss the relative merits of different strategies which modulate both the composition and metabolic activity of the human gut microbiota. Recognising the important role of diet in shaping gut microbiota structure and function and with an emphasis on human studies, examples of foods shown to modulate the gut microbiome and concomitantly, change biomarkers linked to chronic disease risk will be presented.

Recent advances in bariatric surgery have demonstrated remarkable improvements in obesity and associated diabetes. This presentation will discuss the impact of bariatric surgery on gut microbiota structure and function and how these interactions may influence metabolic and inflammatory pathway regulation linked to diabetes and obesity.

Finally, the presentation will examine the potential of faecal microbiome transplant in the treatment of both metabolic and inflammation related chronic diseases and discuss how, together with tailored dietary support, rational faecal microbiome transplant may constitute personalised therapies and nutritional solutions for chronic diseases related to aberrant metabolic and inflammatory pathway regulation.

Novel foods

Klaus Riediger — *Austrian Agency for Health and Food Safety, Austria*

Klaus Riediger studied Chemistry at the Technical University of Graz and finished as Graduated Engineer (Dipl.Ing.) with focus in Food Chemistry. Since 2002 he is expert for official food control in the field of novel foods, food supplements and foods of plant origin at the Austrian Agency for Health and Food Safety. He is member of the EU CAFAB 'Novel Food Working Group', Austrian Codex-Commission Sub Comitees 'Novel Foods and New technologies' as well 'Food Supplements', Austrian 'Abgrenzungsbeirat' (Board on (pharmaceutical) borderline products), Expert for the evaluation of the Austrian Codex-Commission Botanical lists for 'Tea and herbal infusions' and 'Food supplements', Policy Advisory Board (PAB) of the PlantLIBRA EC Project Nr. 245199- 'Botanicals in food supplements', AGES Nano Task Force ('Nanotechnology') and member of the Managing board of the Working group Food, Cosmetics and Consumer Goods of the Austrian Chemical Society.

Novel Food is defined as food that has not been consumed to a significant degree by humans in the EU prior to 1997, when the first Regulation on novel food came into force. 'Novel Food' can be newly developed, innovative food or food produced using new technologies and production processes as well as food traditionally eaten outside of the EU. Examples of Novel Food include agriculture products from third countries (chia seeds), newly produced nutrients (tagatose) or extracts from existing food (rapeseed protein). Novel Food must be safe for consumers and properly labelled to not mislead the consumers.

Traditional foods are safe – we know that from experience, even if it is not scientifically proven. For 'Novel Foods' that is fundamentally different. Because they are new, at least in the European diet, there is no sufficient experience in terms of its safety and tolerability. Whoever wants to market novel foods, must prove scientifically that its consumption is not harmful to health and it does not lead to nutritional deficiencies. For complex foodstuffs, consisting of many individual substances of different composition, it is not easy to demonstrate that they are safe. Therefore some exotic foods have failed approval at this hurdle.

Currently there is a proposal for a revision of the Novel Food Regulation (EC) No. 258/97 which provides a simplified authorization procedure for traditional foods and deals with issues such as nanotechnology and insects as food. In Austria, AGES is responsible for the safety assessment and classification of novel foods and works closely with colleagues from the Novel Food Working Group (CAFAB), a group comprised of Novel Food experts from the Member States together with officials from the European Commission.

Under-used food sources of key nutrients

Nanna Roos — *Department of Nutrition, Exercise and Sports (NEXS), University of Copenhagen, Denmark*

Nanna Roos has extensive research experience in the nutritional contribution from local foods in diets in food insecure population, especially among children. She has been involved in research on aquaculture, fisheries and nutrition in Southeast Asia (Cambodia and Bangladesh), in the documentation of the nutritional importance of small indigenous fish, and the potential of integrating local small fish species in rural fishponds with carp production. This multispecies production system secured better contribution of micronutrients to the families and is now implemented in Bangladesh. She has a continued engagement in research with a focus on the role of local animal-source foods for nutritional status and healthy development in infants and young children. Prof Roos alo led the 'WinFood' project investigating how local foods can be used in processed complementary food products in Cambodia and Kenya. In Cambodia, the project tested a product with local small fish and edible spiders in children age 6-15 months, and in Kenya a similar trial tested a product based on amaranth, termites and small fish. Since 2012 she has been increasingly engaged in research on the potential of insect rearing as an alternative animal food source. She now leads the research project 'GREEiNSECT – Insects for green economy' (2014-2017), in which the potential of developing an insect rearing sector in Kenya for supply of food (rearing of crickets) and animal feed (rearing of fly larvae) is investigated through collaborative reseach with Kenyan and international partners.

Poor quality of diets is the main underlying cause of malnutrition in food insecure populations. Diets lack diversity and nutrient dense foods, in particular animal-source food (e.g. meat, fish, egg, milk). Despite some progress in reducing undernutrition, 127 million children are predicted to be chronic undernourished (stunted) in 2025. New approaches to improve dietary quality in food insecure populations are needed to accelerate the reduction in undernutrition.

Among the key nutrients commonly being limited in these diets are iron, zinc and calcium, along with other micronutrients. Vitamin A is also often low, while vitamin A deficiency is largely controlled through supplementation. Deficiency of essential fatty acids are not well documented but are likely to affect large population groups with diets dominated by staple foods,

and low in total fat and animal-source foods. Efforts are required to identify food sources with high nutrient-density, in particular key nutrients like iron and zinc. Limited or no intakes of animal-source foods are found to be a key problem in many low-diverse diets leading to undernutrition.

In many cultures there is a tradition for using foods from wild sources. While some foods from wild sources are well documented for nutritional contribution – such as fish – many are not. Among wild foods, edible insects may in particular be an overlooked and under-used source of important nutrients. Globally, more than 2,000 insect species are recorded as edible. Most are collected and traded in informal food systems and consumption and nutritional contribution lacks to be documented.

Nutritionally, insects vary with species but can in general be regarded as an animal-source food roughly equalizing meat with regards to macronutrients, by mainly contributing protein and fat. Insects appear to be good source of n-3 and n-6 fatty acids, while long-chained PUFA (C22) seems to be limited in most species. Contents of some micronutrients, especially iron and zinc, are high in many insect species, and are likely to be a valuable source. Bioavailability of minerals remains to be systematically documented. The potential of mass-rearing of selected insect species has received considerable attention. Production systems for species of crickets, mealworms and grasshopper are being developed for food, and fly larvae are produced for animal feed. E.g. in Thailand more than 20,000 farmers produce crickets for consumers in domestic and regional markets.

Nutrition challenges ahead: using agro-biodiversity for healthier diets within sustainable food systems

Gina Kennedy — *Bioversity International, UK*

Gina leads a research programme to understand the role of agricultural biodiversity in improved nutrition and health and serves as the Bioversity International focal point for the CGIAR Research Program on Agriculture for Nutrition and Health. She develops, leads and manages nutrition research activities and projects on the use of agricultural biodiversity, particularly in the context of smallholder farmers in developing countries. Her activities also include research design, implementation and analysis of primary research questions related to the role of agricultural biodiversity, nutrition and sustainable diets. She also identifies and pursues opportunities for funding and provides technical assistance to other CGIAR Research Program leaders working on the diet diversity and diet quality cross-cutting Intermediate Development Objective. Prior to joining Bioversity Gina worked for the Nutrition Division of FAO on food-based indicators for use in food and nutrition security programs, nutrition assessment and nutrient requirements. She also managed a research project studying the impact of food security interventions combined with nutrition education on use of locally available foods to improve nutrient intakes, micronutrient status and growth of young children. Prior to working for FAO, she worked for GIZ in Guinea, West Africa and the public health system in the Republic of Kiribati.

Feeding the global population of 9 billion by 2050 in a way that provides a nutritionally adequate and safe diet that is also sustainable environmentally is one of the principle global challenges of our day. There is mounting evidence that our food systems are not providing the right types of foods to ensure nutritionally balanced diets or healthy populations (Chicago Council on Global Affairs, 2015). About 800 million people suffer from insecure food supplies (FAO, IFAD and WFP 2014). Moreover, the patterns and determinants of inadequate nutrition are changing, with countries facing complex and interrelated malnutrition burdens (IFPRI 2014). Many of them experience a combination of chronic undernutrition (stunting), micronutrient deficiencies (e.g. iron deficient anemia), and overweight and obesity in their populations, with women and children being the most vulnerable.

Diet-related non-communicable diseases including high-blood pressure, stroke, diabetes and coronary heart disease, with a concomitant increase of health care costs create a significant and rising nutrition related health and economic burden. The environmental toll of our global food system is also concerning. The global food system is a major contributor to large environmental footprints, including biodiversity loss, greenhouse gas emissions, water shortages, ecosystem pollution, and land degradation (Millennium Ecosystem Assessment 2005, Rockstrom et al. 2009).

The global trend of malnutrition and environmental degradation is aggravated by the fact that national food systems are converging globally, reflected in progressively uniform, less diverse food supplies with emphasis on a few key cereal and oil crops (Khoury et al. 2014). Eating patterns are dominated by processed staple foods, sugar and oil missing important food groups including fruits, vegetables, pulses, nuts and animal source foods. In response to these challenges, Bioversity International supports with partners, a research portfolio that studies agri-food value chains and local food systems, with three principal focus areas: i) on the demand side, focus is on a 'whole diet' approach to dietary diversification for improved nutrition and health; ii) on the supply side, emphasis is on diversified production systems capitalizing on a broad range of biodiversity to improve resilience of livelihoods and landscapes; iii) in the enabling environment, identifying gaps, conflicts or synergies between, public and private policies.

Plenary session: Science, innovation & society

This session will explore the key developments that may affect EFSA's work, in particular the scientific guidance that EFSA develops and uses. The four topics cover areas which are the subject of major scientific research and development and are expected to affect the development of regulatory assessment methodology. These themes will provide the basis for further discussion in the subsequent breakout sessions.

CHAIRS

Jean-Louis Bresson — *Université Descartes & Hôpital Necker – Enfants Malades, France*

Jean Louis Bresson was trained both as a physician and a scientist at Paris University. He is now a paediatrician at Necker – Enfants Malades hospital and professor of nutrition at Descartes medical school in Paris. His research interests are specifically related to nutrition in childhood and during pregnancy. He set up Necker – Enfants Malades hospital's clinical research centre, which stands as an interface between the clinical ward and basic research units. He was a member of the NDA Panel at EFSA (2003–2012).

Anthony Hardy — *European Food Safety Authority (EFSA), Scientific Committee, Italy*

Tony Hardy is currently Chair the Scientific Committee of the European Food Safety Authority (EFSA), having previously chaired the Plant Protection Products and their Residues Panel for 9 years since the establishment of EFSA. He is a zoologist, environmental chemist and ecotoxicologist. Retired from a career in public science at the Central Science Laboratory of the Ministry of Agriculture, Fisheries and Food, which later became the Department for Environment, Food and Rural Affairs in the UK, he carried out research and management in agricultural science and the environmental impact of farming and pesticides. He has been involved in national and international pesticide and food safety risk assessment committees for more than 35 years.

RAPPORTEURS

Frank Boelaert — *European Food Safety Authority (EFSA), Italy*

Veterinary public health officer with background in veterinary inspection service, animal health management and development and implementation of zoonotic disease eradication and control programs. Doctor in Veterinary Medicine, Master of Science in Biostatistics, Doctor in Veterinary Science, Diplomate of the European College of Veterinary Public Health. Twenty-five years overall experience. Resident during three years in Africa. Active involvement in studies, research, training, extension and surveys. Extensive computer, statistics and epidemiology training. Since 2005 employed by EFSA. As a senior scientific officer I am leading a team within the Unit on Biological Hazards and Contaminants, which coordinates the annual collection of information on zoonoses, zoonotic agents, antimicrobial resistance and foodborne outbreaks from the EU Member States. The team is responsible for collation, analyses and reporting of this data and it produces the annual EU Summary Report on Zoonoses. The Unit is supporting the statistical and epidemiological analyses of EU-wide baseline surveys for zoonotic agents in animal populations and foodstuffs. As a team leader I am accountable for the Project Management, Scientific Excellence of the Project Deliverables (Unit and/or External Reports), Career Development and Appraisal of the staff, Development of Procurements and Grants, Evaluation of Declaration of Interests of external experts, External Representation and Relations in the field of Zoonoses Data Collection at EU-level.

George Kass — *European Food Safety Authority (EFSA), Italy*

George Kass was trained as a biochemist. He received his PhD in biochemical toxicology from the Karolinska Institute in Stockholm in 1990. After a post-doc at the Swiss Federal Institute of Technology in Zurich, he returned to the Karolinska Institute as Assistant Professor. In 1994 he moved to the University of Surrey in the UK as Lecturer (Associate Professor) in Molecular Toxicology and was subsequently nominated Professor of Toxicology. He has been with EFSA since 2009 where he is Deputy Head of the Food Ingredients and Packaging Unit. George Kass was awarded a DSc from the Karolinska Institute and the University of Turku, in Finland, and he holds or has held visiting posts with the University of Surrey, the University of Newcastle, the University of Turku and the University of Rome. He has published over 100 papers and abstracts in the field of Toxicology. A substantial part of his research has focused on the molecular mechanisms of drug toxicity and on liver injury. He has been an invited speaker at many national and international conferences and currently is Associate Editor of the journal Toxicology and Applied Pharmacology.

SPEAKERS

Food and health: the role of intestinal micro-organisms in human health

Anne Salonen — *University of Helsinki, Finland*

Anne Salonen is a senior researcher and docent at University of Helsinki, Finland. She has multidisciplinary training in biosciences and PhD in microbiology (2004). Her research activities are focused on the composition and activity of the intestinal microbiota in health and disease (T2D, NAFLD, irritable bowel syndrome) with special interest in reciprocal interactions between diet and the gut microbiota.

Human gastrointestinal tract is inhabited by complex consortium of microbes, the microbiota. Aside from the human genome, our capacity to digest and metabolise foods is also determined by the intestinal microbiota. They encode our 'other genome' with millions of genes, vastly surpassing the coding capacity of the human genome. Gut microbes mainly contribute to the breakdown and bioconversion of dietary components that are not degraded by our own digestive system, such as most plant-based complex polysaccharides and phytochemicals. The microbial metabolites provide energy but also act as signalling molecules that generate systemic immune and metabolic responses and hence can profoundly affect human physiology and health. If is noteworthy that the type and biological activity of the bacterial metabolites released in our gut heavily depend on diet. For example, colonic fermentation of dietary fibre results in production of short chain fatty acids (SCFAs) of which butyrate and propionate have well-documented beneficial effects on gut and systemic health. On the other hand, bacteria can convert dietary protein into metabolites that increase risk for atherosclerosis and colorectal cancer. Hence, intestinal bacteria appear pivotal in mediating the health effects of foods. Due to their profound digestive role as well as plasticity of the gut microbiome as opposed to the human genome, intestinal microbes have recently gained considerable attention on nutritional research.

There is growing interest to understand how diet affects the intestinal microbiota and how this translates to host health. One of the major challenges in the field is the high individuality of the microbiota composition and its individual-specific dietary responses. Similarly, the high variation of host responses is a challenge in nutritional research and practise. We and others have recently started to study the microbiological basis of the individual dietary responses. In our proof-of-principle study we found that categorization of the study subjects to dietary responders and non-responders allows identification of microbiota features that are specific to responders, both in terms of the microbiota and most importantly, of the anticipated host parameters such as metabolic health markers. If such diagnostic microbiota signatures can be validated in further studies, they will provide a radically new way to understand diet-health relationships and approach personalized nutrition.

New developments in our knowledge of neurodegenerative disease

Pierluigi Nicotera — *German Center for Neurodegenerative Diseases, Germany*

Pierluigi Nicotera, a renowned scientist and leading international expert in the field of neuronal cell death, was appointed Scientific Director of DZNE in April 2009. Prof. Dr. Dr. Nicotera was trained in General Medicine and Cardiology at the University of Pavia, Italy. He obtained his PhD at the Karolinska Institute in Stockholm, where he worked subsequently as associate professor. From 1995 to 2000 Nicotera headed the division of Molecular Toxicology at the University of Konstanz and was then appointed Director of the UK Medical Research Council Toxicology Unit. His research has been centred on the molecular mechanisms that lead to neuronal demise following chronic and acute insults. Loss of neuronal synaptic connections and apoptosis play central roles in neurodegenerative diseases.

As the proportion of older people increases, the burden of neurodegenerative disorders progressively grows. By 2040, neurodegenerative diseases are going to be the second leading cause of death after cardiovascular diseases. It is therefore appropriate to produce a major effort in translational research aimed to understand, prevent and cure neurodegenerative diseases. The most sensible option to address the health problem that society will be facing with increased ageing is to address both fundamental mechanisms of disease and lifestyle factors including nutrition that modify the risk of developing dementia or other forms of neurodegeneration. Nutrition and metabolic changes can affect both neuronal function and survival. In addition, complex modulation of the immune system by metabolic factors can be a key determinant of neurodegeneration. I will discuss the interplay of these factors and their impact in age-related disorders.

Key developments in the research on reproductive endocrinology

Richard M. Sharpe — *University of Edinburgh MRC Centre for Reproductive Health, UK*

Richard Sharpe is based in the MRC/University Centre for Reproductive Health in Edinburgh where he heads a research programme on developmental disorders of (mainly male) reproductive health. He is a Professor in Edinburgh University's College of Medicine & Veterinary Medicine. His expertise and research interests cover sexual differentiation, development and puberty (and disorders thereof), fetal programming, endocrinology, the effects of lifestyle (smoking, obesity, diet, use of personal care products), drugs and environmental chemical exposures on reproductive development and function. He is increasingly interested in the inter-relationships between reproductive and wider aspects of health in relation to diet, obesity, inflammation and aging. He serves/has served on numerous advisory bodies in Europe and elsewhere, and as a member of Council for the Society for Endocrinology and as co-Chair of their Special Interest Group on 'Endocrine disruptors'. He is a Deputy Editor of Human Reproduction. He has published more than 350 papers and has an H index of 74 (ISI) 82 (Google).

All of life is geared to reproduction, as has been the case throughout our evolution. As a consequence, development and function of the reproductive system in both sexes is coordinated/integrated with all bodily systems to ensure that reproduction is optimally timed and executed. The primary mechanism via which this integration and coordination is achieved is via the production and action of sex steroid hormones – testosterone and other androgens in men, estrogens and progesterone in women. However, other body systems have also to 'talk' to the reproductive system and this is also achieved via the production of hormones, examples being leptin from fat cells, insulin from the pancreas and osteocalcin from bone. These systems feed back information on developmental and functional status either directly (by effects on the gonads) or indirectly (via the brain), and the complexity and multiplicity of these feedback systems is continuing to be uncovered via

research. Arguably the biggest recent discovery has been of the Kisspeptin system in the brain, which represents a key pathway via which the level of energy stores and metabolic function are able to exert over-riding control of the reproductive system, in particular in females. An outline of how and why such systems operate will be presented.

The other major development in reproductive endocrinology has been the discovery of the important 'programming' effects of sex steroids on the fetus, in particular of testosterone/androgens in males. To understand the importance of these, the course of normal phenotypic development of males and females will be discussed so that the pivotal role of androgens can be understood. The 'set-up' programme is to develop as a female, so that to become a male there has to be an intervention to modify this programme, and this modification is achieved via androgens. This has to occur during a critical phase in very early fetal development (1st trimester in humans) that is termed the 'masculinisation programming window (MPW)'. If there is insufficient androgen exposure in the MPW, males are likely to develop one or more reproductive disorders and to have smaller reproductive organs; these changes are irrecoverable. Conversely, for optimal female development, minimal exposure to androgens during the MPW is required. If there is supranormal androgen exposure of females in the MPW they are likely to develop reproductive disorders, and with severe over-exposure may masculinize to some extent. In both males and females, adult-onset reproductive disorders that are thought to originate from these fetal events involving androgen exposure, are remarkably common.

The occurrence of reproductive disorders in men and women is invariably associated with adverse changes to other body systems, some of which (eg obesity, type 2 diabetes) are themselves very common. This is yet another reflection of the integration of all body systems with reproduction and highlights that maintaining optimal reproductive health is arguably one of the most important goals to achieve to ensure a long, healthy life. This has added importance when it is considered that optimal reproduction also literally sows the seeds for healthy development of the resulting offspring (ie the next generation).

Understanding complex mechanisms in determining adverse and beneficial health effects with nutrition/diets: from basic science of hazard identification to the concept of 'One Health–One Planet'

James Trosko — *Michigan State University, USA*

He (a) demonstrated UV-induced DNA damage & repair in normal human cells; (b) discovered a lack of DNA repair in syndromes of xeroderma pigmentosum & Cockaynes syndromes.; (c) demonstrated that tumor promoters inhibited gap junctional intercellular communication; (d) demonstrated that chemopreventive agents prevented the down-regulation of GJIC by tumor promoters; (e) demonstrated that the anti-cancer drug, SAHA, caused the increase GJIC; (f) demonstrated the mechanism by which chemopreventive agents enhanced GJIC and restored growth control; (g) co-discoverer of normal human adult stem cells (kidney, breast; pancreas); (h) demonstrated that Oct4A gene is expressed in adult normal human stem cells and that Oct-4A could be used as a biomarker for human cancer stem cells; (i) hypothesized that alteration of the quantity and quality of adult stem cells is responsible for aging and the chronic diseases of aging; (j) showed that Blooms syndrome generated mutations via 'errors in DNA replication' rather than 'errors in DNA repair'; (k) provided a nutritional/dietary mechanism for the Barker hypothesis, in that during development, agents, that cause a modulation of the numbers of adult, organ-specific stem cells, could modify the risk to diseases later in life.

To meet the objectives of this conference, I must broaden the debate beyond food safety risk assessment. This includes the role of gut microbiome, human evolution, toxicology, carcinogenesis, stem cell biology and epigenetics. I will challenge paradigms on the mechanisms of toxicities, caused by natural and synthetic chemicals, in and on foods; introduce a hypothesis that integrates biological & cultural evolution contributing to human diseases; demonstrate that divergent diseases share common mechanisms of modulated cell-cell communication; and put risk assessment of food safety into a 'One Health' concept[1].

The challenges are: (a) chemicals do not 'cause' mutations that contribute to hereditary and somatic acute and chronic diseases, but rather, they either alter stem cell numbers in utero or the selective proliferation of spontaneously mutated stem cells by epigenetic mechanisms[2]; (b) human adult stem cells are targets for food-related epigenetic toxins/toxicants[3]; (c) the basic mechanism, modulated by food chemicals, is an integrated 'extra-, 'intra'- and 'inter'- cellular communication system that homeostatically regulates cell proliferation, differentiation, apoptosis and senescence[4]; and (d) understanding biology of development, concepts of toxicology, the roles of slow biological evolution with rapid cultural evolutionary changes in food production, processing and distribution, with cultural differences of both human biology and human diets, together with psychological, social, cultural behaviors, religious, political and economic factors[1]. As a result, there will never be a universal nutritional/dietary proscription, due to genetic, gender, developmental states and other dynamic forces.

The concept of ONE HEALTH relates to the inter-connectedness of ecological, environmental, animal and human health. It is linked through the production, processing and preparation of foods, with the collision of slow biological evolution of genes needed to convert foods into energy for life and reproduction, with the laser-speed changes in cultural evolution. This has created a missmatch of the appropriate genetic backgrounds to deal with the nutritional/dietary exposures of 7 billion human beings, cause by an imbalance of access to clean water, air , soil, and to climate change. These changes are causing political, economic and psycho-social effects, as well as a Diaspora of both people and foods.

The classical 'hazard assessment to risk assessment' causal chain demands we understand the mechanisms of chemicals, in and on foods. Mechanisms of toxicity of food components, of food supplements, of food contaminants, and of the gut microbiome, as well as how these mechanisms interface with the pathogenesis of human diseases, are emerging. Today, we know there are three fundamental mechanisms of toxicity, namely: (a) mutagenesis; (b) cytotoxicity, and (c) epigenetic alteration of normal gene expression.

The most significant cause of nutritional/dietary chemicals' effects on human health was thought to be their mutagenicity. While mutagenesis does occur in humans and is responsible for many human diseases, such as cancer, the means by which mutations can occur is by (a) 'errors of DNA repair' of genomic DNA damage, or by (b) 'errors of DNA replication' on a non-damaged DNA templates. Because of the use of faulty assays and the misinterpretation of false positive results, many natural and synthetic toxins and toxicants have been classified as 'mutagens'. However, ignored claims that these chemicals worked by 'epigenetic' mechanisms has been supported by newer claims[5].

The second form of toxicity is that of cell death. While this also is a real event that, under rare circumstances, high concentrations, exposure of genetically-predisposed, or of young or immunologically predisposed and of the very old, can chemicals lead to serious illnesses and death of the individual.

'Epigenetic toxicity' has emerged as a significant concept that must be integrated in 'Risk Assessment'. To understand 'epigenetic' mechanisms, the pathogenesis of human carcinogenesis can serve as a model. Most cancers, except teratomas, are the result of a multi-stage, multi-mechanism process or the 'initiation'/'promotion'/'progression' concept. Initiation step is an irreversible step taking place in a single cell of any organ, most likely by a mutation caused either by an error of DNA repair (i.e. xeroderma pigmentosum syndrome) or by an error of DNA replication (i.e. Blooms syndrome)[6]. Promotion, on the other hand, is an epigenetic mechanism, which is threshold-dependent, species-, gender- and organ-specific. It must occur after initiation, for long periods of regular exposures, has oxidative stress-related properties, and occurs in the absence of 'anti-promoters'[7].

Since birth defects, cancers, immunological reactions, reproductive- and neuro-toxicities do occur after exposures to various nutritional/dietary conditions, a hypothesis that will be introduced is that these different pathologies share a common underlying fundamental requirement for homeostatic control of cell proliferation, cell differentiation, cell death , and senescence, namely, gap junctional intercellular communication[8]. From the single fertilized egg, 1-2 trillion

cells are produced at birth, with over two hundred differentiated cell types, and three types of cells in each organ, namely, a few organ-specific adult stem cells, the frequent transit-amplifying or progenitor cells and the terminally-differentiated cells. From birth to death, the homeostatic regulation of growth, replacement of cells, and wound repair must be maintained by cell communication. Chemical agents (aflatoxins, TCDD, PCB's, bisphenol A, chemicals in cigarette smoke or grilled red meat, food additives and food supplements, microbiome-related cytokines, etc.) demonstrate properties of tumor promoters or epigenetic agents. The regulatory implications, based on current scientific mechanisms of toxicological action, have not taken into account the characteristics of the epigenetic-acting chemicals. These include timing of exposures, length of exposures, gender and development stages of exposures[9,10], potential interactions of additive, synergistic and antagonistic epigenetic chemicals. There is a need for immediate scientific understanding, in order to be able to extrapolate potential health risks to humans

The modulation of these forms of cell communication will disrupt how these three cells types will behave. Too many or two few of the organ-specific adult stem cells, especially during embryonic/fetal/neonatal development, could cause devastating effects later in life (The Barker hypothesis[11]). Consequently, understanding the influence of nutrition and diets on epigenetic mechanisms of the developing embryo/fetus and neonate is critical, because this is the only time in human development where interference of cell-cell communication is not 'reversible'.

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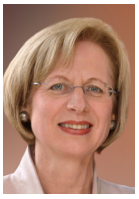
Open Risk Assessment: methods and expertise

EFSA and other scientific advisory bodies recognise there is a need to improve the transparency and openness of scientific assessments in line with today's normative and societal expectations. With this in mind EFSA has launched a number of activities to help it produce more robust, transparent and open scientific assessments. Open scientific assessment can be defined as a decision support process where there is not only full transparency but also an interaction with the outside world on the data, the methodologies used and the outcome. In this context, the framing of the scientific question posed by the requester (in most cases a decision-maker/stakeholder) is important to ensure that the question accurately reflects the problem to be addressed and that it is agreed and clearly expressed prior to the start of the assessment. Methodology, expertise, analyses and information needs should ensure the assessment is fit for purpose and appropriately tailored to answer the question. The session will explore the future challenges and the latest thinking on openness that can assist EFSA to move beyond dialogue to sustainable stakeholder interaction.

CHAIRS

Elke Anklam — *European Commission, Joint Research Centre, Belgium*



Elke Anklam is a chemist by education with specialisation in food, organic and radiation chemistry. After having obtained her PhD from the University Hamburg, Germany, she worked in various European Research Institutions and was a teaching Professor at the Applied University of Fulda, Germany. Since 1991 she has been working in the European Commission's Joint Research Centre (JRC-EC): from 2006-2012 as Director of the JRC Institute for Health and Consumer Protection (JRC-IHCP) in Ispra, Italy, and since January 2013 as Director of the JRC Institute for Reference Materials and Measurements (JRC-IRMM) in Geel, Belgium.

Robert Doubleday — *Centre for Science and Policy, University of Cambridge, UK*

Rob Doubleday has been Executive Director of CSaP since September 2012. Previously Rob established CSaP's research programme. His research interests include the role of science, evidence and expertise in contemporary societies, in particular the relationship between scientific advice, public policy and democracy. His research develops collaborative methods of working with scientists and engineers on the public policy dimensions of their research. In 2010 Rob spent a year on secondment to the Government Office for Science, working on policies to promote engagement between academia and government. Prior to this Rob was the principal investigator of a three-year Wellcome Trust funded project that studied the policy and public dimensions of nanotechnologies. He has published widely on expert advice and public policy, public engagement with emerging technologies, and on public policy dimensions of scientific knowledge. Rob's recent publications include Future Directions for Scientific Advice in Whitehall (CSaP/Alliance for Useful Evidence, 2013). Rob has degrees in Chemistry (Imperial College, London) and Science and Technology Policy (SPRU, University of Sussex). He has a PhD in Geography and Science & Technology Studies from University College London and studied at the Harvard Kennedy School on a Fulbright Scholarship. Rob is also a Senior Research Associate in the Department of Geography at Cambridge and an Honorary Research Associate at the Centre for the Study of Environmental Change, University of Lancaster.

Hiroshi Satoh — *Food Safety Commission, Cabinet Office, Government of Japan*

Hiroshi Satoh, Professor Emeritus of Tohoku University, was appointed Chair of Food Safety Commission Japan in 2015, having been previously Deputy-Chair of this commission and Chair of Expert Committee on Chemicals and Contaminants. He has been engaged in the study on the health effects of mercury and its compounds, especially methylmercury for long time. He graduated from Tohoku University School of Medicine in 1974 and was given a PhD in medical sciences in 1979 by Tohoku University Graduate School of Medicine. He experienced post-doctoral work at the University of Rochester, NY, USA. Returning to his home country, he became an assistant professor at Fukushima Medical College, and later an associate professor at Hokkaido University School of Medicine and Graduate School of Environmental Science. From 1989 to 2011, he was a professor at Tohoku University School of Medicine. His research interest has been how environmental exposure to mercury affects human health. His investigation on this subject includes animal experimental and epidemiological studies. A birth cohort study planned and executed in his locality, examining effects of fetal exposure to methylmercury and other environmental contaminants, stimulated the establishment of Japan Environment and Children's Study, an ongoing national study by Ministry of the Environment. From April 2011 to June 2012, he

was a vice-president of National Institute for Environmental Studies. In addition, Dr Satoh is actively involved in the field of environmental health and public health taking important roles, such as a member of Science Council of Japan, Central Environmental Council of Japan, and Health Sciences Council of Japan.

Reiner Wittkowski — *Federal Institute for Risk Assessment (BfR), Germany*

Reiner Wittkowski is, since 2003, the vice-president of the Federal Institute for Risk Assessment (Bundesinstitut für Risikobewertung, BfR) in Berlin, which is an institution of the Federal Ministry of Food and Agriculture in Germany. After completing the study of Food Chemistry at the Technical University of Berlin, Germany, he attained his doctorate in Food Chemistry (Dr. rer. nat.) at the Technical University of Berlin. In 1986 he received a scholarship from the German Society for Research (Deutsche Forschungsgemeinschaft) with which he did postdoc studies at the University of California, Davis, USA. In 1995 Professor Wittkowski completed postdoctoral studies in Food Sciences and Biotechnology at the Technical University of Berlin. In 1990 he became Head of Dept. for 'Wine and other Beverages' at Max-von-Pettenkofer-Institute of the Federal Health Office (institution preceding BfR). Subsequently, he became Head Dept. for 'Analytics' at BfR. Professor Wittkowski is an Adjunct Professor for Food Chemistry at the Technical University of Berlin as well a member of the Wine Research Committee of the Federal Ministry of Food and Agriculture (BMEL). He is also a member of the Board of Trustees of the Stiftung Warentest and an Advisory Board member of the Committee for Food and Agricultural Products Standards (NAL) of the German Institute for Standardisation (DIN). He is also the second representative for Germany at the Advisory Forum of the European Food Safety Authority (EFSA). From 2003 to 2009, Professor Wittkowski held the offices of President and Vice-President of the International Organisation of Vine and Wine (OIV), Paris. He is currently Honorary President of the OIV and has received several awards for his work in various countries.

RAPPORTEURS

Didier Verloo — *European Food Safety Authority (EFSA), Italy*

Didier Verloo graduated as a veterinarian in 1995 and started his professional career at the Institute of Tropical Medicine in Antwerp on the development and interpretation of diagnostics for human and animal trypanosomosis (sleeping sickness). During the following years he built up experience in biostatistics, epidemiology, risk analysis and test validation and worked from 2000 to 2005 for the Belgian government as a veterinary epidemiologist and risk assessor. Since 2005 he is based in Parma, Italy, working for the European Food Safety Authority (EFSA) where he is heading the Assessment and Methodological Support Unit since 2008. This unit, consisting of about 20 people with different backgrounds, leads and supports the development, implementation and review of evidence-based risk assessment and decision-support approaches in all fields within EFSA's remit.

Tom Meyvis — *European Food Safety Authority (EFSA), Italy*

Tom Meyvis graduated in 1994 from Ghent University as a bio engineer in Environmental Technologies and finished his PhD in Pharmaceutical Technology in 1998, developing hydrogel matrices for controlled vaccine delivery. He continued his research career in the area of polymers and specialised in applications for technical textiles during his 5 years at the Belgian Research Centre for Textiles (Centexbel). During that period he also managed 'Innovation Stimulation Programs' aiming to bring new technologies to the textile sector and to bridge the gap between industry and academia. In 2006 he became director of the Centre for Applied Research and Services of the Ghent University College, managing national and international research projects and promoting scientific services for industry. It was at that time, also being responsible for the Laboratory of Occupational Hygiene, that he discovered risk assessment. In 2010 he followed his wife to Parma and from 2012 he has been a scientific officer in EFSA's Application Desk, focussing on the improvement of the relation with applicants and the application procedures, especially in the area of pesticides. At EFSA he further developed his knowledge of risk assessment and became intrigued by the possibilities of technological development to improve the reliability of risk assessment by making optimal use of worldwide available data, knowledge and resources.

KEYNOTE SPEAKER

From 'Science in Society' to 'Science with Society'?

Gerard H De Vries — *University of Amsterdam, Netherlands*

Gerard H de Vries (1948) was Professor of Philosophy (Chair) at the University of Maastricht, the Netherlands (1987–1997), and Professor of Philosophy of Science (Chair) at the University of Amsterdam (1997–2013). From 2006 to 2015 he was Council Member at the Scientific Council for Government Policy (WRR), the think-tank for long-term policy issues of the Dutch government in The Hague. He was Dean of the Netherlands Graduate School in Science, Technology and Modern Culture (1988–1997). He is a Fellow of Wolfson College, Cambridge (UK). Gerard de Vries' work is chiefly concerned with the social, political and ethical aspects of contemporary science and technology. At the Scientific Council for Government Policy he conducted studies that led to advisory reports on a.o. risk policy (2008) and food policy (2014).

The world of risk analysis and risk governance is facing both internal and external challenges. Internally, it has to learn to better deal with uncertainties; externally, it has to face declining trust in both science and governance in the media and in (parts of) the general public. Various proposals have been forwarded – and to some extent already implemented – to meet these dual challenges, including better risk communication and extending public involvement in risk analysis and governance. However, they leave the heart of the matter untouched. We need to rethink what is science, what is society and what is the place of research in public life.

SPEAKERS

Regulatory impact assessment using socio-economic analysis

Tomas Öberg — *European Chemicals Agency (ECHA), Finland*

Tomas Öberg is since early 2012 chairman of the Committee for Socio-Economic Analysis (SEAC) at the European Chemicals Agency (ECHA) in Helsinki, Finland. He is currently on leave from his professorship in environmental science at Linnaeus University, Sweden. Prior to joining ECHA he was deputy head of unit and managed the team supporting the Scientific Committee of EFSA. Before that Dr Öberg has had a long career in both the public and private sectors, working mainly with environmental protection and chemical safety. His previous research focussed on risk analysis, in particular novel methods for assessment of risk and uncertainty, and the environmental chemistry of PBT substances.

Socio-economic analysis (SEA) plays an important role in the assessment of restriction proposals and applications for authorizations under the REACH regulation. The assessment of impacts through socio-economic analysis considers the health and environmental benefits, the associated costs and other socio-economic impacts of a regulatory action. Experience gained by the ECHA Committee for Socio-economic Analysis (SEAC) indicates that the

assessment of impacts on health and environment is an area requiring further development.

The risk assessment provided in the restriction dossier or the chemical safety report is normally presented as risk characterization ratios. This approach only allows a determination of no risk when from the intended uses; it cannot directly be transferred to an assessment of impacts from an uncontrolled risk. This is one factor that makes it challenging to undertake a risk-benefit evaluation.

Epidemiological studies could in principle help in health impact assessment, and dose-response and likelihood of effects may also be possible to estimate with a sufficiently elaborated toxicological profile and realistic exposure estimates. A shift from qualitative professional judgments to semi-quantitative or quantitative assessments will also require suitable means to handle and describe both natural variability and knowledge uncertainty.

A further challenge to the assessment of risk and socio-economic impacts stems from substances that are persistent, bioaccumulating and toxic (PBT) or very persistent and very bioaccumulating (vPvB). Progress in developing a framework for socio-economic analysis of these PBT/vPvB substances will also be discussed.

The role of crowdsourcing in risk assessment

Steven Drew — *InnoCentive, USA*

InnoCentive, born out of Eli Lilly in 1998, pioneered the business use of crowdsourcing to seek solutions and insights to scientific and technology problems. Today, InnoCentive provides their clients with access to a vast growing global community of registered Solvers with extended outreach to the readers of Nature, Scientific American and The Economist; this Crowd numbers millions. InnoCentive, with their challenge methodology and challenge platforms, uniquely offer their clients the wingspan to crowd-source from their employees, invitational crowds and this vast on-demand global crowd to solve business, policy and technology challenges. Some 3,000 Challenges have been posted across industry, discipline and purpose. Steven Drew, as VP for Business Development in Europe, has been working for InnoCentive for three years, and during this period has helped many leading companies launch the crowdsourcing concept to obtain innovative and diverse solutions from the Crowd. Steven has 12 years of experience in the innovation arena including 8 years leading the UK and North European operations of Invention Machine, a provider of problem solving, content and research software solutions.

Crowd Labour is increasingly being accepted and used by consumers and industry to more diversely service an increasing array of our requirements. More specifically, Open Innovation, has embraced the power of crowd labour to bring diversity and speed to providing new ideas, concepts, techniques and solutions to help overcome industry problems. Challenge Driven Innovation, a form of Open Innovation, allows us to reach out to a vast and growing global community of people, to promote their participation and offer incentives for the best solutions. Challenges can take many forms meaning we can put them to many purposes, whether we are looking for a new concept, seeking global insights and studies, trying to overcome a sticky problem, exploring disruptive landscapes or analysing big data. This presentation will explore the use of Challenge Driven Innovation and the role crowdsourcing can play in Risk Assessment. We will look at what the crowd looks like, the ways in which we can engage the crowd, the types of solutions we can ask of them and the strategies for running challenges. We can ask Solvers from the crowd to conduct studies to feed into risk assessments, to identify models that can be applied to safety assessments, to visualise acquired datasets for diverse or alternative observations, or to derive algorithms to better analyse data. There will be examples to bring the opportunities home to the audience.

User motivation and knowledge sharing in idea crowdsourcing

Miia Kosonen — *Mikkeli University of Applied Sciences, Finland*

DSc (Econ. & Bus. Adm.) Miia Kosonen is an experienced researcher, lecturer, trainer and entrepreneur at CoSoMe specializing in online co-creation. She has been studying the social realms of the Internet since 2003, in collaboration with leading industries, focusing on how to make social-media based communities and crowdsourcing initiatives to evolve and succeed. Previously she has worked as a Senior Researcher, lecturer and Project Manager at Lappeenranta University of Technology, being involved in several research projects about knowledge-based and networked innovation. Her research interests are in the areas of knowledge management, innovation management, knowledge creation, online collaboration and communication, crowdsourcing, skills swarms, online communities and social media. She has published in Computers in Human Behavior, Service Industries Journal, International Journal of Innovation Management, International Journal of Technology Marketing, International Journal of Management Practice, and Knowledge and Process Management, among others.

In idea crowdsourcing, an organisation seeks creative inputs from volunteer users. Crowdsourcing can be defined as 'the act of taking a task traditionally performed by a designated agent (such as an employee or a contractor) and outsourcing it by making an open call to an undefined but large group of people' (Howe, 2008). It could be seen as one method of co-creation, user innovation, and more broadly, open innovation. Yet, the innovative input calls for people who actively participate. Knowledgeable individuals do not automatically share their ideas and insight: they expect some kind of benefit. Building on motivation theories and Uses & Gratifications (U&G) approach, we conducted a web-based survey within IdeasProject, an open innovation and brainstorming community dedicated to harvesting ideas. Based on a sample of 244 users, our research showed that the key driver of knowledge-sharing intentions was made up of two intrinsic motivations — social integrative benefits and learning benefits. We also found that recognition from the host organization affects intentions to share knowledge. From the management point of view, the relative importance of social integrative benefits calls for better facilities available for users to be able to help each other in formulating and developing their ideas. Learning and creativity could be inspired by feedback from professionals and experts, while also providing insight on the features of the current task.

How to support decisions with online collaborative models?

Jouni Tuomisto — *National Institute for Health and Welfare (THL), Finland*

Jouni Tuomisto has a medical background (Lic. Med. 1992 and Dr. Med. Sci. 1999 from the University of Kuopio). He started with toxicology but after the doctoral degree focussed on risk assessment and decision analysis as a post-doc in Harvard School of Public Health in 2000–2001. Subsequent topics have been environmental health assessments of fine particles and dioxins and benefit-risk assessments of food. Mathematical methods became more and more important in the work. In 2006, he started to develop new methods for policy support together with Mikko Pohjola. They developed open assessment, which assesses health and other impacts of policies using open work processes where anyone can participate but specific rules for contributions are applied. In 2014 this was further expanded to open policy practice, which covers – in addition to assessments for policy support – also recommended practices for decision making, implementation of decisions, and evaluation and management of these decision processes. Tuomisto has implemented these methods and practices in numerous assessments related to decisions about environmental health. To support these assessments and decision making, he has also developed a web-workspace Opasnet, which is freely available for similar assessments in other decision support processes. He has given university courses about open policy practice and trained students to use Opasnet in their work as environmental scientists.

Transparent decision-making process and openness is a mega-trend. A lot of good quality scientific data is becoming openly available in national and international databases (en.opasnet.org, avoindata.fi, data.gov.uk, thegovlab.org). Also there is a clear societal strive to provide and use open source models and evidence-based decision making and support their open use and development.

However, models alone are not enough. Collaborative, multidisciplinary practices are needed to solve relevant societal problems or questions. The processes must be reliable, reproducible, and transparent to support these studies effectively and efficiently. Shared practices, tools, data, working environments and concerted actions are the way forward to improve science and decision support. Practices are needed for mutual communication: experts answering policy questions in a defendable and useful way rather than just pushing out data; decision makers more clearly explaining their views using evidence; the focus being on end-users. ICT tools support this transformation.

Open data and open models change the way we think about evidence-based decision making: it is collaborative, dynamic information collection work that produces a quantitative description of the topic in the form of an online collaborative model. An outdated practice is to rely only a long chain of static information products such as scientific articles, reviews, expert reports, policy papers, and finally decision recommendations. Such a model is more complex than just an online calculator or a piece of text written with online tools. Rather, it is a discussion forum for relevant topics aiming to resolve disputes. It is a scientific platform to present hypotheses and attempt to reject them based on data. It is a database for up-to-date quantitative estimates about important variables. It is a modelling environment optimising decision options based on the variables. It is a resource centre for re-using existing information in similar new cases. And finally, it is a forum for discussions about values and objectives that should be used to choose or reject decision options.

Platforms, tools, and practices for doing all that exist already, but they are not systematically used. I will present how it can be done. We can update our own practices as decision makers, experts and stakeholders to better utilise their potential for added value in societal decision making related to food and other topics.

Extracting evidence from unstructered data: potential applications of IBM Watson for RA

Cameron Brooks — *IBM Watson Group, Public Sector Solutions, USA*

Cameron Brooks is the Director of Public Sector Solutions for IBM's newly formed Watson Group, leading the business and go-to-market strategy for Government and Education clients in Europe and worldwide. In this capacity, Cameron works closely with worldwide leaders in government agencies and educational institutions to help them leverage IBM's transformational capabilities in cognitive computing to address their most pressing challenges. Cameron has been employed at IBM for 19 years, and has worked with Public Sector for over 12 years. In his previous role, he was the Public Sector Leader for IBM's Middle East & Africa organization, driving the growth of IBM's business with Government, Healthcare and Education clients across this emerging market. In other Public Sector roles, Cameron has served as Director of IBM's Government Healthcare business, leading the company's transformational strategy of this market segment, and he was instrumental in growing IBM's Smarter Cities initiative from its inception several years ago, as Director of the Smarter Water Management program. Cameron has also successfully led the growth of several new businesses and emerging technologies within IBM. In one of these roles, he was Program Director for the IBM Blue Gene supercomputer, working with a worldwide team of business and technical executives to build and deliver one of the top computational platforms in the world. Cameron started his career in technology development for the Microelectronics Division of IBM's Systems & Technology Group. Cameron holds a BS degree in Electrical Engineering from the University of Waterloo, Canada, and MS and PhD degrees in Electrical Engineering from the University of Michigan. He also holds an MBA degree from the New York University Stern School of Business. Cameron has been issued six US patents and has authored over 20 technical papers. In 2010, he was recognized with the Black Engineer of the Year Award for Professional Achievement in Industry. He is married, and a father to two girls and resides in London, United Kingdom.

Abstract not provided.

Implementing the risk profile: the German risk assessor's experience

Mark Lohmann — *Federal Institute for Risk Assessment (BfR), Germany*

Mark Lohmann has a scientific background in biochemistry and bioinformatics. He worked six years as a manager of the Cologne University Bioinformatics Center (CUBIC). Beside project coordination he was involved in research and teaching activities mainly related to in silico simulation of metabolic networks. From 2006 to 2010 he worked as a project manager in the field of bioprocess engineering and was head of the lab for food sensory science at the Technology Transfer Center Bremerhaven, Germany. During this period Dr Lohmann was involved in the development and integration of novel technologies to improve safety, transparency and quality assurance of the food supply chain as well as in the exploration of psychological and metabolic factors that affect sensory perception. Since 2010 he has been working as the head of unit risk research, perception, early detection and impact assessment at the German Federal Institute for Risk Assessment (BfR) in Berlin. His research interests include the realization of target group-oriented risk communication, the development of methods to determine the influence of social-psychological factors on public risk perception as well as the implementation of systems for the detection of emerging risks.

Risk assessment is the analysis of a risk by means of scientific methods. This includes the identification of a potential risk source; the qualitative and/or quantitative evaluation of adverse health effects that could arise from the risk source under consideration; the establishment of a dose-response relationship as well as an assessment of the degree of human exposure. The results are at least partially based on calculations or mathematical models, and the risk potential is described in mathematical or statistical terms based on these methods.

However, sole quantification of risk information usually leads to an excessive reduction in complexity to a few statistics that have very limited value for laypersons. Also, the likelihood of misinterpretation increases. Consequently, as a means of conveying scientific assessments in a clear and concise manner, a risk chart may be used as a visualization tool to clarify the relationship between probability and extent of damage using color gradations. So far, the already existing 'risk matrix' is considered as a well-functioning form of risk representation. However, due to its restriction to the two-dimensional plane, the risk matrix's informational value is too low to exhaustively characterize the extent of risk potential and uncertainty of data.

Therefore, it was the BfR's aim to develop a chart which, by omitting non-essential aspects of risk assessment to make it easier for the reader to quickly recognize the circumstances and main characteristics of the risk assessed in official opinions with regard to food-, biological-, chemical-safety as well as safety of consumer products. This chart is structured as a table containing the following five characteristics: affected groups of persons; probability and severity of impaired health in the event of exposure; validity of the available data; and possibilities for consumers to control the risk through such measures as avoidance or caution. The risk profile which is part of a large number of BfR-Opinions published on the internet was developed in various internal and external test phases in cooperation with scientists from various disciplines. Currently, it is in the process of being updated, taking into account evaluations by different stakeholders especially focusing on the usability of different verbal expressions defining the term 'probability'.

Novel chemical hazard characterisation approaches

Scientific and technological advances are revolutionising biology and toxicology, as well as regulatory risk assessment, making available a huge number of new tools to investigate chemical effects. Complex endpoints can be predicted by using integration of evidence where information and evidence can be incorporated flexibly. In addition, one of the main themes in current research is the need to move away from animal testing toward the use of in vitro methods, in agreement with the 3R concept. One of the roadmaps of the new toxicology paradigm is represented by Tox21, which indicated the need for researching, developing, validating and translating innovative chemical testing methods that characterise toxicity pathways. This session will present the most up-to-date tools for performing hazard assessment using integrated and alternative testing strategies. The topic is of great relevance considering the new perspectives and developments of toxicology, shifting to innovative approaches which are likely to have an impact on risk assessment procedures.

CHAIRS

William Slikker Jr — *Food and Drug Administration/National Center for Toxicological Research, USA*

William Slikker Jr is the Director of FDA's National Center for Toxicological Research (NCTR). He received his PhD in Pharmacology and Toxicology from the University of California at Davis in 1978. Dr Slikker holds Adjunct Professorships in the Departments of Pediatrics, and Pharmacology and Toxicology at the University of Arkansas for Medical Sciences. He has held committee chairmanships or elected offices in several scientific societies, including the Teratology Society (serving as President) and the American Society for Pharmacology and Experimental Therapeutics (Chair, Developmental Pharmacology Section and member of the Program Committee) and co-founder and past President of the MidSouth Computational Biology and Bioinformatics Society. He is currently Associate Editor for NeuroToxicology and Toxicological Sciences. He is the past President of The Academy of Toxicological Sciences, the Society of Toxicology (Presidential term ended 2013), and the recipient of the 2014 George H. Scott Memorial Award from The Toxicology Forum. Dr Slikker currently serves as the Co-Chair of the Global Coalition for Regulatory Science Research (GCRSR). He has authored or co-authored over 300 publications in the areas of transplacental pharmacokinetics, developmental neurotoxicology, neuroprotection, systems biology, and risk assessment. He has also served on several national/international advisory panels for the International Life Sciences Institute (ILSI)/Health and Environmental Sciences Institute (HESI), Chemical Industry Institute of Toxicology (CIIT) Centers for Health Research, Environmental Protection Agency (EPA), National Institute of Environmental Health Sciences (NIEHS), National Academy of Sciences (NAS), National Institutes of Health (NIH) and World Health Organization (WHO).

Emanuela Testai — *Istituto Superiore di Sanità – Department of Environment and Primary Prevention, Italy*

Highest degree in Biological Sciences – biochemical branch (University of Pisa, 1981). Current Position: Senior/Executive Scientist – Head of the Mechanisms of Toxicity Unit at Istituto Superiore di Sanità (Rome, Italy). Previous experience at the National Research Council – Institute of Mutagenesis and Differentiation in Pisa (doctoral training), then at ISS as young researcher. Her expertise is focused on mammalian toxicology and human health risk assessment, associated to exposure to natural and synthetic chemicals, carrying out both research and regulatory activities. Main research interest: molecular mechanisms of toxicity; toxicokinetics and toxicity of xenobiotics; enzymology of drug-metabolizing systems in hepatic and extrahepatic tissues; in vitro methods as alternative to animal testing, metabolic biomarkers of individual susceptibility; molecular epidemiology; risk assessment associated to exposure to cyanotoxins and study of cyanobacterial communities. Scientific Leader of National/International Research Projects, granted by the Italian Ministry of Health, EU, National Research Council. Member of the Scientific/Organizing Committee of around 20 national and international scientific meetings. Active participation to 6 Twinning Projects financially supported by EU. Member of DG SANCO Scientific Committee for Health and Environmental Risks (SCHER;2004–2012) and Scientific Committee of Emerging and Newly Identified Health Risks (SCENIHR); of the National Committees for Plant Protection Products and Biocides; expert in OECD Test Guidelines Program, expert in EFSA working groups, GLP Inspector, consultant for the Italian Ministry of Health. Contract Professor at the University of Rome 'La Sapienza' (2004–2007); Teacher in several Master Courses, supervisor of several doctoral thesis. Author of more than 90 peer-reviewed papers and book chapters; reviewer for high reputation journals in the field of toxicology and risk assessment.

RAPPORTEUR

Manuela Tiramani — *European Food Safety Authority (EFSA), Italy*

Manuela Tiramani is, since January 2015, head of the FEED unit (ad interim) at EFSA. From 2005 to 2007 she was a scientific officer (mammalian toxicology) in the Pesticides unit of EFSA; and from 2007 to 2014, she worked as team leader of the mammalian toxicology team responsible of the toxicological assessment of plant protection products and of the exposure assessment for operators, workers, bystanders and residents at EU level.

KEYNOTE SPEAKER

The frontiers of predictive toxicology

Thomas Hartung — *Johns Hopkins University, USA*

Thomas Hartung, MD PhD, is Professor of Toxicology (Chair for Evidence-based Toxicology), Pharmacology, Molecular Microbiology and Immunology at Johns Hopkins Bloomberg School of Public Health, Baltimore, and University of Konstanz, Germany; he is also Director of their Centers for Alternatives to Animal Testing (CAAT, http://caat.jhsph.edu) with the portal AltWeb (http://altweb.jhsph.edu). CAAT hosts the secretariat of the Evidence-based Toxicology Collaboration (http://www.ebtox.com) and the industry refinement working group. As PI, he heads the Human Toxome project funded as an NIH Transformative Research Grant. He is the former Head of the European Center for the Validation of Alternative Methods (ECVAM), Ispra, Italy. He has authored more than 430 scientific publications.

Over the last few years, a discussion has started addressing the shortcomings of both the animal tests and our traditional cell culture work. This might actually serve as a door opener for new approaches making evident the limitations in an objective not only animal welfare driven way. At the same time, a mechanistic, molecular research has evolved, which is effectively relying to large extent on methodologies which substitute or complement traditional animal tests. The biotechnology and informatics revolution and their commercialization over the last decades has made such technologies broadly available, standardized and useful. Novel approaches toward a more organo-typic cell culture ('human-on-chip') open new avenues to replicate human physiology in the test tube, often based on human stem cells from healthy donors and patients.

Regulatory toxicology, which can serve as an example for current developments, has begun to embrace these new approaches. Major validation efforts have delivered the evidence that new approaches not necessarily lower safety standards and can be integrated into regulatory safety assessments, especially in integrated testing strategies. In the US, especially the NAS vision report for a toxicology in the 21st century and its most recent adaptation by EPA for their toxicity testing strategy and the Endocrine Disruptor screening program (EDSP21) have initiated a debate how to create a novel approach based on human cell cultures, lower species, high-throughput testing and modeling.

The common theme is a mechanism-based approach (Adverse Outcome Pathways, AOP). The Human Toxome project (http://humantoxome.com), which aims to develop a comprehensive knowledge-base of molecular perturbations (pathways of toxicity) is a prime activity here. However, major parts of toxicology have not yet found in vitro solutions. Beside the technical development of new approaches such as organo-typic cultures or systems toxicology, a case is made that we need both conceptual steering and an objective assessment of current practices by evidence-based toxicology. The Evidence-based Toxicology Collaboration (http://www.ebtox.com) currently pilots systematic reviews in order to adapt them for toxicological method evaluation. The concept of mechanistic validation is proposed as a way forward to quality-assure new cell-based tests.

SPEAKERS

Alternative and integrated testing strategies

Horst Spielmann — *Freie Universität Berlin, Germany*

Horst Spielmann, born April 3, 1942 in Lublin/Poland, retired head of ZEBET (National German Centre for the Documentation and Evaluation of Alternatives to Testing in Animals) at the BfR (Federal Institute for Risk Assessment) in Berlin, Germany. He was Professor for Regulatory Toxicology at the Freie Universität Berlin, Germany, and State Animal Welfare Commissioner of the State of Berlin, Germany. Since 1988 he has been chairperson and member of management teams (MT) of successful international validation studies of non-animal methods for toxicity testing.

After EU Dir. 86/609 for the protection of animal used for scientific purposes had been adopted by EU member states in 1990 the EU Commission established the EU Center for the Validation of Alternative Methods (ECVAM) at the JRC in Ispra/Italy, which had the mission to validate non-animal method for regulatory testing. To meet the requirements of the EU Cosmetics Directive ECVAM focused its validation activities on replacing animal safety tests for local toxicity test on eye and skin. From the failure of the first international validation study tests to replace the Draize rabbit eye irritation test it became apparent that a complex in vivo toxicity test cannot be replaced by a single in vitro toxicity test but that several assay may be needed, each of which covering different in vivo endpoints of the assay to be replaced.

Therefore, integrated testing strategies (ITS) were developed for skin and eye irritation, starting with physico-chemical studies, validated in vitro assays and followed by in vivo testing in a reduced number of animals. Due to a major breakthrough in biotechnology human reconstructed epidermis (hRE) models became commercially available for toxicity testing and also human full skin models and more recently human cornea-like epithelium (RhCE) models. In collaboration with the OECD program for the testing of chemicals ECVAM and other government validation centers designed novel Integrated Approaches on Testing and Assessment (IATA) for toxicity testing without any conformational testing in animals. More recently the adverse outcome pathway (AOP) approach was used to analyze the major pathways underlying all mayor areas of toxicity and in vitro assays were developed for each of the pathways. This approach has successfully been used to replace skin sensitization testing in animals by an in integrated in vitro testing strategy with human cells and tissues, each of which is covering one of the essential pathways underlying of the rather complex mechanism skin sensitization. The new toxicity tests, which do not require testing in animals anymore and which are being used by OECD member countries, should also be used by EFSA and all other international food agencies to provide safer food for all citizens around the globe.

The study of modes of action: the AOP

Ellen Fritsche — *IUF – Leibniz Research Institute for Environmental Medicine, Germany*

Ellen Fritsche studied medicine and achieved her physician PhD at the University of Düsseldorf in 1998. During this time she also started her education in general toxicology. From 1998 to 2001 she spent her first postdoc at the National Institute of Environmental Health Sciences, Research Triangle Park, NC, USA where she worked on genetic polymorphisms. During her second postdoc in Germany she widened her focus on dermato- and neurotoxicology. Therefore, she became an expert on arylhydrocarbon receptor function in skin and developed a human in vitro model for developmental neurotoxicity testing, respectively. Today she is a group leader at the IUF (Group of sphere models and risk assessment) and her research focuses on developmental neurotoxicology by applying neural stem/progenitor cells and induced pluripotent stem cells. The species differences in cellular signal transduction and responses to

neurotoxicants between rats, mice and humans form her current main research topic. This knowledge is currently generated for building AOPs for developmental neurotoxicity. Her international recognition is demonstrated by serving on several editorial boards. Furthermore, she is chair of the CEFIC-LRI external scientific advisory panel and member of the European Commission Expert Group: Horizon 2020 Advisory Group for Health, Demographic Change and Wellbeing as well as member of the EFSA Scientific Panel on Plant Protection Products and their Residues. Moreover, she is Vice-President of the European Society for Alternatives to Animal Testing (EURSAAT) and board member of the ACT (Alternatives Congress Trust). Ellen Fritsche has been awarded for her work several times. Among these awards, the most outstanding one was the CEFIC LRI-Innovative Science Award (2006). She also received the PHORA research award of the German Society for Photobiology (2007) and the Research Award for the support of methodological work from the German Federal Ministry of Food and Agriculture.

Regulatory toxicology in the twenty-first century encounters numerous challenges. For one, there is the need for assessing the hazard potential of a large number of chemicals. At the same time animal use, costs and time required for chemical testing is supposed to be reduced. This circumstance requires a paradigm shift in toxicological testing from apical endpoint testing in animals to a mechanism-based assessment of toxicity using alternative methods. The National Research Council of the USA has recently proposed such a procedure. For positioning toxicity data generated by alternative methods into a regulatory context, the adverse outcome pathway (AOP) concept has been suggested and is supported by the OECD. An AOP is a conceptual framework for organizing existing knowledge concerning the predictive and/or causal linkages between measureable/observable biological changes that are essential to the progression from a molecular initiating event to an adverse outcome considered relevant to regulatory decision-making. Here, current state-of-the-art on AOP building and application will be presented.

In vitro data and in silico models for predictive toxicology — The SEURAT project

Elisabet Berggren — *European Commission, Joint Research Center, Italy*

Elisabet Berggren works currently as a scientific coordinator at the Systems Toxicology Unit and the European Reference Laboratory for Alternatives to Animal Testing (EURL ECVAM). The Systems Toxicology Unit assists in the development of a new and more efficient safety assessment of chemicals based on in vitro, in silico and in chemico methods. The aim is to develop new predictive methodologies more relevant to human health, encouraging innovation and avoiding animal testing. Elisabet is also contributing to the coordination of SEURAT-1, the largest EU initiative ever on alternative testing, focussing on toxicity testing for repeated dose toxicity and funded by European Commission (FP7) and Cosmetics Europe. Elisabet started to work for the European Commission in 1996, and she was responsible for the Technical Committee of Classification and Labelling of Dangerous Chemicals at the European Chemicals Bureau during many years. She was involved in the negotiations of the Globally Harmonised System, its implementation within the EU through the CLP (Classification, Labelling and Packaging of substances and mixtures)Regulation and development of the CLP guidance document. She also contributed to the negotiations of the Rotterdam Convention and its EU regulatory implementation. Elisabet made her PhD in physical chemistry at Stockholm's University in 1991. In her academic career she primarily focussed on the development of theoretical dynamic models for liquid crystals and biological relevant systems.

SEURAT-1[1] is a major European research consortium established to evaluate the safety of chemicals considering repeated exposure in humans without using animals. It is a public-private partnership co-financed by the European Commission's FP7 Health Programme and Cosmetics Europe. SEURAT-1 combines the research efforts of over 70 European universities, public research institutes and companies. The SEURAT vision is to fundamentally change the way we assess the safety of chemicals, by superseding traditional animal experiments with a predictive toxicology that is based on a comprehensive understanding of how chemicals can cause adverse effects in humans[2]. The SEURAT strategy is to adopt a toxicological mode-of-action framework to describe how any substance may adversely affect human health, and to use this knowledge to develop complementary theoretical, computational and experimental (in vitro) models that predict quantitative points of departure needed for safety assessment. One of the SEURAT-1 objectives is to demonstrate a multiple level proof-of-concept (theoretical, methodological, and application)[3] for repeated dose systemic toxicity but in principle also applicable to other endpoints. Firstly we develop a theoretical Adverse Outcome Pathway (AOP) describing the key events of the biological process initiated by a chemical stressor[4,5]. Secondly we need a testing strategy for toxicity prediction, based on AOP knowledge relevant to the toxicity to be predicted. Typically a combination of in vitro, in silico and in chemico data is needed to trigger selected key events. Finally the results of testing strategies in combination with already existing data (physical chemical information, animal or human in vivo data or other) and biokinetic modelling, could provide sufficient evidence to support chemical safety assessment.

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QSAR and computational tools

Emilio Benfenati — *Istituto di Ricerche Farmacologiche Mario Negri, Italy*

Emilio Benfenati is head of the Laboratory of Environmental Chemistry and Toxicology at the Mario Negri Institute, Milan, Italy, since 1997. In the laboratory about 35 researchers are active. Previously he has been researcher at Stanford University, California, USA (in that period he also had a collaborative research at the Berkeley University, California, USA). He coordinated 16 European projects (including CAESAR, CALEIDOS, ANTARES, PROSIL, ToxBank, IMAGETOX) and participated in 22 others. Many of them are on toxicity and environmental modelling. His research activities include: toxicity and environmental modelling, molecular descriptors, QSARs, toxicity prediction, environmental management, characterisation and assessment of contaminants, risk assessment; development of QSAR models; analysis of environmental and food samples for pollutants such as dioxins, PCB, PAH, pesticides, endocrine disruptors, industrial pollutants; environmental assessment. He is author or co-author of about 300 papers in international journals and edited a few books.

The field of in silico models is rapidly evolving. A number of models are available for the same endpoint, and new read across tools have been also introduced. This offers a palette of solutions, but at the same time it requires new perspectives for the integration of the multiple values which are generated. The challenge is not simply the prediction of one value for one property, but the complex integration of different pieces of information. Advanced platforms are available offering reasoning related to mechanism of action, exploring similar chemicals defined on the basis of different criteria, and providing quantitative measurements for each step and decision. Thus, the new frontier is producing models which can help the evaluators to get reproducible, understandable, and statistically sound results. Indeed, a major issue in the interpretation and integration of the results is the subjective assessment, which relies on the past, personal experience on the individual experts.

A questionnaire has been done with about 200 replies, asking for the read across evaluation of a number of substances, and results indicated that assessment for the same substance may be highly irreproducible, depending on the approach. The in silico models are not always sufficiently valid. For

this reason, the evaluation of the reliability of each individual prediction is an important issue, and it has been demonstrated to be a necessary component for the correct and safe use of the results. Modern tools provide powerful measurements of this reliability and this is part of the overall strategy of this mature technology. Thus, important components of the in modern silico models are their ability to integrate different perspectives (for instance those arising from different models or modelling tools within a hybrid approach), to provide reasoning and documentation (the simple value as output of a model is not sufficient), and to get a quantitative assessment of the reliability (related to the applicability domain).

This refers to the evaluation of the result for a single substance. A different perspective is when there is the need of prioritization. In this case the evaluation process is of course extended to a large set of chemicals, but in addition multiple endpoints can be addressed. Finally, the purpose of the prioritization has to be defined, if for instance the overall in silico integrated approach has to minimize false negatives or positives.

In vitro and high throughput screening (HTS) assays

Raymond Tice — *NIEHS (volunteer), USA*

Raymond Tice received a PhD in Biology in 1976 from Johns Hopkins University (Baltimore, MD). From 1976 to 1988 he was employed by the Medical Department at Brookhaven National Laboratory (Upton, NY), and from 1988 to 2005 by ILS, Inc. (Durham, NC). In 2005, he joined NIEHS as the Deputy Director of the NTP Interagency Center for the Evaluation of Alternative Toxicological Methods (NICEATM) and in 2009 became Chief of the Biomolecular Screening Branch (BSB) within the Division of the NTP. In this position, he was responsible for directing NTP's high throughput screening program and supporting the US Tox21 effort. Dr. Tice retired in January 2015 and is currently serving as a special volunteer to the BSB and DNTP. He served as President of the US Environmental Mutagen Society and as Vice-President of the International Association of Environmental Mutagen Societies. In 2008, he shared the North American Alternative Award from the Humane Society of the United States and Proctor & Gamble for 'outstanding scientific contributions to the advancement of viable alternatives to animal testing'. In 2009, Dr. Tice received the EMS Alexander Hollaender Award in recognition of outstanding contributions in the application of the principles and techniques of environmental mutagenesis to the protection of human health. Among NIH awards, in 2014, he received NIH Director's Awards as a member of the NIEHS/NCATS/UNC DREAM Toxicogenetics Challenge Team and of the Tox21 Team. He has authored 170 scientific papers and book chapters and edited 4 symposia proceedings.

Launched in 2007, Tox21 is a US multiagency collaborative effort involving the National Institute of Environmental Health Sciences/National Toxicology Program, the National Center for Advancing Translational Sciences, the Environmental Protection Agency's (EPA's) National Center for Computational Toxicology, and the Food and Drug Administration. The objective of this partnership is to shift the assessment of chemical hazards from traditional experimental animal toxicology studies to one based on target-specific, mechanism-based, biological observations largely obtained using HTS and high content in vitro assays with the ultimate aim of improving risk assessment for humans and the environment. More specific goals are to identify patterns of compound-induced biological response to characterize toxicity/disease pathways, prioritize compounds for more extensive toxicological evaluation, develop models predictive of adverse health effects in humans, and evaluate population variability in sensitivity to toxic compounds.

The Tox21 Program has profiled a diverse library of >10,000 chemicals, including mixtures, across a set of nuclear receptor and stress response pathway assays at 15 concentrations, with each assay run 3 times to improve the robustness of the data. The activity profiles generated have been made public and are being analyzed in terms of structure-activity relationships and biological relevance. Limitations associated with the HTS effort have been identified; these include, for example, the limited pathway coverage (i.e., focus on nuclear receptor and stress response pathways) and the lack of biological complexity (i.e., the use of reporter gene assays using immortal cell lines with limited capability for xenobiotic metabolism). The current focus of Tox21 is to overcome these limitations by incorporating into the testing strategy more physiologically-relevant cell types (e.g., HepaRG cells, embryonic stem and induced pluripotent stem cell-differentiated cell populations) and lower organisms (e.g., zebrafish, *Caenorhabditis elegans*), coupled with high content screening and high throughput transcriptomics platforms to assess chemical toxicity potential. Equally important are continuing efforts to make all data public and to increase stakeholder involvement by establishing formal and informal relationships with investigators and/or organizations interested in contributing to this effort.

Organs-on-Chips: A living platform for generating human relevant data

Remi Villenave — *Emulate, USA*

During his PhD in molecular virology at the Queen's University in Belfast, UK, Remi Villenave developed a new pediatric model of Respiratory Syncytial Virus infection based on differentiated primary airway epithelial cells that recapitulated hallmarks of the disease in children. Following a first post-doctoral training, Remi Villenave acquired a strong expertise in human airway epithelium biology and mucosal immunity applied to 3D modeling of airway tissues and host pathogen interaction. This expertise brought him to the Wyss Institute at Harvard University in 2012, where he participated to the Organs-on-Chip program and led the development of a new Airway-on-a-Chip platform for the modeling and study of human pulmonary obstructive diseases in collaboration with industry partners. As a senior research fellow under the mentorship of Don Ingber, Remi Villenave applied the Organs-on-Chips technology to better understand mechanisms of diseases, study host pathogen interaction and test new therapeutics. Recently, Remi Villenave joined Emulate's founding scientific team to further develop and apply the Organs-on-Chips technology to respiratory and infectious diseases.

This presentation will review our novel biomimetic microsystem technology and its potential applications for evaluating safety, toxicity, efficacy, and mechanism of action for new drugs, agrochemicals, and cosmetics. Organs-on-Chips offer exciting new approaches to attack fundamental questions in biology, and develop smart in vitro surrogates for regulatory sciences that can positively impact human health. We apply microfabrication approaches to engineered cell culture microenvironments that go beyond conventional three-dimensional models by recapitulating the tissue-tissue interfaces, spatiotemporal chemical gradients, mechanical microenvironments, and physiological function of living organs. These Organs-on-Chips are being combined with cultured human cells to elucidate human physiology in an organ-specific context. This technology is poised to achieve new standards in emulating human physiology for use in biological testing to advance product innovation, design and safety across a range of applications in pharmaceutical development, food safety, personalized health, agriculture, and chemical-based consumer products.

Microbiological risk assessment

Microbiological risk assessment (MRA) is a scientifically based process consisting of these steps: (i) hazard identification, (ii) hazard characterisation, (iii) exposure assessment, and (iv) risk characterisation. This short definition hides a complex discipline with a broad spectrum of approaches; from qualitative to quantitative assessments, focused on part of the food chain to the whole chain. This complexity is also reflected in the number and diversity of risk assessment tools currently available. EFSA has conducted several MRAs for which a model was designed to fit a particular question, while other models may become generic tools. In parallel to this, progress has been made in measuring the impact of foodborne diseases on the human population and to use this information to prioritise risks. Work is also being undertaken to deal with uncertainty in these MRAs in a more structured way; this is challenging, particularly when communicating risk estimates to risk managers and the general public. The session will examine topics at the forefront of MRA, looking at lessons learned from applying current methodologies for risk assessment. Aspects that will be considered are the ranking of microbiological risks, rapid risk assessments throughout the food chain, examining the methodological challenges posed and the opportunities that lie ahead.

CHAIRS

Steve Hathaway — *Ministry for Primary Industries, New Zealand*

Following an early academic career in veterinary epidemiology and public health, my interest in practical application of food safety risk analysis in standard development began in the early 1990s and has shaped much of my subsequent work. In the last 10 years I have become increasingly involved in biosecurity and in 2007 I was a major contributor to the FAO Biosecurity Tool Kit that drew strong parallels in application of risk analysis in both the food safety and biosecurity sectors. I have had extensive involvement in the CAC, OIE and FAO in their development of international standards and guidelines on food safety risk analysis. Judgement of the equivalence of different food safety measures in different countries has been a particular interest. Most recently, I have been involved in development of risk models for Trichinella spiralis *in pigs and* Taenia saginata *in cattle as the basis for new risk-based Codex standards. I am chairman of the Codex Committee on Milk and Milk Products and I am a member of the OIE Animal Production and Food Safety Working Group which co-ordinates and informs work on food-borne zoonoses. My current position is Director, food safety and biosecurity science and risk assessment, in the Ministry of Primary Industries in New Zealand. This Directorate primarily provides scientific and risk assessment advice on food safety, animal, plant and nutritional hazards to risk managers in MPI, the Joint Australia New Zealand Food Standards system and external organisations. This sometimes involves a high level of scientific involvement in investigation and response. We are also responsible for scientific substantiation of health claims for foods. Our work is underpinned by an extensive operational research programme whereby we contract external science providers both in New Zealand and overseas to fill gaps in scientific knowledge. My publication record illustrates specific interests in application of a risk-based approach to meat hygiene, control of* Campylobacter *in poultry and control of STEC in cattle. I have worked as a risk analysis consultant for several competent authorities and in 2014 I very much enjoyed working with national experts to draft a new biosecurity policy for the Nepalese government to cover animal, plant and public health.*

Birgit Nørrung — *University of Copenhagen, Denmark*

Birgit Nørrung, DVM, PhD, School Director, School of Veterinary Medicine and Animal Science and Head of Department of Veterinary Disease Biology, Faculty of Health and Medical Science, University of Copenhagen. Birgit Nørrung has published more than 120 papers/proceedings/reports/book chapters in the area of microbiological food safety. Her main research interest is applied research related to risk assessment and control of zoonotic microorganisms and foodborne viruses in the food chain. Birgit Nørrung has great experience with provision of scientific advice and has been a member of EFSA´s Scientific Panel on Biological Hazards for a total of nine years and a member of EFSA´s Scientific Committee for three years.

RAPPORTEURS

Winy Messens — *European Food Safety Authority (EFSA), Italy*

Winy Messens is currently Senior Scientific Officer, Food Microbiologist in the Biological Hazards and Contaminants Unit (BIOCONTAM Unit) of EFSA. In 2010 she moved to Parma to support the work of EFSA's Panel on Biological Hazards (BIOHAZ Panel). She graduated in 1994 as bio-engineer, option Food Technology, at the Faculty of Agricultural and Applied Biological Science of Ghent University (currently the Faculty of Bioscience Engineering), in Belgium. In 2000, she obtained the degree of Doctor in Applied Biological Sciences at the same university on high hydrostatic pressure treatment of cheese. Then she joined the research group Industrial Microbiology and Food Biotechnology (IMDO) at the Vrije Universiteit Brussels (Belgium) for two years coordinating a project on bacteriocin-producing starter bacteria. From 2001 until June 2010, she was scientific attaché at the Institute for Agricultural and Fisheries Research (ILVO, Belgium) where she coordinated several projects related to biological hazards in various foods (such as Salmonella *contamination of hen's eggs and* Campylobacter *contamination of broilers). Before joining EFSA, she was a member of the BIOHAZ Panel for one year.*

Valentina Rizzi — *European Food Safety Authority (EFSA), Italy*

Valentina Rizzi is presently Senior Scientific Officer in the Biological Hazards and Contaminants Unit (BIOCONTAM Unit) of EFSA. Since the beginning of 2015, she has been working in the coordination of the EFSA's team leading the BIOHAZ Panel and the Networks on Microbial Risk Assessment (MRA) and Transmissible Spongiform Encephalopathies (TSE). In 2013 and 2014 she coordinated the EFSA's project on the collection of molecular typing data of some foodborne pathogens. From 2008 to 2014 she worked in the area of zoonoses data collection and in the production of the European Union Summary Reports on zoonoses and foodborne outbreaks. She was involved in several projects on data harmonisation, web reporting and controlled terminology management. From 1998 to 2008, she worked at the Istituto Zooprofilattico Sperimentale dell'Abruzzo e del Molise (IZSAM) in Teramo (Italy), in the Laboratory of hygiene of food of animal origin, non-animal origin and feedstuff, where she was involved in the coordination of research projects, teaching activities for veterinarian and practitioners, laboratory testing of food and feed samples, and consultancy for the implementation of HACCP plans. In 1995 she obtained the degree of Doctor in 'Pathology of small ruminants' at the University of Veterinary Medicine in Pisa, Italy. She graduated in 1991 in Veterinary Medicine at the University of Pisa, in Italy.

KEYNOTE SPEAKER

World Health Organization estimates of the global burden of foodborne disease, 2010

Arie Havelaar — *University of Florida, USA*

Arie Hendrik Havelaar is as a professor in the Animal Sciences Department and the Emerging Pathogens Institute of the University of Florida, Gainesville, FL, USA. Before moving to the United States in 2014, Arie Havelaar worked at the National Institute for Public Health and the Environment, Bilthoven, the Netherlands, and at the Institute for Risk Assessment Sciences, Utrecht University, Utrecht, the Netherlands, to which he still is affiliated. His research focuses on quantitative approaches to foodborne diseases and prevention. Recent activities on the epidemiology of foodborne diseases include estimating the true incidence of foodborne illness, attribution of human disease to food and other pathways, estimating the disease burden using Disability Adjusted Life Years as a summary metric of public health and estimating cost-of-illness. Quantitative microbial risk assessment studies include method development with a special interest in dose-response modeling, the impact of acquired immunity and uncertainty analysis. Farm-to-fork modeling of pathogens in animal food chains is a basis for evaluating the public health impact of interventions, cost-benefit and risk-benefit analysis, and decision support modeling. Arie Havelaar will participate in the Global Food Systems Institute at the University of Florida with a special interest in emerging zoonotic risks in relation to (global) food chains and their interaction with drivers of change (climate change, globalization, demography, technology, consumer demands, regulations etc.).

Illness and death from diseases caused by contaminated food are a constant threat to public health and a significant impediment socio-economic development worldwide. Recognizing the absence of global and regional estimates of foodborne diseases and the need for such estimates to guide public health policy, the World Health Organization (WHO) launched the 'Initiative to Estimate the Global Burden of Foodborne Diseases' in 2006 and established the Foodborne Disease Burden Epidemiology Reference Group (FERG) in 2007.

FERG will provide the first estimates of the global foodborne disease (FBD) incidence, mortality, and disease burden in Disability Adjusted Life Years (DALYs) caused by 31 foodborne hazards by the end of 2015. These will include bacteria and protozoa causing predominantly acute diarrheal diseases (11 hazards); bacteria and protozoa causing invasive infectious diseases (7 hazards); helminths (3 cestodes, 2 nematodes, 5 trematodes including the broad group of 'intestinal flukes'); and diseases induced by 3 chemical hazards. Estimates of incidence, duration, mortality and sequelae were based on systematic reviews, complemented with other literature sources and expert inputs. For each hazard that was not considered 100% foodborne, the proportion of the disease burden caused by foodborne transmission was estimated from a structured expert elicitation. Selected results from reviews underlying FERG estimates will be presented.

The diversity of foodborne hazards and regional differences in priorities suggest the need for consideration of these estimates at the national or even subnational level. As one of its aims FERG has fostered national studies of the burden of FBD, and four pilot studies have been conducted. The estimates developed by the WHO initiative will be invaluable for countries where local data gaps inhibit the development of a full picture of FBD.

Due to data gaps and limitations, the FERG estimates are conservative, yet it is apparent that the global burden of foodborne diseases is considerable, affecting individuals of all ages, but particularly children under 5 years of age and those living in low-income regions of the world. By incorporating these estimates into policy development at national and international levels, all stakeholders can contribute to improvements in safety throughout the food chain.

SPEAKERS

Methodology and uncertainty impact on risk ranking of microbiological hazards: present and future

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 BS degree in Agriculture Engineering from the Agricultural University of Athens, Greece, in 1997 and PhD (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 the EFSA Panel on Biological Hazards. 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 cantered on the microbiological quality and safety of fresh and processed food products, predictive microbiology, microbial risk assessment, stochastic modelling 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.

In a science-based food safety management system, resources should be deployed in a manner that maximizes the public health benefit achieved through risk reduction. Risk ranking has been recognized as the proper starting point for risk-based priority setting and resource allocation, because it would permit policymakers to focus attention on the most significant public health problems and develop strategies for addressing them. An overview of the recent activities of the EFSA Panel on Biological Hazards (BIOHAZ Panel) on risk ranking will be presented with specific emphasis on two recent opinions related to the development of a risk ranking framework on biological hazards and a risk ranking toolbox for the BIOHAZ Panel. The scope of the first opinion is to develop a standardised risk ranking conceptual framework to ensure consistency and transparency. This framework shows how the interaction between the risk managers and the risk assessors in the definition of the risk ranking purpose and the communication of the risk ranking results should be encouraged. It also provides the ability of adopting the appropriate risk ranking methodology by selecting different options at each stage. In the second opinion, available tools relevant to risk ranking of biological hazards in foods are identified and assessed using two examples. A systematic approach for the evaluation of the performance of risk ranking methodologies is also presented through a case study on the comparison between stochastic, deterministic and ordinal scoring approaches. In addition, methodologies for the identification of the most important uncertainty sources and their incorporation in risk ranking models are discussed.

Improving the usability and communicability of burden of disease methods and outputs: the BCoDE toolkit application

Alessandro Cassini — *European Centre for Disease Prevention and Control (ECDC), Sweden*

Alessandro Cassini is a medical doctor by training, with a specialisation in Public Health, Epidemiology and Preventive Medicine at 'la Sapienza' University in Rome and an MSc in Health Policy Planning and Financing from LSE and LSHTM. Before joining ECDC in 2009, he worked for a private consultancy firm in London, advising mainly on matters related to market access, health technology assessment and overall appraisal of unmet needs. Before becoming Expert Antimicrobial Resistance and Healthcare-associated Infections last year, Alessandro has led the Burden of Communicable Diseases in Europe (BCoDE) project with the remit of estimating and expressing the burden of communicable diseases and related conditions by means of composite health measures (e.g. DALYs). Alessandro's main areas of interest are infectious disease epidemiology, assessment of surveillance systems, economic impact and forecasting of infectious diseases, social and economic determinants of health, disease threat risk ranking. Moreover, Alessandro enjoys relating his epidemiological competencies with health policy decision-making by researching ways to bridge the communication and technical gap between risk assessors and managers (knowledge translation). Hence, the implementation of enhanced visualization tools embedded in the BCoDE toolkit. Occasionally, Alessandro goes back to the field and has been recently involved in two missions in West Africa in response to the Ebola outbreak.

Burden of disease methodologies through computation of evidence-based composite health measures allow for estimation and prioritisation of their impact on population health. The Burden of Communicable Diseases in Europe (BCoDE) is a project led and funded by the European Centre for Disease Prevention and Control (ECDC) with the purpose of estimating in DALYs the impact of communicable diseases (CDs), including foodborne diseases. Further objectives include informing and empowering stakeholders for the comprehension and use of composite methodologies. A stand-alone application for calculation of DALYs was developed in C++ using Qt C++ toolkit, version 4.8.4. All computations are implemented in C++ and the interface is HTML with JavaScript. Each selected communicable disease generates a model visible as a graphical outcome tree and based on literature reviews concerning its natural history. By default, users input age-group and gender-specific annual cases of disease from a preferred data source. Possible editions include inputting multiplication factors adjusting for underestimation, population data, life expectancy and any parameter of the outcome tree (disability weights, transition probabilities and durations). Further development of the tool will allow building a custom outcome tree. BCoDE toolkit outputs are bestowed through interactive graphs and tables, and present crude and per 100 000 incidence, deaths, DALYs, years of life lost due to premature mortality (YLL) and years of life lived with disability (YLD), with uncertainty intervals. Disease specific results include impact of acute illness versus sequelae, gender and age-group estimates. Aggregated results are displayed as bubble charts (DALYs per 100 000 population) plotted against mortality, incidence, and DALYs per case. Interactive tables and bar charts ranking diseases and uncertainty are produced and exported. The aim of the software is to assist users in applying the proposed BCoDE evidence-based approach for estimation of the burden of infectious diseases, including for risk ranking purposes and disease prioritisation. Eventually, choices concerning data input allow national experts to assess availability and quality of data, identify gaps, and generate additional information on national surveillance systems. Moreover, multiple visualisation options focus on facilitating communication between data generators and users, ultimately fostering its value in health policy formulation.

The contribution of typing methods to risk assessment

Flemming Scheutz — *Statens Serum Institute, Denmark*

Flemming Scheutz is the head of the World Health Organization (WHO) Collaborating Centre for Reference and Research on Escherichia *and* Klebsiella *since 1993. The centre functions as the Danish National Reference Centre for typing of* E. coli *and* Klebsiella*. His main interests are identification, characterization, subtyping, and epidemiology of pathogenic* E. coli*, including Diarrhoeagenic* E. coli *and Extraintestinal pathogenic* E. coli *(ExPEC). The centre is contracted as the microbiological support laboratory for the European Centre of Disease Prevention and Control (ECDC) and organizes the annual External Quality Assurance (EQA) programmes for the serotyping and virulence typing of VTEC. Recent EQA programmes have focussed on the application of a vtx subtyping protocol developed in collaboration with six international research and reference laboratories (9). Using this vtx subtyping protocol his team has been able to demonstrate that HUSEC is almost exclusively associated with subtype vtx2a (I). The WHO Centre has a large strain collection of approximately 75,000* E. coli *strains, most of which are clinical isolates representing almost any possible sero- and virulence type as far back as 1951. These isolates can be provided for specific research projects to study the evolution and virulence factors of pathogenic* E. coli*. The most recent examples include the discovery that ExPEC strains associated with urinary tract infections (UTI) can harbour EAEC genes (6,7). Ongoing Research Primary Investigator (PI) in a project entitled 'A New Virulence Repressor of Gram-Negative Bacteria: Potential Protection Against Disease'. The Danish Council for Independent Research, Medical Sciences; together with Post Doc Nadia Boisen. This project includes whole genome sequencing (WGS) of a minimum of 100 Enteroaggregative* E. coli *(EAEC) strains from different parts of the world. Development of web tools for characterization of* E. coli *strains using WGS, together with PhD student Katrine G.*

To assist in the assessment of the ranking of microbiological risks associated with different *E. coli* pathotypes they have been classified into Extraintestinal Pathogenic *E. coli* (ExPEC) associated with extraintestinal infections (urinary tract infection, sepsis/meningitis) and Diarrhoeagenic *E. coli* (DEC) associated with enteric/diarrhoeal disease. DEC are subdivided into different pathotypes based on their adhesion/colonization mechanism and the toxins produced: Enteropathogenic *E. coli* (EPEC), Enterotoxigenic *E. coli* (ETEC), Enteroinvasive *E. coli* (EIEC), Enteroaggregative E. coli (EAEC), Shiga toxin (Stx)-producing *E. coli* (STEC) (or Vero cytotoxin (VT)-producing E. coli (VTEC)), Diffusely Adherent *E. coli* (DAEC), and Adherent Invasive *E. coli* (AIEC). For the majority of these pathotypes a minimum number of factors (molecular and/or phenotypic) can be used to define the pathotype. The plasticity of the *E. coli* genome combined with the ability to acquire and exchange mobile genetic elements and cassettes can result in crossover strains, for which it may be difficult to predict the risk. A plethora of different methods are in use and a major challenge in the characterization of *E. coli* is the comparability of the results generated by these different techniques and methods. STEC is the pathotype most often associated with food borne outbreaks and severe disease. To assist in the clinical and public health risk assessment of STEC, a basic and primary definition of HUSEC (HUS associated *E. coli*) for first line public health action is proposed. Accurate and rapid typing of pathogens is essential for effective surveillance and outbreak detection. *E. coli* sero- and virulence typing can now be performed solely from Whole Genome Sequencing (WGS) data, providing faster and cheaper typing than current routine procedures. WGS-typing is a superior alternative to conventional typing strategies and foreseen to greatly improve our ability to perform rapid risk assessments throughout the food chain.

Challenges in risk assessment for viruses

Marion Koopmans — *Erasmus MC Viroscience Department, Netherlands*

Marion Koopmans (DVM, PhD) is Head of the Department of Viroscience at Erasmus MC Rotterdam, the Netherlands. She is also Professor of virology at the Laboratory for Infectious Diseases of the National Institute of Public health. Her responsibilities include reference diagnostics, syndrome surveillance and emergency preparedness for viral diseases, including research aimed at improving the response capacity of a public health lab. Her research interests focus on enteric viruses, food-borne infections, emerging disease preparedness and infections at the human-animal interface, with a particular focus on unravelling mechanisms underlying possible emergence of new health threats and optimizing the early detection and response. She is initiator and coordinator of a network of laboratories with responsibility for norovirus surveillance that agreed to share data and sequences internationally. She has authored over 300 papers in peer reviewed journals.

The recent estimate from the World Health Organization (WHO) has concluded that viruses contribute substantially to the global burden of food-borne disease. In addition, viral disease outbreaks can impact heavily on the food sector, resulting in shifts in the market with potential consequences, as for instance observed when new influenza viruses emerge. The currently recognized priority food-borne pathogens are noroviruses and hepatitis A, but additional viruses continue to be detected. Globalization of food and travel, coupled with rapid increases in the global population and the resulting pressure on the environment has its downsides in the form of emerging infectious disease outbreaks, often arising from spillover from a zoonotic reservoir. Food production – handling and – preparation are recognized risk factors for such incursions, and may lead to food scares even if the primary mode of transmission of novel pathogens may not be by the fecal oral route. Current food quality control monitoring relies on bacterial indicators, as virus detection methods are technically demanding and not yet widely available. An additional challenge is the highly variable nature of (emerging) viral diseases, requiring rapid adaptation of detection methods with the need for careful revalidation of food detection methods. Risk assessment for (emerging) viruses has many parallels with that for other pathogens, but specific challenges are the lack of routine surveillance in humans and products to provide data for attribution, the lack of cell culture systems to address questions about stability and infectivity, and the great viral diversity of which the clinical impact is only partly understood. This is true for the known endemic foodborne viruses, but particularly challenging when new viruses emerge. Ideally, the potential public health risks of newly recognized viral diseases should be assessed prior to species jumps, and some efforts have been done to provide such assessments for novel influenza viruses. Recent examples, however, have confirmed the unpredictable nature of viral diseases, and the challenge remains.

Approaches to deal with uncertainty in emergency assessments: the case of the EHEC outbreak in 2011 in Germany

Gordon Müller-Seitz — *Technical University Kaiserslautern, Germany*

Gordon Müller-Seitz is Chair of Strategic Management, Department of Business Studies and Economics at the TU Kaiserslautern, Germany. His research and consulting focus on dealing with risks and uncertainties in the case of disease outbreaks, technological/innovative disruptions as well as collaborating and coordinating in interorganizational emergency networks and projects. His work has been applied at globally operating organizations and has appeared in research journals and practitioner outlets.

Based upon two recent studies on the EHEC outbreak I was involved in (Müller-Seitz 2014; Berthod et al. 2014), I contrast calculable risks from uncertainty – the latter understood here as the unexpected and non-calculable as underresearched phenomenon despite its societal and organizational relevance and omnipresence. I elaborate upon how organizations practise uncertainty in the face of large-scale outbreaks of disease in Germany between 2000 and 2012 with an emphasis on the EHEC outbreak in 2011. Towards this end, I identify two overarching forms of practice, namely, reducing (i.e., coping with unforeseen incidents) and inducing (i.e., championing an overarching cause) uncertainty. I show that actors use both these forms of practice, which constitute the basis of the framework introduced herein, intentionally or unintentionally depending on the differing and sometimes conflicting objectives of the organizations involved.

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Müller-Seitz G, 2014. Practising uncertainty in the case of large-scale disease outbreaks. Journal of Management Inquiry, 23(3): 276-293.

Drivers for emerging issues in animal and plant health

The history of agriculture includes many developments related to animal and plant health that have had a major impact on humans. At the same time, human activities have often driven the appearance of emerging issues. The more humans expand the footprint of the global population, encroach onto natural habitats, alter these habitats to extract resources and intensify food production, and move animals, people and commodities and the pathogens they carry, the greater the potential for infection and spread of pathogens and pests to emerge or re-emerge. Producing food plays a major role in this. The risk of emergence of pests and pathogens has increased as a consequence of global changes in the way food is produced and consumed. In addition, climate change is likely to increase pressure on the availability of food. It will also provide newly suitable conditions for invasive species, pests and pathogens. Population displacements due to economic, political and humanitarian crises represent another set of potential drivers for emerging issues. The overlapping drivers of human, animal and plant diseases and environmental changes point towards the concept of 'One Health'. It underlines the urgent need to understand the influence of human behaviour and incorporate this understanding into our approach to emerging risks. For this we face two major challenges. One is cultural; the second is methodological. We have to look at systems not under the narrow angle of specific hazards but for their dynamics, and a spectrum of potential outcomes in terms of risk. And we have to make sense of the vast amounts of data that are available in the modern age. The aim of this session is to prepare for the cultural and methodological shifts needed in our approach to emerging risks.

CHAIRS

Guy Poppy — *Food Standards Agency and University of Southampton, UK*

Professor Guy Poppy took up his role as the FSA's Chief Scientific Adviser in August 2014. He will also continue with his research in global food security at the University of Southampton, where he is Professor of Ecology and the Director of Interdisciplinary Research. Professor Poppy has significant research experience in food systems and food security and has advised governments around the world on these issues. He has published over 100 peer-reviewed papers including a number of highly cited articles on risk assessment, risk analysis and risk communication. He is currently a member of the Research Excellence Framework (REF2014) panel assessing the quality of agriculture, food and veterinary science in the UK. A graduate of Imperial College and Oxford University, Professor Poppy previously worked at Rothamsted Research, becoming Principal Scientific Officer. He left in 2001 to join the University of Southampton where he has been Head of Biodiversity and Ecology and, more recently, Head of Biological Sciences. As the FSA's Chief Scientific Adviser, Professor Poppy will provide expert scientific advice to the UK government and play a critical role in helping to understand how scientific developments will shape the work of the FSA as well as the strategic implications of any possible changes.

Jan Schans — *Netherlands Food and Consumer Product Safety Authority (NVWA)*

Jan Schans has been a senior officer for plant health at the National Plant Protection Organization (NPPO) of the Netherlands for 18 years. As of 2015, he is senior advisor for food chain risk analysis at the Office for Risk Assessment and Research at the Netherlands Food and Consumer Product Safety Authority (NVWA). He has worked on many aspects of plant health, notably pest risk assessment and the evaluation of risk reduction options, design and coordination of the national phytosanitary surveillance program, and the coordination of research for plant health development. He has been a member of the EFSA Panel on Plant Health for 9 years, serving as the Chair of the Panel in its first mandate (2006–2009), and a member of the EFSA Standing Working Group on Emerging Risks. He received an MSc in Plant pathology (1983) and a PhD in Agricultural and Environmental Sciences (1993), both from Wageningen University, the Netherlands.

RAPPORTEURS

Franck CJ Berthe — *European Food Safety Authority (EFSA), Italy*

Franck Berthe is Head of the Animal and Plant Health Unit at EFSA, based in Parma, Italy. Within the Department for Risk Assessment and Scientific Assistance, the core activity of his group is to assess animal and plant health in relation to production systems and farming practices. His work connects primary production, ecosystem and public health. Franck's group provides scientific advice to the European Union risk managers and decision makers on a wide range of risks at the human-animal-ecosystem interface. The contributions of the Animal and Plant Health Unit on Q-fever, Schmallenberg, bluetongue virus and Xylella fastidiosa emphasise the importance of emerging risk along the food chain. Franck's group currently works on drivers of the Ebola virus spill-over from animal compartment. Prior to coming to Italy in 2007, Franck was Associate Professor at the Atlantic Veterinary College (UPEI) and Canada Research Chair in Aquatic Health Sciences. His multi-disciplinary program aimed at exploring host pathogens relations in their environment. From 1994 to 2004, he has led active research in aquatic animal health at the French institute for the exploitation of the sea (IFREMER) in France and overseas territories of French Polynesia and New Caledonia. A native of France, Franck received his doctorate of veterinary medicine degree from the Veterinary School of Maisons-Alfort, and a PhD degree in molecular parasitology from the University Blaise Pascal, Clermont Ferrand. He has a diploma in bacteriology (molecular taxonomy and epidemiology) from the Pasteur Institute, Paris. Franck's contribution was recognised by an OIE Meritorious Award in 2012.

Katinka De Balogh — *Food and Agricultural Organization (FAO), Italy*

Katinka de Balogh studied veterinary medicine in Berlin and Munich and obtained her doctorate in tropical parasitology from the Tropical Institute of the University of Munich in 1984. She specialized in tropical animal production and health in France and in Veterinary Public Health (VPH) in the Netherlands. After a short career as a zoo veterinarian in the Rotterdam Zoo she moved to Africa where she worked for over nine years as a district veterinary officer (DVO) in rural Zambia and lecturer at the veterinary faculties of Lusaka (Zambia) and Maputo (Mozambique). After also having spent two years at the World Health Organization in Geneva, in 1997 she started to work as a lecturer in VPH and international coordinator at the veterinary faculty in Utrecht, the Netherlands. Since 2002, she works at the Food and Agriculture Organization of the United Nations (FAO) in Rome, Italy, and presently leads the VPH activities of the organization and is the FAO focal point on One Health and its linkages with the World Organisation for Animal Health (OIE) and the World Health Organization (WHO).

Stefano Pongolini — *Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia-Romagna, Italy*

Stefano Pongolini, DVM, obtained a post-graduate specialization degree in Avian Pathology from the University of Milan in 1996 and in Veterinary Legislation from the University of Parma in 2002. He is a Veterinary Officer for animal health and food safety at the Institute of Zooprophylaxis of Lombardy and Emilia-Romagna where he is Director of the Section of Parma since 2010. He is the head of the Regional Laboratory of Emilia-Romagna for surveillance of food-borne diseases since 2012 and has been member of the Emerging Risks Exchange Network of EFSA since 2010. His present research interests are in the field of molecular epidemiology of food-borne agents along the food chain and in the area of disease modelling for optimization of surveillance strategies and crisis preparedness in animal health. From 1994 to 2009 he served as Veterinary Officer at the Section on Modena of the same Institute working as microbiologist and pathologist. He had the technical and managerial responsibility of the Regional TSE Laboratory of Emilia-Romagna from 2001 to 2009.

Jane Richardson — *European Food Safety Authority (EFSA), Italy*

Jane Richardson has a degree in Molecular Biology from the University of Edinburgh (1994) and a PhD in Microbiology from Imperial College of Science, Technology and Medicine (2000). She spent three years training as an Analyst/Programmer developing patient management systems for the Welsh Radiology Service. In 2003 she joined the UK Health Protection Agency in the North West of England. In her role as epidemiology and surveillance analyst she worked on both infectious diseases and chemical exposure events, developing expertise in the use of routine health data to support exposure assessments for contaminated land incidents and airborne releases. In 2007 she joined EFSA in Italy. Combining her scientific training with experience gained in an IT environment she has worked as a senior database manager on a number of multinational monitoring and surveillance programmes.

KEYNOTE SPEAKER

People, animals, plants, pests and pathogens: connections matter

William Karesh — *EcoHealth Alliance, USA*

William Karesh is the Executive Vice President for Health and Policy for EcoHealth Alliance. He is also the President of the World Organisation for Animal Health (OIE) Working Group on Wildlife and chairs the International Union for the Conservation of Nature (IUCN) Species Survival Commission's Wildlife Health Specialist Group, a network of hundreds of wildlife and health experts around the world. He serves as the inter-project liaison for the USAID Emerging Pandemic Threats PREDICT-2 program. Dr Karesh has pioneered initiatives focusing attention and resources on solving problems created by the interactions among wildlife, people, and their animals. He coined the term 'One Health' to describe the interdependence of healthy ecosystems, animals and people and the term has been adopted by many organizations, including the United Nations, in local and global health efforts. Dr Karesh has created dozens of initiatives to encourage linkages among public health, agriculture and environmental health agencies and organizations around the world. He has personally lead programs and projects in over 45 countries, covering terrain from Argentina to Zambia. In addition to his work in the private non-profit sector, Dr Karesh has also worked for the USDA, US DOD and DOI. He serves on the World Health Organization's Roster of Experts, as a consultant for the Food and Agriculture Organization of the UN, and on the 4-person Steering Committee of OFFLU (OIE-FAO Network of Expertise for Influenzas). Dr Karesh is internationally recognized as an authority on the subject of animal and human health linkages and wildlife. He has published over one 160 scientific papers and numerous book chapters, and written for broader audience publications such as Foreign Affairs and The Huffington Post.

People, animals, plants, pests and pathogens: connections matter. Nowhere in the world are the health impacts from both emerging and endemic zoonotic diseases more important than in developing countries, where daily work and livelihoods are highly dependent on natural resources. With globalization and international travel, disease movement is now rapid and the natural barriers to disease spread from points of origin are becoming meaningless. A significant proportion of natural or agriculturally based resources harvested or produced in developing countries are further processed or consumed in more economically advanced countries, providing regular routes for hitch-hiking organisms. Many developing countries have little to no capacity for diagnosis of endemic zoonoses nor for detecting disease emergence from wildlife and domestic animals prior to spread to humans. Meanwhile, zoonotic diseases are estimated to continue to cause over one billion cases of human disease annually. While the linkages of human, animal, and environmental health is at the heart of the One Health approach, an increasingly important prism through which governments, NGOs, and practitioners view public health, we still have three critically important challenges facing us: 1) we need a much broader and deeper knowledge of what underlies disease emergence and spread, 2) we need to better target our surveillance efforts to maximize available resources, and 3) we need to greatly improve awareness and local capacity for preventing endemic and emerging zoonoses.

SPEAKERS

Relations between pathogens, hosts and environment: joining the dots

Matthew Baylis — *University of Liverpool, UK*

Matthew Baylis studied Zoology at Oxford University, followed by a PhD, also at Oxford, on the ecological interactions of lycaenid (blue) butterflies and ants. This work was undertaken in Australia and Princeton University in the US. His first postdoctoral post was at the Tsetse Research Laboratory, University of Bristol, but he was permanently seconded to Kenya, where spent 4 years in the wilderness of Galana Ranch near Malindi. In 1993 he joined the Institute for Animal Health, Pirbright Laboratory, to study African horse sickness in Morocco and, later, southern Africa. In 1998, now at the Compton Laboratory, he started to work on TSEs (scrapie) in sheep in the UK. In 2003 he became the head of the Division of Epidemiology in the Institute for Animal Health. In 2005 he took up the Chair in Veterinary Epidemiology at the University of Liverpool. With fellowship funding from the Leverhulme Trust, in 2007 he established the Liverpool University Climate and Infectious Diseases of Animals group (LUCINDA). LUCINDA is a multidisciplinary group of postgraduate and postdoctoral researchers investigating how climate change will impact on infectious diseases; with a broad focus on bluetongue, mosquito-borne arboviruses and helminths. Research by the LUCINDA group led to the creation of the Enhanced Infectious Diseases Database (EID2). In 2010. Dr Baylis became head of the Department of Epidemiology and Population Health in the Institute of Infection and Global Health.

Human food-borne pathogens are microbes present in food-producing animals or the wider environment that we acquire by eating contaminated food. Understanding the risks and drivers of emergence of food-borne pathogens requires, therefore, extensive knowledge of the types of microbe present in animals and the environment, which might become infectious to humans. This is, however, sadly lacking, as there is significant under-representation of animal and environmental pathogens in the scientific literature. For example, studies

of pathogen diversity find over forty percent more pathogen species known in humans alone than in up to fifty domestic animal species combined. A key research challenge is to identify the characteristics of animal pathogens that give them the propensity to be infectious to humans – in other words, to be zoonotic. Are some pathogen types (such as bacteria, viruses) more or less likely to spread from animals to humans? Are we humans most likely to acquire zoonotic pathogens from the animals that we eat (livestock), the ones we share our homes with (pets) or the ones we are most similar to genetically (primates)? And is the transmission route important? Are we more likely to share food-borne pathogens with animals, than those that are transmitted by direct contact, by aerosol or by sexual contact? A novel pathogen/host database, the Enhanced Infectious Disease Database (EID2) is being used to address such questions. Data on pathogens and hosts are acquired using automated procedures from online data sources, such as the metadata uploaded with gene sequences, and the abstracts of biomedical papers. At present the database holds information on 1,606 human pathogens, of which half are zoonotic, and 1,038 pathogens of domestic animals. Using network analysis, the human and animal hosts (dots) can be joined by the pathogens they share, with the strongest joins for those that share the most pathogens. Joining the dots creates pathogen networks that can then be used to assess which types of host are the major source of pathogens for humans; and the network properties can be compared for different types of pathogen and those with different properties, such as transmission route.

Discovering novel pathways of cross-species pathogen transmission

Tony Goldberg — *University of Wisconsin-Madison, USA*

Tony Goldberg's research and teaching focus on the ecology, epidemiology and evolution of infectious disease. His work combines field and laboratory studies to understand how disease-causing agents are transmitted among hosts, across complex landscapes, and over time. This involves tools ranging from field ecology to molecular epidemiology, combined with methods from the social sciences to understand the root drivers of disease emergence in 'real world' settings. The focus is not only on diseases emerging in the world today, but also on predicting the conditions under which future diseases might emerge. Most of Dr Goldberg's projects are place-based, focusing on well-defined ecosystems at fine spatial and temporal scales. This includes numerous projects around the world that use evolutionary and epidemiological tools to track the movement of pathogens, from viruses to bacteria to protozoa and fungi. The overall goal of is to discover generalized mechanisms that govern pathogen transmission, evolution, and emergence, and to improve the health and well being of animals and humans while helping to conserve the rapidly changing ecosystems that we share.

Emerging methods in epidemiology have the potential to inform a comprehensive assessment of how pathogens might move from natural settings into and throughout human societies. The search for new disease transmission pathways could be independent of the pathogens themselves, which may be known, lurking undiscovered in reservoirs, or not yet even evolved. Moreover, multiple pathogens probably traverse common transmission pathways, such that disrupting these pathways would have public health impacts beyond individual diseases. The benefit would be a level of 'pandemic preparedness' not fully attainable by other approaches. Many undiscovered transmission pathways involve human cultural practices that bring people into contact with reservoirs or vectors at times in places that are especially suitable for subsequent human-to-human transmission. Some pathways may not presently be traversed by any known pathogens, making them invisible to traditional epidemiological approaches. To discover them would require a re-focusing of efforts by epidemiologists and social scientists working together.

Broad brush analysis of livestock disease drivers, ecology and pathogen evolution

Jan Slingenbergh — *Independent advisor*

Jan Slingenbergh, a veterinarian with postgraduate training in parasitology and entomology, employs himself for disease ecology projects, as Independent Advisor, since May 2012. Before that, he was Head of the FAO Emergency Prevention System for animal health. During his 20+ years in FAO HQs in Rome he also formed part of the Influenza management team, created early 2004 to face the H5N1 HPAI panzootic. Prior to that he acted as the focal point in the Secretariat of the AU/FAO/IAEA/ WHO Programme Against African Trypanosomoses and, also, led the FAO international screwworm programme. His main field work was in Sub-Saharan Africa: Nigeria, Benin, Mozambique and Ethiopia. His scientific work covers the ecology of infectious and parasitic diseases. In this regard, he played a significant role in clarifying the important role of rice-duck farming in Eastern and South-Eastern Asia in the epidemiology of avian influenza. He also established the details of the Eurasian small ruminant street, a major dispersion route for FMD and other pathogens connecting South Asia with West Asia/Europe. A more recent highlight concerns his work as editor and main author of the 2013 FAO flagship publication 'Changing Disease Landscapes'. His current focus is to establish and predict how pathogens with different trait profiles and corresponding disease ecology each respond differently to the human induced alterations in the pathogen-host-environment interplay. Findings comprise the rationale for the enhanced role of animal viruses as infectious disease agents and of bacteria as the cause of a growing number of food hazards.

With animal origin pathogens posing a growing number of public health and food safety concerns, the disentangling of the underlying disease ecological dynamics has become a matter of some urgency. Here, I argue that while livestock viruses affecting respiratory and enteric tract epithelia may readily turn more pathogenic in the face of livestock mass-rearing, the long term evolutionary pull remains towards an ever increasing intimacy with the host, with pathogenic viruses interfacing the diversity of commensalistic and endoviruses. Intimate pathogenic viruses may circumvent the outer defence lines, cause quasi-inapparent infection and yet infiltrate inner-body organs and vital systems, causing life-long infections that are vertically transmitted, selecting for ever greater host specificity. Host radiation thus appears more of a feature typical for opportunistic myxoviruses. However, given the enhanced ecological perturbation at the interfaces of the livestock, wildlife and human host domains, long term inter-domain and inter-species barriers are breaking down, permitting spill-over and species jumps also by the intimate pathogenic viruses. Livestock bacteria reside chiefly on the skin and mucosal tracts, supporting the host health rather than being harmful. Novel forms of clinical disease may appear when a bacterium succeeds in infiltrating inner-body environments, with virulent Q fever emerging in intensive goat dairy systems as a main, recent example. Arguably more important still is the evolution of new strains and toxines showing up in the enteric tract environment of fast growing food animals. Also the use of antibiotics as growth promoter interferes with the functioning of the enteric tract microbiome, metabolism and immune system. Indeed, most modern livestock diseases and food safety challenges appear the result of an intensification process that is driven too far.

Horizon scanning for emergence of new viruses in animal and public health

Paul Gale — *Department of Epidemiological Sciences, Animal and Plant Health Agency (APHA), UK*

Paul read Biochemistry and studied membrane protein interactions using spectroscopy for his D.Phil. For 13 years he worked in the water industry and environment fields developing risk assessments for pathogens in drinking water, sewage sludge and composts. In 1997, he represented the Environment Agency as an expert witness at a Public Inquiry on BSE in drinking water. He developed a risk assessment for exotic livestock viruses in composted catering waste for the Animal By-Products Regulations 2003. Since 2006 he has worked at the APHA where

his interests include factors influencing the emergence of new pathogens. In addition to continuing his work on pathogen risks from waste recycling he has led a work package within a European project to map the impact of climate change on the risk of Crimean-Congo haemorrhagic fever. In 2010, he convened a workshop with Royal Holloway, University of London, and funded by the European Science Foundation to develop new approaches for horizon scanning for viruses in animal and public health. He has also developed various risk assessments focusing on the entry of livestock viruses into GB.

Horizon scanning, as applied here, is the approach to predict the next virus to emerge in animal and/or public health, both in terms of its route(s) of transmission and its origins (i.e. country and source reservoir). The emergence of a pathogen generally involves a combination of events together with a change in key drivers, typically socio-economic, environmental, climatic and/or zoological factors. Central to horizon scanning, therefore, is the construction of 'complex scenarios'. In a novel approach developed at a European Science Foundation-funded workshop in 2010, complex scenarios in the form of 'spidergrams' were produced by randomly linking factors which may directly or indirectly affect the emergence of viruses. The factors were chosen from a database under eight header categories (as defined by the workshop's participants). Many thousands of scenario chains can be produced by this method and most may be irrational. However, the approach enables the testing of combinations not previously considered but which would be tested in nature. While it may not be possible to develop quantitative risk assessments for each combination, the approach provides a discussion focus for scientists of different disciplines and may help address identification of 'unknown-knowns' and even 'unknown-unknowns'.

Mapping complexity: visualising a world of change

Tommaso Venturini — *Sciences Po médialab, France*

Tommaso Venturini is associate professor and research coordinator at the Sciences Po médialab. He is the leading scientist of the projects EMAPS (www.emapsproject.com) and MEDEA (projetmedea.hypotheses.org) and his research activities focus on digital methods, Controversy Mapping, Social Modernization. He teaches Controversy Mapping (http://controverses.sciences-po.fr/archiveindex/), Digital Methods, Data Journalism and STS at graduate and undergraduate level. He has been trained in sociology and media studies at the University of Bologna, completed a PhD in Society of Information at the University of Milano Bicocca and a post-doc in Sociology of Modernity at the Department of Philosophy and Communication of the University of Bologna. He has been visiting student at UCLA and visiting researcher at the CETCOPRA of Paris 1 Pantheon Sorbonne.

Controversy mapping is a teaching/research methodology developed in the field of STS (Science and Technology Studies) to deal with the growing intricacy of socio-technical debates. The aim of controversy mapping is to open up the black-boxes of science and technology and expose the complexity of their construction. Not to debunk them, but to show the amazing amount of work required to build them and to associate more and more actors to such work. The political aim of controversy mapping is to provide innovative methods for approaching scientific and technical disputes. Instead of worrying about the fact that the public is exposed to disagreement, controversy mapping asks what advantages can be drawn by rendering controversies more 'readable'. Instead of worrying about how science might be contaminated by political interference and lamenting the fragmentation of society, controversy mapping asks under what conditions can public intervention enhance scientific discussion and what tools can be harnessed to help citizens navigate controversies. The metaphor of mapping is itself controversial. Though few of their pages resemble to standard geographical maps, controversy atlases have the same objective of classic travel atlases and they struggle with the same difficulties. They aim at providing as much detail as possible on a region while remaining compact and legible; at respecting the unique characteristics that define a territory, while translating them in a standard visual language. Only, the objects that these atlases are not (in most cases) geographical territories, but rather issues or matters of concern. In this sense, the words 'atlas' and 'map' should be taken somewhat metaphorically (in the same way as one can talk about botanical or medical atlas). Not so much because our territories are discursive more than geographical, but because the quality and the standardization of our maps is still far from that of proper atlases. Such objective is pursued – by collaborating with experts from different camps in the debate, – by exploiting digital data and computation to follow the weaving of techno-scientific discourses – and by using design to make such complexity readable for a larger public. Related research projects: MACOSPOL (mapping controversies on science for politics) 2007–09, EU FP7; DMI (Digital Methods Initiative) – MEDEA (mapping environmental debate on adaptation) 2011–14, ANR CEP&S; FORCCAST (formation à la C.C. pour l'analyse de science).

A vision for a global operations room

Mike Catchpole — *European Centre for Disease Prevention and Control (ECDC), Sweden*

Mike Catchpole is a medical doctor who has worked in infectious disease epidemiology and response at the national and international level since 1991. He is Chief Scientist at the European Centre for Disease Prevention and Control (ECDC). Prior to that he was Director of Public Health at England's national Centre for Infectious Disease Surveillance and Control, and was the UK member of the ECDC Advisory Forum from 2007 until 2014. He has 20 years' experience of management of communicable disease surveillance and response, including leadership of the national epidemiological response to the 2009 influenza A(H1N1) pandemic in England, developing and managing the surveillance systems for the 2012 London Olympics and the surveillance systems that have been instrumental in driving the dramatic reductions in MRSA and C. difficile *in England. His primary research interests have included HIV and other sexually transmitted infections, the wider health effects of major incidents, and public health information systems development. He has also been a member of the steering groups for a number of European projects, and chaired the Steering Committee of the European Programme for Intervention Epidemiology Training (EPIET) from 2001 to 2006. He has academic appointments, as a visiting professor, at Imperial College London and City University London*

The need and the expectation that control and prevention will involve coordinated action across scientific, organisational, geographical, and political boundaries is greater today than at any time before. It is now possible for a person to travel from one side of the globe to the other in less than the time that it takes for most infectious diseases to become symptomatic following exposure to a source of infection. The experience of SARS, the 2009 influenza pandemic, Middle East Respiratory Syndrome coronavirus (MERS-CoV), and gastrointestinal outbreaks such as that associated with sprouting fenugreek seeds in Germany, has demonstrated the need for rapid and coordinated international action to control outbreaks and emerging infections. International legislation and agreements, such as the EU Decision on Serious Cross-Border Threats to Health (2013) and the International Health Regulations (2005), reflect the need for formalised threat detection and response coordination arrangements at the international level. This presentation sets out a vision for how such coordination could be delivered at the global level, through a global operations room. The purpose of a global operations room would be to ensure that appropriate decisions are made and appropriate actions taken in response to emerging threats. The function would be to bring together the appropriate information and the appropriate expertise, and to provide the technical and organisational infrastructure to support threat detection and response coordination. There are many questions and challenges that need to be addressed in setting up such an operations room, including: the establishment of the political mandate and mutual trust required to ensure effective coordination between national and supra-national authorities; creating the network of expertise required to enable the operations room to deal with the full range of threats requiring global coordination; developing the technical infrastructure for threat detection and communication, and for coordination of response; and standard operating procedures for escalation and de-escalation of threat response, and for command and control arrangements in risk management. The presentation will provide examples of operations rooms models adopted or proposed by national international authorities, and consider the technological opportunities and issues related to the detection and response to threats at a global level.

Advancing environmental risk assessment

Maintaining a healthy environment and conserving biodiversity are major goals of environmental protection, as they contribute to human wellbeing and economic prosperity through the provision of ecological services, including ecosystem services. Biodiversity is also intrinsically valuable and, therefore, worth protecting. Environmental risk assessment (ERA) of regulated stressors such as plant protection products, genetically modified organisms and feed additives is an important safeguard to ensure the desired level of protection of the environment and biodiversity. ERA evaluates the potential adverse effects on the environment of certain actions, and has become an important support to regulatory decision-making. Significant advances have been made in the field in recent years. However, ERA still faces a number of challenges such as the integration of multiple stressors and harmonisation of risk assessment approaches across disciplines. This break-out session explores potential avenues to the further advancement of ERA. The following issues are discussed: harmonising approaches to make protection goals operational; paying greater attention to the relevance and quality of scientific studies to support ERA; and moving towards integrating ERAs to take account of multiple stressors in the environment.

CHAIRS

Helmut Gaugitsch — *Umweltbundesamt (Environment Agency Austria), Austria*

Helmut Gaugitsch studied technical chemistry and food chemistry at the Technical University of Vienna, with graduation as PhD in technical chemistry and food chemistry. He joined the Environment Agency Austria in 1991 and worked on ecological risk assessment, monitoring and coexistence of genetically modified organisms (GMO) since. Presently, he serves as Head of Unit Landuse & Biosafety at the Environment Agency Austria. His main areas of work are: reviewing European GMO notifications concerning risk assessment and monitoring of GMOs; coordination and preparation of studies on environmental effects of GMOs; assistance development and implementation of national, regional and international Regulatory Frameworks for Biosafety; National Focal Point for the Cartagena Protocol; environmental effects of landuse, agriculture and forestry.

Jock Martin — *European Environment Agency (EEA), Denmark*

Jock Martin has been involved in environmental statistics, indicators and assessments since 1991. Initially for six years at the Department for the Environment in the UK, where he was an author of the first national indicator report on sustainable development. He was also EEA National Focal Point (NFP) for the UK from 1991–1997. Since joining the EEA in 1997 he has been strongly involved in building capacities in the EEA's network of 33 member countries, and designing data flows and core indicators. He has also authored several integrated assessment reports focused on environment, well-being and economic interactions (e.g. green economy), and is presently Head of Integrated Environmental Assessments (IEA) at EEA. Lead author of the European state of the environment and outlook (SOER) 2015 report.

RAPPORTEURS

Yann Devos — *European Food Safety Authority (EFSA), Italy*

Yann Devos is a senior scientific officer at the GMO Unit of EFSA, where he is involved in the risk assessment of genetically modified (GM) plants and the development of risk assessment guidelines. He has an MSc in Biology, a supplementary degree in Environmental Sciences and a PhD in Applied Biological Sciences. He has more than 10 years of experience in the environmental risk assessment of GM plants and their associated farm management practices.

Agnès Rortais — *European Food Safety Authority (EFSA), Italy*

Agnès Rortais is a scientific officer at the Scientific Committee and Emerging Risks Unit of EFSA since 2008. At EFSA, she is developing an integrated conceptual framework for the holistic risk assessment of multiple stressors in bees. This framework is based on experimental, monitoring and modelling approaches. She is also supporting the development of harmonised approaches in the area of environmental risk assessment. She has a PhD in tropical

ecology and biology and she worked seven years as a bee researcher (in the areas of risk assessment of pesticides, population genetics, collective behaviour and calibration of field monitoring tools for the use of bees as bioindicators of the environment).

Reinhilde Schoonjans — *European Food Safety Authority (EFSA), Italy*

Reinhilde Schoonjans, Risk Assessment Scientist at EFSA since 2005, provides scientific input to the work on overarching elements of environmental risk assessment for EFSA's Scientific Committee, as well as on nanomaterials in food/feed and on cloning of farmed animals. Previously, she developed risk assessment guidance for GM plants used for non-food or non-feed purposes, such as for the production of medicinal or industrial products. She is a molecular biologist, holding a PhD from the University of Ghent, where she performed research on bispecific antibodies for cancer immunotherapy. In October 2005, she qualified as European Patent Attorney for the European Patent Office in Munich. She is an enthusiast of scientific communication and enjoyed media trainings organised by the European Commission.

Franz Streissl — *European Food Safety Authority (EFSA), Italy*

Franz Streissl is a senior scientific officer in the Pesticide Unit of EFSA. The focus of his work is on developing guidance documents for pesticide risk assessments (e.g. Guidance document on the risk assessment for bees). In the past he was responsible for assessment of environmental risks of pesticides at national and European level. He has contributed to a large number of governmental reports, presentations at conferences and workshops. He has an in-depth knowledge of the regulatory framework and risk assessment procedures of pesticides.

KEYNOTE SPEAKER

An introduction to ERA: advances and challenges

Alan Gray — *Centre for Ecology and Hydrology (CEH), UK*

Alan Gray is a Research Fellow with the Centre for Ecology and Hydrology in the UK. He retired in 2003 as Director of CEH's Dorset laboratory, following more than 35 years' research in plant ecology and genetics, and more than 200 publications – mainly on plant ecology, gene flow, population ecology and genetics and the conservation genetics of natural populations of plants. Following a BA in Biology and Philosophy and a PhD in Genetics at the University of Keele, he joined the Nature Conservancy's Merlewood Research Station in 1968, moved to ITE Norwich in 1970 and to Dorset in 1976, where he was appointed Head of Furzebrook Research Station in 1993 and Director CEH Dorset in 1999. He has been a Visiting Professor at the Universities of Southampton, Bournemouth, Stockholm and Groningen and has held a number of senior committee posts including Chairman of the Governing Body of the Institute of Grassland and Environmental Research, President of the Estuarine and Coastal Sciences Association and Vice President of the British Ecological Society. He is currently President of the International Society for Biosafety Research. Alan has been involved in risk assessment for GMOs since 1990 undertaking research and providing independent advice to the UK Government. He was, first, a member (1994–1999) and then Chairman (1999–2003) of ACRE (the Advisory Committee on Releases to the Environment) – the UK's statutory advisory committee on GMOs. Since retirement he has continued his involvement with the biosafety of GM crops (e.g. on the UK GM Science Review), with science governance and peer audit, also recently completing, with a co-author, a Flora of British Grasses. In recent years he has taken part by invitation in GM Biosafety workshops in Argentina, South Africa, Brazil, Canada, Chile, India, Indonesia, Italy, South Korea, Mexico, New Zealand, Russia, Slovenia, Sri Lanka, Switzerland, Ukraine, Uganda, the USA and Vietnam.

Although shaped to some extent by the different approaches taken by different national jurisdictions, the development of Environmental Risk Assessment (ERA) in support of decision-making in agricultural biotechnology has displayed a number of clear trends. By way of introduction to the break-out session this presentation traces the development of ERA since the early days of building regulatory systems for the cultivation of genetically modified (GM) crops. As well as identifying some of the significant advances made, I will explore some of the challenges which remain.

A noticeable early trend was the reduction to a realistic number the list of potential harms, and therefore risks, which might result from growing GM crops. Challenged initially to imagine even the most improbable outcomes, scientists and regulators have commonly framed ERAs to address fewer but more substantive questions. Typical of these are the so-called 'five pillars' of the Canadian regulations which cover problems which might arise from the weediness or invasiveness of the GM plant or a GM/wild relative hybrid, harm to non-target organisms including humans, the GM plant becoming a plant pest and the potential impact on biodiversity. Other jurisdictions (e.g. the EU) have longer lists but several of the low-probability risks, such as those from horizontal gene flow or the emergence of novel viral disease from recombination in virus-resistant plants, now rarely feature as part of the ERA.

A second advance, one covered in detail in later talks, has been the development of research which is targeted to address specific questions raised by the risk assessment (need-to know versus nice-to-know). It has taken ecologists a relatively long time to fully accept the importance of this development (possibly for reasons not unrelated to the availability of funding to investigate extremely interesting questions) and to appreciate that much of the data needed to make a risk assessment may already be available, for example from the phase of plant characterisation during product development. Whilst the challenge remains to only undertake research which is relevant and hypothesis-testing, perhaps a greater challenge is to know when to stop. Judging the extent to which further scientific evidence will contribute to a better risk assessment is not a trivial undertaking, especially when often unpredictable or variable socioeconomic factors such as farmer behaviour are dragged into the assessment of overall risk of harm.

Arguably the most significant advance (noticeably present from the beginning in some regulations), and one which has made better-targeted research possible, is the structuring of the ERA in a way which addresses potential harms at the start of the risk assessment process – in other words the development and adoption of a 'problem formulation' approach. By asking ourselves at the outset what it is that we do not want to see harmed, what we want to protect, and envisioning ways in which it might be harmed, we focus immediately on the questions which lie along the pathway to harm. Problem formulation, also covered in later talks, has empowered clearer thinking about the challenge of translating policy aims, usually expressed as broad normative concepts such as sustainability, into a set of operational protection goals. Challenges remain, not least in bridging the gap between such generic policy objectives and the generally more specific and limited outcomes of individual pieces of science. Advances made easier by an ecosystem services approach are helping to pin together such individual studies, not only to tackle the problem of multiple stressors but also to arrive at more clearly defined protection goals and more testable hypotheses regarding risk.

In some ways the biggest challenge faced by ERA is maintaining its force and relevance to decision-making in agricultural and environmental policy. This will be achieved, one suspects, not by making it more complex or more finely calibrated, but by making clear what questions it can answer, what its limitations are and where residual uncertainties remain. Unless, in future decision-making, putative risks are balanced by potential benefits and questions about the 'safety' of GM crops are uncoupled from all the frequently emphasised but entirely contingent 'harms' associated with increasingly industrialised farming, ERA for GM crops will be expected to provide answers to questions which lie outside its boundaries. ERA is a part, not all, of the decision-making process.

SPEAKERS

The ecosystem service approach to make protection goals operational

Lorraine Maltby — *University of Sheffield, UK*

Lorraine Maltby is Professor of Environmental Biology at the University of Sheffield, UK, and has published extensively in the fields of freshwater ecology and ecotoxicology. Her research aims to understand how ecosystems respond and adapt to environmental stressors, including pollutants. This understanding is fundamental to the successful protection and management of ecosystems and the contributions they make to human well-being. Lorraine has served on several UK government advisory committees and has chaired the Environment Panel of the UK Advisory Committee on Pesticides. She has also served on EFSA's expert group on ecotoxicology, which produced the 2010 scientific opinion on protection goals. Lorraine is currently a member of the European Centre for Ecotoxicology and Toxicology of Chemicals (ECETOC) Scientific Committee and the UNEP Scientific Expert Group on Chemicals and the Environment.

What do we want to protect, where do we want to protect it and how long do we want to protect it for? These questions are fundamental to formulating the protection goals that risk assessment addresses. However, the answers – everything, everywhere, forever – are not tenable and therefore a transparent and robust framework for deciding what to protect and where to protect it is required. Ecosystems provide humans with many benefits (i.e. ecosystem services), including our basic needs of food, water, shelter and clothing. The EU Biodiversity Strategy recognizes the importance of ecosystems in providing benefits to humans and has stated that 'By 2020 … the degradation of ecosystem services will be halted'. This presentation will explore how an ecosystem services framework can be used to develop specific protection goals and will outline the challenges and opportunities of operationalising protection goals within environmental risk assessment. It will lay out the rationale for a holistic, spatially defined approach that generates options for prioritised bundles of ecosystem services provided by agro-ecological landscapes. The focus will be on arable landscapes and plant protection products, but reference will be made to other regulated stressors within EFSA's remit (i.e. GMOs, feed additives, invasive species).

Ecosystem services and biodiversity

Steve Wratten — *Bio-Protection Research Centre, Lincoln University, New Zealand*

Steve Wratten is Professor of Ecology at Lincoln University as well as being on the staff of the Bio-Protection Research Centre. He has studied and worked in the Universities of Reading, Glasgow, London, Cambridge and Southampton in the UK. His research largely concerns the understanding and enhancement of nature's services (ecosystem services – ES) on farmland. He is the world leader in biological control of pests and is currently working on using ecological techniques to reduce the decline in populations of pollinators. Pollination is an ES. One of the clear outcomes that his research has delivered is the Greening Waipara programme (http://bioprotection.org.nz/greening-waipara). This involves adding flowering native and non-native plants to vineyards to improve the efficacy of pest biological control agents. His work in the MBNZT involve helping the schools and gardeners to enhance butterfly populations on their land. Butterflies also have ES value, which is their aesthetic quality. His current research includes a strong emphasis on understanding and enhancing ecosystem services in farmland. This includes work on a giant hybrid grass called miscanthus. This can be used for production of renewable liquid fuels, but unlike other biofuel feedstocks, it delivers at least 16 ecosystem services within the farm and, beyond it, to society as a whole.

The global agroecosystem faces many hazards, ranging from the use and mis-use of insecticides, fungicides and herbicides to soil degradation, fossil-fuel use and over-use of fertiliser and water and very high transport costs, as well as food wastage. Consequently, it contributes to climate change, with the global food chain producing up to one-third of global fossil-carbon emissions. That in turn represents a further hazard, affecting what crops can be grown successfully in the future, where and at what levels of production. The basis of this is that if appropriate functional biodiversity is deployed in and around farmland, ecosystem functions are strengthened and as a result, ecosystem services are delivered. These services provide benefits within the farm and outside it, the latter to society as a whole. The enhanced biodiversity, however, may not itself be native to the study system nor indeed have innate conservation value. This can be partly addressed by identifying species which confer a wide range of ecosystem services (ES) to the farm and beyond, ideally including conservation, aesthetic and wellbeing benefits.

Agroecology can double world yields in a decade (Olivier de Schutter, UN Special Rapporteur, 2010) but conventional agriculture, even including the possibly oxymoronic concept of 'sustainable intensification', cannot do that. This presentation will outline how ecosystem services such as biological control of pests, enhanced soil carbon and reducing fertiliser needs in agriculture can be easily and greatly enhanced, through the deployment of existing and simple Service Providing Units (SPUs). Examples of SPUs in current use by farmers worldwide will be given, along with how food-safety risks associated with agriculture can be minimised.

ERA vs Ecological research — The importance of good problem formulation

Joe Smith — *Advisor in Government, Science and Regulation, Australia*

Joe Smith is an independent advisor in science and regulation, specialising in biotechnology and agricultural chemicals. He has a diverse background spanning over 35 years' experience in public and private sector roles involving scientific research, services, regulation policy. He was Australia's Gene Technology Regulator from 2009 to 2014. In this role, he was the Australian Government statutory office holder responsible for leading the national regulatory system for gene technology. He was strongly committed to delivering a transparent, scientifically robust, effective and internationally respected system for gene technology regulation. Joe has extensive experience in various other senior government regulatory and related roles, including as Chief Executive Officer of the Australian Pesticides and Veterinary Medicines Authority, Director of the Therapeutic Goods Administration Laboratories and as Australian Government Analyst. He continues to be actively engaged in international collaborative, standards setting, and harmonisation activities through forums such as the OECD, FAO/WHO and ISBR, and in building cooperation and assisting capacity development with counterpart regulators in other countries. He has a PhD and BSc (Hons), is a Fellow of the Royal Australian Chemical Institute and a Graduate of the Australian Institute of Company Directors.

Decisions on whether to approve environmental releases of genetically modified (GM) crops, and under what conditions, are widely based on detailed environmental risk assessments (ERAs). The broad framework for these ERAs is provided in legislation and regulations which reflect the expectations of governments and citizens in individual countries.

ERA is not ecological research. The data that are required and used in ERAs for GM crops are key to their effectiveness and to ensuring they remain clearly focussed on identifying and realistically evaluating possible risks. The type and extent of the data that are useful (rather than merely of interest) in conducting the ERAs is determined by the important first step of problem formulation which distils factors such as legislated protection goals and the risk assessment context, methodology and endpoints into explicit questions to be considered.

Data used in ERA of GM crops can come from many sources – for example, from the scientific literature or from data supplied by the applicant seeking approval for a particular crop. The challenge for regulators conducting ERAs is to ensure that the data they request and use are both reliable and actually relevant to the risk assessment and not simply information that is of general interest to satisfy irrelevant curiosities. In this regard, for example, the value of excessive reliance on small (and often poorly understood) variations at the molecular level can be questioned. Regulators also need to have in place robust mechanisms for evaluating the quality and value of different information and addressing uncertainty in reaching conclusions based on a weight of evidence approach to risk assessment. Some information available in the wider literature is, at best, irrelevant to the risk assessment and, at worst, scientifically flawed and misleading.

This presentation explores the difference between need-to-know and nice-to-know data, and in particular the role of good problem formulation in determining the usefulness and relevance of different types of data in conducting regulatory risk assessments of GM crops.

Methods used to assure high quality studies for ERA — Non-target arthropod testing of transgenic plants as a case study

Jörg Romeis — *Agroscope, Institute for Sustainability Sciences, Switzerland*

Jörg Romeis is the head of the Biosafety Research Group at Agroscope, Institute for Sustainability Sciences (ISS), in Zurich, Switzerland. In addition he is a lecturer at the University of Bern and an adjunct professor with the Chinese Academy of Agricultural Sciences. Jörg has an MSc and PhD in biology and has been trained as an applied entomologist with a focus on biological control and multi-trophic interactions. He has more than 14 years of experience in non-target risk assessment of genetically modified (GM) crops and in particular in the design and execution of non-target laboratory studies. The focus of his research has been on the effects of Bt-plants, such as maize and cotton, on valued non-target arthropods including predators, parasitoids, pollinators and decomposers. In addition to primary research, he has also been actively involved in developing testing protocols and guidelines for risk assessment. In total, this work has resulted in more than 100 peer-reviewed publications. From 2003 to 2013, Jörg Romeis served as convenor of the working group 'GMOs in Integrated Plant production' of the Western Palaearctic Regional Section (WPRS) of the International Organization for Biological Control (IOBC). Within this working group he has organised and led several international expert panels to develop a rigorous approach to evaluate potential non-target risks of GM crops, to propose design criteria for laboratory studies with non-target organisms, and to develop criteria for the selection of surrogate test species.

Environmental risk assessment of genetically modified (GM) plants is designed to answer very specific questions about the potential risks of introducing such plants into the environment. A common concern among regulatory authorities is the potential for a GM plant to have adverse impacts on non-target arthropods (NTAs) that are valued for the ecosystem services they provide. This risk is particularly evident for GM plants that express insecticidal proteins to control specific pests such as Bt-transgenic plants encoding for insecticidal Cry proteins. A typical risk hypothesis to be addressed for Bt crops is that the expressed protein is not toxic to valued non-target organisms (NTOs) at the concentration present in the field. This hypothesis is commonly assessed following a tiered approach that progresses from lower tier laboratory studies using relevant surrogate species under worst-case exposure conditions to more realistic but less controlled higher tier studies including field studies if required. Laboratory studies have a good ability to detect adverse effects of e.g. insecticidal proteins on NTAs. However, reliable test systems need to adhere to a number of study criteria to avoid erroneous results, i.e. false negatives and false positives, that will be discussed. Adhering to these principles and recommendations increases the confidence in the results of laboratory studies and thereby reduces data requirements for stressors that pose low risk. It will also facilitate study reproducibility, peer review of tests by others, and will benefit regulatory authorities by enhancing the quality of information generated for use in risk assessment. Furthermore, high confidence in the study results is a precondition for the acceptance of data across regulatory jurisdictions and should encourage agencies to share useful information and thus avoid redundant testing. It is essential that risk assessors be discriminating about the legitimacy of the studies that they consider during the evaluation process, irrespective of their source. This evaluation should include both the quality of the study itself as well as its relevance to the risk assessment process as described in regulations and associated guidance.

Multiple stressors: bees as a case study

Jeffrey Pettis — *USDA-ARS Bee Research Laboratory, United States*

As a research Entomologist in the USDA-ARS Bee Research Laboratory in Beltsville, Jeff Pettis leads a broad research effort to improve colony health by limiting the impact of pests and diseases on honey bee colonies. His research areas include; IPM techniques to reduce the impacts of parasitic mites and disease, effects of pesticides and pathogens on queen health and longevity, host-parasite relationships and bee behavior. Dr. Pettis serves on several international committees concerning bee health and is frequently interviewed by the media for his opinions on worldwide pollinator declines. He received an undergraduate and MS degree from the University of Georgia and his doctoral degree in Entomology from Texas A&M University in 1992.

Ecological risk assessment currently does not consider simultaneous exposures that often happen when bees forage over a wide area. How can multiple exposures be assessed? This is not a hypothetical question but rather a question that must be considered when trying to protect pollinators in a diverse environment. Risk assessment can be a complicated endeavor considering the range of life histories, from solitary bees to large honeybee colonies. Traditionally honey bees have served as surrogates for all pollinators but recent research with bumble bees and honey bees show that honey bees can absorb more pesticide exposure while the same exposures dramatically impact the growth and reproduction of the smaller bumblebee colonies.

Another variable to consider in risk assessment is how relevant is lab generated data on individual honey bees to the honey bee colony and or other pollinators? Risk assessment has historically taken a tiered approach with individual honey bees subjected to a single active ingredient in a contact LD50 cage bioassay. When these test yielded an LD50<11ug/bee the chemical would then move to higher tier testing. But higher tier test are complicated by using larger honey bee colonies and the need to guarantee exposure results in semi-field test of short duration or open field testing which is too often conducted using minimal plot size.

Lastly, it has become apparent that sub-lethal effects must be considered but adequate endpoints are hard to define. How can regulatory bodies consider multiple lines of evidence in a weighed manner? The answer of how to test and or assess multiple stressors is not clear cut. Questions will be raised concerning 1) the use of models as a tool in risk assessment, 2) the validity of extrapolation from individual results to the superorganism and improved sub-lethal assays and 3) a recently published field study that tested only a single compound but with three different pollinators will be presented and discussed. Lastly, a study of multiple exposures will be presented and discussed that utilizes an epidemiological approach to test the effects of multiple stressors to identify those stressors that had a significant interactive effect with pathogen levels in honey bees.

The reality is that bees are exposed to multiple stressors and it is difficult to test for a single effect without confounding factors such as diseases, pests, nutrition or other factors playing a part in bee health. Bees operate in the real world and there must be a means to accurately assess the factors that directly affect bee health. The ability of large honey bee colonies to absorb impacts from stressors and continue to grow makes the extrapolation from individual bees to colony level effects difficult. Smaller colonies such

as *Bombus* may be better suited to field studies as they have shorter flight ranges allowing smaller field plot size and recent published studies indicate that they are indeed sensitive to sub-lethal exposures. Comparative studies using *Apis*, *Bombus* and *Osmia* or *Megachilidae* are a good starting point but there is a need for even more sophisticated methodologies. Perhaps with more complex models, improved statistical methods and perhaps more sensitive test methods we can accurately assess multiple stressors of bees and other pollinators.

Poster session

Open Risk Assessment: data

1. Use of clustered data models to infer compliance to Commission Regulation of foodstuff in Europe, with application to *Listeria monocytogenes*

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ABSTRACT

Objectives: One of the food safety criteria, as described in Chapter 1 of Annex I of the Commission Regulation (EC) No 2073/2005, for ready-to-eat foods able to support the growth of *L. monocytogenes*, state that out of 5 units comprising the sample none of the sample units should give values over 100 cfu/g. This criterion is used here as the definition for 'compliance'. The objective is to develop predictive models to infer compliance. **Materials and Methods:** A probability model for compliance is defined based on the beta-binomial model for clustered binary data. It depends on the probability for a random sample unit from a random sample to exceed the limit of 100 cfu/g and on the within sample correlation. **Results:** Based on estimates for the proportion of samples with counts exceeding the level of 100 cfu/g, the model-based approach allows constructing confidence intervals (CI) for the compliance probability. At the date of testing at the end of shelf-life: for fish, the 95% CI for the compliance probability, accounting for the unknown within-batch correlation in a conservative way, is given by (0.890,0.987); for packaged heat-treated meat products by (0.964,0.997) and for cheese products by (0.988,1.000). In any situation, compliance increases with the level of correlation. **Conclusions:** It is shown how a predictive model for compliance can be developed. The methodology allows evaluating the effect of different sampling designs and the within sample correlation. Disclaimer: The present abstract has been produced and adopted by the bodies identified above as author(s). This task has been carried out exclusively by the author(s) in the context of a contract between the European Food Safety Authority and the author(s), awarded following a tender procedure. The present abstract is published complying with the transparency principle to which the Authority is subject. It may not be considered as an output adopted by the Authority. The European Food Safety Authority reserves its rights, view and position as regards the issues addressed and the conclusions reached in the present abstract, without prejudice to the rights of the authors. **Acknowledgements:** The authors would like to thank the EFSA staff members Marios Georgiadis, Frank Boelaert, Pia Mäkelä (former Head of the EFSA Unit on Biological Monitoring), Gabriele Zancanaro and Giusi Amore for the support provided to this scientific output.

2. Exposure assessment within a Total Diet Study: a comparison of the use of the pan-European classification system FoodEx-1 with national food classification systems

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ABSTRACT

Objectives: A Total Diet Study (TDS) consists of selecting, collecting and analysing commonly consumed foods purchased at retail level, processing the food as for consumption, pooling the prepared food items into representative food groups, homogenizing the pooled samples and analysing them for harmful and/or beneficial chemical substances. TDSs are commonly designed at national level and aim to cover the overall diet of the population, in order to assess the dietary exposure to hazardous chemical substances of interests by the population of a certain country. To assess dietary exposure, a food classification system is needed to link existing food consumption data with the analytical data obtained in the TDS. In this study a comparison was made between the use of national classification system and the use of FoodEx-1, a food classification system recommended by the European Food Safety Authority (EFSA). **Materials and Methods:** The work was performed using data of six European countries; Belgium, Czech Republic, France, the Netherlands, Spain and the UK. For each of the countries population's exposure to contaminant A (organic compounds) and/or contaminant B (inorganic compound) was assessed by the Monte Carlo Risk Assessment (MCRA) software using the national classification system and FoodEx-1 for food consumption data and for TDS laboratory results. **Results:** The exposure assessment obtained when applying FoodEx-1 was not always the same as the results of the exposure assessment obtained with national codes. Overall, minimal differences between exposure assessment results using national classification and FoodEx-1 classification were observed. This observation was made as well as for contaminant A as for contaminant B. **Conclusions:** Minimal differences between both approaches were observed for the organic as well as the inorganic compound. In general, the conclusion of the risk assessment will be similar for both approaches, however this is not guaranteed for all types of contaminants. FoodEx-1 proved to be a valuable hierarchic classification system in order to harmonise exposure assessment based on existing TDS results throughout Europe.

3. Data collection for the EFSA's Chemical Hazards Database

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ABSTRACT

The EFSA's Chemical Hazards Database aims at mapping the hazard data included in the EFSA documents (opinions, statements and conclusions) adopted by its panels (NDA, CONTAM, FEEDAP, AFC, CEF, ANS and PPR) on risk assessments on food and feed. The structure of the database is devised to store these features: chemical identification, document descriptors, hazard identification, and hazard characterisation/risk characterisation. A suitable ad hoc accessory IT platform has been developed by the contractor to aid and support the activity of data collection, entry and submission. This infrastructure provides the means for collating and storing the data in a temporary local database, that is then exported and submitted to EFSA in due time. A number of control mechanisms at the data entry level are automated

to guarantee high quality of deliverables and reduce unintended errors. Two different approaches are followed to minimise errors that may occur during data entry: a) automatic verification of data quality at the data entry level, where a number of rules (e.g. drop-down lists, compulsory fields) governs and steers data collation; b) manual revision of the collected data that is first randomly checked and secondly analysed to search possible issues according to a number of empirical rules. About 1500 documents have been screened, reporting about 8000 assessments, registering more than 4000 chemicals, and summarizing at least 7500 toxicity studies (including critical studies used to derive health-based guidance values or margin of safety/exposure). Such data that is first collected, stored, and quality-checked is then exported from the contractor's local repository and finally submitted in XML format to the Data Collection Framework (DCF) of EFSA. The DCF ensures that the dataset is compliant with EFSA IT standards and will enable future sharing with EU Member States, international bodies (e.g., FAO/WHO) and third parties (e.g., US-EPA).

4. Occurrence of aflatoxin B1, aflatoxin M1 and ochratoxin A in different food products: a preliminary study

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ABSTRACT

Objectives: Aflatoxin B1 (AFB1), aflatoxin M1 (AFM1) and ochratoxin A (OTA) are mycotoxins responsible of many different toxic effects. The International Agency for Research on Cancer classified AFB1 as carcinogenic to humans (Group 1), AFM1 and OTA as possible human carcinogen (Group 2B). In this research, we analyzed samples of flour and milk, whether conventional or organic, for the presence of AFB1 and AFM1 respectively. Furthermore, we tested samples of salami for the presence of OTA. **Materials and Methods:** In the present study, 90 samples of flour and 58 samples of milk, purchased from local stores in the surrounding of Bologna, were analyzed for the presence of AFB1 and AFM1 respectively. As regards OTA, analysis were performed on 50 salami from Veneto and 55 from Sicily. For analysis of AFB1 in flour, a fast and sensitive method was developed and validated, while for AFM1 and OTA, immunoaffinity-based techniques were used. The analyses were performed by high performance liquid chromatography with fluorescent detection (HPLC-FD). **Results:** AFB1 was found in 13 samples of corn flour (4 organic and 9 conventional ones). These results confirm the high incidence of contamination of corn compared to wheat, as reported in literature. The levels of AFB1 in the positive samples ranged between 0.17-3.75 µg kg-1, but only in one sample the limit defined at Community level (2 µg kg-1) was exceeded (data already published). Aflatoxin M1 was detected in 35 milk samples (11 organic and 24 conventional). The levels of contamination were very low and ranged from 9 to 26 ng kg-1, below the legal limit of 50 ng kg-1 set out at EU level. In salami, OTA was found in 10 samples, 5 from Veneto and 5 from Sicily (2 samples, one for each region, exceeded the Italian guideline level of 1 µg kg-1). **Conclusions:** The considered foods are safe as regards the presence of AFB1, AFM1 and OTA. AFB1 levels in flour and AFM1 levels in milk did not show differences between organic and conventional products. Although the reduced number of samples does not allow us to draw conclusions at national level, the present study seems to confirm the effectiveness of the Italian programs of monitoring and surveillance for ensuring that chemical residues in food do not pose a threat to human health. Given that a good food safety can also be ensured by continuous and numerous checks, this research will continue involving the same and other Italian food products.

5. Design of a risk-based AFM1 surveillance plan upon previous monitoring data

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ABSTRACT

Objectives: Aflatoxins are toxic fungal metabolites found in foods and feeds, they are known to be genotoxic and carcinogenic, they may both cause acute and chronic toxicity in humans. Aflatoxin M1 (AFM1), the monohydroxylated derivate of aflatoxin B1 (AFB1), occurs in milk from dairy cows fed an AFB1-contaminated diet; it may be subsequently transferred into derived dairy products. They are both classified as carcinogenic to humans and included in Group 1 by IARC. A survey was conducted in NW Italy (Liguria, Piedmont, Valle d'Aosta) to estimate the factors associated with AFM1 contamination level in commercially available raw and pasteurized milk. **Materials and Methods:** From 2008 to 2014 a total of 2,998 milk samples were analyzed. 1,505 samples were collected from dairy farms, 1,082 from raw milk vending machines, 285 from cheese farms and 126 from retail markets. Considering geographic position 2,287 samples were from Piedmont region, 515 milk samples from Liguria and 196 from Valle d'Aosta. A screening analyses step was performed using an Enzymatic Linked Immunosorbent Assay kit. Confirmation on suspected positive samples was carried out using High Performance Liquid Chromatography method. Association between AFM1 contamination and each factor (years, average temperature, seasonality and geographic position) was evaluated by a logistic regression model. Data were also used to classify dairy farms into 3 risk classes: AFM1 <1 ng/kg, AFM1 1-49 ng/kg and AFM1 ≥50 ng/kg. **Results:** Data indicated that 2.37% of the whole samples was contaminated with AFM1 in concentration levels ranging from 1 to 208 ng/kg. The 0.47% of the samples was higher than the limit accepted by EU (50 ng/kg). Raw milk showed highest AFM1 contamination (2.03%) while pasteurized milk registered a 0.34%. Piedmont region (OR 2.7; CI95% 1.3-5.7) and the year (OR 2.4; CI95% 1.5-4.0) were also associated with AFM1 contamination. A total of 421 farms was classified as AFM1 <1 ng/kg, 33 as AFM1 1-49 ng/kg and 7 as AFM1 ≥50 ng/kg. **Conclusions:** Only 0.47% of samples were not compliance within EU regulation, showing a low risk for the consumers. The annual higher prevalence observed in 2013 (0.9%) is probably linked to the previous dry summer (increase of 2.3 C° compared to average temperature). Based on the results of our study, a futurable risk-based surveillance plan should take into account the geographic position and the whether condition; in addition, the farm risk class should be considered.

6. Risk assessment of enteric viruses in ready-to-eat vegetables

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ABSTRACT

In the last few years, there was an increasing demand for leafy green vegetables and their ready-to-eat (RTE) salads since consumers changed their eating habits. Nevertheless fresh leafy green vegetables and their RTE salads are recognized as a source of food poisoning outbreaks in many parts of the world, in particular, fresh produce increasingly has been implicated in viral disease outbreaks. The aim of the study was to investigate the presence of enteric pathogenic viruses (Hepatitis A-HAV, and Norovirus genogroup II-NoV GII) in RTE vegetables available on the Italian markets. 5 imported and 25 not imported RTE salads samples were collected during 2014. The viral RNA was extracted following a normalized International Standard Organization (ISO) extraction method before further detection using One-step RT-PCR. Positive samples were confirmed by sequencing. The total incidence of NoV GII and HAV was 13,33 % (4/30) and 3,3 % (1/30), respectively. These results highlight the high occurrence of enteric viruses in foodstuffs that can be eaten raw or after a moderate technological processing or treatment. The determination of the potential risk factors for the contamination and cross-contamination of vegetables through supply chain, from production, processing and point-of-sale, remains an important challenge for the future. This study confirmed

for the first time in Italy the presence of Hepatitis A virus in 'ready to eat' vegetables.

7. Quality and safety assessment of plant food supplements: the role of the Istituto Superiore Di Sanità

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ABSTRACT

Introduction: The observed substantial increase, for over the last two decades, in the use of plant food supplements (PFS) based on herbs/botanicals and their products, especially extracts, should be adequately accompanied by quality and safety control. This includes in particular identity/authenticity, contaminants, residues and other not legally or undesired substances as well as not allowed or not declared treatment (irradiation). A proper quality control must also include the analysis of a broad spectrum of potential physiologically active phytochemicals and biomarkers as well. **Objectives:** On the basis of the acquired experience within the official control activities, this presentation aims to describe emerging quality and safety issues through the evaluation of results obtained from a selected number of the most significant cases examined by the Istituto Superiore di Sanità (I.S.S.). **Materials:** The products reported in this study were Plant Food Supplements marketed in Italy. These products were checked by I.S.S. because of: found not in compliance with existing regulations by the official laboratories, confiscated by the Anti-adulteration Squad of the Carabinieri (NAS) for suspected adulteration, replacements, falsifications, involved in cases of adverse effects. **Methods:** Samples were checked by chromatographic techniques: e.g. Thin Layer Chromatography (TLC), High Performance Liquid Chromatography (HPLC), High Performance Thin Layer Chromatography (HPTLC). To detect irradiation treatment PFS and/or their ingredients were checked by luminescence techniques such as Photo-stimulated luminescence (PSL) and Thermoluminescence (TL). **Results:** In this presentation will be reported the analytical results related to the content of bioactive compounds not in compliance with the labeling, detection of not allowed substances (pharmacologically active compounds), identification of not admitted plants, analysis of plant food supplements suspected of adverse reactions. The results of the detection of irradiation treatment in plant food ingredients will be also reported. **Conclusions:** The obtained results highlight important issues related to the quality and safety of this category of products. The growing volume of products and sales call for a more formal premarketing assessment and an enforcement of the surveillance system.

8. Shiga toxin-producing *Escherichia coli* in meat and raw milk in Sicily (Italy)

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ABSTRACT

Directive 2003/99 EC on the monitoring of zoonoses and zoonotic agents, identified Shiga Toxin-producing *E. coli* (STEC) as public health priorities to be monitored. Indeed, according to EFSA and ECDC, an increasing trend in illness cases has been reported in the EU in recent years, with approximately 6000 cases confirmed in 2013. Moreover, in the first quarter of 2015, RASFF reported 16 notifications for STEC in 3 cheeses and 13 meat. Data on STEC prevalence focus mainly on cattle and beef products, which are the primary vehicles of these pathogens. In this context, the aim of this study was to investigate the prevalence of STEC in Sicily, considering also the role of food products of species other than bovine. During 2014 and January 2015, a total of 136 samples were analysed by PCR Real Time (ISO/TS 13136:2012). In detail, 81 were meat samples (beef, pork and sheep meat) and 55 were raw milk (bovine and ovine milk). Firstly all samples were tested for the main virulence genes *(Stx1, Stx2, eae),* and then positive samples were subsequently tested for serogroup-associated genes. PCR screening results showed 8 positivity for STEC virulence factors (5.8%), specifically in 6 meat samples (7.4%) and 2 milk samples (3.6%). PCR B confirmed the presence of O157 in 3 positive pork samples, while serogroup O145 gene was detected in 1 positive bovine meat. Serogroups of the other 4 positive samples were not identified, presumably due to the presence of other serogroups, different from those targeted with the PCR method. These preliminary results show only the indirect evidence of the presence of STEC, as positive samples were not confirmed by cultural isolation, as reported in other studies. Despite the high sensitivity of molecular methods, isolation of strains is still essential, although critical because of difficulties in developing culture media specific and/or differential for the diverse STEC serogroups. In fact, molecular methods may overestimate the effective contamination by detecting dead cells, free DNA or phages in the enrichment broth. It is noteworthy the potential role of pork meat in the transmission of STEC, besides confirming bovine as a reservoir. Indeed, presumptive O157 were found in 10.3% of pork samples, highlighting the need to further investigate their contribution to STEC epidemiology.

9. Radiochemical control of food destined for export into the Russian territory: two years activity of National Reference Centre for the Detection of Radioacivity in Feed and Foodstuff

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ABSTRACT

Objectives: Cesium-137 and Strontium-90 are two important radiochemical contaminants which may be present in different food supply chains as a consequence of nuclear fallouts, weapons testing, and incorrect wastes management. Due to their high water solubility (137Cs) and long half-life (90Sr) they are considered harmful contaminants. Following the EC and Russian protocols, which establish maximum levels for several contaminants (including 137Cs and 90Sr) in different foodstuffs, the Italian Ministry of Health developed a monitoring plan for the control of food destined for export into the Russian territory. As a contribution for radiological risk assessment, in this work, the recent control activity (2013-2014) developed by Italian National Reference Centre for the detection of Radiactivity in Feed and Foodstuffs, concerning the determination of 137Cs and 90Sr in different foodstuffs is described. **Materials and Methods:** The radiochemical determinations were carried out by gamma spectrometry (137Cs) and liquid scintillation counting (90Sr). These methods are accredited by Italian Organism for Laboratory Accreditation ACCREDIA and they allow the quantification of two radionuclides up to sensibility equal to 0.006 Bq L-1 (90Sr in liquid matrices), 0.008 Bq Kg-1 (90Sr in solid matrices) and 0.7 Bq Kg-1 (137Cs). **Results:** The following types of foodstuff were collected in 5 Italian Regions (Lombardy, Piedmont, Veneto, Emilia Romagna and Lazio): fresh meat (13.0%), meat product (8.6%), ripened cheese (23.2%), unripened cheese (20.1%), raw milk (24.6%), milk powder (4.3%), pasteurized milk (5.7%), animal feed (0.5%). 137Cs: none of the analysed samples showed a quantifiable (C>LOQ = 0.7 Bq Kg-1) activity concentration of 137Cs, confirming a good level of food safety from this point of view. 90Sr: the employed methods are characterised by high sensibility. Consequently, it is possible to determine low contamination levels. The following mean levels were detected: meat products (0.02 Bq kg-1), ripened cheeses (0.12 Bq kg-1), unripened cheese (0.05 Bq kg-1), raw milk (0.04 Bq kg-1), milk powder (0.06 Bq kg-1), pasteurized milk 0.05 Bq kg-1), animal feeds (0.06 Bq kg-1). **Conclusion:** The recent control activity developed by Italian National Reference Centre for the detection of Radiactivity in Feed and Foodstuffs demonstrated that contamination levels of 137Cs and 90Sr in foodstuffs destined for export into the Russian territory are well lower than established limits.

10. Human biomonitoring linked to dietary and life-style surveys improves risk and benefit assessment quality

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ABSTRACT

Objectives: In performing risk- and benefit assessments for food safety, high quality exposure data are of vital importance, but hard to reach due to a number of uncertainty factors. Therefore, as a complement, human biomonitoring (HBM), which measures actual levels in e.g. blood and urine, could be performed. By combining HBM with dietary and other important data, possible associations are studied. **Methods:** In the latest Swedish nationwide dietary survey, Riksmaten 2010-11, a subgroup (n=300; 30% participation rate) of the randomly chosen adults (18–80 years) donated blood and spot urine samples. For dietary assessment, 4-day food record and questionnaire form were completed. Among other substances, the samples were analysed for fatty acids in phospholipids, PCB and chloropesticides, heavy metals, brominated flame retardants, perfluorinated alkyl-substances (PFAS), phthalates, bisphenol A and several mycotoxins, including deoxynivalonol (DON). Associations between body levels, diet and life-style factors were studied by statistical analyses. **Results:** BM data show that levels of the studied substances in most cases are quantifiable in serum/urine. Dietary associations were observed between levels of PCBs, PFAS or methyl mercury and consumption of fish, between lead levels and intakes of game and alcohol and between n-3 fatty acids in phospholipids and corresponding dietary fatty acids and fish intake. DON was associated with the intake of total cereal grain, whole grain, breakfast cereals and porridge. In case of non-dietary factors, cadmium levels were related to smoking, while phthalate levels associated with residential factors. **Conclusion:** By linking HBM to the latest Swedish dietary survey, human levels of both nutrient and toxic compounds were studied. In most cases, detectable levels were found in blood/serum and urine and, in spite of a limited number of participants, associations to both dietary and non-dietary factors were observed. Advantages with continuous HBM linked to national dietary surveys are the population-based approach, the high quality of dietary data to be used in statistical analyses, the possibility to separate dietary from other type of sources, and, upon iteration, the production of time-trend analyses. At the same time, the national HBM approach in dietary risk- and benefit assessment needs to be further developed and harmonized in order to relate obtained data to internationally agreed health-based values.

11. Dietary intake and risk assessment of phthalates in the Belgian population

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ABSTRACT

Objectives: Phthalates are one of world's most used groups of plasticisers. Some of them are suspected to cause endocrine disrupting activities in humans. For this purpose, we estimated and evaluated dietary exposure of the Belgian population to four phthalates (diethyl phthalate (DEP), di-n-butyl phthalate (DnBP), benzylbutyl phthalate (BBP) and di(2-ethylhexyl) phthalate (DEHP)). **Materials and Methods:** For Belgian adults, two dietary intake estimation methods were considered: one based on phthalate levels measured in food products using gas chromatography-low resolution-mass spectrometry and one based on predicted levels using the newly developed EN-forc model (ENvironmental Food transfer model for ORganic Contaminants). For Belgian infants and preschoolers, only measured levels were used. To assess the health risks associated with dietary phthalate exposure, intake estimates were evaluated against available tolerable daily intakes (TDIs). **Results:** All dietary intake assessments revealed that Belgian people are exposed daily to phthalates. The assessed exposure was the highest for DEHP and DnBP. Depending on the considered scenario, dietary exposure to DEP, DnBP, BBP and DEHP in Belgian adults represented <0.1, 1-12, <0.1-1 and 3-28% of the corresponding TDI values, respectively (calculated from P50 and P95 intakes). With respect to Belgian preschoolers, dietary exposure to DEP, DnBP and BBP was low, being respectively <0.1, 2-3 and <0.1% of the corresponding TDIs. However, for DEHP, the 95th percentile of the worst case scenario represented 58% of the TDI value. This percentage is within the range of observed contribution percentages of the diet to integral DEHP exposure in European children, namely 50-90%. For Belgian infants, almost the same results as for Belgian preschoolers were obtained regarding dietary DEHP exposure: intake estimates amounted to 30-54% of the TDI, which is in line with the contribution percentage of food ingestion to integral DEHP exposure in European infants, namely 50-96%. **Conclusions:** In conclusion, it can be stated that no TDI exceedances are to be expected regarding dietary exposure to DEP, DnBP, BBP and DEHP in Belgian adults, even when the contributions of other exposure pathways (e.g. inhalation and use of personal care products) are taken into account. However, for the younger age categories (i.e. Belgian infants and preschoolers), there might be a chance that – in specific situations – the TDI of DEHP will be exceeded.

12. Changes in proteomic profile can lead to differentiate fresh/frozen-thawed musky octopus

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ABSTRACT

Objectives: Providing information about the preserving method of a fish product is of enormous importance for the perception of its quality by the final consumer. The substitution and sale of frozen-thawed fish labeled as fresh is a widespread and difficult to unmask commercial fraud, and a potential danger for the health of consumers. Histology is a valid method to differentiate fresh from frozen-thawed fish, but not for all species, in particular for cephalopods. Two-dimensional electrophoresis could help to identify markers for rapid screening of samples to find fraud, as we did on tissue samples of musky octopus *(Eledone moschata)*. **Materials and Methods:** Ten samples of certainly fresh *Eledone moschata* were divided into two aliquots, the first was immediately processed to extract proteins, while the second aliquot was frozen at -20 °C for 72 hours and subsequently thawed and processed to extract proteins. Samples were provided from a fish market by an official veterinarian. 10% homogenates in lysis buffer were centrifuged and supernatant were quantified with Bradford method. An aliquot of 360 µg ml –1 of total protein per sample was added to 150 µl of a buffer solution. Samples were loaded by cup loading system on rehydrated IPG Strips (pH3-10NL, 18 cm) for isoelectric focusing (IEF). Before transfer to polyacrylamide gel for the second dimension, IPG strips were washed both with reducing and alkylating solution. The second dimension was performed with homemade 8–16% polyacrylamide gels. After electrophoresis, gels stained with Blue Silver. Images were acquired and subsequently analysed with BioNumerics® 2D Gel Types software. Protein maps were then compared to detect qualitative and quantitative differences between samples from the two groups. **Results:** About 280 spot were detected in each map and preliminary analysis showed a significant reduction of two spots in all ten samples, with a molecular weight of about 45-50 kDa and an isoelectrical point of about 6,5-7. **Conclusions:** The present study provides a 2DE comparative analysis to differentiate frozen-thawed from fresh musky octopus. Clearly, further analysis on a larger number of animals will be necessary to verify the potential of the two spots as biomarkers. If data will be confirmed, an identification of the proteins of interest by mass spectrometry will open the way for future studies to develop a rapid and accurate test, providing a useful tool for producers and official authorithies.

13. Biomonitoring of mycotoxin exposure through biomarker analysis: deoxynivalenol as a potential risk for the belgian population?

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ABSTRACT

The aim of the BIOMYCO study was to assess human mycotoxin exposure based on the direct measurement of urinary biomarkers in samples of the Belgian population. Morning urine of 155 children (3-12 years) and 239 adults (19-65 years) was collected according to a standardised study protocol. These urine samples were analysed for the presence of 33 urinary mycotoxins and their metabolites. Nine out of 33 potential biomarkers were detected whereby deoxynivalenol (DON), DON-glucuronides (DON-GlcA), deepoxy-deoxynivalenol-glucuronide (DOMGlcA), ochratoxin A, citrinin and dihydrocitrinone were the most frequently detected. DON15GlcA was the main urinary metabolite found in 100% of the samples and for the first time DOMGlcA was detected in urine of children. A risk assessment was performed by comparing the estimated dietary intake of DON with its tolerable daily intake (TDI) whereby the dietary intake of DON was estimated using the urinary concentrations. The estimated intake for DON varied between 0.02–25.47 µg/kg.BW/day for children and 0.01–15.65 µg/kg.BW/day for adults. This could imply a health risk as 1 to 74% of the cases exceeded the TDI for DON (depending on the approach applied to calculate the intake). These results are much higher than previous estimations reported in Europe. Differences in exposure could be explained to the different dietary habits between different countries and the higher occurrence of DON in temperate climates. In order to perform more accurate estimations, research needs to be done to collect information about the human metabolism of mycotoxins. Furthermore, a standardized protocol is needed to perform exposure assessments and to calculate the estimated exposure through urinary levels in order to compare different estimates and to evaluate the variability amongst people. In general, risk assessment based on these data indicate a potential concern for a number of individuals whereby young children need special attention because of the relatively higher food intake. **Acknowledgements:** This research was financially supported by the Federal Public Service Health, Food Chain Safety and Environment (grant number RT 11/2 – BIOMYCO) and the Special Research Fund of Ghent University. Isabelle Sioen is financially supported by the Research Foundation Flanders.

14. Pyrrolizidine alkaloids and relative N-oxides evaluation in italian honey from the Veneto region by LC-MS/MS

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ABSTRACT

Pyrrolizidine alkaloids (PAs) are the most common class of natural toxins (approximately 350 types), synthesised by a wide variety plant species and the presence of PAs in food and feed clearly represents a potential risk to human and animals. In this study, an analytical method by liquid chromatography coupled to mass spectrometry (LC-MS/MS) was applied to evaluate and characterise PA and their N-oxides (PANOs) content in Italian honey. Echimidine, heliotrine, lycopsamine, senecionine, seneciphylline, retrorsine and the corresponding PANOs were considered. For the matrix clean-up and the extraction of PAs/PANOs, the solid phase extraction non-polar/cation-exchange polymeric cartridges, PLEXA 200 mg/6 mL, (Agilent Technologies, USA) were adopted. Analyses were performed by a LTQ XL ion trap, equipped with a heated electrospray ionisation probe, operating in positive ion mode (Thermo Fischer Scientific, San Jose, CA, USA). The linearity of the calibration curves was evaluated in the range of 0.25-50 µg kg-1, the mean recovery ranged from 82.70 % to 104.16 %; LOQ was 0.25 µg kg-1 for all PAs/PANOS, fully satisfying the limit stated in the document 'Summary report of the standing committee on plants, animals, food and feed held in Brussels on 1 July2014'. 147 honey samples from the Veneto region were analysed, according to their geographical origin: 60 samples were from Area A (Padova, Treviso and Belluno), 51 from Area B (Northern Vicenza) and 36 from Area C (Southern Vicenza). Analyses showed that 69% of the honey samples were negative for all PAs/PANOs monitored, 31% were positive. In all positive samples the contribute from target PANOs was negligible (range 0.39-1.52 µg kg-1). The total content of PAs/PANOs monitored in the Veneto honey ranged from 0.25 to 64.44 µg kg-1. These results are not consistent with previous studies about honey samples from other European countries. Taking into account the indication from CONTAM Panel (EFSA 2011), based on the Margin of Exposure (MOE) approach, a honey sample must not exceed a PAs/PANOs total content of 21 µg kg-1; only 3% of our samples exceed this limit. Moreover, preliminary results showed an influence of two important factor as season (summer > spring) and territory (hills > lowland) on the total content of PAs/PANOs in honey. Honey produced from local beekeepers of Veneto region is not a real threat to health, while a monitoring program of PAs/PANOs in honey is highly desirable to risk assessment.

15. Deoxynivalenol in herbal teas: Current situation in Serbia

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ABSTRACT

Deoxynivalenol (DON) is a mycotoxin belonging to the group of trichothecenes, produced by various *Fusarium* species. It contaminates mainly grains and cereal-based food and feed. However its good solubility in water makes a presence of deoxynivalenol in herbal teas a great risk for human health, due to leaching from tea bag into the hot water during a tea preparation. In order to determine the current situation in Serbia, 50 samples of various herbal teas from 10 different domestic producers were collected from market. Samples were anlyzed using enzyme immunoassay for quantitative analysis (R-biopharm) and Elisa reader. Results ranged from 294 µg/kg to 1120 µg/kg. The highest contamination occured in teas made from mint leaves. If we take into a consideration sample preparation and the usual tea preparation procedure results could also be expressed per cup (200 ml): ranging from 29,4 µg/cup to 112 µg/cup. A provisional tolerable daily intake (TDI) for DON was set in 2002 by the Scientific Committee for Food (SCF) at 1 µg/kg body weight (b.w.) per day. At the moment maximum limit (ML) for deoxynivalenol in tea is not set.

16. The importance of accurate recipe calculation for policy making

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ABSTRACT

Objectives: Accurate food composition data is fundamental e.g. for research within epidemiology and in policy making by for example EFSA, Codex, OECD, WHO and national agencies. The aim of this project was to improve accuracy in food composition data by developing and validate a standardized method to calculate nutrient content in foods. **Material and methods:** A new general method for recipe calculation was developed; an important improvement was to standardize the procedure to reduce errors and to update factors for loss or gain of water and fat as well as retention factors for micronutrients. Foods and dishes in the Swedish national food composition database were recalculated (n=507). To validate the method dishes (n=34) were analysed using accredited methods at ALS Scandinavia and EuroFins. Energy and content of carbohydrates, water, ash, fat, protein, dietary fiber, fatty acids, vitamin C, folate, vitamin D, Fe, Ca, P, Se, I and Na were determined. Calculated nutrient content was compared with analysed nutrient content using the Wilcoxon's signed rank test (Minitab version 15.1). The importance of accurate recipe calculation was evaluated by recalculating and evaluating energy and nutrient intake from the dietary survey 'Riksmaten adults 2010-11' (n=1797) using the new validated method. **Results:** The results from the validation of the new method showed good agreement for macronutrients (0.98-1.10). However, the validation showed a non-significant overestimation for some micronutrients (agreement ranging from 1.00-1.22), for example about 15%

for iron. New evaluation of 'Riksmaten adults 2010-11' did not affect intakes of macronutrients as agreement was satisfactory (0.98-1.04). However, for micronutrients significant changes were found. For example intakes of vitamin A, vitamin D, iron, selenium and zinc were significantly lower, whereas intake of vitamin E was significantly higher. **Conclusion:** Today, average intake of vitamin D is below recommendations in many European countries, e.g. in Sweden intake is about 95% of the average requirement (AR, 7.5 µg per day). Furthermore, intake of iron for women in childbearing age is also about 95% of the average requirement (AR, 10 mg per day). With the standardized method for recipe calculation the intakes of those critical nutrients are even lower. This show how fundamental accurate food composition data is for adequate policy making.

17. Implementation of the first German Total Diet Study

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ABSTRACT

Food borne diseases are one of the major health problems worldwide. In the 21st century food processing and environmental impacts are getting increasingly in focus of dietary health concern. National food surveillance programs are monitoring high risk foods in a targeted approach. However, a more comprehensive monitoring with representative data for a whole population is needed to get a realistic picture of the overall exposure to chemicals and nutrients. Therefore, Germany started the first national TDS in 2015. The German TDS involves analysis of nine substance groups ('modules'), which will lead to a modular structure. Environmental contaminates have been chosen as 'core module' due to their ubiquitous occurrence. The core food list (max. 350 food groups) will be established through national food consumption surveys covering the age groups between 0.5 - 80 years. The estimated 25.000 single food items will be purchased considering regions, seasons, and production (organic vs. conventional). Centralized in Berlin, they will be prepared as consumed, homogenized, deep frozen and then delivered to contract laboratories for analysis. The stratification strategy for the core module will serve as basis for the modules veterinary drugs, mycotoxins, perfluorinated tensides (PFTs), pesticides and nutrients. The modules process contaminants, food additives and substances migrating from food packaging will require a more refined level of pooling according type of preparation, authorization and food packaging respectively, compared with the core module. In total the sampling of ~ 60.000 single food items is expected to cover each module. This innovative approach of using overlapping modules in a TDS will lead to the best possible balance between scientific requirements and financial and structural capacities. On the national level, results are highly relevant to close existent data gaps in concentration for contaminants, especially for processing related substances (e.g. process contaminants), of which a TDS is the only methodical tool to assess exposure. The results will be the basis for risk management decisions as well as for risk communication and can advise the food monitoring program to ensure food safety in Germany. On the European level, data can provide support for EFSA opinions in relation to foodborne risks.

18. Analytical advances in food-technology by establishment of a 14-C food–technology lab and kitchen

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ABSTRACT

Analyzing the fate and metabolism of ingredients or environmental and technical contaminations in complex matrices, such as food, is a major challenge in modern food processing. By using radioactively labelled substances any remains, metabolites and fragments can easily be monitored along the food processing chain. One major interest is the formation of flavour compounds and their fate during processing. Fraunhofer IME has set up a fully equipped laboratory kitchen and laboratory-scale food-processing facilities to be run under radioactive conditions. The core technical units are assembled around the four fields: 1. Domestic/restaurant kitchen; 2.Fruit technology, beverages, wine, oil and respective microbiology (presses, separators, mills, fermenters and pasteurizers); 3. Cereals, milling, baking technology (diverse mills, flexible extruder, separators, fermenters); 4. Meat and meat technology (masticator, cutter, kettle, injector, piston filler). This approach allows us to monitor the fate of agrochemicals and other contaminants as well as the fate and generation of flavouring compounds and the optimization of processing strategies. With these facilities on the other hand, we aim to identify precursors of desired and undesired cues and to optimize food processes to yield tastier, healthier and more sustainable food products.

19. Study design of KiESEL – a food survey of infants, toddlers and other children up to 5 years

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ABSTRACT

KiESEL is a cross-sectional study on food consumption of children in the age of 6 month up to 5 years. It is a module of the KiGGS study on health of children and adolescents in Germany, conducted by the Robert Koch-Institute. KiESEL collects representative data of 1000 children throughout Germany and is conducted by the Federal Institute for Risk Assessment (BfR) in the years 2014 to 2018. The data gained by KiESEL will update the consumption data among German children collected in 2001/2002 by the VELS-study (University of Paderborn), and will further replenish the considered age-group by 5 years old children. Therefore it will provide an actual and comprehensive data basis that will be used for exposure assessment, as part of risk assessment of Germany's youngest consumers. For collecting consumption data, parents and child care workers are asked to fill out a 3-day dietary record and some weeks later another 1-day dietary record. Further to written instructions, the participants will obtain explanations and support from interviewers. During home visits they conduct a parental interview about dietary habits (e.g. breastfeeding, formulas, dietary supplements etc.) including a food frequency questionnaire to cover also seldom eaten foods which are assumed to contain high amounts of undesirable substances. Moreover, the children's weight and height are measured in a standardized manner. The parents are trained to measure food which is eaten at home with a standardized kitchen scale. Foods that are eaten outside the home for example in children's day care or in restaurants are estimated via a picture book. The picture book contains images of various foods in different portions. Moreover, it includes a register of foods that are not shown as pictures to guide parents in selection of the best substitute picture. Returned dietary records are controlled with regard to contents. In case of missing information participants are contacted again to close the information gaps. The consumption data is coded with the software EAT which is also linked to Foodex. The results of the pretest (2014) lead to modifications regarding increasing compliance of the child care workers by simplification of dietary records and offering incentives for their institutions. Finally, the provided data can be analyzed in terms of nutritional aspects as well as food safety and exposure assessment of children and they will be provided for EFSA's use.

20. Prevalence and antimicrobial resistance of *Salmonella* spp. in environmental samples from poultry houses

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ABSTRACT

Prevalence and antimicrobial resistance of *Salmonella* spp. in environmental samples from poultry houses *Salmonella* spp. is one of the major pathogens causing enteric zoonotic around the world. Data collected by European Food Safety Authority (EFSA) in 2012 reported 91034 salmonellosis cases in Europe and 4181 in Spain. This places *Salmonella* genus as the second

responsible of zoonosis after *Campylobacter* genus. Thus, the aim of this work was study the prevalence and antimicrobial resistance of *Salmonella* spp. isolated from environmental samples from poultry houses. A total of 6000 samples were analyzed according ISO 6579:2003/A1:2007 and strains identified to serovar level by White-Kauffmann scheme. Antimicrobial minimum inhibitory concentrations (MIC) were determined using the microdilution broth method in accordance with the Clinical and Laboratory Standards Institute (CLSI). Twenty antibiotics were tested: ampicillin, cefotaxime, chloramphenicol, cinoxacin, ciprofloxacin, doxycycline, enoxacin, gentamicin, kanamycin, levofloxacin, minocycline, nalidixic acid, neomycin, spectinomycin, streptomycin, sulfamethoxazole, sulfisoxazole, tetracycline and trimethoprim. *Salmonella* spp was detected in 1,15% (69/6000) samples. The strains were: 16 *S.* Typhimurium , 10 *S.* Anatum , 6 *S.* Infantis , 4 *S.* Newport , 3 *S.*enterica serovar 4:b:-, 2 *S.* Bardo, 2 *S.* Ndolo , 1 *S.* Canada , 1 *S.* Cerro and 1 *S.* Seftenberg of strains belong to subspecie enterica. And 20 *S.* arizonae , 2 *S.* salamae , 1 *S.* diarizonae of another subspecies. All strains were susceptible to gentamicin, kanamycin, levofloxacin and trimethoprim. The number of resistance *Salmonella* strains were: 30 to sulfamethoxazole, 15 to doxycycline, 15 to nalidixic acid, 14 to streptomycin, 13 to minocycline, 12 to ampicillin, 11 to chloramphenicol, 7 to polimixin B, 6 to Spectinomycin, 5 to cinoxacin, 5 to ciprofloxacin, 5 to tetracycline, 4 to sulfisoxazole, 1 to cefotaxime, 1 to enoxacin. The number of antimicrobial resistance of subspecie *enterica* were significantly higher (P<0,05) than other subspecies and presented multidrug resistant. In conclusion, serovars from subspecie *enterica* are more prevalent in poultry houses and carry more antimicrobial resistance than other subspecies. In addition multidrug resistant is only present in subspecie enterica. Thus, *Salmonella enterica* subspecie *enterica* strains, from the first step of food chain, could represent a major public health problem.

21. *[Poster withdrawn]*

22. Prevalence and pathogenic potential of *Escherichia coli* isolated from a dairy production chain in Piedmont, Italy

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ABSTRACT

Objectives: The presence of *Escherichia coli* was investigated along a dairy production chain comprised in a small geographical area. Twenty farms were selected that conferred the milk to a single dairy plant producing an Italian blue veined cheese. Different samples were collected to identify putative dissemination pathways and assess the pathogenic potential of E. coli. **Materials and methods:** Samples from milk, milking filters, feed and cattle feces were collected on four sampling rounds. All samples were tested to detect *E. coli*, O157:H7 and extended-spectrum β-lactamases (ESBLs) producing strains. PCR was used determine phylogenetic groups and investigate the presence of 6 intestinal and 8 extra-intestinal virulence factors. **Results:** A total of 329 samples was collected: feces (n=74), milking filters (n=73), maize silage (n=70), unifeed (n=65), and milk (n=47). *E. coli* was detected in all feces samples; in 52% of milking filters, 6% of maize silage and 1,5% of unifeed. Milk samples were always negative. Among the 288 confirmed *E. coli* isolates, O157:H7 and ESBL-*E. coli* were never detected. Phylogroup A grouped 228 isolates (79%), while 39 were in B1 (14%) , 12 in D (4%), and 9 in B2 (3%). We detected the intimin gene in 13 isolates (4,5%, representing 8 samples); hemolysin in 11 isolates (3.8%, 8 samples); heat-stable enterotoxin and verotoxin 2 in one isolate each, respectively (0,3%). Isolates belonging to B2 and D were tested for the presence of extraintestinal virulence factors and six isolates were defined as extraintestinal pathogenic *E. coli*. **Conclusions:** The presence of *E. coli* in milking filters and feed suggests that fecal contamination might have occurred. Even if Good Agricultural Practices (GAP) and Good Hygienic Procedures (GHP) are in place, occasional dissemination of the microorganism can still occur. Thus, primary producers should regularly be made aware of the importance of GHPs. In spite of a high detection rate of *E. coli*, the prevalence of pathogenic *E. coli* in the tested population was limited. These findings suggest that the considered production chain can be considered 'safe' in respect to the general *E. coli* population.

23. Estimating inorganic arsenic dietary intake using biomarkers of exposure and duplicate diets

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ABSTRACT

Objectives: This study aims to estimate inorganic arsenic (iAs) dietary exposure in residents of As-rich areas of Latium (Italy) by means of a duplicate diet approach and biomonitoring. **Materials and methods:** Subjects (n=26) resident in the study area provided duplicate servings of all foods consumed in 3 consecutive days, keeping separate solid and liquid foods. Samples of drinking water, urine and toenails were collected from each subject. [iAs] was measured in dietary samples as described elsewhere (1). Toenail arsenic was used as long-term biomarker, reflecting chronic exposure over several months, whereas the sum of urinary iAs and the methylated metabolites MMA and DMA (iAs-met) was used as biomarker of short-term exposure (1-2). The relationships between log[iAs-met] in urine, log[As] in nails and iAs intake from the 3 diet components (solids, liquids, water) were analyzed by linear regression models, adjusting for age, sex and BMI. **Results:** The mean iAs intake was 0.22 µg/kg bw/day (SD 0.16). Solid foods were the main source of iAs (64% vs liquid component 17% and drinking water 18%). The proportion of iAs intake from the solid and liquid components was inversely related to the intake from drinking water. The median (range) of [iAs-met] in urine and [As] in nails were 11.7 µg/L (4.5-21.2) and 170 ng/g (54-665), respectively. The log[As] in nails and urine were correlated with the solid component of iAs intake (coef 2.5%, CI95% 1.0-3.9; coef 1.1%, CI95% 0.1-2.2, respectively). A correlation was also observed between log[iAs] in urine and the iAs intake through drinking

water (coef 3.2%, CI95% 0.1-6.3). Subjects using drinking water with higher As concentrations (11-18 µg/L) showed an average intake of 0.41 µg/kg b.w./day and an average [iAs-met] in urine of 17 µg/L. **Conclusions:** Duplicate diet studies are useful to estimate dietary exposures at the individual level and to identify which diet components contribute most to the total intake of iAs. Speciated As in urine and total As in nails are reliable biomarkers of dietary exposure; using both biomarkers is important to define the role of diet components. The present approach supports the characterization of iAs exposure in European scenarios, including the assessment of potential health risks in vulnerable subgroups. (1) Ann Ig 27:39 (2) Pure Appl Chem 84:203

24. 'alimentos PT.ON.DATA' – Information management system for the analytical data of food chain official control

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ABSTRACT

Objectives: To extend and upgrade the national information management system for the analytical data of the food chain official control 'alimentos PT.ON.DATA' to include all the data from contaminants, additives, pesticide residues and zoonotic agents, under the scope of the 'Pilot project on the implementation of SSD2 in the frame of the electronic transmission of harmonised data collection of analytical results to EFSA'. **Material and methods:** The system uses EFSA standards, Standard Sample Description (SSD) ver. 2 data model and catalogues, Guidance on Data Exchange (GDE) ver.2 and FoodEx2 food classification system and is an upgrade of the system developed under article 36 project 'Implementation of Electronic Transmission of Chemical Occurrence Data in Portugal', that contains a database with more than 40000 entries from official control (2009-2013) and uses SSD1, GDE1 and FoodEx1 as standards. The National Health Institute Doutor Ricardo Jorge and the General Directorate of Food and Veterinary Affairs, responsible for data transmission, contracted with EFSA the implementation of SSD2. Afterwards, a cooperation protocol was created with National Competent Authorities and Public Laboratories as partners of the project, namely Economic and Food Safety Authority, National Agrarian and Veterinary Research Institute and Portuguese Sea and Atmosphere Institute. Partners are defining new 'alimentos PT.ON.DATA' requirements and functionalities and harmonising sample collection forms. **Results:** The current system includes functionalities for data upload, mapping and validation, create XML files, search, statistics and administration tools. The upgrade includes functionalities to monitor sampling and execution of control plans in real time and also to report to different authorities, like EC, EFSA and Portuguese government. All sample collection forms were harmonised in order to fulfill SSD2 requirements. **Conclusions:** EFSA standards and its support to Member States contributed to foster national IT infrastructures and to increase data quality. The development of 'alimentos PT.ON.DATA' is supporting the country in the: i) electronic transmission of data to EFSA; ii) coordination of multiannual control plan (real time sampling monitoring and execution); iii) harmonisation of sampling records; iv) risk assessment associated to diet; v) management of the national food and feed safety system; vi) definition and implementation of policies for food and feed safe.

25. Gain edge from HACCP data to survey M1 aflatoxin in milk for human consumption

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ABSTRACT

M1 aflatoxin (AFM1) is a carcinogenic mycotoxin produced by several fungi that may contaminate crops and enter the food chain. The maximum level of AFM1 in cow milk for adults consumption is set at 50 ng/kg milk by EU legislation, but additional measures to reduce risk, such as alerts at lower AFM1 threshold, may be adopted at national and regional level. Good production practices to avoid AFM1 contamination are in place along the milk production chain and two risk mitigation points are setup at crop harvest and before milk is pasteurized or processed. In the Friuli Venezia Giulia Region (north-east Italy) surveillance of AFM1 in milk for human consumption is in place since 2011. Risk mitigation rely on testing at least twice a month all milk producers using milk tank from single or multiple herds. Not compliant milk tanks are discharged from human consumption and, together with alerts from threshold higher than 30 ng/kg, trigger public health (PH) authorities for actions to ensure good breeding practices are back to normal. Checking AFM1 content in milk tank is within the HACCP scope and tests are performed by private laboratories authorized by the regional PH authority. In 2013, crops regionally harvested were overall highly contaminated with AFM1. Despite management of non-compliant milk tanks and of >30 ng/kg alert, the overall burden of AFM1 in feed affected importantly the regional milk industry. To increase sensitivity, in 2014 the AFM1 surveillance was based on results from all milk testing. A data collection system (i.e. data model, reporting, curator and warehouse) was centralized and analysed monthly. Different commercial ELISA test were used by laboratories and 40% records were reported below different detection limits. We utilized all data available by imputing AFM1 contents reported below the detection limit with predictions from a model trained on available data. Simple indicators of centrality (median and weighted mean) were chosen to monitor deviance from the baseline AFM1 content in the regional milk production and were made available to stakeholders with friendly visualization (heat-map and boxplot). In 2014, indicators identified increasing contents of AFM1 in late spring despite low numbers of not-compliant milk tanks were reported. We concluded that surveillance of AFM1 in milk gains edge from all data available from HACCP allowing early detection of deviance from the regional baseline of AFM1 content in milk.

26. Evaluation of the acrylamide content in potato crisps from Spanish market – Trends from 2004 to 2014

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ABSTRACT

Acrylamide is a process contaminant generated in several foods during cooking as a consequence of the Maillard reaction. According to the European Food Safety Agency (EFSA), processed potatoes together with coffee and cereal based food, are the main sources of exposure to acrylamide in the diet. Specifically French fries and potato chips have been reported to contribute 56% of the total acrylamide intake in the Western diet of the adolescents. Due to this fact, authorities and producers have invested great efforts in mitigating acrylamide formation and reducing acrylamide concentration in processed potatoes. In the present study the acrylamide content of commercial potato crisps marketed in Spain in 2014 was determined. Results were compared with previous data reported by our research group in 2004 and 2009 with the aim of evaluating the trend since 2004 and the effectiveness of the mitigation strategies over the last years. The dietary exposure to acrylamide by Spanish population through the potato crisps intake was also estimated. Two different batches of 40 potato crisps brands from 18 producers were analyzed in September 2014. Acrylamide content ranged from 108 to 2180 µg/kg, with an average value of 630 µg/kg and a median of 556 µg/kg. Data revealed a continuous decreased trend from 2004 to 2014. In 2014, potato crisps showed

an average acrylamide content 14.8% lower than previous data reported in 2009 (740 µg/kg) and 57.6% lower than those from 2004 (1484 µg/kg). In a similar way, the observed signal value (90th percentile) decreased from 2270 µg/kg (2004) and 1377 µg/kg (2009) to 1169 µg/kg in 2014. These data confirm the effectiveness of the mitigation strategies implemented by the Spanish industrial sector. However, the 17.5% of the samples registered values higher than the indicative value recommended by European Commission for potato crisps (1000 µg/kg). The dietary exposure to acrylamide by Spanish population through the potato crisps intake was estimated to be 0.035 µg/kg body weight/day. In conclusion, although exposure has decreased over the last ten years, it is necessary to continue efforts to reduce acrylamide content in potato crisps since it is technically realistic.

27. EASIS (Endocrine active substances information system): a data management system based on an international reporting standard for regulatory & scientific purposes

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ABSTRACT

Chemicals that interfere with the endocrine system can potentially have adverse effects on both humans and wildlife. A wide range of chemical substances are under scrutiny for endocrine disrupting properties, including plastic additives in consumer goods and medical devices, pesticides and by-products of industrial processes A large number of chemicals have already been assayed for their ability to interact with the endocrine system in numerous separate initiatives. However, the resulting data reside in various incompatible databases and different formats. This heterogeneity in terms of data storage and retrieval restricts the ability of regulatory, industry and academic stakeholders to fully utilize available information for assessing risks connected to endocrine activity. To address this issue, the 'Endocrine Active Substances Information System' (EASIS) was created. It is based on the OECD Harmonised Templates (OHT), ensuring compatibility with major international data collection undertakings, such as REACH. The system contains data from a pre-existing DB created by the European Commission's Environment Directorate General – now complemented with data from more recent studies (2006-2013). The ca. 600 chemicals in EASIS (drugs, pesticides, industrial chemicals, consumer product chemicals, and new chemical entities) were tested in binding, reporter gene, cell proliferation, and in vivo assays in different species. The EASIS aims to provide read (query, download) access for the public, and write (submit, publish) access for registered members of a managed stakeholder community. It aims to centralize available data and stimulate further research in the field of endocrine active substances serving as a focal point collecting and making available studies on experimental systems, as well as results of field studies and human epidemiology data. The EASIS contributes to the recent efforts to move towards a mechanistic-based approach to prediction of toxicity, capturing mode of action based data along with apical adverse effect data in a structured knowledge base, facilitating the development of predictive pathway-based models for complex systemic toxicity outcomes.

28. Assessment of lead and cadmium residues in farmed fish in Machakos and Kiambu counties, Kenya

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ABSTRACT

Fish have long been known as a cheap source of proteins and a source of essential heavy metals that can be toxic at higher concentrations. Lead (Pb) and cadmium (Cd) concentrations were determined in muscle, gonad, liver and brain of tilapia fish caught from fish ponds in Machakos and Kiambu counties in Kenya. A total of 217 fish samples were randomly sampled from the two counties. Acid digestion method and atomic absorption spectrophotometer were used for analysis. Heavy metal concentrations varied significantly depending on the type of tissue analyzed. Generally, the highest concentration of Pb was detected in brain and the liver. Fish organs contained Pb in the following order: brain > liver > muscle > gonad, while Cd followed the order: brain > liver > gonad > muscle. Kiambu County recorded higher concentration of the studied heavy metals compared to Machakos County. Lead and Cd content in both counties studied exceeded the maximum allowable limit. The study recommended controlling industrial and agricultural effluents into surface water and proper sitting of ponds to minimize the risk of contamination of farmed fish by heavy metals.

29. A baseline microbiological hazard evaluation in mountain dairy products from North-eastern Alps

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ABSTRACT

Objectives: In the recent years the interest of consumers against mountain dairy products (MDP), generally manufactured from raw milk using traditional technologies, has greatly increased. However, a proper evaluation of the risk related to the consumption of these products does not seem possible yet, since data available are few and often refer to particular productions as PDO (Protected Designation of Origin) cheese and not necessarily representative of all MDP. The aim of this work was to evaluate the main microbiological hazard associated with MDP consumption. **Materials and methods:** Data were collected for research projects or control programmes performed by the competent Authorities from 2006 until 2014 summer season in typical dairy facilities of the Italian alpine pastures, so-called 'malghe', of the Veneto region and Trento Province. Food safety criteria such as *Salmonella* spp., *Listeria monocytogenes* and staphylococcal enterotoxins, and hygiene process criteria such as *Escherichia coli* count and coagulase-positive *Staphylococci* (CPS) count were evaluated in more than 2000 MDP samples (cheese, butter and ricotta) according to ISO standards. **Results:** *Salmonella* spp. was detected in only 3 products, while *L. monocytogenes* occurred in 1.9% of tested samples: 23 cheese, 2 butter and 2 ricotta. Staphylococcal enterotoxins were found in 9.5% of cheese, in one butter and in one ricotta. According to a durability study conducted on cheese naturally contaminated by staphylococcal enterotoxins, we observed that, in case of high initial contamination (SP>1,34), enterotoxins could persist even after 28 months of storage. Cheese samples were non-compliant for contamination level more than 100.000 cfu/g for *E. coli* in 17,4% and for CPS in 14%. Butter samples exceeded the limit of 100 cfu/g required by Regulation EC 2073/2005 for *E.coli* in 12.4%, whereas CPS concentration ≥ 10.000 cfu/g was detected in 35.4% of butter samples. **Conclusions:** Our data can be assumed as representative of MDP obtained by traditional methods in the malghe. The results suggest that the main microbiological hazard is represented by the possible presence of staphylococcal enterotoxins due to high contamination of CPS. Good hygienic practices could not be enough to reduce risks, considering that malghe are not always well-equipped. Therefore, further control options (such as technological practices) should be considered to prevent risks related to the consumption of MDP.

30. Natural plant food supplements and food coloring agents derived from *Vitis Vinifera*: a new source of human exposure to ochratoxin A

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ABSTRACT

Objectives: Red grape pomaces are rich in natural polyphenols that comprise numerous pigments. Thanks to their antioxidant properties, they maintain and promote health and reduce the risk of cardiovascular diseases. Grape pomaces are increasingly being used as starting material to produce plant food supplements (PFS), food coloring agents and tartrates. But these byproducts could be naturally contaminated by ochratoxin A (OTA), a mycotoxin with nephrotoxic and carcinogenic effects. In this study, we analyzed 24 commercial PFS, 13 food coloring samples, and 4 leavening agents derived from *Vitis vinifera* using an improved HPLC–FLD method for OTA determination (1). **Materials and methods:** Commercially products derived from *Vitis vinifera* were purchased from retail stores in South Italy or from e-commerce for a total of 41 samples. Each sample was extracted with organic solvents, purified with immunoaffinity column and analysed by HPLC/FLD (1). The identity and concentration of OTA in the purified extracts of two food supplements containing low levels of OTA were confirmed by LC–MS/MS. The method was robust, precise, accurate and applicable to all tested samples. Results of recovery and repeatability experiments were in the range of 87–102% and 2–4%, respectively; the values of limit of detection (LOD) and limit of quantitation (LOQ), calculated as signal-to-noise ratios of 3:1 and 6:1, were 0.50 µg/kg and 1.16 µg/kg, respectively (1). **Results:** OTA was found in 75% of PFS samples at levels of <1.16–20.23 µg/kg and in 69% of food coloring samples at levels of <1.16–32.00 µg/kg. The four commercial leavening agents containing tartrates were negative for OTA (1). **Conclusions:** In Europe the maximum levels of OTA in several food commodities are in force (2) but not for PFS and food coloring agents derived from *Vitis vinifera*. The high incidence of OTA contamination in these products suggests an amendment of the regulation to include them. We previously reported that OTA can occur at high levels (up to 849.10 µg/kg) in grape pomaces (3). These findings makes imperative to control for OTA contamination all grape pomaces destined to the preparation of edible products. (1) Solfrizzo et al. J. Agric. Food Chem.2015, DOI: 10.1021/acs.jafc.5b00326. (2) EC No.1881/2006 of 19 December 2006 and subsequent amendments. (3) Solfrizzo et al. J. Agric. Food Chem. 2008, 56, 11081–11086.

31. Consumption safety of pastries, confectionery and potato products as related to fat content in Poland

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ABSTRACT

Study aim: To determine the content of the main groups of fatty acids in pastries, confectionery and potato products, paying special attention to trans fatty acids and the products of fat oxidation and hydrolysis, as factors affecting the safety of consumption. **Methods:** A total of 157 products were collected in Poland in 2009 – 2010. In fats extracted from samples products of oxidation, and hydrolysis were assayed using peroxide (PV), anisidine (AnV) and acid (AV) values. The fatty acid (FA) composition, especially the trans FAs (TFA) content were determined by gas chromatography. When assessing the TFA intake, the Household Budget Surveys were considered. **Results:** Highest content of fat was found for wafers with filling and crisps (32.3 and 29.3%, respectively). In 4 out of 9 groups of pastry and confectionary products studied, the quality of fat was decreased due to an excessive oxidation, as evidenced by a substantial content of secondary products of fat oxidation. The extracted fat was rich in SFA (on average, 50 g SFA/100g FA) except fries and mixes. A great diversity of TFA content in fat of the products was found (0.1-24.8 g TFA/100 g FA). Wafers were characterized by the highest average content of TFA in the group of pastries (1.94 g TFA/100 g of product). Products of natural origin supplied 0.496 g of TFA per day, and those of industrial origin about 1.5 g. Conclusions: The estimated average TFA consumption, about 2 g/day, does not seem to affect health. However, an excessive consumption of pastry and confectionery products may present a risk, due not only to TFA but to a high consumption of toxic secondary oxidation lipid products as well. Moreover, as 75% of TFA in the diet were isomers of industrial origin, their further limitation and monitoring their level in food seems highly recommendable.

32. Inorganic arsenic in rice-based products for infants and young children

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ABSTRACT

Arsenic (As) is ubiquitous in the environment and inorganic arsenic (Asi) is a non-threshold human carcinogen. Other than cancer, human exposure to Asi has been associated to diverse health problems, which may be exacerbated with early life exposure. The main sources of human exposure to Asi are water and food. Asi in water is tightly regulated. However, there are no EU or USA standards for As in food, despite the fact food sources dominate the Asi exposure, especially rice and rice-based products. More Asi data of rice-based products consumed by infants and young children is needed to accurately define risk assessment for one of the most vulnerable subpopulation, and set regulations for Asi content in food to protect them. In this study, therefore, As speciation was measured in 29 commercial baby rice, 53 commercial rice cereals and 97 commercial rice crackers form the EU market, and compared to 85 commercial baby rice, 105 rice cereals and 199 rice crackers included in a US FDA survey on Asi in rice and rice-based products, and the findings put in context of exposure risk to infants and young children. All rice-based products included in the study were freeze-dried, and then powdered with a rotatory ball-mill. The powdered samples were weighted in polypropylene centrifuge tubes and 10ml of 1% nitric acid was added. Then samples were microwave digested in batches that included rice Certified Reference Material and blanks. Ultimately, As species were analysed using ion chromatography with ICP-MS. The Asi concentration in the rice-based products studied reached up to 0.268, 0.323 and 0.212 mg/kg for baby rice, rice cereals and rice crackers, respectively. Similar Asi concentrations have been found compared to the US FDA survey. The estimated Asi exposure showed values within the BMDL01 range identified by EFSA, which highlights that the risk cannot be excluded for infants and young children consuming rice-based food. Infants and young children consuming rice-based products are exposed to high levels of Asi that may adversely affect long-term health. This could be worse when these rice-based products represent a major part of the diet and Asi contribution is accumulated. There is an urgent need for regulatory limits on Asi in the EU and USA for food, especially for rice-based products in order to protect the most vulnerable subpopulations.

33. Levels of selected flame retardants and organochlorine pesticides in Slovak breast milk

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ABSTRACT

Objectives: The aims of this study were to estimate levels of seven polybrominated biphenyl ethers (PBDEs: BDE-28, 47, 99, 100, 153, 154, 183), 2,2',4,4',5,5'-hexabromobiphenyl (HBB-153), hexachlorobenzene (HCB), petachlorobenzene (PeCBz) and hexachlorocyclohexane (HCH) isomers in breast milk samples in Slovakia. Received data were used for estimation of daily infant intake (DI) of each group of pollutants. **Materials and methods:** Breast milk samples from 53 women were collected in eight regions of Slovakia in 2005-2006. After extraction and clean up, PBDEs and HBB-153 were separated on Rtx®-1614 capillary column and determined using high resolution mass spectrometer (DFS) coupled to Trace GC Ultra gas chromatographs (Thermo Scientific, Germany). HP 6890 Plus GC (Hewlett-Packard, USA) coupled to a

MAT 95XP (ThermoFinnigan, Germany) using DB-5MS column was used for separation and determination of α, β, γ - HCH, PeCBz and HCB. **Results:** Total median concentrations of ∑PBDEs, HBB-153, PeCBz, HCB and α, γ, β - HCH (ng.g-1 lipid weight) in the human milk samples were 0.5, 0.01, 0.2, 61.6, 0.2, 0.5 and 16.5 respectively. Estimated median values of DI (ng.kg-1, body weight assuming 5 kg body weight of infant in average) for PBDEs, HBB-153, PeCBz, HCB and α, γ, β - HCH were 1.8, 0.05, 0.8, 212, 0.7, 1.8 and 60.3, respectively. The data from this study showed several strong significant associations mainly between HBB-153, PBDE-153 and BMI and also between β – HCH and age. **Conclusions:** The present results are focused on concentrations of seven PBDE congeners, HBB-153, PCB-153, HCB, PeCBz and two isomers of HCH as well as on estimation of daily infant intake of those compounds. Although this study is limited by number of volunteers, it describes the level of contamination in Slovakia as well as its specific regions by individual organohalogen pollutants. The major added value of this study is determination of HBB-153 levels and the estimation of its daily infant intake, since there has been no study investigating this group of pollutants in Slovak breast milk up to present.

34. Risk assessment for lead in Cyprus & the use of IMPRORISK model

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ABSTRACT

Objectives: The objectives of the present work were to estimate the dietary lead intake of the adolescent population in Cyprus, to carry out the relevant risk assessment and to determine which food groups are the major contributors to the dietary lead exposure. **Materials and methods:** Dietary lead exposure was calculated by a deterministic approach using the IMPRORISK model, an empirical distribution model. Specifically the dietary lead intake was determined by matching lower, middle and upper bound mean occurrence data of lead in Cyprus with mean daily consumption and body weight for each individual (Childhealth survey of Cyprus) at level 2 of the EFSA FoodEx food categories [1]. Middle bound mean exposure was used to establish a relative ranking for the contributions of the different broad food categories of FoodEx. **Results:** Average lead dietary exposure ranged from 0,35 to 0,59 μg/kg b.w./day for mean consumers and 0,61 to 0,87 μg/kg b.w./day for high consumers. The broad category 'Grains and grain-based products' had the highest contribution to dietary lead intake. **Conclusions:** The above exposure estimates are below or exceed (for high consumers) the BMDL10 intake value for nephrotoxicity (0,63 μg/kg b.w./day) and are below the BMDL01 intake value for cardiovascular effects (1,50 μg/kg b.w./day). These findings are consistent with the relevant EFSA estimations for Cyprus [2]. References: [1] EFSA 2011. Evaluation of the FoodEx, the food classification system applied to the development of the EFSA Comprehensive European Food Consumption Database. EFSA Journal 2011; 9(3):1970. [27pp.] doi:10.2903/j.efsa.2011.1970. [2] EFSA 2012. Lead dietary exposure in the European population. EFSA Journal 2012; 10(7):2831. [59pp.] doi:10.2903/j.efsa.2012.2831

35. Aflatoxins' occurrence in cereals from Albania, exposure assessment to different population groups

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ABSTRACT

Objective: The aim of the study was to determine the occurrence of aflatoxins B1, B2, G1 and G2 in wheat and maize from harvesting year 2014 in Albania, and to estimate the daily exposure to adult population through wheat- and maize-based products. Aflatoxins, the most distinguished family of mycotoxins are produced mainly by fungus *Aspergillus flavus* and *A. parasiticus*, found especially in peanuts and cereals. Aflatoxins are natural carcinogenic compounds, and AFB1 is classified by IARC as the most naturally carcinogenic compound (Group 1). Consequently, there is no established Tolerable Daily Intake (TDI) for aflatoxins, and the consumers' exposure is preferred to keep in 'as low as is reasonably achievable' (ALARA) levels. **Materials and methods:** Wheat and maize samples were analyzed with multi-mycotoxin method using LC-MS/MS. The exposure assessment to aflatoxins for different population groups was calculated. Probable Daily Intake (PDI) for three population groups: infants (0-3 years), children (5-12 years) and adults (18-65 years) were obtained through integration of obtained aflatoxin analysis data combined with the food consumption assumption of population groups with body weight (b.w.) of 10, 25 and 70 kg, respectively. Three scenarios were evaluated based on: I) data from mean concentrations of positive samples, II) data from mean concentration of all analyzed samples where 'non-detect' was replaced by LOD/2, and III) maximum concentration found. **Results:** The incidence of sum of AFBs in wheat was 37.1% (13 of 35 samples), within the interval range 0.21 – 0.37 μg/kg, concluding that no sample exceeded the EU ML (4 μg/kg) for the sum of AFBs. In maize, the incidence of AFB1 contamination was 74.2%, and range 0.32 – 3550 μg/kg. The AFB1 detected levels in maize surpassed in 41.9% of all samples the EU maximum level (5 μg/kg). **Conclusion:** According to FAOSTAT, wheat and wheat-based products contribute by 93.3%, while the maize and maize-based products with less than 4% of the total cereal consumption in the country. Taking into account all three scenarios, the infants' group resulted most exposed regarding both commodities. In the first scenario the AFB1-PDI from wheat was 0.011 μg/kg b.w. /day (infants), 0.004 μg/kg b.w. /day (children), and 0.002 μg/kg b.w. /day (adults). The AFB1-PDI from maize was much higher 0.431 μg/kg b.w. /day (infants), 0.172 μg/kg b.w. / day (children) and 0.062 μg/kg b.w. /day (adults). In the third scenario the PDI, calculated with maximum detected levels reached the value of 3302 μg/ kg b.w. /day (infants) with maize. In conclusion, the main AFB1 exposure to all three groups of population originated from maize due to high level of contaminations. The incidence of AFs contamination to wheat was in non-alert situation for this harvesting year, while the maize contamination reveals high levels of AFs concentration.

36. Cross sectorial collaboration to prevent foodborne outbreaks

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ABSTRACT

Objectives: Promote collaboration and data sharing between ministries of health and education, municipalities and local authorities in order to contribute to prevention/control of FBO in Portugal. **Methodology:** A) Analysis of FBO investigation data, accordingly WHO and EFSA guidelines, in order to identify the consumer's bad practices that contributed to FBO occurrence and elaboration of a Manual of Consumer Good Practices. B) Promotion of collaboration between ministries of health and education, municipalities and local authorities, in order to implement the Manual of Consumer Good Practices as educative material. **Results and Discussion:** A) Considering the 84 FBO that occurred in Portugal between 2009-2013 in which food products were analysed in INSA laboratories, the majority occurred in domestic kitchens (29%) and canteens (26%) and the major food vehicle was mixed meals (70,3%). Considering EFSA's code system, FBO contributive factors were Inadequate time/temperature Storage (25,8%), Cross-contamination (19,1%), Inadequate heat treatment (14,6%), Infected food handler (14,6%) and Unprocessed contaminated ingredient (4,5%). To tackle these factors through food safety education targeted to consumers risk, we elaborated a Manual of Consumers Good Practices (from market to fork) including 8 chapters: 1)Food hazards, 2)FBO occurrence in Portugal, Europe and USA 3)Risk of infection at home, 4)FBO prevention, 5)How to improve food safety at home (care with shopping, storing and preparing food, maintaining cleanliness, separating raw and cooked food, 6)Cook the food thoroughly and keep food safe temperature, 7)Precautions eating away from home, 8)What to do in a suspicion of FBO and 9)Food preparation: identification and control of hazards. B) To prevent FBO across EU we need health literacy improving trough 'whole-of-society' and 'health in all policies' approaches, promoting collaboration between all stakeholders to include the Manual of Consumers Good Practices as part of educative materials. Conclusion: The FBO investigation data can be used as scientific evidence to guide the collaboration between all stakeholders in order to develop inform policies and strategies to change consumer risk behaviours for prevention of the occurrence of FBO, and minimize their human, social and financial burden.

37. Horizon 2020: shaping the EU research and innovation agenda in the food safety and related domains

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ABSTRACT

Horizon 2020 is the biggest EU Research and Innovation programme ever put into place by the European Commission, with a budget of nearly 80 billion euros covering the period 2014–2020. It sits over three pillars: excellence in science, industrial leadership and societal challenges. The latter addresses seven areas covering the key policy priorities of the Europe 2020 strategy tackling areas of major concern for society. Societal Challenge 2 on 'Food security, sustainable agriculture and forestry, marine and maritime and inland water research, and the Bioeconomy' has a budget of about 3.8 billion euros. It addresses the whole food chain and related services from primary production to consumption. Consumer health, food and feed safety, animal health and welfare, and plant health are addressed in a holistic manner integrated with food systems sustainability. Cross-cutting issues with Societal Challenge 1 on 'Health, demographic change and wellbeing' are present, making evident the relevance of food beyond one single area. The Horizon 2020 work programmes include calls for proposals on a range of research and innovation topics. They are published biannually. During 2014 and 2015, food safety topics considered the control of infectious epidemics and food-borne outbreaks through the rapid identification of pathogens, the assessment of the health risks of combined human exposure to multiple food-related toxic substances, and the biological contamination of crops and the food and feed chains. Animal health and welfare are included in topics aimed at tackling losses from terrestrial animal diseases and diseases of farmed fish and molluscs, and via a dedicated subtopic of the European Research Area. A topic on native alien pests in agriculture addressed plant health. Strategic programming is needed in order to identify key priorities within societal challenges, a task where the EU Member States in the Horizon 2020 Programme Committee have an important role. The work programme for 2016 and 2017 will show the commitment in place to continue advancing our understanding of food safety and related issues which are important for citizens in the EU and elsewhere. [This abstract shall neither be binding nor constructed as constituting commitment by the European Commission. For info on the research and framework programme Horizon 2020 visit: http://ec.europa.eu/research]

38. The First Hong Kong Total Diet Study (2010-2014)

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ABSTRACT

Objectives: To estimate dietary exposures of the Hong Kong population and various population subgroups to a range of chemicals, including contaminants and nutrients, and assess any associated health risks. **Materials and methods:** Over a hundred of substances such as persistent organic pollutants, pesticide residues, metallic contaminants, processing contaminant, mycotoxins and nutrients, was included in the study. 150 food items, which represented the Hong Kong people's diet, were collected and prepared 'as consumed' for analysis. The analytical results of the chemicals were then combined with food consumption data captured from the Hong Kong Population-based Food Consumption Survey on 5,008 people aged 20-84 to estimate the dietary exposures of the average (mean) and high (95th percentile) consumers of the population using an in-house developed web-based computer system. **Results:** Minerals More than 60% of the adult population had the dietary intake of sodium above the recommended maximum level of intake for sodium for adult established by World Health Organization while for calcium and iron, more than 90% and 80% of the adult population were below the respective recommended intakes. Acrylamide The margin of exposure (MOEs) of the dietary exposures to acrylamide of the average and high consumers were below 10,000. It may indicate human health concern. Stir- fried vegetables were found to be the major food contributor (44.9% of the total exposure). Methymercury The average consumers of women aged 20-49 (childbearing age) had dietary methylmercury exposure well below the HBGV. However, about 11% of this group had dietary methylmercury exposure exceeded the HBGV which may indicate that methylmercury exposure during pregnancy is a public health concern due to potential health risks to the foetus. Other contaminants (i.e. persistent organic pollutants, pesticide residues, metallic contaminants and mycotoxins) The dietary exposure estimates of general adult population were below their respective HBGVs. **Conclusions and recommendations:** Results suggested that the general adult population was unlikely to experience major undesirable health effects caused by exposure to most of the contaminants and nutrients covered in the TDS. However, there is a need to continue monitoring the exposures to contaminants and nutrients especially acrylamide, methylmercury and sodium of the Hong Kong population.

Weighing evidence and assessing uncertainty

39. Meta-analysis to better integrate human variability in toxicokinetic: CYP2D6-related uncertainty factors

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ABSTRACT

Conventionally, a 3.16-fold default uncertainty factor has been used in risk assessment to cover human variability in toxicokinetics. The objective of this work is to show how data collection and meta-analysis can be used to inform human variability in tokicokinetic and refine uncertainty factors. This work focuses on CYP2D6 which a major human polymorphic metabolic pathway. Published pharmacokinetic studies in Caucasian and Asian extensive (EMs) intermediate (IMs) and poor metabolizers (PMs) for CYP2D6 have been analyzed using data primarily related to chronic exposure (oral clearance and area under the plasma concentration time curve) and acute exposure (peak concentration). A multi-level hierarchical Bayesian model has been proposed to meta-analyze these data integrating all quantifiable sources of variability, including inter-compound and inter-study variability. This work highlights large inter-individual variability in kinetic within the Caucasian EMs (coefficients of variation above 50%). A slightly weaker inter-individual variability was found within Caucasian PM (coefficients of variation around 35%) and within Asian population (coefficients of variation below 40% for EMs and IMs). Comparisons between Caucasian EMs and PMs and Asian EMs and IMs revealed an increase internal dose for PMs and IMs with a ratio compared to EMs that ranges from 1 to 34 and from 1 to 5. Uncertainty factors were calculated for each subgroup and for the two populations taking the prevalence of PMs and IMs into account through Monte-Carlo simulations. It results that none of the subgroup would be cover by the 3.16-fold default factor. Indeed, a 11-fold CYP2D6-related factors would be necessary to cover 95% of Caucasian and Asian populations. Moreover, an exponential relationship was found between the percentage of CYP2D6 metabolism in EMs and the uncertainty factors for PMs. Such relation combined with in vitro method could be used to establish chemical-specific uncertainty factors or as input to for kinetic models used in risk assessment. This work highlights how published data can be combined into a weight of evidence approach to better assess variability and uncertainty in human risk assessment. Indeed, pathway related uncertainty factors derived in this work could be proposed to replace the default uncertainty factor in case of single exposure. Such approach could also be extended to investigate human variability in case of exposure to chemical mixture.

40. Communication of scientific uncertainty: international case studies on the development of folate and vitamin D Dietary Reference Values

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ABSTRACT

Objectives: Transparent evidence-based decision making has been promoted worldwide to engender trust in science and policy making. Yet, little attention has been given to transparency implementation. The degree of transparency (focused on how uncertain evidence was handled) during the development of folate and vitamin D Dietary Reference Values was explored in three a priori defined areas: (i) value request; (ii) evidence evaluation; and (iii) final values. **Materials and methods:** Design: Qualitative case studies (semi-structured interviews and desk research). A common protocol was used for data collection, interview thematic analysis and reporting. Results were coordinated via cross-case synthesis. Setting: Australia and New Zealand, Netherlands, Nordic countries, Poland, Spain and UK. Subjects: Twenty-one interviews were conducted in six case studies. **Results:** Transparency of process was not universally observed across countries or areas of the recommendation setting process. Transparency practices were most commonly seen surrounding the request to develop reference values (e.g. access to risk manager/assessor problem formulation discussions) and evidence evaluation (e.g. disclosure of risk assessor data sourcing/evaluation protocols). Fewer transparency practices were observed to assist with handling uncertainty in the evidence base during the development of quantitative reference values. **Conclusions:** Implementation of transparency policies may be limited by a lack of dedicated resources and best practice procedures, particularly to assist with the latter stages of reference value development. Challenges remain regarding the best practice for transparently communicating the influence of uncertain evidence on the final reference values. Resolving this issue may assist the evolution of nutrition risk assessment and better inform the recommendation setting process.

41. Consumer willingness-to-pay for farm animal welfare: a systematic review and meta-analysis

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ABSTRACT

Objectives: Farm animal health and welfare (FAW) is an important area of public policy, and increasingly so from a demand perspective. A meta-analysis and systematic review was conducted to establish the publics' willingness-to-pay (WTP) for FAW, piloting a tool to assess the uncertainty and strength of evidence in consumer decision-making. Data synthesis and integration methodologies are becoming increasingly important in ensuring that information from the ever expanding body of literature is presented in a more useable format, enabling policy makers and researchers to assess both the strength of evidence and uncertainty. The application of robust methodologies ensures only reliable data are included. **Materials and Methods:** Multiple databases were searched to identify relevant studies. Following a two stage screening process using a set of pre-determined inclusion criteria, 70 studies with 59 unique populations were included in the final analysis, with the strength of evidence and uncertainty for each study being assessed. Meta-regression based on random effects meta-analysis explored heterogeneity in relation to animal species, welfare measures, socio-demographic and socio-economic characteristics. Akaike's Information Criterion (AIC) was used to minimise over fitting. Sensitivity analyses were conducted to assess the risk of bias where appropriate. A cumulative meta-analysis was conducted to establish changes in WTP over time. **Results:** Variation in WTP estimates were reported in relation to animal species and participant characteristics. Reporting standards of studies were mixed. Only 65% of studies reported statistics to enable the weighting of evidence. 25% failed to report socio-demographic characteristics that would enable in-depth exploration of the data. **Conclusions:** A positive WTP for FAW was demonstrated, varying in relation to a number of factors. An evidence gap was highlighted in relation to WTP for specific animal production diseases associated with the intensification of production, along with the need for more consistent reporting standards.

42. The importance of an integrated approach for genotoxic testing

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ABSTRACT

The main goal of hazard identification is the realistic assessment of the potential risks a compound might have. Testing for genotoxicity is an essential part of hazard identification and is defined as the process in which the structure and/or information of the DNA gets altered. A range of in vitro and in vivo assays have been developed to identify substances which could trigger genotoxicity. The present study demonstrates the importance of a more integrated approach in the investigation of genotoxic potential of compounds, here okadaic acid (OA) and azaspiracid (AZAs). In contrast to other studies two additional non-genotoxic assays were included to reduce false positive/negative results by eliminating possible alternative explanation for the observed DNA damage, such as overt cytotoxicity and apoptotic processes. To investigate genotoxicity in the present study, DNA fragmentation was detected using the COMET assay. Additionally, the Trypan Blue Exclusion assay was used to provide information on possible cytotoxicity and cell number. Flow cytometer analysis was included to detect the possible involvement of apoptotic processes. In house data for all endpoints were established using positive controls. Three different cell lines, Jurkat T cells, CaCo-2 cells and HepG-2 cells, representing the main target organs, were exposed to OA and AZA1-3 at different concentrations and exposure times. Data obtained from the COMET assay showed an increase in DNA fragmentation for all phycotoxins, indicating a modest genotoxic effect. However, the data obtained from the Trypan Blue Exclusion assay showed a clear reduction in cell viability and cell number at the concentrations where DNA fragmentation was observed, indicating the involvement of cytotoxic and/or apoptotic processes. This was further supported by data obtained by flow cytometer analysis. All phycotoxins showed signs of early/late apoptosis. Therefore, the combined observations made in the present study indicate that OA and AZA1-3 are not genotoxic per se. Apoptotic processes appear to make a major contribution to the observed DNA fragmentation. Genotoxic testing is a key component of risk assessment and results of this study stress the importance of using a more integrated approach, including cytotoxicity and apoptosis studies to avoid false positive results due to other factors than direct DNA strand breaks.

43. Assessing multiple sources of cadmium exposure in an Italian population

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ABSTRACT

Objectives: Cadmium (Cd) is a heavy metal representing a serious environmental hazard to the human. Even though food and cigarette smoking are usually by far the main sources of exposure, outdoor air pollution could be an additional important source to be taken into account. Main anthropogenic sources of outdoor air cadmium are non-ferrous metal industrial production and fossil fuel combustion, followed by ferrous metal and cement production, and waste incineration. The aim of our study was

to assess the influence of outdoor air pollution on a biomarker of cadmium exposure. **Material and Methods:** Outdoor exposure to particulate matter ≤10 µm (PM10) from motorized traffic was assessed for fifty subjects randomly selected from Modena municipality residents, aged 35-70. We geocoded the residence of these subjects and modeled the corresponding ambient air PM10 concentration using the CAlifornia LINE Source Dispersion Model version 4 (CALINE-4) as a proxy of environmental air Cd level. We compared these estimate with the serum Cd, measured with inductively coupled plasma – sectorfield – mass spectrometry. Information on smoking habits and cadmium dietary intake were collected with a semi-quantitative food frequency questionnaire in order to assess possible confounding factors. We used both crude and multivariate linear regression models to determine the influence of outdoor PM10 levels, smoking and dietary Cd intake on serum Cd. **Results:** Median values (25th–75th) for serum and dietary Cd were 40.60 ng/l (30.05 - 53.5) and 13.36 µg/die (10.45 - 16.77). Crude β-coefficients were 0.617 (95% CI -0.194–1.428, P=0.133), 0.026 (-0.827–0.829, P=0.952) and 6.962 (-0.022–13.945, P=0.051) for PM10, diet and smoking, respectively. Adjusted values were 0.463 (-0.365–1.292, P=0.266), -0.036 (-0.866–0.793, P=0.930) and 6.057 (-1.175–13.289, P=0.099), respectively. **Conclusion:** In our population the most important factor influencing Cd serum content appears to be cigarette smoking, followed by outdoor air pollution (measured by PM10 levels) and lastly diet, possibly for the limitations of dietary assessment methodology. In addition, other unmeasured factors could have influenced serum Cd content, such as a slow release from liver and kidney due to long term exposure.

44. Effects of chemical mixtures in hymenoptera, reptiles and amphibians and variability associated

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ABSTRACT

Animal and ecological populations are subject to a multi-chemical exposure which may have adverse effects. The risk assessment of chemical mixtures raises several questions and it would be necessary to develop methodological tools that could improve the evaluation. The systematic review methodology was used to gather data on the effects of chemical mixtures in hymenoptera, reptiles and amphibians, and to make their assessment. Sixty-one publications mostly studying mortality parameters and reproductive and developmental toxicity in response to pesticides' mixtures were included (29, 12 and 20 studies for hymenoptera, reptiles and amphibians, respectively). If the most often measured effect in the context of chemical mixtures was the dose or response addition, synergistic effects were also found in significant number, mainly for pesticides' mixtures. Antagonistic effects were also reported, especially for mixtures of pharmaceuticals. Variability and magnitude of interaction were estimated according to different criteria (e.g., kinds of chemicals in association, toxicological targets…). These results were then compared with those of several groups of species like birds, molluscs, fishes, worms… , representing different ecological compartments and various relevant trophic levels to assess inter-species variability. Specific variability factors associated to the estimation of magnitudes of effects following combined exposure to multiple chemicals could be applied to specific groups of chemicals and/or species showing the most important variability, in order to improve hazard assessment.

45. Evidence synthesis and GMO impact assessment

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ABSTRACT

The European regulatory framework for the market authorization of genetically modified organisms (GMO) and products derived thereof demands a comprehensive risk assessment. Even though a large body of GMO safety data has been accumulated, it is difficult for stakeholders, risk assessors and the general public to fully overview the evidence base. The EU-funded research project GRACE (GMO Risk Assessment and Communication of Evidence) identifies the need to impartially compile existing evidence of potential impacts, including risks and benefits, caused by the deliberate release of GM plants on human/animal health, the environment and socio-economy. The evidence synthesis performed in the frame of GRACE is based on the outcomes of national, EU and international research activities. Systematic evidence synthesis approaches are already established in other research fields to support evidence-based decision-making. They represent powerful tools to collect, evaluate and summarize accessible research results in order to address a specific scientific question in a transparent, reproducible and unbiased manner. Thus, their adaption to and implementation in the impact assessment process for GM plants and products derived thereof aims to increase the transparency and supports the updating of science-based decision-making processes. GRACE aims to identify and to integrate the most appropriate evidence synthesis approaches in a unified framework for the impact assessment of GM plants by drawing on and adapting existing concepts and general guidelines. A set of research questions referring to health, environmental and socio-economic impacts is reviewed by GRACE and stakeholders are actively involved at both planning and interpretation stages. Review teams and stakeholders are supported in preparing and using reviews through the open-access database CADIMA (Central Access Database for the Impact Assessment of Crop Genetic Improvement Technologies) that mirrors the entire evidence synthesis process. The database will be permanently established beyond the lifetime of the GRACE project.

46. Weighting evidence can shape results: how to deal with 0 estimates in proportion meta-analysis

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ABSTRACT

Objectives: Systematic reviews are effective tools to provide qualified overviews of scientific evidence. In case of specific (closed framed) questions they allow a quantitative synthesis of data through meta-analysis. Proportion (one arm) meta-analysis can provide useful estimates to fill data gaps in risk assessment models but some problem could arise in combining studies with high heterogeneity, different sample size and comprising 0 estimates. Aim of this work is to analyse different transformations of prevalence data for the synthesis of literature. **Materials and Methods:** A dataset of data describing the prevalence of Toxoplasma in cattle was used for this exercise. Meta-analysis was performed with R ('metafor' package [1]). As the use of raw prevalence proportions are problematic for several reasons [2] applying three different transformation (logit, arcsine and double arcsine) on both raw data and data corrected according to 0.5 continuity correction [3,4]. For each transformation, forest plot with and without moderators (geographic area and analytical technique), funnel plot, cumulative meta-analysis (ordered by decreasing sample number), qq-plot, radial plot and sensitivity analysis were performed and compared. **Results:** The prevalence estimate is strongly dependent on the applied transformations. 0.5 continuity correction has a marked effect due to the presence in the dataset of studies with small N and 0 estimates. This influence is even higher if moderator or subgroup analysis is performed as results depend on the distribution of 'problematic' studies in different groups. **Conclusions:** As meta-analytical methods rely on the weight of single studies, variance represents a critical parameter, and its calculation has to be addressed carefully. Particular problems arise when datasets comprise studies with 0 estimates that are at risk of outweighing. Our work highlights the importance of transforming data with arcsine or double arcsine functions that give a more reliable weight to studies and do not need correction for 0 estimates. References[1] Viechtbauer W. Conducting Meta-Analyses in R with the metafor Package. J Stat Softw 2010;36. [2] Barendregt JJ, et al. Meta-analysis of prevalence. J Epidemiol Community Health 2013;67:974-8. [3] Cox DR, and Snell JE. The analysis of multivariate binary data. CRC; 1989. [4] Andreano A, et al. Measures of single arm outcome in meta-analyses of rare events in the presence of competing risks. Biom J 2015;00:1–12

47. A Bayesian approach to reduce uncertainty in the aquatic effect assessment of realistic chemical mixtures

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ABSTRACT

Species in the aquatic environment differ in their toxicological sensitivity to the various chemicals they encounter. In aquatic risk assessment, this interspecies variation is often quantified via species sensitivity distributions (SSDs). SSDs are typically constructed based on a sample of toxicity data reflecting the relative sensitivities of individual species, such as effective concentrations for 50% of the individuals (EC50s). If the concentration of a chemical in the environment is known, SSDs can be used to predict the fraction of species exceeding the endpoint considered. This is the so-called potentially affected fraction of species, or PAF. The PAF cannot only be calculated for single chemicals, but also for a mixture and it is then referred to as the multi-substance PAF (msPAF). To aggregate the individual contributions of single chemicals into an msPAF, the principles of response addition and concentration addition can be followed, or a hybrid form of the two in which concentration addition principles are followed for chemicals with the same toxic mode of action (TMoA), and response addition principles for (groups of) chemicals that have a different TMoA. The confidence that can be attributed to an msPAF depends, among other things, on how certain the parameters of the underlying SSDs can be estimated from the available data, i.e., how well the sample of test species represents the community of interest. Because the available data are often limited, optimal use of information is essential to reduce uncertainty involved in the assessment. In the present study, we show that the confidence intervals on the estimated potentially affected fraction of species after exposure to a mixture of chemicals at environmentally relevant surface water concentrations can be extremely wide if only toxicity data on the assessed chemical are considered using a traditional frequentist approach. As an alternative, we propose a Bayesian approach, in which knowledge on the toxicity of chemicals other than those assessed is incorporated in informative prior distributions, based on prior hypotheses assuming that the SSD parameters estimated for (a subset of) substances with sufficient available toxicity data, are representative for the range of possible SSD parameters of the chemicals of concern. A case study with a mixture of 18 pharmaceuticals demonstrates that this Bayesian approach results in less uncertain estimations of the multi-substpotentially affected fraction.

48. Cumulative risk assessment of organophosphorus, carbamate, pyrethroids and pyrethrins through vegetables consumption in spain

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ABSTRACT

Introduction: Pesticide use in the European Union is intensive and about 19% of the products used has its origin in Spain. One possible consequence of pesticide use is the presence of these residues in treated products and finally in the food chain. Objectives: The study was carried out to estimate the dietary exposure to organophosphorus (OPs), carbamate and pyrethroid and pyrethrin pesticides of the population of Valencia Region, and to evaluate the risk associated with this exposure. **Materials and methods:** Residues data for 84 pesticides were obtained from the Valencia Region monitoring programme 2007-2011, including 752 samples of vegetables selected based on the consuming habits. A questionnaire-based dietary survey was conducted in 2010. Dietary data were collected through a 24-hour recall in which 1478 subjects (from 6 to 95 years) were asked. A deterministic assessment was performed to assess the dietary exposure. Two scenarios were assumed for left-censored results: the lower-bound (LB) scenario, in which unquantified results (below the LOQ) were set to zero and the upper-bound (UB) scenario, in which unquantified results were set to the LOQ value. Cumulative exposure was also addressed because people are daily exposed to more than one compound via the diet and if these compounds could have the same mechanism of action. The conventional way of assessing the risk individually may lead to an underestimation. Relative potency factor approach was applied to estimate the cumulative dietary exposure to OPs, carbamate, pyrethroids and pyrethrins, using acephate, oxamyl and deltamethrin as index compound, respectively. **Results:** Ocurrence: A total of 488 samples (65%) had levels above the limit of quantification for any of the pesticides analyzed. Of these, only 14 (2%) exceeded the maximum residue limits established by law. Risk Characterisation: For the cumulative risk characterisation, the cumulative EDI was compared by the ADI of the corresponding index compund (acephate, oxamyl and deltamethrin for OPs, carbamate, pyrethroids and pyrethrins, respectively. **Conclusion:** In Spain, only the exposure of great consumers (P95) of vegetables could exceed the ADI of the index compounds in OPs and carbamates. Several parameters can affect the uncertainty of the calculated exposure such us the use of variability factors, processing factors and the target nature of the monitoring data.

49. Evidence-based hazard assessment of chemical mixtures

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ABSTRACT

The occurrence of interactions between chemicals in mixture poses several challenges to scientists and risk assessors. The current legislation is currently moving from assessments carried out on individual chemicals to approaches accounting for co-exposures (e.g., interaction-based hazard index approach). In this work, we aimed to support evidence-based human hazard assessment of chemical mixtures by providing quantitative information on both kinetic and toxicological effects of multiple chemicals. At a first step, we collected, using extensive literature searches/systematic review methods, pharmaco/toxicokinetic (PK/TK) and pharmaco/toxicodynamic (PD/TD) information on potential interactions between selected chemicals. Quantitative information was then consolidated via meta-analyses to quantify magnitudes of interaction and their inter-individual variability for both PK/TK and PD/TD dimensions. Our results allowed the estimation of the magnitudes of combined effects according to groups of compounds, metabolic pathways, toxicological targets…, and the derivation of probabilistic variability factors. As a conclusion, this work illustrates how human kinetic and toxicity data can be incorporated into the risk assessment process of combined effects of chemicals. Better estimation of variability and uncertainty, as well as possible prediction of magnitudes of effects of chemicals in mixture, can be set using these kinds of data and approaches.

50. Quantifiable uncertainty in Bayesian (microbial) risk assessments

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ABSTRACT

Objectives: Some examples are presented from various risk assessment projects in 2001-2015 where Bayesian statistics was applied to quantitative risk assessments (e.g. salmonella and campylobacter). The use of Bayesian methods has been gradually spreading as a formal method for quantifying uncertainties in food safety risk assessments and elsewhere. However, challenges range from sheer computational obstacles to lack of training. The examples deal with latent variables, nuisance parameters, missing data, and evidence synthesis. **Methods:** The essential nature of Bayesian methods lies in the updating of the existing state of uncertainty to a new state of uncertainty, by using the newly observed data as evidence. Formally, this follows from the Bayes' theorem in probability calculus. However, as the founding father of Bayesian statistics, Dennis Lindley, said: 'The main danger with (Bayesian) methods is that they are used too automatically'. **Results:** Lessons learnt? Naturally, when data contain little evidence, the obtained posterior distribution will not be much different from the prior distribution we started with. Interpretation and communication of the results may be challenging and requires assessing how much of the results actually were due to data. It is technically possible to build models with redundant parameters that may not be identifiable at all. The 'estimation' of such parameters would rely solely on the chosen prior, and the inherent un-identifiability would be hidden behind shallow prior assumptions. It is not enough to ask if the assumptions, reflected by chosen prior distributions, match the preconceptions of expert opinions, but do the results contain anything

more than the same preconceptions? In some situations, no amount of data can outweight the prior. It would then be misleading to communicate the results as model based estimates obtained from the data but rather an expert opinion. **Conclusions:** Attention should be paid to careful separation of assumptions and evidence, and to quantifiable and unquantifiable uncertainty. Skillfully used, and combined with sensitivity analysis and model comparisons, Bayesian methods can provide very flexible tools for evidence synthesis, and even for quantifying uncertainties that at first may seem unquantifiable!

51. Cost-effectiveness of alternative strategies for surveillance of BSE and classical scrapie in Great Britain

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ABSTRACT

Objectives: Active surveillance for bovine spongiform encephalopathy (BSE) and classical scrapie is compulsory in the EU, but their incidence has declined consistently in GB in recent years. The objective of this study was to explore alternative cost-effective surveillance strategies to save resources without compromising the ability to detect new or re-emerging threats in a timely way. **Methods:** In order to align with current policy objectives, a consultation was held with policy makers. It was decided that scenarios to be considered should include changes to the proportion/number of animals sampled and, for scrapie, changing the ratio between abattoir and fallen stock testing. Outcomes to indicate the effectiveness of surveillance were agreed, e.g. the time until a significant increase in prevalence is detected. Outcomes were modelled for each scenario using a bespoke algorithm. A stochastic economic model was used to estimate the cost of each scenario, and incremental cost-effectiveness ratios (ICERs) were used to describe cost-effectiveness. ICERs are often used in human health to evaluate proposed interventions. In this case, a proposed reduction in spending is compared to an increase in an undesirable outcome, therefore we use a 'reduction-acceptability' ratio or RA-ICER, calculated as per: -(Cp-Cb)/(Op-Ob) where C=cost; O=outcome; p=proposed scenario and b=baseline. **Results:** Reducing the proportion of fallen cattle tested for BSE from 100% to 75% is the most cost-effective strategy, with mean cost savings of £716,000 pa. Testing 50% of fallen stock results in a mean cost saving of £1.43 million pa, but with reduced cost-effectiveness. Policy makers must decide whether this cost-effectiveness is within an acceptable limit. Cost-effectiveness thresholds are already well defined in human health settings. For scrapie, testing only fallen stock produced the most cost-effective strategy with a mean saving of £104,000 pa. Reducing the number of fallen stock tested by half resulted in a mean saving of £283,000 pa but was less cost-effective. **Conclusions:** This method was developed in consultation with stakeholders in response to the need to downscale surveillance while accepting some reduction in effectiveness. Cost-effective alternative strategies were identified and RA-ICERs enabled the explicit comparison of alternative policy options. Policy makers may consider defining a threshold of cost-effectiveness acceptability, applicable to other diseases and settings.

Expertise for the future

52. 'Safe Food' Project – Food safety for children

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ABSTRACT

There is a growing number of declarations due to food-borne disease, making them a public health concern, requiring efforts towards preventing them in children, promoting child health and healthy lifestyles. The present study focuses on a population of 3339 students with ages between 10 and 15 years in the Great Lisbon area. It integrates the 'Safe Food' project, created by the Division of Food Risks of the Economy and Food Safety Standards Authority (ASAE), as part of their Food-Risk Communication Strategy. The objectives of this study are to provide education sessions on food safety, and to assess the degree of student knowledge before and after the sessions. Students exhibited most knowledge on the topics of labelling and hand-washing, and least knowledge on the topic additives. The percentage of increase in correct answers was calculated, revealing significant improvements in knowledge—as high as 126% and 475% in some questions. The analysis of the evolution of 'don't know' answers yielded a low repetition rate, with migration to the correct answer above 98%. This study illustrates the effectiveness of educative interventions in this age segment, and establishes baseline knowledge for further research, especially the assessment of their long-term effects.

53. Perception and awareness of the European Union food safety framework

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ABSTRACT

Objectives: The aim of the study was to register the response of different social groups of the Turkish public opinion to specific parameters of food safety issues and to compared them with EU citizens. **Materials and methods:** Ten specific questions on the perception of food safety were answered by three groups of subjects. The first group (Turkish Educated Group, TG) was from Turkish academic and administrative staff (242 persons). The Europeans (73 university students and professors) attending an Erasmus Intensive Programme on Food and Feed Safety (IPRASAFF, 2012) was the second group (EG). The third group consisted of randomly selected subjects living all over Turkey (250 persons, Turkish Public Group, TPG). **Results:** The majority of the respondents was aware which authority is responsible for food safety at national level but did not clearly understand how to make food complains (mostly made to food companies instead of public institutions). The concept of food safety and subsequent behaviours were greatly different among socialdemographically different classes at both national and international level. The TPG showed a higher concern (compared to TG and EG) towards the intrinsic safety of the food products and probably to the family safety (i.e., food terror law, ban of unauthorized slaughters, less trust in open-air markets, severe control of wholesalers, publication of the producers implicated in frauds, publication of the results of the public controls). The manufacturer name and price were important for the Turks, the food label for EG. 'Food safety' was associated to 'quality control' and 'healthy life' by the TG and EG groups; however, the TPG understood it as 'healthy life' and 'food terror'. Individuals with higher education showed a high interest in the food package. Halal certification was highly appreciated by both TG and TPG. **Conclusions:** Since the consumers sometimes give more weights to the negative than the positive information, this proves that the perception is a complex issue mediated by individual and social factors. GMOs are rated as highly unknown risks due to their unknown consequences. In the present study, both Turkish educated and public groups highly rated the GMOs as risky, while the European educated group did not. Thus, the public awareness of Turkish public about recently introduced aspects of food safety related to the EU accession negotiation must be improved.

54. Food risk-benefit assessment: an emerging scientific discipline

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ABSTRACT

The impact of food consumption on human health has been studied for decades in the scientific fields of microbiology, nutrition and chemistry. However, to assess the overall impact of food on human health, a tri-disciplinary approach is necessary. Food consumption can lead to negative

and also to positive health effects. For these two reasons, Food Risk-Benefit Assessment (RBA) has recently emerged as a new scientific discipline. The first objective of this study was to review what has been done thus far in RBA in terms of food applications and methodologies associated with food consumption. Based upon this synthesis, the second objective was to develop an integrative and multi-disciplinary framework in which to build a food RBA. To perform the analysis, 70 articles dealing with RBA studies and 34 dealing with the RBA methodological framework were gathered from databases, websites and scientific journals. Although studies were relatively diversified to face various issues regarding the 'farm to fork' supply chain, the most frequent research topic was the assessment of fish consumption with a comparison of nutritional beneficial effects and chemical adverse effects. In addition, the majority of studies were not fully quantitative but based on the comparison of consumers' exposure to risks and benefits with safety reference values, such as tolerable weekly intake for chemicals and reference daily intake for nutritional parameters. The methodology undertaken so far in RBA has been mainly based on the traditional risk assessment process yet broadly applied in the fields of chemistry or microbiology. However, this emerging field is not only the addition of a risk assessment and a benefit assessment but a more complex, integrative and multi-disciplinary strategy, requiring the harmonization and comparison of different health impacts, often expressed in different ways, for different scenarios of consumption. Subsequent work will focus on the development of quantitative RBA based on an assessment of the overall impact of food on human health. An integrative, multi-disciplinary and quantitative RBA will be a valuable tool to address future challenging issues in terms of balancing food safety and nutrition. The authors would like to acknowledge the technical training from the Erasmus+ Intensive Programme 'Quantitative Tools for Sustainable Food and Energy in the food chain' of the European Union, Project No: 2014-1-MT01-KA200-000327.

55. Food Safety in Domestic Environment : why we need a transdisciplinary approach

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ABSTRACT

The improvement of food safety in the domestic environment requires a transdisciplinary approach, involving interaction between both the social and natural sciences. This approach is applied in a study on risks associated with *Campylobacter* on broiler meat. First, some web-based information interventions were designed and tested on participant motivation and intentions to cook more safely. Based on these self-reported measures, the intervention supported by the emotion 'disgust' was selected as the most promising information intervention. Its effect on microbial cross-contamination was tested by recruiting a set of participants who prepared a salad with chicken breast fillet carrying a known amount of tracer bacteria. The amount of tracer that could be recovered from the salad revealed the transfer and survival of *Campylobacter* and was used as a measure of hygiene. This was introduced into an existing risk model on *Campylobacter* in the Netherlands to assess the effect of the information intervention both at the level of exposure and the level of human disease risk. We showed that the information intervention supported by the emotion 'disgust' alone had no measurable effect on the health risk. However, when a behavioral cue was embedded within the instruction for the salad preparation, the risk decreased sharply. It is shown that a transdisciplinary approach, involving research on risk perception, microbiology, and risk assessment, is successful in evaluating the efficacy of an information intervention in terms of human health risks. The approach offers a novel tool for science-based risk management in the area of food safety.

56. Law and politics for food safety: what role for the European Union?

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ABSTRACT

Regulation (EC) 178/2002 of the European Parliament and the Council of the EU of January 28th 2002 defined the General EU Food Law. It introduced a common legal framework at EU level relating to general principles and legal requirements of foods. The regulation also instituted the European Safety Authority, and fundamental procedures for food safety. Since then, the action of the European Union relating to food safety requirements is increasingly evolved, taking into account both the health and consumers' interests and rights, and the good functioning of the internal market. The present proposal would aim to present an overview of the principles, standards and procedural requirements fixed at EU level in order to familiarize the non specialized public with the institutional approaches and methods implemented at the EU level. How European Union deals with, for example, crisis management? Which Institutions are legitimate to intervene and how they do it? What is the role of Member States and how they coordinate their respective action with EU Institutions? These questions are sometimes unknown by the general public and the present proposal will try to clarify this aspects. First part conclusions' will underline possible challenges and/or gaps. Further to the internal action on food safety, the European Union, as one of the biggest actors on the global food market, have an important role in promoting an integrated approach to food safety in international forums. Particularly in promoting health and food safety common approach to global trade negotiations on food and agriculture. Consequently, the second part of the present proposal will make reference to the European Union action on food safety in its 'dress' as Global Actor in the context of the World Trade Negotiations on Food and Agriculture, underlining, inter alia, coherence/incoherence between internal and external action of the EU, progress and future challenges for EU law and regulations.

57. Health risk assessment of chemicals – A commentary on requirements for the provision of training

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ABSTRACT

Training programs on chemicals and health risks exist within different European Organisations and Universities, but currently there are no agreed European standards on the training of chemical health risk assessors. The need for practical training has been recognised in meetings on need for risk assessment training organized by DG SANCO (DG SANCO, 2007, 2008). Data collected in the market study 'Preparation of a mapping of existing courses relevant to the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH) and the classification, labelling and packaging (CLP)' available from universities, other academic institutions and professional organizations within the European Economic Area (EEA) commissioned by the European Chemicals Agency (ECHA) to the University of Milan, support the limited number of comprehensive full courses available throughout Europe offering the necessary training requirements in human health risk assessment (University of Milan, 2012). The requirements for the provision of training in the field of human health risk assessment of chemicals draw on the experiences gained from many training initiatives throughout Europe, for example training qualifying for EUROTOX European Registered Toxicologist (ERT) and Guidance on Risk Assessment Advanced Training Programme (RAAP), the EU-funded-projects: European Toxicology Risk Assessment Training (TRISK) and Risk Assessment and Management – European Training Programme (Risk Assets). Despite such interest, however, it appears that a comprehensive training course that truly focuses on all steps in the risk assessment process and provides both theoretical and practical training is not harmonized throughout Europe. Assuming all goes well and the EN 16736 'Health risk assessment of chemicals - Requirements for the provision of training' will be approved in a final formal vote, a European standard will be available and adopted in

late 2015 by all CEN member states. The draft standard specifies that ideal courses in health risk assessment of chemicals should be multidisciplinary and cover toxicology, epidemiology, exposure assessment, risk characterization, ethics and quality control and implications for risk management and risk communication. Also applied training with hands on experience such as case studies or examples of concrete risk- assessments should be part of these courses.

58. Veterinary Day One Competences: an overview on the different approaches

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ABSTRACT

Veterinary professionals play a key-role in ensuring food safety due to their multi- and interdisciplinary background. To meet the worldwide challenges and to be able to act in accordance with law, particular factual knowledge, competencies and skills are required from new (Day One) veterinary graduates. Many organizations constituted different 'Day One Competences' lists to combine the above requirements with appropriate experience. Authors overviewed and compared the above lists of the European Union (EU) and international veterinary organizations. The EU legislation delineates the framework for veterinary professional education while putting less emphasis on highlighting each aspects of food hygiene; it stresses competence in all veterinary fields, adequate knowledge, the importance of skills and good practice. In the core curriculum of veterinary professional programs, food hygiene and technology, inspections and control of foodstuffs of animal origin, and practical experience appears. EU legislation also covers the requirements for becoming an official veterinarian. Official veterinarians should pass a test that also involves topics on food safety: the principle, concepts and methods of risk analysis. Several international veterinary organizations promote 'Day One Competences' to meet both public and private sectors requirements towards day-one veterinary graduates. All of them list contribution to public health and food safety/food hygiene and its control as part of the core curriculum. Some also stresses the importance of theoretical and practical food inspection. Only one of the organizations articulates the importance of the food chain approach and risk-based slaughter inspection while it recommends comprising the principles of risk analysis in the epidemiology courses. The development of a standardized 'Day One Competences' list might enhance the harmonization of worldwide veterinary professional curricula and also might be a good tool to emphasize the complex, multidisciplinar and chain approached nature of the field. In the elaboration of the standardized competences list the European Food Safety Authority should also be involved.

59. Keeping food safety challenges on current and future agendas: The European Association for Food Safety, SAFE consortium

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ABSTRACT

The European consumer trusts that the food supply is safe. Companies and official agencies spend hundreds of millions of Euros every year to maintain the high level of food safety that we enjoy. Nevertheless, food safety crises do occur, often resulting in sickness and loss of life and certainly causing consumers to lose faith in food providers and regulators. The SAFE consortium (SAFE) works to keep food safety high on the agenda of funding bodies, regulators, government, industry, researchers and the general public, partly by drawing the attention of research programmers to the key current issues relevant to food safety. SAFE encourages scientific discussion by publishing White Papers such as 'Keeping Food Safety on the Agenda', in 2013, and more focused position papers such as 'Update on Microbial Safety of Fresh Produce', in 2012. The consortium offers a unique contribution to food safety research discussions through workshops and seminars, most recently 'Keeping Safety and Integrity in the Food Chain: A View from Many Sides' in March 2015. This event brought together representatives from research, universities, industry, consumer groups, regulators and assessors to share expertise and further develop an integrated vision of where food safety is and where it needs to be. SAFE also participates in research projects such as TRACK-FAST, which focused on education of the food scientists of the future. SAFE builds partnerships among organisations with different missions to further a single agenda: Food safety for today and tomorrow. SAFE members choose the food safety topics that the consortium focuses on and all SAFE members are invited to participate in member meetings where all SAFE voices are heard. SAFE is a non-profit international association registered in Belgium in October 2002. Members are organisations that do food safety research & development, that are managerially independent and of high scientific excellence. In 2015, members include national research institutes, private research & development foundations, university faculties, industry with food safety interests and governmental food safety authorities. The SAFE network covers, in an interdisciplinary way, all fields of food safety, from food microbiology to new technological approaches, food security and protection of the European consumer. SAFE receives no government or other funding but is self-sustainable based on the contributions of its members, and fully independent.

60. Analysis of Nutrition Education Within the Compulsory Schooling in Czech Republic

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ABSTRACT

The average annual health expenditure per capita in Europe has, in the last 15 years, more than doubled. The rapid rise of health-care expenditures predicts strong economic utilization across the European Region and State economics of all Member States. Lifestyle changes such as diet and exercise continue to remain the most effective way to prevent long term health care complications which ultimately reduces the financial burden of associated health care costs. Inappropriate diet and lack of physical activity are the highest correlating factors to the obesity epidemic. Along with heavy alcohol consumption and smoking, these four lifestyle choices contribute the most to the development of chronic non-communicable diseases. According to WHO (2014) these diseases kill more than 36 million people worldwide annually, with more than 9,000,000 deaths occurring before the sixtieth year of life. In 2007 the Health Education was implemented in to the curriculum of youth between the ages of 3 to 19, to improve the health of Czech students. The issue of health promotion and proper nutrition is now an essential part of primary and secondary educational process. The main research objective of this study was to analyze and to evaluate the current phase of implementation of Nutrition Education within compulsory (lower secondary) education in the Czech grammar schools (CzGS). Headmasters of 348 different CzGS were asked to push Health Educators or Nutrition Educators to fill in the on-line Teacher Questionnaire about Nutrition Education implementation. 113 teachers (102 women and 11 men) averaging 45.3 years old and averaging 21.5 years of teaching completed the questionnaire. The results shows among others that Nutrition Education is the most frequently taught within Health Education, Family Education, Natural Sciences or Physical Education classes. The most frequently lectured topics related to nutrition are Principles of Healthy Eating, Nutrition and Healthy Lifestyle, Nutrition and Non-communicable Diseases, and Eating Disorders. 82.6% of educators search information for lesson preparation online using the Internet. The most widely used educational tools in classes are the Internet, videos, magazines, school kitchen equipment, food containers, and real food samples. The results of this study could be an initiative for Nutrition Education curriculum revision and the results provide the background information for designing of new Nutrition Education Program for CzGS.

61. Knowledge gaps and research needs in the evaluation of the effects of GMOs

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ABSTRACT

Objectives: This poster aims to identify knowledge gaps and future research needs on the effects of genetically modified organisms (GMOs). It will present the results of a mapping exercise of existing research activities on the effects of GMOs in the EU, and the main outcomes of a workshop with relevant experts and stakeholders, European institutions, and civil society organizations (CSOs). **Methods:** This first step was a mapping of existing research activities and knowledge regarding the health, environmental and socio-economic effects of GMOs in the EU. Based on this mapping exercise, a workshop, involving 23 experts and stakeholders from the academic, Member State and EU agency, CSO communities, was held in Milan in 2014 aiming to define a comprehensive list of research needs regarding the effects of GMOs in the areas of human and animal health, environment, and techno-economics and societies, as well as requirements for sharing of available research capacities and existing infrastructures. **Results:** Based on the information of 320 mapped projects, the workshop allowed to define a long list of research needs covering 40 issues in the area human and animal health (in the areas of food and feed safety, nutritional value, toxicity and allergenicity). A total of 67 research needs were identify in the area of environment (in the areas of biodiversity, soil, water, plant pest and diseases, air, ecosystem services and climate change). The long list of research needs developed in the area of techno-economics consisted of 70 items (in the areas of costs, profitability, coexistence, legislative framework, socio-economic, macro-economics, and yields). **Conclusions:** As a consequence of the myriad of research initiatives, there is a need to significantly enhance the alignment of the research programmes of the individual MSs, in order to avoid duplication of work in these areas, to leverage complementarities, and to enhance cooperation between scientists. This should be done improving the involvement of stakeholders (e.g., industry, farming organisations, NGOs, EU and national competent authorities, academia) in the shaping of future research agendas and programmes, in order to make these research programmes more meaningful to the end-users of the scientific results, and to increase legitimisation of research trajectories and ownership.

62. The Role of Marie Curie (MSCA) Actions in shaping the future of Food Safety

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ABSTRACT

Introduction: The Marie Curie Actions or the MSCA Actions within the FP7 and Horizon 2020 framework is the gateway to one of the top-class research funding instruments of European Commission. To this date, MSCA has massively invested and played a crucial role in developing the next generation of early stage researchers in Europe who have done ground breaking research in the inter-disciplinary and complex field of Food Safety thereby securing and shaping the future of Europe and the health of its citizens. **Objectives:** The aim of this Poster is to present an global overview of the role of MSCA Actions in shaping the future of Food Safety. The Poster will highlight how MSCA projects have trained and developed a new breed of early stage researchers in Food Safety and the tangible contributions that they have made so far to the European society at large. **Result:** The Poster will also showcase the unique diversity and inter-disciplinarity of this field by presenting some practical examples and concrete results from projects like IMPRESS (http://www.impress-itn.eu/), CREAM (http://cream-itn.eu/) and HST Food Train (http://hstfoodtrain-itn.eu/). **Conclusion:** This Poster perfectly fits with the session on 'Expertise for the Future'. This Poster will connect MSCA and EFSA and will present how MSCA has/is contributing towards developing the expertise for the future.

63. A Raspberry (Pi) in the LAB: consumer electronics and 3D printing as innovative tools for scientific training and research

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ABSTRACT

The recent advancements in the field of consumer electronics is dramatically expanding the employment of smart, low cost devices for specific needs, beyond the common use. A fascinating approach, particularly in the field of research, prototyping and for educational purpose is represented by the fast growing area of the universal electronic modules. These tools are widely used by schools and enthusiast for learning the basics of electronics, informatics and engineering and to make objects for entertainment, internet-of-things or accomplish specific informatics-oriented tasks. These devices are often 'open source' systems, are quite inexpensive and a wide range of sensors, actuators and expansion modules are already available. The modularity, along with powerful open-source software libraries, makes them perfect tools for research, trial-and-error learning and, more in general, for cross-contamination of science fields. Despite the great potential of such devices, very few efforts to hack and adapt these tools in life science-related fields are reported to date. The results presented herein are focused on application of Raspberry Pi to chemistry analysis. Raspberry pi is a low cost (30€) single-board credit-card sized computer developed with the main aim to advance the education of adults and children in the field of computers, computer science and related subject. The Raspberry Pi card and its CMOS camera, assembled with 3D printed parts and inexpensive opto-electronic components, was employed to build cheap readers for absorbance and fluorescence for food and medical analysis. The software and the user-friendly GUI that manage the whole analysis was developed entirely using Python and some free python packages. In order to test the absorbance-reader for diagnostics-oriented applications, a DPPH-based assay for the evaluation of scavenging activity of antioxidant molecules was carried out using the Raspberry Pi-based device. The analysis of bottled beverages performed in triplicate was compared with a conventional laboratory-based approach showing no significant differences. The reader for fluorescent assay was employed to read fluorescein in nano-molar concentration and was able to detect positive samples of the foodborne virus norovirus after PCR amplification. The approach presented herein is a valuable tool for low-cost, citizen involvement in science and will help the DIY approach in science, opening new opportunities in research and education.

64. The future of Georgian food risk assessment

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ABSTRACT

Objectives: The research aims to determine the potentials and opportunities for food risk assessment in Georgia using methods of gap analysis. This will be a valuable contribution to communications between various stakeholders regarding future results of food risk assessment. The results of this research will also constitute an important tool and reference point for policy development related to food risk assessment in Georgia. **Materials and Methods:** Materials and methods will help determine real problems of food risk assessment in Georgia and develop corrective measures. Data will be collected using methods such as opinion polls of different focus groups (representatives of governmental institutions, the Council of Scientific Risk Assessment, experts, foreign experts, and representative public samples) regarding the current state of food risk assessment in Georgia. This data will be collected and analyzed using statistical data processing and adjustment methods. **Results:** Results will identify deficiencies in risk assessment in Georgia, a system that remains ineffective despite recent activities for improvement. For example, food-related health risks to the population are not adequately analyzed. In addition, current legislation does not obligate relevant governmental authorities to obtain qualified scientific opinions necessary for the implementation of risk

analysis, as well as for elaborating a methodology for risk assessment, and for scientific and technical research data analysis. Without these practices and laws in place, comprehensive risk assessment is impossible. Introducing this project's research results to the public and professional groups will promote integrated and effective approaches resulting from identifying and addressing problem areas, as well as developing relevant preventive actions. **Conclusion:** Experienced, expert human resources are very important for the development of risk analysis capabilities in a country, yet there is a very scarce amount of appropriately-qualified specialists in Georgia, where a satisfactory application of scientific principles and risk analysis at the national level in food safety has not yet been reached. Obtaining and researching relevant data on the existing situation in Georgia, as well as data regarding the experience and approaches of EU countries, will contribute to an accurate and comprehensive approach to food risk assessment and its future in Georgia.

65. From 1993 to 2015 : more than 20 years of a EU Reference Laboratory activity for networking key national reference laboratories for the food safety control in the European Union – Shaping the future of control and expertise of VMP residues in food

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ABSTRACT

An overview of the food security activities engaged by one of the early-designated European Union Reference Laboratories (EU-RL) for the control of residues of veterinary medicinal product (VMP) in food-producing animals will be presented. Will be particularly displayed the step-by-step building of the networking of the EU-MS NRLs from the early 15 Member State countries as of the 1994 EU state-of-play to the years 2000s completion with new NRLs from Eastern Europe acceding countries to the EU stepping up to 24, then 27 and now 28 Member States. Will also be explained the categories of activities implemented throughout the 20-years+ combined actions. Several food security expertise conducted by the network of EU-NRLs to help solving periods of food crisis will be exemplified. Especially that kind of food crisis triggered after evidence of abuse for EU-banned veterinary substances and discovery of their presence as residues in animal derived imported food commodities. Also will be tentatively pictured a shaping for the future of this networking expertise to help monitoring the security of food from animal origin throughout the EU as it could be offered after the opening to several innovative technologies.

Nutrition challenges ahead

66. Genetic and nutritional influence in the incidence and aggressiveness of prostate cancer in Spanish population

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ABSTRACT

Objectives: The main aim of this study is to prove the relation between the genes RNASEL, ELAC2 and MSR1 in the aggressiveness and progression of prostate cancer and how diet can interfere the aggressiveness and progression of this cancer. **Materials and Methods:** 322 men (mean ages 66-75 years) with PSA values above 4ng/ml that fulfil the requirements for a prostatic biopsy. We have samples from blood, fresh tissue and FFPE tissues from men with and without PCa. DNA and RNA will be extracted by organic procedures. Genotyping will be performed by TaqMann Technologies (Life Technologies) and a 10% of the samples will be confirmed by Sanger sequencing. Expression analysis will be made by Real-Time PCR using SYBR® Green (Life Technologies). The diet effect will be studied by an adhesion to Mediterranean diet questionnaire that will gather data of semiquantitative food frequency and each participant's food consumptions. The dietary frequency intake will be analyzed by the dietary software program Novartis-Dietsource v. 1.2 that will be used to convert foods into nutrients. **Results:** Preliminary results gather patients with more aggressive clinical parameters (TNM above 2 and Gleason score ≥ 7) in individuals with variants AA and CC in rs486907 and rs2127565 in respectively with an increased risk of PCa; as well as, less aggressiveness parameters in men with a rich diet of vegetables and fruits**. Conclusions:** Despite the information available, a common consensus on which nutrients may be beneficial and which could be harmful in PCa is lacking. Therefore more research is needed to elucidate the effects of consumption of particular nutrients and how this relates to PCa. Mainly diet and genes interaction we find to be very relevant in this cancer because of the high variability in incidence rates due to differences in race, geographic area or age as well as differences in environmental factors, such as place of work, nutrition and lifestyle.

67. Exposure assessment and risk characterization of 3-monochloropropane-1,2-diol (3-mcpd) from oils, fats and fried foods in Brazil

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ABSTRACT

Fatty acid esters of 3-monochloropropane-1,2-diol (3-MCPD esters) are processing contaminants that can occur in refined vegetable oils and fats at significantly high concentrations. Dietary exposure to 3-MCPD esters has been considered a priority food safety issue since free 3-MCPD can be potentially released through the action of gut lipases, representing a public health concern. This work describes a preliminary estimate of 3-MCPD intake from oils, fats and fried foods in Brazil as well as the evaluation of the potential risks to health. The daily intake was assessed using a deterministic approach by combining measured levels of 3-MCPD esters in selected food samples (oils, fats, fried snacks, instant noodles, French fries) with national food consumption data provided by a survey on the Analysis of the Individual Food Consumption in Brazil carried out from 2008 to 2009. The levels of 3-MCPD esters were determined in 180 samples by an indirect method based on acid transesterification and analysis by gas chromatography-mass spectrometry. For risk assessment purposes, the estimated intake was compared to the provisional maximum tolerable daily intake (PMTDI) of 2 µg/kg body weight (bw). Mean intakes of 0.06 and 0.15 µg/kg bw/day were estimated for the Brazilian population using mean and P95 levels of 3-MCPD esters, respectively. Oils and fats were the most important contributors, accounting for 46% of the total intake. Within subgroups, the highest exposure was observed for adolescents aged 14-18 years old (0.10 and 0.21 µg/kg bw/day for average and high consumers, respectively). Considering the total population as well as the investigated subgroups, the estimated intakes represented 2-5% and 5-10% of the PMTDI for average and high consumers, respectively, which suggests a low concern to human health.

68. SCD5-induced oleic acid reduces melanoma metastatization: importance of 'good' fatty acids

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ABSTRACT

Acronyms: FAs (Fatty Acids), SCD1/5 (Stearoyl-CoA desaturases 1/5), OA (Oleic Acid). **Objectives:** Cutaneous melanoma is the fastest increasing cancer worldwide. Highly proliferating neoplastic cells require a continuous supply of lipids with a correct molecular composition of fatty acids (FAs) in order to form new cellular membranes. SCD1 and SCD5 Stearoyl-CoA desaturases, two endoplasmic reticulum integral proteins, catalyze the introduction of the first double bond in cis-delta9 position of saturated palmitoyl-CoA and stearoyl-CoA to obtain monounsaturated palmitoleoyl-and oleoyl-CoA FAs. Here, we have analyzed the expression and function of SCD5 and its products in melanoma development and progression. **Materials and methods:** Expression studies (obtained by western blot, real time PCR and immunofluorescence) and in vitro biological assays were performed according to standard procedures. Quantification of FAs was evaluated by gas chromatography/mass spectrometry analysis. For in vivo studies, xenografted nude mice were utilized to assess SCD5 functional role on tumor growth and metastatic potential. **Results:** Our data showed the antimetastatic function of SCD5 and of its main byproduct Oleic Acid (OA). SCD5 and OA expressions were inversely related respect to melanoma progression. In addition SCD5 restored expression and/or OA supplementation in metastatic melanoma cell lines were able to block the secretion of pro-tumoral proteins leading to reduced tumorigenicity. Notably, in vivo models confirmed a significant SCD5-dependent impairment of metastatic dissemination. Finally, our data indicated a SCD5- or OA-dependent modification of intracellular pH (pHi<pHe) towards a more physiological condition that might balance the acidic extracellular tumor environment associated with less aggressive phenotypes. **Conclusions:** This study supports a protective role of SCD5 and, more important, of its enzymatic product Oleic Acid against melanoma metastatic spreading, a finding offering explanation for the beneficial Mediterranean diet.

69. Edible macroalgae: a source of mineral intake

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ABSTRACT

Global food needs are increasing significantly due to world's population growth. One of the greatest challenges to answer this concern is to explore the natural resources bearing in mind sustainable approaches, taking into account social, economical and environmental sustainability. Macroalgae are ecological and economically important resources of the marine biodiversity. They have a great potential in the food industry and their direct consumption in western diets has increased. They are known as excellent sources of minerals and bioactive compounds with recognized benefits for human health. Nowadays, aquaculture industry provides 95% of the consumed macroalgae. This production system allows an efficient control and selection of the best conditions to obtain outstanding quality products. This work aims to characterize the elemental fraction of 13 commercial edible macroalgae from different geographical origins (Galicia, Spain; Japan; and France). Selected samples included 5 brown species (Laminaria japonica; Undaria pinnatifida; Hijikia fusiforme, Himanthalia and Fucus vesiculosus), and 4 red species (Porphyra tenera; Eisenia arborea; Eisenia bicyclis; and Palmaria palmata), obtained in the market. The determination of the essential elements (Ca, Mg, P, Fe, I, Mn, Cu, Zn, and Se) was performed by ICP-MS and EAA. In general, samples presented high contents of Ca, Mg, P, Fe and Zn. Brown algae presented higher mineral contents of Ca, Mg, Fe, I and Se, when compared with the red ones, except in the case of P, which displayed superior amounts in the red species. Cu amounts were similar in both groups. The results show differences in mineral composition between brown and red macroalgae and within the same species from different geographical origins. These findings can be explained by environmental and seasonal changes, factors that directly influence the composition. Due to the high contents in minerals, macroalgae can contribute to attain mineral dietary recommendations.

70. A standardised evaluation of dietary monitoring tools for risk assessment in Europe

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ABSTRACT

Objectives: To compare results on dietary intakes obtained through six assessment tools based on the same batches of consumption days in a ring-trial using standardised methodology. **Materials and Methods:** A ring-trial protocol was applied in 24-hour dietary recall and food record methods used in Estonia, Italy, Latvia, Portugal, Spain and Sweden. Ten consumption days including 256 foods, beverages and dietary supplements (with several being reported in repetition) were constructed, standardised and translated to national languages, and were subsequently reported by 'subjects' trained to eliminate variability introduced by study participants. Statistical analysis involved the evaluation of the methods' repeatability and relative accuracy in capturing quantitative as well as qualitative information on dietary intake. **Results:** In most, but not all, cases tools adequately recorded items which were repeatedly reported identically and discrepancies revealed inconsistencies in the implementation of study protocols. The dietary assessment methods performed inefficiently in capturing qualitative characteristics of food items (e.g. fat or salt content; additives, cooking methods), indicating that improvements need to be made in the food descriptors used. The relative accuracy of results was practically independent of the number of integrated databases and the variety of available portion measurement aids. Results did not support the assumption that tools with multiple food quantification means may perform better in a dietary survey. Notwithstanding the possibility of unpredicted variance in the 'subjects' reports, several sources of uncertainty were identified, with the interviewer and food record keeper being primarily responsible for the inconsistencies observed. **Conclusions:** In a ring-trial mimicking laboratory conditions as closely as possible and involving six European dietary assessment methods, the relative accuracy in capturing qualitative characteristics of food items was low. Improvements in the integrated food descriptors and adequate training of the survey personnel could improve the quality of the collected data. The abstract describes methods and results of a project coordinated by the Hellenic Health Foundation and supported by EFSA (Service Contract: CT/EFSA/DCM/2012/02; financial support: 289.300€). The opinions expressed are those of the Contractor only and do not represent the Authority's official position.

71. A novel approach to improve the nutritional status of modern people

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ABSTRACT

Nowadays, the global food industry must implement new politics of action to meet the rising consumer demand for healthy foods and to provide solutions for sustainable agriculture and food technology. Progresses in food processing technologies could enhance global food safety and contribute to efficient global food distribution, while reducing food wastes and minimizing high rates of poor nutrition globally. Obesity, diabetes and cardiovascular

diseases are interrelated with poor nutrition being one of the main factors that link them all together. While one of the food security strategies is directed toward changing the consumer behavior in the durection of 'healthy' foods, it might be more efficient in the shortest term to solve this nutritional issue by enriching processed foods with dietary fiber obtained form agri-food wastes. The research is focused on the development of fruit fillings fortified with dietary fibers for future application in bakery as one of the possible ways to improve nutritional status of modern people. The objective of this research work was to fortify fruit fillings with a natural heat-stable blend of two dietary fibers – long-chain chicory inulin and pectin on the basis of experimental design technique. Fortification of the fruit fillings with inulin/pectin mixture was carried out based on the satisfaction of 30-50% of the adult daily requirement of dietary fibers with prebiotic properties from the consumption of 100 grams of product. The remained amount of inulin in fruit fillings for claiming their prebiotic properties was found by both calorimetric assay and chromatographic analyses for fructose determination conducted on SHIMADZU LC-20AD high-performance liquid chromatograph equipped with refractive index detector RID-10A, by using acetonitrile-water (80:20) as mobile phase. It was established that 80-90% of the initially added inulin remains in fruit filling compositions in form of FOS that can claim the prebiotic properties of the finished product. It was also confirmed by validation experiments that heat-stable, physicochemical and sensory parameters of fruit fillings with inulin-pectin blends were improved compared to control samples (fruit fillings without complex). Given the importance of global agri-food losses, obtaining functional ingredients from food wastes for fortification of food products could be a way of both reducing global food wastes and minimizing the cost of producing foods with health benefits.

72. 'Expose the gaps not the consumer': The need to implement all aspects of nutrition regulations

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ABSTRACT

This study explores consumer protection issues arising from two key gaps in nutrition regulation – nutrient profiles and maximum safe levels. Claims on foods are regulated in Europe1 but nutrient profiles provided for in legislation have not been set. Nutrient profiles would disqualify foods that should be limited in a healthy diet ('treat foods') from making claims. Addition of micronutrients to food supplements is regulated in Europe2 but maximum safe levels provided for in legislation have not been set. Maximum safe levels protect consumers from adverse effects of excessive consumption of micronutrients. Foods fitting the definition of a 'treat food' in Ireland3 were identified from composition data (n202)4. The micronutrient composition of these foods was compared with the value required to meet a 'source of' claim for that micronutrient (i.e. 15% labelled daily reference intake /100g). Three quarters of these 'treat foods' (n149) were found to be a 'source of' at least one micronutrient. Currently there are 164 authorised health claims that can be made on these foods (April 2015). For example, a potential legal claim for these foods is: 'sherbet sweets contain magnesium which contributes to the maintenance of normal teeth'. Food supplements notified as being on the Irish market between 2007 and 2014 (n9483) were assessed to identify vitamin D containing products (n1435). Daily amounts of vitamin D provided were examined according to year notified. Laboratory testing for a limited range of products (n51) facilitated comparison with labelled values. From 2007 to 2014 there was a significant increase in both the number of vitamin D supplements available and the daily amount of vitamin D provided. Some supplements exceeded the adult tolerable upper level for vitamin D (n11) and laboratory testing found that products providing higher amounts were less likely to be within acceptable tolerance levels for labelling. These regulatory gaps urgently need to be closed to ensure consumer protection. Authorising claims without nutrient profiles in place misleads the consumer about health benefits of foods. Safe maximum levels for micronutrients need to be established to protect consumers against excessive intakes. 1.Regulation 1924/2006/EC on nutrition and health claims 2.European Directive 2002/46/EC on food supplements 3.FSAI (2011). www.fsai.ie/recommendationsforhealthyeatingguidelinesinireland.html 4.FSA (2002) McCance and Widdowson 6th Edition (CoF IDS)

73. Jellyfish: old Eastern food becomes the Western novel food

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ABSTRACT

Background: Since the 1960s substantial increase of migration fluxes to Western Europe led to the consequent introduction of new ethnic foods, such as jellyfish from Southeast Asia. In addition, climatic and anthropogenic causes are shaping dramatic changes in marine biodiversity and food sources across world oceans, including the Mediterranean Sea, where every year an increasing number of jellyfish are recorded and washed up ashore. Among them, several species show biologic and nutritional features similar to the Asiatic edible species, can be equally cultured, and may be therefore regarded as good candidates as alternative food or feed sources, or for nutraceutical and bioactive compounds isolation and exploitation. **Objectives:** This work is addressing the potential use of Mediterranean jellyfish as natural biological resources of interest for several biotechnological and production sectors. **Materials and Methods:** We investigated three common species of the Mediterranean Sea: Aurelia sp., Cotylorhiza tuberculata and Rhizostoma pulmo, and their protein contents were qualitatively and quantitatively determined. Different jellyfish body parts were subjected to aqueous or hydroalcoholic extraction, followed by sequential protein enzymatic hydrolyses. The antioxidant capacity of proteinaceous and not proteinaceous extracts and hydrolyzed proteins was assessed by TEAC (Trolox Equivalent Antioxidant Capacity). assay. The potential of the gelatinous biomasses as fresh or 100 °C heated material was also evaluated. Finally, the antiproliferative activity of the hydroalcoholic extract of C. tuberculata was assayed in vitro on MCF-7 breast cancer cells. **Results:** More than 40% of the jellyfish dry biomass is composed by collagen and other pepsin-digestible proteins. Hydroalcoholic extracts and the protein-hydrolyzed fractions from all the three jellyfish revealed appreciable antioxidant activity. The hydroalcoholic extract of C. tuberculata exhibited significant anti-proliferative activity on MCF-7 cells. **Conclusions:** Mediterranean jellyfish biomasses represent a still unexplored source for human food and feeds; in addition, the abundance of collagen, peptides and other bioactive molecules may represent a valuable source of natural compounds of nutraceutical, cosmeceutical and pharmacological interest. Intervention is therefore needed to bridge the gaps related to food safety assessment, insufficient scientific knowledge, data or data access.

74. Risk assessment of certain cognitive functions reduction in preschool children in relation to the fatty acid composition of the diet

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ABSTRACT

Objectives: Analysis of fatty acid (FA) composition of diets of preschool children and risk assessment of low level consumption of ω-3 polyunsaturated fatty acid (PUFA) on cognitive function. **Materials and methods:** Daily diet, consumption levels of ω-6 and ω -3 FA, cognitive abilities of 186 children aged 5-6 years was studied. Study of energy and some nutrients level in the diets was performed using the analytical method (menu-layout), the 24-hour recall and laboratory tests. The cognitive abilities (memory, attention) were assessed with the figured tables, which consists of an alternating sequence of 7 figures (circle, square, flag, etc.). Children were asked to put certain signs in the 3 specified figures for 2 minutes. Estimation of the test was carried out by counting the number of scanned figures, mistakes, calculation of productivity index. **Results:** Analysis of the diet FA composition showed an excess of saturated FA (34.2 ± 0.53 g/day), or 15.2% of energy intake (%E) when these

FA should provide no more than 8% of the daily energy intake for this age group, according to FAO/WHO. However, the content of PUFAs in the diet was 5.3%E, which correspond to the recommended consumption level of PUFAs in Belarus (5-10%E), and slightly below the international recommendations (6-11%E). Total ω-3 PUFAs amount in the childrens' diet was 1.05 g/day or 0.49%E, and was below the FAO/WHO recommendations (0.5-2%E). The content of ω-6 PUFAs corresponded to the levels set out in FAO/WHO (2.5-9%E), and was 10.7 g/day or 4.9%E. Cognitive function study has shown that ω-3 PUFAs intake at the level 0.6-1.0%E leads to a significant increase in the number of scanned characters and growth productivity index compared to children whose diet was contained less than 0.5%E ω-3 PUFAs. The mistakes number in the first group were significantly less than in children with the ω-3 PUFAs consumption less than 0.5%E. The content of ω-3 PUFAs more than 1.0% E didn't lead to further increase the number of scanned characters **Conclusions:** Daily diet of preschool children contain an excess of saturated FA and is poor for ω-3 PUFAs. Consumption of ω-3 PUFAs at the level of 0.6%-1.0%E by preschool children can reduce the risk of cognitive decline.

75. The 'hit and miss' of voluntary folic acid food fortification in Ireland

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ABSTRACT

During the 1980s, voluntary folic acid food fortification was introduced in Ireland and as a result the rate of pregnancies affected by neural tube defects (NTDs) declined. However, in 2008, high blood folate levels found in children and older adults (non-target population groups) were related to the widespread voluntary folic acid food fortification in place(1). As a result the proposed introduction of mandatory fortification of foods with folic acid was postponed(2). In Ireland, recent research reports the rate of NTDs has increased since 2009(3). This study examines the changes in voluntary folic acid food fortification in 2007 compared with 2014, which may contribute to the increase in NTDs. In both 2007 and 2014, all foods voluntarily fortified with folic acid on the Irish market were identified in supermarket surveys. These surveys included the number of different types of folic acid fortified foods as well as the amounts of folic acid in different brands. Daily meal plans (n7) that provided 400µg folic acid using folic acid fortified foods were developed in 2007, and were re-examined in 2014 to estimate changes in the amount of folic acid provided. The numbers and types of folic acid fortified foods and the amounts of folic acid in different brands identified in the 2014 survey were lower compared with the 2007 survey. This was particularly significant for fat spreads, while levels in cereals remained comparable at the two time points. All seven daily meal plans developed in 2007 to provide 400µg folic acid provided lower amounts in 2014, with differences ranging from 6-365µg/day. This study shows that the numbers, types and brands of foods voluntarily fortified with folic acid (along with the level of folic acid provided) are continuously changing in Ireland, making it very difficult for women of childbearing age to meet their needs. More critically, some women in the target group are completely excluded from the benefits of voluntary folic acid fortification due to their food choice. Introduction of a mandatory folic acid fortification programme of a staple food consumed by most women (e.g. bread), would address this public health issue and be easier to monitor. This would require legislative controls on voluntary food fortification. References: 1. Flynn MAT et al. (2008) Proc Nutr Soc 67, 381–389 2. FSAI (2008) https://www.fsai.ie/uploadedfiles/folic_acid.pdf 3. McDonnell R et al. (2014) J Public Health 37, 57-63.

76. Are glycemic index and glycemic load associated with risk of cutaneous melanoma? A case-control study in an Italian population

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ABSTRACT

Objectives: Glycemic index (GI) and glycemic load (GL) measure different characteristics of carbohydrate intake. A recent systematic review and meta-analysis of observational studies showed that high GI and GL diets are associated with moderately increased risk of cancer at several sites, such as colon-rectum, breast, and prostate. Taking into account a recently reported association of melanoma risk with insulin resistance, a condition where diet, and specifically dietary carbohydrates, might play a role, we analyzed the possible association of GI and GL with melanoma risk in a population-based case-control study. **Materials and Methods:** All patients diagnosed with cutaneous melanoma in the years 2005-2006 in five provinces of the Emilia Romagna region (northern Italy) were recruited at the Dermatologic Units of these provinces. The cases were matched according to sex, age and province of residence with population controls drawn from the National Health Service Emilia-Romagna directory. Each study participant compiled a food frequencies questionnaire specifically developed within the European Prospective Investigation into Cancer and Nutrition (EPIC) study for the Northern Italy population. Average dietary GI and GL were then calculated for each subject, and GI of food items containing carbohydrates was obtained from the Italian Glycemic Index Table. **Results:** Cases (n=380) and controls (n=719) were divided into quintiles according to their GI and GL intake. A direct association between melanoma risk and GL emerged in unadjusted logistic regression analysis with odds ratio=1.5 (95% confidence interval 1.02-2.30) for the highest versus the lowest quintile of GL score (P trend 0.074). This effect also persisted in multivariate analysis adjusting for several potential confounders (phototype, sun skin reaction, sunburns history, saturated fats, vitamin C, vitamin D, fiber, total energy intake, body mass index and education). Sex-stratified analysis indicated that the GL effect was stronger in females (odds ratio=2.8, 95% confidence interval 1.22-6.26, P trend 0.047). There was no evidence of a relation between GI and melanoma risk. **Conclusions**: We observed in this population, and specifically in women, an association between GL and melanoma risk. These results need further investigation in order to better clarify the role of diet, and specifically of GL and GI, on melanoma risk.

77. Impact of a high-oleic sunflower oil bovine feeding regimen on the fatty acid profile of milk and dairy products

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ABSTRACT

The recommendation to reduce saturated fatty acid (SFA) intake to <10% of total energy is a key public health strategy aimed at reducing the burden of cardiovascular disease (CVD). Since this dietary target is still exceeded by the majority of the UK population, there is an urgent need to find novel approaches for reducing SFA intake. Natural alteration of the fatty acid (FA) profile of ruminant milk, through plant oil supplementation, provides a sustainable means of producing SFA-reduced dairy products. This approach also helps to prevent SFA from re-entering the food chain. The current research examined the impact of high-oleic sunflower oil (HOS: 83 g/100g monounsaturated FA (MUFA)) bovine feeding regimen on the FA composition of milk and dairy products. The habitual total mixed ration diet of multiparous Holstein-Friesan cows was supplemented with HOS (1kg/cow/d) for a 21-day period to produce fat-modified milk. Subsequently, the fat-modified milk was

used to produce cheddar cheese and butter. Fat-modified and commercially available (control) milk, cheese and butter samples were analysed in triplicate for FA composition by gas chromatography (GC-FID). When compared with the control products, fat-modified milk and dairy products had a lower SFA, higher cis-MUFA and higher trans-FA (TFA) concentrations. There were 16.4, 9.7 and 6.5g/100g total FA difference in SFA, cis-MUFA and TFA between the fat-modified and control milk. Most of the reduction in SFA was in myristic (14:0) and palmitic acids (16:0), with a concomitant increase in oleic acid (18:1c9). These data illustrate that HOS supplementation of the bovine diet is an effective strategy for enhancing the FA profile of dairy products. Although increases in ruminant produced TFA (predominantly trans-10-18:1 concentrations) were observed following supplementation, these isomers are distinctive from trans-9-18:1 that are rich industrially produced TFA. This research formed part of the RESET (REplacement of SaturatEd fat in dairy on Total cholesterol) project that will determine the effect of fat-modified dairy consumption on holistic markers of cardiometabolic health in free-living individuals with moderate CVD risk (NCT02089035). This work was funded by the Medical Research Council, Arla Foods and AAK UK.

78. In vitro antibacterial 'sugarbag' pot-honeys of Australian stingless bees

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ABSTRACT

Background: Medical research has recently refocussed on honeys to identify potential drug leads. Honeybees (*Apis mellifera*, Apinae) collect nectars of *Leptospermum* species (Mertaceae) to produce the medicinal honeys 'mañuka' in New Zealand and 'jellybush' in Australia, containing the antibacterial methylglyoxal (MGO) (Beitlich et al. 2014). Australian stingless bees *Tetragonula carbonaria* (Hymenoptera: Apidae: Meliponini) produce 'sugarbag-pot-honeys' that Australian Aboriginal people use as gastrointestinal cleansers. While their antioxidant and antimicrobial properties have been reported (Boorn et al. 2010), the bioactive factors remained unidentified. Whether Australian stingless bees honey could originate from *Leptospermum* sp. was unknown, thus we investigated the chemical and antibacterial properties of these honeys (Massaro et al. 2014). **Material and Methods:** Honeys were harvested from beehives in South East Australia sites where *Leptospermum* species were abundant or absent (control). Liquid–liquid extractions yielded the phenolic concentrates for the analyses by Liquid Chromatography (UPLC-UV-ESI-HR-MS) and Gas Chromatography Mass Spectrometry (GC-MS). UV absorbances of MGO and peroxide contents in honeys were quantitated against standard calibration curves by spectrophotometric techniques. MGO was derivatised with hydroxylamine under acidic conditions; hydrogen peroxide levels were determined using dianisidine in a horseradish peroxidase assay. In vitro antibacterial assays were conducted in triplicates against reference strains of *Staphylococcus aureus* by agar diffusion (Fig. 1) and broth dilution tests. Controls were ethanol (negative) and phenolic standard solutions (positive). **Results:** The phenolic extracts averaged to 5.87 mg/100 g within the raw honeys, and their main chemical constituents were 3-phenyllactic acid, lumichrome, diglycosylflavonoids and norisoprenoids. No MGO or other chemical markers of *Leptospermum* nectars/honeys were found. Hydrogen peroxide was 155.8 µM in honeys, therefore below the bactericidal concentration of 760 µM, while the phenolic extracts were bactericidal at 1.2–1.8 mg/mL. The total antibacterial activities of sugarbag-pot honeys were partially ascribed to contents of hydrogen peroxide as well as phytochemicals identified within the extracts. **Conclusions:** The reported chemical and antimicrobial properties of Australian bee honeys can foster to develop medicinal applications and functional foods.

79. Production and evaluation of complementary food based on maize (*Zea mays*) and bambara groundnut (*Vigna subterranea*)

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ABSTRACT

Background: The study was aimed at formulating a complementary foods based on maize and bambara groundnut with a view of reducing malnutrition in low income families. **Methods:** The blends 70% maize, 30% bambara groundnut were biochemically evaluated for proximate, minerals, amino acids profile, and antinutritional factors, using proprietary formula ('Nutrend') as standard. Antinutritional factors, amino acids, microbiological properties and sensory attributes were determined using standard methods. **Results:** For Protein, the results were 15.0% for roasted bambara groundnut maize germinated flour (RBMGF), 13.80% for cooked bambara groundnut maize germinated flour (CBMGF), 15.18% for soaked bambara groundnut maize germinated flour (SBMGF) ; values for maize flour and nutrend had 10.4% and 23.21% respectively. With respect to energy value, RBMGF, CBMGF, SBMGF, maize flour and nutrend had 494.9, 327.58, 356.49, 366.8 and 467.2kcal respectively. The percentages of total essential amino acids in the composition of the blends were 36.9%, 40.7% and 38.9% for CBMGF, SBMGF and RBMGF, respectively, non-essential amino acids contents were 63.1%, 59.3% and 61.1% for CBMGF, SBMGF and RBMGF respectively. The mineral content, that is, calcium, potassium, magnesium and sodium, of formulated samples were higher than those obtained for maize flour and Nutrend. The antinutrient composition of RBMGF and CBMGF were lower than of SBMGF. The rats fed with the control diet exhibited better growth performance such as feed intake (1527g) and body weight gain (93.8g). Moreover, the protein quality parameters such as total digestibility, biological value, net protein utilization were comparable within the same range as that of Nutrend. For the microbial status, microflora gradually changed from gram negative enteric bacteria, molds, lactic acid bacteria and yeast to be dominated by gram positive lactic acid bacteria (LAB) and yeasts. Yeasts and LAB growth counts in the complementary food varied between 4.44 and 7.36 log cfu/ml. LAB number increased from 5.40 to 7.36 log cfu/ml during fermentation. Yeasts increased from 4.44 to 5.60 log cfu/ml. Organoleptic evaluation revealed that the foods were well accepted. **Conclusion:** The application of bambara groundnut fortification to traditional foods can promote the nutritional quality of African maize - based traditional foods with acceptable rheological and cooking qualities.

80. Regulation of body energy homeostasis by selected retinoids

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ABSTRACT

General objective of this study was to investigate the effects of Vitamin A related compounds on the processes involved in white adipose tissue (WAT) biology and on the body energy homeostasis, in particular: 1) the effects of all-trans retinoic acid (ATRA) on skeletal muscle secretoma, 2) ATRA effects on mitochondriogenesis and the induction of WAT oxidative metabolism and thermogenesis in adult animals; 3) the effects of vitamin A and β-carotene (BC) supplementation during the suckling period, which can affect the susceptibility to chronic metabolic alterations later in life. In vivo experiment on rats and mice as well as different in vitro experiments were performed. Study of the effects of ATRA on the secretoma of skeletal muscle was addressed using ATRA-treated myotubes and ATRA-treated adult mice. The induction of mitochondria quantity was estimated by staining them and by measuring mitochondrial DNA. To study the effects of early vitamin A supplementation rat pups at weaning were given daily oral dose of vitamin A in the form of retinyl ester (RE) or BC. Upon sacrifice serum and tissues were collected and retinoid and carotenoid levels, gene expression and tissue morphology were measured. In adult mice, ATRA induced skeletal muscle expression and

secretion of myokines, which were proved to exert effects upon adipose tissue, including browning, and induce mitochondria biogenesis and oxidative phosphorylation/thermogenic capacities in mature white adipocytes. Early vitamin A supplementation (as RE) affects WAT development in young rats in a way that adipose cells retain increased proliferation potential, which correlates with a reduced expression of adipogenic markers. Although BC absorption by suckling rats was confirmed by significantly increased serum and liver levels (not detected in the iWAT), its supplementation in the same period did not affect adipose tissue development in young rats. Taken together, these results contribute to the understanding of the effects of vitamin A metabolites on different elements of energy expenditure control processes that contribute to the organism's energy homeostasis and could be used in the prevention and treatment of obesity and associated metabolic disorders.

81. Pandan leaves 'Vanilla of The East' as potential natural colorants and flavorants

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ABSTRACT

Objectives: Pandan leaves are a promising source of natural colorant, as well as a natural flavourants. In this work, a general screening of norisoprenoids and carotenoids as their precursors was performed. It is a fact that, several harmful synthetic colorants and synthetic ingredients are still illegally used in Indonesia. Rhodamin B was found in tomato ketchup products, where methanyl yellow were used in several traditional food in street vendor in Indonesia. So, it is an 'urgent call' to find a promising source of natural colorant that can act also as natural flavour like pandan leaves in Indonesia for replacing harmful synthetic ingredients. **Materials:** Fresh pandan leaves were collected in Indonesia kept in air-tight plastic bags were used for the investigation. Chemicals and solvents were analytical grade. **Methods:** Carotenoid were identified by RP-HPLC and free volatile analysis in pandan leaves by HS-SPME GC- MS **Results:** Several carotenoids as natural colorants in pandan leave extracts were identified and compared to the retention time and maximum absorbance. β-Carotene and lutein, major carotenoids in carotenoids extract from pandan leaves, were found in HPLC chromatogram . The major carotenoids content of pandan leaves consisted of 1.52 ± 0.56 mg/kg D.W of lutein and 1.58 ± 0.21 mg/kg D.W of β-carotene. Several minor carotenoids were identified including violaxanthin, neoxanthin, lutein, lutein epoxide, α-carotene and zeaxanthin in carotenoids extract from pandan leaves. The free volatiles profiles in pandan leaves were investigated by HS-SPME GC-MS. Several norisoprenoids as the target compounds were derived from carotenoids as precursors. Several norisoprenoids were identified in pandan leaves e.g. α-ionone, β-ionone, and β-cyclocitral based on database GC-MS System and Kovats Index. Although several carotenoids were identified in pandan leaves extract, only a few possess the structural requirements to produce several norisoprenoids observed in this study. Carotenoids with the correct structural features to act as norisoprenoids precursors include α-carotene and β-carotene. **Conclusion:** Based on our finding, several carotenoids and norisoprenoids were identified in pandan leaves. With the double function of pandan leaves as natural colorants and natural flavour in foods, it is likely that pandan leaves with their abundance and easy to cultivate can become a good source of natural ingredients in foods with the economic and healthy benefits.

82. The high content of macromolecular antioxidants in the most consumed fruit and vegetables in European countries reveals that the current nutritional approach to dietary antioxidants is partial

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ABSTRACT

Objectives: Strong cumulative evidence supports the role of dietary antioxidants in disease prevention and health promotion. However, studies on dietary antioxidants focus exclusively on soluble antioxidants (vitamins A, C and E, selenium, and certain carotenoids and phenolic compounds). Recent findings show that plant foods also contain macroantioxidants or nonextractable polyphenols, which consist of polymeric polyphenols as well as single polyphenols linked to polysaccharides or proteins (Saura-Calixto. J. Agric. Food Chem., 60, 2012, 1195-200). Macroantioxidants reach intact the colon producing absorbable metabolites and are associated with specific health-related properties (reviewed in Pérez-Jiménez et al. Nutr. Res. Rev., 26, 2013, 118-29). The aim of this study was to determine by the first time the macroantioxidant content and profile in fruit and vegetables (F&V), in order to estimate macroantioxidant intake from F&V. **Materials and Methods:** Data from national food consumption surveys were used to identify the most consumed F&V in Spain, Netherlands, France and Germany. Nine and fifteen vegetables were selected. Macroantioxidants and soluble antioxidants content were evaluated by different methodologies, including their profile by HPLC-MS analysis. These data were used to estimate the daily intake of macroantioxidants from F&V. **Results:** Mean soluble antioxidant content in the selected F&V was about 600 mg/100 g dw, while mean macroantioxidant content was about 800 mg/100 g dw, showing the major contribution of macroantioxidants to total antioxidant content. The HPLC-MS profile of macroantioxidants was evaluated by the first time in individual F&V, identifying several hydroxybenzoic acids, hydroxycinnamic acids, flavanones and flavonols. Macroantioxidant intake from F&V in the four selected countries was over 200 mg/p/day, an amount similar or higher than soluble antioxidants intake. Intakes in the different countries will be presented. **Conclusions:** This study shows that F&V commonly consumed in Europe contain major amounts of macroantioxidants, which significantly contribute to the daily intake of antioxidants. While up to the moment studies on dietary antioxidants have been partial, considering only soluble antioxidants, a whole approach, including the ignored fraction of macroantioxidants, is needed in order to have a better understanding of the role of antioxidants in nutrition and health.

83. Substantiating and regulating health claims in different jurisdictions: possible implications for public health and innovation in the food industry

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ABSTRACT

Objectives: Nutrition is recognised as an important factor influencing the growing incidence of non-communicable diseases. While we are experiencing a global rise in obesity, specific populations are still at risk of nutrient deficiencies. Functional foods could support these nutritional challenges, if appropriately formulated and available to those in need. To ensure non-misleading labelling, many countries regulate the use of health claims on such foods. Research in the REDICLAIM (REduction of DIsease risk CLAIMs on food and drinks) project compares how health claims are being substantiated and used in different jurisdictions, focusing on the advantages and disadvantages of different regulatory models. **Materials and Methods:** The jurisdictions were selected to have well-documented regulation and use of health claims and included the EU, USA, Canada and Australia/New Zealand. Desk research and key informant interviews were used to establish the processes used, including regulatory criteria for the substantiation and use of health claims. **Results:** In all of the selected countries health claims need to be substantiated with generally accepted scientific evidence. However, substantial differences exist in the use of health claims on foods. Notable differences relate to the regulation of nutrient function claims, the rating of the overall composition of final foods (nutrient profiling), and extent to which there is stimulation of R&D activities in the industry. In some jurisdictions, claims can be substantiated with the confidential use of proprietary data; in the EU there are cases where 5-year protection of such proprietary data was granted and well used by petitioners. **Conclusions:** New health claims can be a driving force for innovation, yet significant investments in R&D are needed. The health claims substantiation process should therefore be well defined and efficient to support innovation and global competitiveness in the food

industry. On the other hand, reasonable mechanisms should be established to ensure that functional foods are indeed supporting public health. The work was supported by the European Union's FP7 programme (FP7-603036) and by the Slovenian Research Agency (P3-0395). The funding are not liable for any use that may be made of the information contained.

84. Insect kibbles for eco-sustainability of pet food

Prola L, Pagani E, Schiavone A

ABSTRACT

Objectives: Source of protein included in feed was identified as the major contributor to land occupation, acidification, climate change, energy use, water dependence and competition with human food. Pet food production increases each year (globally +4% in the last year) with a global production of over than 17 mln of tons and, in Europe, it is a market of more than 130 mln EUR. From an environmental point of view, insects are sustainable and they show low scores on carbon footprint. In addition, insects are efficient food converters because they do not use energy to maintain a high body temperature. Due to the only recent interest in the use of insects as an alternative protein source, the nutritive properties are not well known. Previous studies on the nutritive composition of insects have focused on human nutrition, and most of those insects demonstrate a good composition as human food. Insects exhibit great potential for development as a standard ingredient in animal feeding. The objective of our research project was to evaluate the potential use of insect meals in dog feeding as first step to the industrialization of petfood production from Insect meals. Our data on palatability, digestibility and nutrititional adequacy shows the potential of these meals as source of protein for pet food industry. **Materials and Methods:** We evaluated chemical composition and toxicological aspects of meals of different insect species. For each meal we produced 4 different diets: Extruded diet with substitution of 100% and 50% of conventional protein with protein of insect meal; Extruded diet with substitution of 100% and 50% of conventional protein and fat with protein and fat from insect. Kibbles were administered to 10 adult dogs of the same breed. Health status of dogs were monitored by a Veterinarian each day. Feaces were collected and were analyzed for chemical analysis of nutrients. **Results:** Our data on palatability, digestibility and nutritional adequacy of insect meals in dog food shows the potential of these meals as a perfect source of protein for pet food industry used alone without other source of conventional proteins. **Conclusions:** This study reveals the importance of eco-sustainable sources of proteins in pets because they require a high percentage of protein in their diet. Currently, insects are being considered as a new protein source for pet food industry.

85. D-Fagomine reduces weight gain and high glycaemia while attenuating the gut microbiota imbalance induced by an energy-dense diet

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ABSTRACT

Objectives: WHO global estimates that more than 1.9 billion adults were overweight (of these over 600 million were obese) and over 42 million children under the age of 5 in 2013-2014(1). Obesity and its related parameters have been linked to an imbalance in the proportions of the major subgroups of distal gut microbiota(2). D-Fagomine is a natural iminosugar present in buckwheat with bioactivity over microbiota(3). The goals of this work are: - To establish if D-fagomine reduces weight gain, fasting glucose and insulin induced by diet at long-term in vivo - To explore if this D-fagomine effect is linked to the attenuation of the gut microbiota disequilibrium. **Materials and Methods:** Rats (n=29) were fed a high-fat high-sucrose diet (HFHS) supplemented (or not) with 0.096% w/w of D-fagomine or a standard diet for 6 months. Blood glucose concentrations were measured by the enzyme electrode method using a blood glucose meter. Plasma insulin was measured using a rat/mouse insulin ELISA kit. The levels of total bacteria, and Bacteroidales, Clostridiales and Lactobacilliales (three of the major gut microbiota subgroups) were determined in fecal DNA by quantitative real-time polymerase chain reaction (qRT-PCR). **Results:** Compared to the body weight of the standard group, rats fed HFHS supplemented with D-fagomine gained less weight (20% gain) than those fed HFHS (29% gain). Energy intake was independent of the D-fagomine supplementation throughout the intervention. D Fagomine prevented the increase in both fasting blood glucose ($P<0.05$ vs HFHS) and plasma insulin concentrations induced by the high-energy-dense diet. Already after 1 week on HFHS diet the relative abundance of excreted Bacteroidales significantly ($P<0.01$) decreased while no significant changes were detected in the other subgroups. The animals supplemented with D fagomine presented a proportion of Bacteroidales significantly higher than those in the HFHS group from week 3 to the end of the study. As a decrease in the relative population of Bacteroidetes has been related to the obesity1, D-fagomine may be reducing weight gain by keeping bacterial populations close to those corresponding to a lean phenotype. **Conclusions:** D-Fagomine counteracts the effect of an energy-dense diet on the population of intestinal Bacteroidales and this may be related to a reduction in weight gain. 1 http://www.who.int/mediacentre/factsheets/fs311/en/ 2 Turnbaugh PJ et al. Nature 2009 3 Ramos-Romero S et al. Obesity 2014.

86. Comparison of the environmental impact of omnivorous, ovo-lacto-vegetarian, and vegan diet

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ABSTRACT

Objectives: The foods we produce and consume may affect our health and well-being, but for sure have a great impact on the environment. However, there is a lack of information about the real impact of specific food choices on parameters associated to environmental impact. The aim of this study was to determine the environmental impact of omnivorous, vegetarian, and vegan diets in the real-life context of an Italian small cohort of volunteers. **Materials and methods:** In an Italian observational multicentre cohort study, 153 volunteers were enrolled (51 omnivorous, 51 ovo-lacto-vegetarians and 51 vegans, matched for gender, age, BMI and smoking habits). Food intake was monitored with a 7 days dietary record. The European Institute of Oncology database was used to calculate nutritional values. The Barilla Center for Food and Nutrition database was used to evaluate environmental impacts, taking into account three indexes: carbon footprint, water footprint, and ecological footprint. **Results:** The qualitative analysis of food patterns confirmed that each volunteer was on the diet effectively declared. Energy intakes were similar among the three diets: 2471 ± 366 kcal in omnivorous, 2393 ± 314 kcal in ovo-lacto-vegetarians, and 2326 ± 324 kcal in vegans. Considering the food categories, the intake of legumes, vegetables, fruit and dried fruit was significantly the highest for vegans and the lowest for omnivorous. Consequently, the omnivorous choice generated significantly worse carbon and ecological footprints ($p< 0.001$) when compared to other diets, whereas the water footprint was significantly lower for the ovo-lacto-vegetarian choice ($p< 0.001$) with respect to other diets. **Conclusions:** A plant-based diet, especially the ovo-lacto-vegetarian approach, represents a clear environmental advantage. To reach an environmentally sustainable scenario, animal-based foodstuffs should be partially replaced with legumes, cereals, fruits and vegetables, according to nutritional guidelines. To maximize these effects, seasonal and locally grown foods should be preferred. Educating people to make little changes in their dietary behaviours could be a key action towards the diseases prevention as well as the environment preservation.

87. HPLC and ITEX/GC-MS techniques in combination with chemometric methods for the discrimination of hop varieties

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ABSTRACT

Introduction: Given some malpractices of mixing/falsification of hops varieties with the purpose of obtaining higher value for bitter acids, and thus to obtain better value quotation during commercialisation, the identification of the hops variety is a stringent matter. Brewers need to keep standard quality of beer by using standard quality raw materials, and they expect the identification methods to be simple and well reproducible (Jelínek et al., 2010). In agro-food chemistry, the application of chemometric methods for the characterisation or classification of products according to origin, quality or variety is very atractive and has already been widely used (Gad et al., 2013). Objectives: In the present work we explore the use of HPLC and ITEX/GC-MS methods in combination with Principal Component Analysis (PCA) and Cluster Analysis (CA), for the possibility of the classification of hop samples according to the three different varieties, grown in Romania: Hüller Bitterer (HB), Aroma (AR) and Magnum (MG). **Materials and Methods:** The amounts of particular and total α and β-acids, ratios between total amounts of α- and β-acids, ratios of cohumulone and colupulone in alfa-and beta-acids and the Lead conductance value of hops determined according to the instructions in Methods 7.7 and 7.4 in Analytica EBC, were used as the input parameters to the chemometric methods. The chromatographic matrix (volatile profile) was subjected to cluster analysis (CA) with the Euclidean distances and principal component analysis (PCA) with cross-validation (full model size and centre data). **Results:** The cluster analysis affords a dendrogram which demonstrates differences between the hop cultivars based on bitter acids and volatile oils content. PCA and CA both gave comparable results for the separation of the hop samples according to the varieties. According to PCA, the first two principal components explained 98% (bitter acids) and 96% (volatile oils) of the variance of the data, showing a good discrimination between the samples and types (cones and pellets). **Conclusions:** The application of the combination of the analytical techniques with the chemometric approach can be very useful in the discrimination of the hop varieties based on their composition of bitter substances and volatile oils.

88. Comparative assessment of Real Time PCR approaches to detect pistachio allergen coding sequences

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ABSTRACT

Objectives: Real Time PCR allows the detection of DNA markers indicating the presence of food allergens. The goal of this work is to set up a reliable and suitable Real Time-based detection assay for pistachio allergen coding sequences. In addition, it is aimed to analyse how the detectability of pistachio DNA targets in complex food products is affected by high temperature and/or high-pressure treatments. **Materials and Methods:** Two different Real Time-PCR assays to detect pistachio, Sybr Green and DNA probe based detection have been tested. Primer pairs, designed from the information available in the NCBI, have been used to amplify and clone the allergen-coding sequences in different pistachio varieties. We have analysed the multiple alignment of the sequences obtained in the lab, and their homologous in related species (Genbank sequences), in order to design specific primers and probes for each allergen. The specificity of each method has been assessed by RT-qPCR with the DNA obtained from pistachio and other plants commonly used as food ingredients. The sensitivity to detect and quantify pistachio has been investigated on spiked wheat flour with defined pistachio contents (0.5 – 100,000 ppm). For evaluating the effect of thermal processing (with and without pressure) on DNA detection, raw and treated (boiled and autoclaved) pistachio samples have been tested by RTqPCR. Practical applicability of the qPCR method has been tested by the analysis of several commercial foodstuffs. **Results and Conclusions:** The method comparison allows us to set up the most solid and suitable Real Time-based test for the detection of pistachio allergen sequences. According to our results, DNA probe-based RT-qPCR appears to be the most sensible and specific method. The limit of detection of the DNA probed quantification has been determined to be of 1 ppm while for Sybr-Green assay was 10 ppm. Moreover, DNA probes permit a more reliable detection of pistachio DNA in complex food matrices than SybrGreen chemistry. With regard to processing effect on allergen detection by RT-PCR, the data show a reduced amplificability of pistachio DNA after thermal treatment (121°C and 138°C) under pressure (autoclave), nevertheless, this effect is not observed after thermal treatment (100°C) without pressure (boiling). The findings from this study could improve the knowledge on the allergenicity of tree nut proteins, which may be relevant for consumers, regulatory agencies, and food industry.

89. Relation between Methylenetetrahydrofolate reductase 677C>T polymorphism and obesity

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ABSTRACT

Background: The 677C>T polymorphism in gene encoding methylenetetrahydrofolate reductase is related to increased homocysteine (HCys) concentration in plasma, especially when levels of folate are low. Elevated level of HCys is established risk factor for cerebro- and cardiovascular diseases. Thus, individuals with polymorphic variants of *MTHFR* (CT and TT genotypes) could modify the genetic effect with adequate folate intake. Obesity is a multifactorial chronic disease in which a complex interactions between genetic and environmental factors play a crucial role. Previously, it was observed that low folate levels are associated with a high body mass index (BMI) and 677C>T polymorphism is related to obesity in British women. Epigenetic control of gene expression is a potential mechanism by which folate could influence obesity. We performed a systematic review and meta-analysis of available data addressing the associations between *MTHFR* polymorphism and obesity. **Methods:** We searched Pubmed using '*MTHFR* polymorphism', 'obesity' 'overweight', 'BMI', '677C>T polymorphism' as keywords. We included to a study a total number of 1059 individuals with obesity and 1370 healthy controls. Statistical analyses were performed using MedCalc software. Heterogeneity between the studies was evaluated using the Dersimonian and Laird's Q test. In case of significant heterogeneity observed between the studies, the pooled OR was estimated using a random effects model, otherwise, a fixed effects model was used. **Results:** Heterogeneity between the analysed studies observed in case of CT+TT vs CC analysis was not significant, therefore we used fixed effects model to analyse this combination. The frequencies of carrier-state of T allele in subjects with high BMI and those with normal weight were similar (50.9% vs 50.6%, respectively). The pooled analysis showed that carrier-state of T allele of *MTHFR* gene (CT+TT genotypes) is not associate with obesity (p=0.869, OR=0.986 95%CI 0.84-1.16). BMI significantly differentiated both groups (p<0.001). **Conclusions:** The results based on a sizeable groups of subjects with overweight/obesity and normal weight showed that the 677C>T polymorphism in *MTHFR* gene is not related to obesity.

90. Phytosterol oxidation products in enriched foods – the other side of the coin

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ABSTRACT

Hypercholesterolemia is an established risk factor for the development of cardiovascular diseases. A daily dietary intake of 2 g phytosterols/-stanols

results in a reduction of LDL- and total plasma cholesterol of approximately 10%. Therefore, phytosterols and their fatty acid esters were among the first functional ingredients used to enrich foods in order to obtain an additional beneficial health effect. However, owing to their structural resemblance, phytosterols are susceptible to oxidation reactions similar to those known for cholesterol. Comparable to cholesterol oxidation products, several in vitro studies showed cytotoxic and pro-inflammatory effects of phytosterol oxidation products (POPs). The number and the variety of foods enriched with phytosterols/-stanols and their esters are increasing. Yet, information on the contents of POPs is limited. Therefore, the aim of the present study was a systematic investigation of the occurrence and formation of POPs in these types of foods. For this purpose, novel analytical approaches, e.g. on-line LC-GC analysis, have been developed, allowing a sensitive, robust and fast determination of POPs. These techniques have subsequently been employed to generate data on the formation of POPs resulting from common household ways of use and preparation, e.g. storage and electric heating of enriched margarines. The types and the amounts of POPs were shown to be significantly impacted by the time/temperature conditions as well as by the type of enrichment (e.g. phytosteryl esters vs. phytostanyl esters). The obtained quantitative data allowed a substantiated estimation of intakes of POPs to be expected via the consumption of enriched foods. In conclusion, the established approaches can serve as a reliable platform to provide the occurrence data needed for a safety assessment of POPs in enriched foods. They may assist in evaluating potential adverse health effects of POPs in foods and ultimately enable a risk-benefit analysis of the cholesterol-lowering effects of enriched foods.

91. *[Poster withdrawn]*

92. Benefit and risk assessment for human health of increasing potassium by replacement of sodium chloride with potassium chloride in industrial food products

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ABSTRACT

The Norwegian Scientific Committee for Food Safety (VKM) has conducted a benefit and risk assessment of increasing potassium by replacement of sodium chloride with potassium chloride in industrial food products in Norway. The assessment includes intake calculations for three different scenarios; 30:70, 50:50 and 70:30. The calculations were based on the national food consumption surveys 'Norkost 3' (18-70 years), 'Småbarnskost' (2 years) and 'Ungkost' (4-, 9- and 13 years). An intake of at least 3.5 g/day of potassium in adults could lead to decreased risk of stroke (affecting 13 000 individuals/year), and a beneficial effect on blood pressure in individuals with hypertension. Women (59%) and men (30%) had a potassium intake below 3.5 g/day. In the 30:70, 50:50 and 70:30 scenarios, the percentage of women with intakes below 3.5 g/day decreased from 59% to 33, 21 and 13%, and in men, from 30% to 10, 6 and 4%, respectively. Additional intake of 3.0 g potassium/day to the mean food intake is anticipated to be safe for the healthy population. In the 30:70, 50:50 and 70:30 scenarios, women and men with mean intakes have no risk of adverse effects when compared with the anticipated safe level. However, based on the 95 percentile intake, adults will exceed this level in the 50:50 and 70:30 scenarios (women) and in all scenarios (men). Healthy children aged 2, 4, 9 or 13 years with a mean intake of potassium will have no risk of adverse effects in any scenario. However, based on the 95 percentile intake, children will exceed this level in the 50:50 and 70:30 scenarios (4- and 9-year-olds) or in all the scenarios (2- and 13-year-olds). In the 30:70, 50:50 and 70:30 scenarios, the percentage with potassium intakes above the anticipated safe level will increase from 1% to 3, 7 and 15% for women and from 2% to 9, 19 and 30% for men, respectively. Groups vulnerable to increased potassium intake are haemodialysis or kidney replacement patients, patients with moderate renal failure, coronary heart disease or diabetes, patients using drugs increasing hyperkalemia, and infants <1 year old, elderly persons and persons undergoing strenuous physical activity. A full benefit and risk assessment including an evaluation of severity of risks versus benefits was not conducted. However, VKM concludes that it is reasonable to anticipate that the percentage of persons likely to face an increased risk is far greater than the percentage of persons likely to benefit from this measure.

93. Risk management and communication in informal dairy sector in Côte d'Ivoire: Options for sustainable livelihoods

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ABSTRACT

Intervention in food and nutrition is the best investment for our collective future in term of managing co-morbidity in population. This investment should combine agricultural system with health and education. Fermented dairy products (FDP) play an important role for prolonged shelf life, microbial safety and nutrition. FDP was proved to be contaminated in Kenya, Somalia, Mali and Côte d'Ivoire by foodborne pathogens including Staphylococcus aureus and Escherichia coli. Recently, it has been showed that FDP is predominated by a novel Streptococcus infantarius subsp. infantarius (Sii) variant. Sii-produced bacteriocin and fermentation activity could contribute to the suppression of pathogens and possibly mitigate socioeconomic and health risks. However, Sii as member of Streptococcus bovis group is associated with human and animal infections. Therefore, a potential application of Sii as adapted African starter culture for enhanced food safety requires a thorough safety assessment. In order to improve hygiene and quality as well as to increase production for school canteens, urban consumption and sustainable livelihoods, a cross-sectional study was conducted in Korhogo (Côte d'Ivoire) from May to August 2014. The objective was to assess local technologies and the dairy value chain

in relation to Sii prevalence, followed by a participatory stakeholder workshop to validate findings and derive adapted interventions. The study showed that the dairy value chain contributed to livelihoods and household income. About 90% of milk produced (range: 12-44 liters/collector) were sold via collectors, generating daily 6-20 Euros that were shared between herder, collector and vendor. The remaining 10% were consumed within the household. However, dairy production was low and scattered due to informal practices resulting in poor quality of product. Basic hygiene such as cleaning, washing, disinfecting was lacking. Milk quality depreciated with the local practices, access to clean water and energy. Future interventions identified by stakeholders comprised (i) awareness on local dairy hygiene and nutritional value for the population especially school children, (ii) stakeholders organization around cooperative to develop sustainable dairy model (public dairy with private management); (iii) promote healthy milk products for school canteen programme in Korhogo through adapted local dairy technology.

94. Mixtures identification from the second French Total Diet Study

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ABSTRACT

Through their diet, humans are exposed to a wide range of substances which could have adverse effects on health. The total diet studies aim to assess the dietary exposure and the risk for a large number of substances. The risk assessment is limited to single substance or mixtures of congeners with assumed common mechanism, e.g. dioxins and PCBs. However, substances from different chemical families could also interact. That is why the identification of mixtures to which the population is really exposed through their actual food consumption is an essential first step in the risk assessment related to mixtures. A method based on Non-negative Matrix Factorization (NMF) to identify the major pesticide mixtures to which the French population is exposed and their associated diets was developed at ANSES. The present work aims to apply this method to the second French total diet study data on more than 400 substances and data from the French individual and food consumption survey including 2620 subjects. The first step consisted in selecting the substances with contamination values higher than the limit of detection. This selection reduced the number of substances to 153. Second, consumption systems composed of major consumed foods were identified and then combined with the concentration levels of the substances in order to form the main mixtures of substances. Third, a clustering of the individuals was performed to identify groups of individuals with similar consumption patterns and thus profiles of exposure. Six main consumption systems and their associated mixtures were thus obtained. For example, a mixture of 10 pesticides and 5 inorganic contaminants was identified. Exposure to this mixture is related to fruits and vegetables consumed by a subject group aged in mean around 50 years old and composed at 62% of women. Another group composed in majority of young men (31 years old) consumes junk food like sodas, pizzas, sandwiches, hamburgers and is exposed to a mixture of PAHs, acrylamide, inorganic contaminants, mycotoxins, pesticides, and brominated flame retardants. This work shows the diversity of the mixtures to which the population is exposed through its diet. It will help to prioritize the combinations of substances for which it would be crucial to investigate the possible combined effects, as toxicological information about mixtures is essential to carry out risk assessment.

95. Evolution in measuring food intake in Italy towards the harmonization within the EU-Menu program

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ABSTRACT

Objectives: Illustrating the evolution in measuring food intake techniques and the changes required to comply with the needs for harmonizing the methodology in the EU-Menu framework. **Materials and methods:** Desk analysis of survey methods characteristics of three nationwide dietary survey in Italy. **Results:** Representatives surveys have been carried out to estimate dietary pattern in Italy since the 80's at ten years interval times until 2005-06. Household was the sample unit, individual was the unit for analysis. From an indirect estimate (food disappearance data) to direct estimate (food record), from weighing by scales, to visual evaluation (photographic atlas). A number of 10,000 household in the 80's to 1,300 households in 2005-06, but individual food diaries have been administered. Great attention was paid to training of the interviewers in order to ensure an accurate food data collection. Socio-demographic and lifestyle questionnaires were also administered. Representativeness was achieved at level of main geographical areas. Technology played a crucial role in enhancing the real time check of the data, so reducing the time from the completion of the field-work and the publication of the results. The surveys allowed to capture important changes in dietary patterns related to modification in work time scheduling and an increasing offer of composite foods. The relatively low response rate and the trend to self-selection implicit in the substitution of non-respondents with randomly alternative units led to a sort of self-selection of more motivated people. This topic will be addressed in the next study trying to increase the response rate and decrease the self-selection in order to widen the representativeness. **Conclusion:** Currently, there is a need for updating the information for the Italian population then a new study is in preparation. Two waves will be undertaken one for children and one for adults. The studies will take part of the EU-Menu program and have received the support to harmonized the methodology according to the European Food Safety Authority (EFSA) guidance.

96. A diet naturally-rich in polyphenols improves risk factors for cardiovascular disease and diabetes: a controlled randomized trial

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ABSTRACT

Objectives: Dietary polyphenols (PP) are associated with a lower risk of cardiovascular disease (CVD) and diabetes. Abnormalities of glucose and lipid metabolism, and increased oxidation are well-known risk factors for these diseases that may be affected by diet. Therefore, we carried out a randomized controlled study (NCT 01154478) that evaluates the effects of a diet naturally rich in PP on postprandial lipid response, glucose metabolism, and markers of oxidation (isoprostanes) in people at high risk for type 2 diabetes and CVD. **Materials and Methods:** Forty individuals with high waist circumference and any other component of the metabolic syndrome were randomized to one of two isoenergetic diets differing only for PP content: (A) low-PP diet (Control, 365 mg/day) or (B) high-PP diet (PP-diet, 2903 mg/day). Before and after the 8-week intervention, lipid response to a test meal was evaluated. Moreover, insulin-sensitivity (OGIS) was derived from plasma glucose and insulin concentrations during 3-h oral glucose tolerance test. Oxidation was measured as urinary isoprostanes. **Results:** Dietary compliance was optimal in both groups. Polyphenols significantly reduced blood glucose (PP-diet: -16mg/dl vs Control: +30mg/dl), triglycerides in whole plasma (PP-diet: -156mg/dl vs Control: +167mg/dl) and in triglyceride-rich lipoproteins (PP-diet: -79mg/dl vs Control: +60mg/dl). Moreover, an increase of the early secretion of insulin (0-30 minutes; PP-diet: +1.86mg/dl vs Control: -2.13mg/dl) and an improvement in insulin-sensitivity (PP-diet: +32 vs Control: +2.90) were observed. Finally, markers of oxidation decreased (PP-diet: -264ng/dl vs Control: +176ng/dl) after the PP-rich diet ($p<0.05$ vs control for all outcomes). **Conclusions:** A diets naturally-rich in PP reduced blood lipid concentration and improved glucose tolerance, likely through an increase of early insulin secretion and insulin sensitivity. Therefore, a PP-rich diet may favorably influence the risk for diabetes and CVD.

97. Rapid detection of crab allergy using paddle-style dipstick method

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ABSTRACT

This research aimed to develop rapid detection of crab specific IgE antibodies using polystyrene paddle. Polystyrene was used Nunc Immuno Stick system and protein coated onto the surface of polystyrene was the extraction yield of crab meat obtained using phosphate-buffered saline solution. Characterization of crab protein extracts using SDS-PAGE showed that there were 17 protein bands with the molecular weights of 9.1 kDa-155.2 kDa. Optimization results showed that the concentration of crab protein extracts coated onto the surface of polystyrene stick was 0.1 ug/ul in the carbonate buffer, pH of 9.6, 0.5% BSA blocking agent, human sera dilution of 1:10, anti-IgE human antibody labeled with alkaline phosphatase enzyme 1:3000 and BCIP/NBT (5-bromo-4-chloro-3-indolyl phosphate/nitro blue tetrazolium) substrate. Positive results were marked with blue color formation within 20-30 minutes. The test results in sensitivity and specificity of Nunc Immuno Stick polystyrene to detect crab specific IgE antibodies were both 90%.

Open Risk Assessment: methods and expertise

98. Identifying key points for the safety assessment of biotechnological food produced with genetically modified microorganisms

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ABSTRACT

The classic biotechnological use of natural selected microorganisms and/or their derived products have been performed since centuries ago in a wide variety of ancestral cultures resulting in several types of foods that represent different environments and specific conditions of use in various civilizations (e.g. yogurt, kefir, koji, viili, kumis, beer, wine, bread). In this frame, the safety assessment in food derived from microorganisms face nowadays a more complex situation, due to globalization effect that has accelerated the export of food, easy moving people, exchange of traditions and with them the microorganisms linked to the specific food resources. Moreover, industrialization, modern food technologies and concretely microbial genetic modification tools have also accelerated the transformation of microorganisms for obtaining improved substances in shorter time terms that can affect their safety of use. Currently, the role of EFSA is to perform the risk assessment, taking into account all updated different factors that could affect the production of foods and food ingredients by fermentation using genetically modified microorganisms (GMMs) according to a specific regulatory framework and scientific guidance documents. Through an example of an opinion of a genetically modified food enzyme, the assessment and recommendations made by the Panel of experts will be further explained.

99. Risk based communication strategy on zoonoses transmitted by companion animals in Europe

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ABSTRACT

Introduction: CALLISTO is a EU 7FP project investigating zoonoses transmitted by companion animals to humans in Europe, whose final goal was to propose targeted actions to reduce the risk of zoonotic diseases and to promote risk awareness in healthy human-animal relationships. The complexity of the issue and the need to understand the problems posed by these zoonoses not only from a veterinary public health perspective, but also from a medical and socio-political point of view, required the implementation of a detailed communication strategy. **Objectives:** The strategy had to overcome the little co-ordination among pet owners, veterinarians, physicians, industry, associations, policy makers, scientists, the poor concerns of the public on companion animal zoonoses, and address to stakeholders policy and research recommendations arising from the assessment of the risk factors for the spread of 15 paradigmatic diseases, the knowledge and policy gaps, and the sociological aspects of human-animal relationships. **Materials and methods:** The communication strategy was based on the assumption that stakeholders have distinctive interests and languages. A variety of means - available on a web based platform - was used to produce outcomes applicable by stakeholders. The project visual identity was designed to establish an empathic contact with the audience. The communication style aimed at provoking an emotional marking, to create closeness and empathy with the end users. **Results:** Based on the principle that recommending implies dialogue and interaction with the others to produce change and share responsibilities, 29 recommendations were grouped in five main areas (demographics and tracing of companion animals, education and communication, surveillance and control, risk assessment, diagnosis, prevention and therapy) and prioritised using a 3-star ranking based on feasibility and impact on human/animal health. Target user groups were coded and recommendations described according to the same structure: priority, implementation, field, end users. **Conclusions:** The diversity of the communication means, the recommendation classification and prioritisation, and the use of targeted communication strategies facilitated the achievement of the end users. Further investigations could assess the impact of communication on the implementation of the recommended actions.

100. Quantitative data collection about physico-chemical characteristics of RTE cooked meat products for *L. monocytogenes* exposure assessment

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ABSTRACT

Reliability of a QMRA relies, among others, on the availability of quantitative data. Particularly, exposure assessment modules using predictive models about *L. monocytogenes* behavior in food should take into consideration the variability of the intrinsic factors with a significant impact on the growth kinetics. The objective of the present study was to perform a prospective research for a comprehensive characterization of the most relevant intrinsic factors in standard and nutritionally improved cooked RTE meat products, i.e., distribution of pH, aw, concentration of NaCl (%wp), nitrite, organic acids, fat and aqueous content as well as levels of competing microbiota. Data collection was performed through: (i) analyzing in the laboratory a number of representative products of the Spanish retail (n=48), (ii) searching data in international scientific literature (n=20), and (iii) collecting information from product labels (n=351). Analytical results showed mean values (and standard deviation, SD) for pH, aw and NaCl % wp of 6.09(0.24), 0.975(0.004) and 3.9(0.3), respectively. In most of the cases residual nitrite concentration was ≤5 mg/kg, the analytical detection limit. Mean of nitrate levels was 62.77 mg/kg. In products containing lactic acid/lactate, detected levels were between 0.56-20.7 mg/kg. The proportion of fat was highly variable and depended on the type of product ranging from 1% to 40%. LAB counts ranged from 0.5 to 8.2 log ufc/g. Bibliographic data collection from scientific publications usually provided limited information about one or more parameters in relation to physico-chemical characteristics and LAB levels for different types of products produced in different countries. Surveys dealing with physico-chemical characteristics of retail samples are scarce and data was mainly extracted from laboratory experiments with a limited number of products. The screening of the product labels of on sale products showed predominance, among cooked meat products, of pork and chicken deli meats with both standard and salt and/or fat reduced and enabled the estimation of the proportion of products

manufactured with *L. monocytogenes* growth inhibitors. The generated database will enable, through distribution fitting, the characterization of the variability of the intrinsic factors of cooked RTE meat products. This information will be useful to assess their impact on the consumer exposure to *L. monocytogenes* from contaminated products.

101. Stakeholder's views and priorities survey regarding factors limiting the Plant Protection Products (PPPs) Risk Evaluation

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ABSTRACT

Using a web-based tool, we elicit 136 selected stakeholders views and priorities regarding factors limiting the Plant Protection Products (PPPs) Risk Assessment (RA) phase linked on how the knowledge is produced and the way the data from RA are used in risk management and in risk communication. The criteria for expert selection were: different types of expertise, representativeness across different EU member states, types of institutional or group affiliation, and gender. The survey confirms that data on the variability of environmental (and human) exposure in space and time are missing, standard scenarios used for the assessment are not always relevant or are not context specific, statistically-based tools capable to quantitatively assess uncertainties are not available. Models and computational tools are not sufficient to represent the complexity of use and are not flexible to represents different scenarios. User knowledge on models and computational tools is limited; there is a lack of scientific basis or knowledge to express effects in terms that are of relevance for protection of human health (i.e. mortality, morbidity) or the environment (i.e. ecosystem services), lack of available data (epidemiological, nutritional, etc) to assess risk-benefit in the characterisation phase of RAStakeholders have indicated that the results of the RA extrapolated to the whole population may not be representative of societal needs. Attitude toward risk, certain vulnerable groups and immigrant workers and their difficulties to understand proper use are not properly considered. The survey helps us to better understand how the knowledge produced in the RA phase is interpreted and used for establish monitoring programs and mitigation measure and the factors limiting the effectiveness of RM as availability of context specific data, trust in pesticide use data methodology and reliability of data on use, consistency between mitigation measures and local conditions of use and that that risk management responsibility is not clearly distributed between authorities. The analysis confirm the strong influence of socio-psychological factors and the cultural ones on the overall quality and effectiveness risk evaluations and that to the risk assessor are attribute skills and responsibility that are not part of his cultural background process as well as that, due to the growing complexity and multiculturalism of the real world, a cross-cultural and interdisciplinary research is required.

102. The impact of a website for healthy diet promotion on public engagement

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ABSTRACT

Unhealthy diet and poor nutrition have become a scientific, medical, psychological, social and economic concern because of the negative impact on the health of population from low-income and even from high-income countries. In this context, the use of modern tools that promote a healthy diet is becoming more desirable as Internet use continues to increase among the population. However, little is known about the extent to which different types of content from a website on nutrition influence the level of engagement with online users. **Objectives:** To evaluate the level of public engagement with different types of content related to nutrition through a website. **Materials and methods:** The website, NOBEZITATE, was devellopped as a nutritional education tool to increase the level of knowledge on healthy eating habits in population from the Republic of Moldova. The website is linked to social networking platform – facebook and provides information about WHO recommendations; NCDs; the results of studies; videos and photos related to nutrition and healthy diet, etc. **Results:** The Website was apreciated by one thousand two hundred seventy nine people within facebook: 75% of fans are women and 24% of fans are men. There were posted 93 items related to nutrition and healthy diet for a period of 6 months (educational – 39; scientifical – 20; interactive that include videos, photos – 34). The engagement rate in reading the educational posts related to healthy diet is – 73,19 %; scientific results in the area of nutrition – 26,7 %; interactive posts related to nutrition (videos, photos, tests, etc.) – 47,9 %. **Conclusion:** The website, NOBEZITATE, completed a need for a fast, useful and effective nutritional education tool for the population from the Republic of Moldova. The website provided information about nutrition and healthy diet during 6 months. The results showed that the level of public engagement with educational posts is higher than the level of engagement with scientific posts. In this context, the results demonstrate that the type of content influences the level of public engagement.

103. Challenge of One Health and Global Health strategies in Africa: a North-South, South-South networking experience for risk analysis

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ABSTRACT

Introduction: The Network 'Nutrition & food safety and wholesomeness - Prevention, education and research network' (NOODLES, http://www.noodlesonlus.org) is registered as a no-profit organization since 2008. NOODLES is a network crosscutting public institutions, universities, NGOs and social, professional and scientific organizations, with the aim of implementing north-south and south-south strategy towards the translation of scientific knowledge into local proactive policy and movement. **Objectives:** One of NOODLES' major targets is the achievement of a safe and secure nutrition for women in childbearing age, under the Sustainable Food Safety and security umbrella; accordingly, the main objective of the Network is the mitigation of infant morbidity and mortality and the increase of life expectancy at birth. **Methods:** NOODLES protects and supports local food chains, food sovereignty and traditional healthy dietary habits in an increasingly interconnected, 'globalized', food production system. Through its trans-disciplinary and multisectoral approach, NOODLES intends to foster the One Health concept, where protection of environment, farm animals and human well-being are linked and mutually support each other. **Results:** The network allowed the understanding of food production and consumption scenarios in a data poor continent like Africa, from the e-waste piles in food producing lands to the nutritional value of staple foods. Powerful and popular social media showed potential significant role in achieving results in the assessment of risks and benefits, challenges and opportunities of staple food and diets. **Conclusion:** The implementation of the One Health approach in Africa is challenging and complex, but feasible. It will allow identifying more effective actions for Sustainable Food Safety. Acknowledgements: NOODLES thanks the contribution of local communities and the creative participation of researchers and volunteers in shaping information.

104. Bivalve shellfish consumption: engaging experts and stakeholders in risk assessment

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ABSTRACT

Objectives: Food-borne diseases are a serious health problem in many developed countries. Bivalve shellfish are one of the major vehicles since they expose consumers to chemical, microbiological and biotossicological

food risks. Many sanitary control activities of bivalve shellfish are performed in Italy, but information about risks associated with this food is often widespread among expert while consumers tend to be unaware of risks and of strategies for self-protection. The present study was designed to inform consumers on this issue. Two specific goals were set: to identify the main risks associated with bivalve shellfish consumption and to provide consumers with practical advice for a safe handling of this food in the home. **Materials and Methods:** Two participatory research methodologies were applied in order to select targeted scientific information from the exchange and interaction of different viewpoints. The Delphi method was used to identify and select the main risk factors from production to consumption of bivalve shellfish. The Nominal Group Technique was applied to evaluate and to rank the selected risks according to two criteria: the spread and the severity of the danger. Data were analyzed by appropriate measures of central tendency (mean and median) and variability (interquartile range). The level of statistical significance was set at 5% ($\alpha = 0.05$) **Results:** The Delphi method involved 14 Italian scientific experts (biologists, chemists, veterinarians, epidemiologists) and led to the identification of 32 risks. These risks were assessed through the Nominal Group Technique by 68 Italian stakeholders of the seafood chain (manufacturers, inspectors, retailers, restaurateurs/chefs). The risks which emerged as the most dangerous and most common mainly concern: improper harvest of bivalve shellfish (harvest in not permitted/not classified/ contaminated areas); inadequate transport methods both by producers and by consumers; improper handling practices in the home and in the peddling trucks; inadequate storage in peddling trucks, in restaurants and at home; consumption of raw bivalve shellfish in restaurants **Conclusions:** The methodologies applied and the overall data collected in the study led to the publication of guidelines containing practical advice for proper risk management in the home. These guidelines have been disseminated to Italian consumers and to institutions operating in the public health sector and in particular in the food safety area.

105. Risk-benefit assessment of food in Sweden – the development of a working procedure

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ABSTRACT

Objectives: The importance of considering both beneficial and adverse health effects associated with foods and their constituents is increasingly acknowledged. In Sweden, scientifically based risk-benefit assessment of foods is the responsibility of the Department of Risk and Benefit Assessment at the National Food Agency (NFA), which brings together the disciplines toxicology, nutrition and microbiology. A project was initiated with the overall aim to develop a procedure for risk-benefit assessment applicable for practical use at the NFA. Specific objectives were to summarize previous experiences of risk-benefit assessment procedures, to develop a working procedure and to test it in a case study. **Methods:** A literature review of approaches previously used in risk-benefit assessments of foods or well defined constituents of foods was performed. The guidance given by the EFSA Scientific Committee (EFSA Journal 2010; 8(7)) was identified as a suitable starting point for developing a working procedure. In the case study, we assessed potential health benefits obtained by reducing the nitrite levels by 50% and sodium content by 10% in processed meat in a scenario where maximum storage temperature was decreased from 8°C to 5°C. The impact of such changes on risk for microbiological adverse effects was assessed. **Results:** In the previous literature, tiered approaches to risk-benefit assessments have frequently been used. Our procedure for risk-benefit assessment contains: 1) Initial assessment of risks and benefits separately; 2) Enhanced assessment where different metrics for risks and benefits are weighted; 3) Same metric quantitative assessment expressing risks and benefits on the same scale (e.g. Disability Adjusted Life Years, DALYs). The two first steps of the procedure were successfully applied in our case study. A modest reduction in the sodium and nitrite levels of processed meat would only have marginal effects on public health, and would not increase growth of *Listeria monocytogenes* or *Clostridium botulinum* at storage temperature 5°C. **Conclusions:** The described working procedure is based on current understanding of how to perform risk-benefit assessments and is presently being implemented at NFA. Because of the complexity of performing risk-benefit assessments, we have identified a need for interaction between national authorities in EU member states. To initiate collaboration we will host a workshop with the aim to establish a Nordic network.

106. Survey on foodborne-related risk perception during pregnancy

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ABSTRACT

Objectives: Foodborne diseases represent a significant public health problem, particularly in pregnant women. Therefore, food safety education is fundamental to minimize risks. The objective of this study was to assess pregnancy women's awareness regarding food safety practices, foodborne pathogens and diseases, in order to improve preventive strategies. **Materials and methods:** During an overall period of four months (July 2011 and April-June 2012), all the pregnant women attending a medical examination at the obstetrical clinic of the San Paolo hospital (Milan, Italy) were invited to respond to an anonymous and voluntary questionnaire, which consisted of fourteen multiple-choice questions designed to assess their knowledge regarding foodborne diseases and the sources they used to gather information on food safety. The associations between categorical variables were assessed using the chi-square test. **Results:** A total of 218 women, aged 20-47 years, completed the questionnaire. Of the respondents, 100 (45.9%) were Italian and 118 (54.1%) were foreign-born: European (18.9%), African (16.5%), Asian (13.3%) and American (5.5%). Half women (44.9%) obtained a high school diploma. Results showed an incomplete knowledge about foodborne pathogens as well as high-risk foods. Most of the interviewed women (82.6%) considered *Toxoplasma gondii* dangerous, while only 6.9% indicated *Listeria monocytogenes* as harmful. Most of the questionnaire participants (85.3%) identified raw meat a food to be avoided in pregnancy. Cured meat and seafood were identified as high-risk foods by approximately half of the respondents (56.4% and 45.4%, respectively), while less than 2% considered milk consumption as a risk factor. Overall, 54.1% of the pregnant women demonstrated an insufficient level of knowledge, with the highest percentage (86.2%) among the Africans, those with the lower education level. **Conclusions:** This study revealed a clear lack of knowledge in pregnant women regarding food safety practices and foodborne diseases, especially among those with a low level of education. In particular, results pointed out a limited knowledge about listeriosis and its prevention, while toxoplasmosis-related knowledge was generally good. Therefore, information sources regarding prevention of foodborne diseases should be improved, ensuring easy access for women from all population groups, in order to prevent adverse pregnancy outcomes.

107. Nutritional risks and benefits of intense sweeteners: does science meet the expectations of consumers and risk managers?

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ABSTRACT

Objectives: The use and consumption of intense sweeteners (ISs) have risen over the last years, due to consumer concerns linked to overweight and obesity. Nutritional policies aim to reduce sugar intakes and the use of ISs as substitutes for sugar is often suggested in this context. This work aims to assess the overall nutritional risks and benefits of ISs in order to establish recommendations for consumers and risk managers. **Materials and methods:** A systematic analysis of the data currently available in the literature on the potential nutritional benefits and risks of IS consumption has been carried out. **Results:** The use of ISs instead of sugar engenders a short term reduction in caloric intake due to their low calorie content and the lack of

compensation. However, the available data cover insufficient time periods to guarantee that this effect is maintained in the long term. Studies of weight control in adults and children report conflicting associations, and even show a paradoxical positive association with weight gain, although the causality of this relationship has not been established. The consumption of ISs was not shown to have any beneficial effects on regulating blood glucose levels or on prevention of type 2 diabetes. The data available have not established any links between the consumption of ISs and the occurrence of cancer, type 2 diabetes or premature births. A few studies do however highlight the need to obtain further knowledge, especially in specific populations such as pregnant women and frequent IS users. **Conclusions:** This work highlights the lack of relevant data on the potential benefits of IS consumption in the context of their broad, long-standing use in nutrition. No beneficial effects have been shown that might provide grounds for recommending regular IS consumption in adults or children. The available data do not indicate any risks for occasional consumers. However, based on the epidemiological data, it is not possible to completely rule out certain risks in the event of regular, prolonged consumption. Therefore, this general assessment shows that no meaningful data are available that justify encouraging the substitution of sugars by ISs. A reduction in sugar intake should be achieved by limiting sweet-tasting foods in general at an early age. It is also important to avoid consuming artificially-sweetened or sugar-sweetened soft drinks as a replacement for water.

108. Piloting the consensus conference model on food safety issues: a dialog between experts and young consumers

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ABSTRACT

Objectives: Reducing food-borne disease is a public health objective of the European community. Promoting awareness and fostering correct risk-reducing behaviour become important objectives for all organizations dealing with the protection of citizens' health. The aim of this study was to pilot a participatory communication method designed to reduce microbiological risks associated with meat consumption among young consumers. The 'consensus conference' model was applied to food safety issues with these goals: to foster the sharing of knowledge regarding correct practices of meat handling at home between consumers and experts; to design risk communication contents (for the management of microbiological risk in the kitchen) to be disseminated to the target **Methods:** A structured questionnaire and a focus group allowed the identification of young consumers' habits and behaviors in the kitchen. These data was used to design the consensus conferences. The participatory methodology of the consensus conference was applied to create a dialog between consumers and experts (doctors, biologists, veterinarians, chefs and media experts) and to produce shared and well-informed opinions. Three consensus conferences were organized in Italian cities, involving 60 university students (19 and 21 years old) **Results:** The results highlight the importance of engaging young consumers and experts on the topic. The application of the consensus conference model proves to be effective and appreciated by the participants. The overall results were used to design a brief communication content based on the perceptions, behaviours and fact-finding needs of the target. The publication of these results in the form of guidelines promotes the dissemination of targeted communication on the microbiological risks associated to meat consumption and on strategy for risk reduction in daily food handling practices **Conclusions:** An effective risk communication should consider perceptions, behaviors and knowledge needs of the target audience next to the experts' scientific evaluation. Thanks to the consensus conference model the scientific information blend in with the opinion of the target involved, thus providing new knowledge, ideas and patterns of behavior. The dialog with experts proves to be an effective way of learning for young consumers. The active participation of the target group in the construction of the guidelines allows the definition of a communication suited to specific information needs.

109. The role of empowerment in online health communities in the doctor-patient relationship

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ABSTRACT

Background: The proliferation of social media applications such as online health communities (OHCs) has enabled social interaction between experts and public. OHCs present an important tool for information exchange. For this reason, they have become vital source of bridging capital for individuals who may find themselves in possible risk situations, such as being involved in food related incidents, possibly exposed to hazardous substances, or contemplating an illness etc. Certain projects can be identified at a national as well as a global level that are dedicated to measuring various dimensions of OHCs. The results of these projects indicate that a large number of online users are seeking emotional support and information related to health risk. Objectives: The present study investigates the potential of OHCs to empower patients and how this influences different empowerment outcomes (changes in the relationship with the experts, changes in communication with the experts, changes in health-related thinking and behaviour, etc.). **Material and Methods:** With a help of the administrators of the largest OHC in Slovenia, 'Med.Over.Net', we published the link to an online survey and invited approximately 7500 users to participate. 742 respondents completed the online questionnaire. We analysed the gathered data using statistical package SPSS Statistics 17.0 for social science research. Potential of the OHCs to influence empowerment outcomes (for those who choose to use them) was tested by means of multiple linear regression. **Results:** Literature review revealed following empowerment processes taking place in OHCs: receiving social support, helping others, finding a positive meaning, useful health information exchange, communication with experts. The results of our analysis showed that two of revealed processes (finding positive meaning in the OHC and communication with experts on the OHC) had a significant impact on doctor-patient relationship, positive attitude, sense of control and health-related behaviour (including dietary changes). **Conclusions:** With the ubiquitous nature of OHCs in today's society, health and food safety risk experts should consider taking advantage of these platforms to provide information and engage the public. The results of this research could be used to inspire a more web friendly mind-set amongst experts.

110. Assessing the added value of animal feeding trials with whole food/feed for GMO risk assessment

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ABSTRACT

The new Implementing Regulation (EU) No. 503/2013 asks for the mandatory performance of a 90-day rodent feeding study to determine the toxicological potential a certain genetically modified (GM) plant might pose to human or animal health. The design of animal feeding trials using whole food/feed as a test substance is largely based on test guidelines provided by the OECD to evaluate the toxicological potential of chemicals. The major difference between these two approaches relates to the maximal incorporation level of the tested entity, being restricted by ensuring for the balance of the respective diets. Thus, when using whole food/feed, the tested entity can only be administered in much lower doses when compared to single substances. Consequently questions are raised regarding the sensitivity. This concern is frequently brought up as a ready to use argument supporting the need for the performance of long term studies in order not to miss any adverse affect possibly caused by GM plant consumption. In a coordinated action, the EU-funded projects GRACE (GMO Risk Assessment and Communication of Evidence) and G-TwYST (GMP-Two Year Safety Testing) and the French project GMO90+ aim to assess the added value of subchronic and chronic toxicology and carcinogenicity studies, as well as of alternative in vitro and in silico approaches for GM plant risk assessment by using MON810 and NK603 maize varieties as test material. Throughout the whole planning and interpretation stages of the three projects, special emphasize is placed on active stakeholder

involvement to increase transparency and to strengthen the relevance of the gathered results from a broader societal perspective. Following the projects` transparency obligation, all data will be made publically available via the open access database CADIMA (Central Access Database for the Impact Assessment of Crop Genetic Improvement Technologies). The regulatory implication of the gathered results is underlined by a review clause implemented in the abovementioned Regulation, stating that 'the Commission shall monitor the application of this Regulation, the developments in scientific knowledge on replacement, reduction and refinement of animal use in scientific procedures and the publication of new guidance from EFSA. The Commission shall in particular monitor the outcome of the research project called GRACE'.

111. Holistic risk assessment: Enabling food policy

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ABSTRACT

Food contamination can be intentional or unintentional and may cause food safety issues depending on the nature of the contamination or simply cause a food product not to meet specification. Intentional food crime includes wider food fraud, economically motivated adulteration (EMA), and ideological food contamination. Various types of food frauds generate different levels of monetary gain and successes are dependent on how well the 'fraud' has been carried out and if detection actually occurs. Traditional risk assessment is based on known criteria: the hazard (agent that can cause harm or substitution); the severity (degree of harm including financial or economic penalty) of the hazard if it occurs; and the likelihood (frequency) of the hazard to occur. Traditionally food contaminants (biological, chemical and physical agents that can cause harm), food allergens and food quality and food issues are assessed independently but increasingly there is a requirement to consider a more holistic approach. This poster aims to introduce a Food Crime Assessment Model (FCA) into methods of traditional risk assessment. As part of a holistic approach to risk assessment, understanding of behavioural science, fraud forensic accounting, as well as the likelihood for profit, detection levels, and the potential for deliberate contamination to cause harm are interlinked to create the FCA. Therefore traditional risk assessment methodology that informs food crime policy must follow this holistic approach that integrates social science considerations with an understanding of biological and physical science.

112. Strengthening risk analysis process within the National Food Safety and Quality System in Chile

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ABSTRACT

Acronyms: ACHIPIA Chilean Food Quality and Safety Agency NFSQS National Food Safety and Quality System RAP Risk Analysis Process RA Risk Assessment RM Risk Management. Government program has defined the strengthening of food safety institutionality as a priority. For this purpose, a draft law that creates the NFSQS has been elaborated, and ACHIPIA would be in charge of conducting it. The NFSQS comprises the set of policies, programmes, and standards executed by competent authorities. One of the aims of ACHIPIA is coordinating and supporting RAP. **Objectives:** The objective is to propose a general model of risk analysis to be implemented in the National Food Safety and Quality System of Chile. **Methods:** A proposal of a general model of risk analysis has been designed considering international recommendations. The proposal has two phases: designing phase, which has been divided in three stages (proposal, plan implementation, and procedures design), and implementation phase, which involves three stages, validation, start-up, and evaluation and adjustment. **Results:** Regarding designing phase, the RAP will be described in terms of its procedures, ACHIPIA´s role, other coordination and support organisms, such as Board of Directors, Interinstitutional Body, Scientific Committee, and participation channels for key stakeholders. ACHIPIA´s role in the RAP will be focused on risk identification, prioritization, assessment and communication, for which purpose it has developed instruments to support this process, such as the Food Safety Observatory, Food Safety Alert Network, Integrated Food Laboratories System, and Expert Panels. Hazard identification will be supported by the Food Safety Observatory and Expert Panels. Risk and hazard prioritization will be based on methodologies previously agreed with competent authorities. RA will have three complementary instruments: expert panel opinions, scientific studies, and RA. Risk communication will occur through the whole RAP. Inspection authorities are the risk managers, and ACHIPIA will coordinate them through the establishment of interinstitutional working groups named National Integrated Programmes, which will act as a bridge between RA and RM. **Conclusions:** The proposed general model of risk analysis comes under a process of institutional modernization, which is intended to provide a systemic food chain approach and an institutional integrated strategy for addressing hazards through an efficient RM for Chile.

113. Consumers perception of food safety: results of web-based questionnaires

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ABSTRACT

Objectives: The aim of this work was to evaluate the knowledge and attitudes related to food safety of consumers using questionnaires section of IZSalimenTO website. **Materials and Methods:** Data collected by 4 questionnaires related to botulism, food safety during pregnancy, hepatitis A virus, and food borne diseases were analyzed. Questionnaires were available on IZSalimenTO website (www.izsalimento.izsto.it/palimenti/) for a month, were anonymous, and proposed 6 multiple-choice questions. For each questionnaire the number of correct answers were calculated and used to classify the respondents into three classes: good (≥83% right answer), satisfactory (34-82%) and poor knowledge (≤33%). **Results:** Among 118 respondents, 77% were female, 80% aged between 21-45 years, and 61% graduated. Botulism questionnaire recorded 48 respondents, 62% with satisfactory knowledge. 94% knew that *C. botulinum* can produce toxins in low acid home-canned food, 75% was aware that it is not inactivated by freezing and 54% knew that honey is at high risk for infant under 1 year of age. Food safety during pregnancy recorded 27 respondents, 81% with poor knowledge. As for food at high risk for *Toxoplasma*, 82% indicated raw meat but only 15% indicated raw vegetables; 74% believed that it can be inactivated by washing vegetables with sodium bicarbonate or chlorine based solutions. Only 29% knew that is advisable to avoid blue cheese, besides raw milk cheese (70%). Hepatitis A virus questionnaire recorded 26 respondents, 54% with satisfactory knowledge. 46% identified correctly high risk foods; 68% was aware of hepatitis A vaccination. Food borne diseases questionnaire recorded 26 respondents, 53% with good knowledge. 94% of respondents identified homemade tiramisu and mayonnaise at risk of *Salmonella*. 76% identified raw milk cheese, blue cheese and smoked fish at risk for *L. monocytogenes*. 76% was aware that raw fish preparations are safe after freezing fish at – 20°C for 24 hours. **Conclusions:** Although website users represent a part of consumers population, the website may be considered a useful tool to assess consumers' knowledge and to plan proper educational programs. The information collected show a lack of knowledge about important food safety behaviour, despite the high educational level of the respondents. On-line available educational interactive tools on food safety can play an important role in spreading correct behaviours among consumers.

114. Selecting the most appropriate risk ranking methods

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ABSTRACT

Introduction: Governmental organisations, food safety authorities and food industry would like to prioritize their inspection of feed and food safety hazards using risk-based approaches. Various methods are available for risk ranking of hazards ranging from full risk assessment to the application of expert judgment. **Objectives:** The aim of the current study was to systematically review risk ranking methodologies for prioritisation of food and feed safety hazards based on their anticipated health impact. **Material and methods:** An extensive literature review was performed to identify and characterize available methods for risk ranking. A systematic approach was followed, using a predefined protocol with keywords and selection criteria. In addition to applications within the food safety area, methods from the socio-economics and environmental fields were also considered 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. In total 10 different groups of methods were published: (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. Then, 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 at hand. The selection is based on both the risk manager's prerequisites as well as data availability. The framework has been applied to a number of case studies to determine the appropriateness of the method. **Conclusions:** 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.
Acknowledgements: This project has been financed by the European Food Safety Authority (EFSA) through a public procurement procedure OC/EFSA/SCOM/2013/01. The opinions expressed are those of the contractor only and do not represent the EFSA's official position.

115. The human factor in risk assessment – The island of Pantelleria as a study model to design preventive educational intervention for food safety

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ABSTRACT

Objectives: Educational campaigns for food safety might have low efficacy on a long run. The island of Pantelleria in Italy was used as a study model to design preventive campaigns for food safety with the active involvement of the 'target population'. **Methods:** First stage: questionnaires were administrated to assess basic knowledge on food safety. Second stage: interactive meetings with students and adult population. Third stage: students were actively involved through a drawing competitions on topics regarding typical foods. **Results:** The results of the questionnaire showed that a good balanced diet is quite diffused. Children at the elementary schools were highly involved during all stages in contrast to teenagers students. An active and interested participation was also present in the students' parent, teachers and local authorities. Some basic concept of food transmitted bacteria such as *Salmonella* or parasites like *Toxoplasma* were diffused in almost 40% of participants. *T. gondii* was particularly well known among females because of the risk related to vertical transmission during pregnancy. **Conclusions:** The model represented by a small Mediterranean island is good to study some general aspects of human behaviour for both risk assessment and the design of effective preventive campaigns for food safety. During all stages, primary school children were more involved than high school students, which suggests that effective educational interventions can be obtained if intended for younger population, particularly children that automatically involve their parents or other close relatives. In small communities, basic knowledge of good nutrition and food security issues is easily disseminated. The organization of a drawings exposition on typical food for elementary schools was a further occasion for interactive educational aspects on food related topics. It is possible to considerer specific target populations as small island inhabitants? More effective educational campaigns when environmental, social and cultural aspects of the target groups are known. The educational intervention for food safety can be tailored on the specific target groups that could also be involved in the campaign organization. **Acknowledgements:** The authors are very grateful for the active collaboration of the community of Pantelleria island. The work has been supported by the RCIZSSi 15/11 project to M.V. from the Italian Ministry of Health.

116. CADIMA: A dissemination portal facilitating synergistic effects between research projects on impact assessment of genetically modified plants

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ABSTRACT

The open access database CADIMA (Central Access Database for Impact Assessment of Crop Genetic Improvement Technologies) is a web portal for the dissemination of research data and more general information associated with the impact assessment of genetically modified (GM) plants. The portal was launched as an activity of the EU-funded project GRACE (GMO risk assessment and communication of evidence) and is currently supported by further three projects. So far, a commitment was made by the EU-funded projects GRACE, PreSto GMO ERA-NET, G-TwYST (GMP-Two Year Safety Testing) and the French project GMO90+ (Genetically modified organism, 90-day to 180-day testing) to use CADIMA when making results publically available. CADIMA will be permanently operated by the Julius Kühn-Institut and financed by public money only. CADIMA offers three major services. (1) It provides a central access point for key information sources supporting the risk assessment of GM crops and comprises a searchable list of the respective documents and webpages. (2) The evidence synthesis service provides tools for the performance and documentation of impartial evidence summaries of GMO impact data (including both risks and benefits) on human/animal health, socio economy and the environment by mirroring the key concepts of two evidence synthesis approaches, namely Systematic Reviews and Systematic Maps. Systematic Reviews are well established tools to efficiently inform evidence-based decision-making processes in medical sciences and their application in the area of food/feed safety assessment was recently promoted by the European Food Safety Authority (EFSA). (3) The open access sharing of (raw) data generated by animal feeding trials with GM plants and alternative in vitro approaches shall facilitate a transparent and detailed documentation of results and support further analyses. CADIMA continuously seeks input from different research projects providing relevant information. Depending on their specific needs, further core areas can be implemented and thus CADIMA represents an ideal tool to support synergistic effects between related projects.

Novel chemical hazard characterisation approaches

117. Neuropeptide Y alterations induced by Okadaic acid could be related to their diarrheic effect

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ABSTRACT

Okadaic acid (OA) and dinophysistoxins (DTXs) are a group of marine toxins that cause diarrheic shellfish poisoning (DSP) in humans and animals. These compounds are produced by dinoflagellates of the Prorocentrum and Dinophysisgenera and can accumulate in filter-feeding bivalves, posing a serious health risk for shellfish consumers. The enteric nervous system (ENS) plays a crucial role in the regulation of the gastrointestinal tract. In addition neuropeptides produced by ENS affects the epithelial barrier functions. To better understand the toxicological effect of DSP toxins, in the present work we have determined OA cytotoxicity in the human intestinal cell line, Caco-2. We also performed neurotoxicological studies with OA by using the human SH-SY5Y neuroblastoma cell line. OA reduces the viability of SH-SY5Y in a dose-dependent way, even though DTX1 is 4 to 5 times more potent than OA. Besides OA modifies the neuropeptide Y (NPY) levels of neuroblastoma cells without affecting the intestinal cell line. This offers a novel approach to establish the OA neuronal action as one of the triggers of their diarrheic effects. Therefore, the OA mechanisms of toxicity that were long attributed only to the inhibition of protein phosphatases, would require a reevaluation.

118. Subchronic Toxicity Study with a GM Maize MON810 Monsanto

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ABSTRACT

Objectives: In a coordinated action, the EU-funded projects GRACE (GMO Risk Assessment and Communication of Evidence) and G-TwYST (GMP-Two Year Safety Testing) and the French project GMO90+ aim to assess the added value of subchronic and chronic toxicology and carcinogenicity studies, as well as of alternative in vitro and in silico approaches for GM plant risk assessment by using MON810 and NK603 maize varieties as test material. Here we describe a 90-day feeding trial with a GM maize MON810 Monsanto variety. **Methods:** The study was conducted in agreement with the OECD method No. 408 and in accordance with GLP (good laboratory practice). Wistar Rcc Han rats were used in Specific Pathogen Free conditions. Dietary treatments represented following groups: 11% GMO, 33% GMO, near-isogenic control, conventional 1 and conventional 2. **Results:** The body weight of the male rats in all five groups increased with time and reached a plateau (i.e. about 410-430 g/rat) at week 11, whereby no statistically significant differences in body weight were observed between the groups during the whole 90 days. The body weight of the female rats also increased time-dependently and reached a maximum (about 250 g/rat) at week 11, while no statistically significant differences in body weight were observed. The haematology parameters, all clinical biochemistry parameters and the differential leukocyte count were similar in male and female rats fed the control, conventional 1 and conventional 2 diets without statistical significant differences. Histological changes were sporadically observed in the control and 33% GMO group. Since no treatment-related changes were observed between the two groups, no further tissue analyses were carried out on other groups. **Conclusion:** The compositional analysis of diets showed that the results differences among diets containing near-isogenic non-GM maize, MON810 Monsanto maize and conventional maize varieties were not statistically significant and can not be considered to harm the health of experimental animals in this study. The research study was supported by the European Commission: 7th Framework Programme for the project GRACE 311957.

119. Characterization of nanomaterials and their analytical determination in food and biological specimens: a challenge and a need for risk assessment of nanotechnology applications

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ABSTRACT

Objectives: Engineered nanomaterials (ENM) exhibit new properties compared to coarser materials with the same composition and in recent years the food industry has been exploring the potential of nanotechnology for novel applications. They comprise food additives, flavourings, food contact materials, novel foods, feed additives and pesticides. Critical issues for risk assessment of trace elements-containing nanoparticles (NPs) in foodstuffs include adequate physicochemical characterisation in food products and under toxicity testing conditions, the assessment of any degradation/solubilisation in the gastrointestinal tract, the understanding of their interactions with tissues, and potential exposure levels. The development of methods for the analytical determination of NPs in food and biological specimens is key for all these aims. **Materials and methods:** Imaging techniques (e.g. TEM, SEM) are essential for the characterization of ENM but can not provide quantitative data for critical parameters (chemical identity, size, size distribution, number and mass concentration of particles) in complex samples, whereas ICP-MS-based approaches can. We used ICP-MS (/MS), single particle-ICP-MS and AF4-UV-MALS-ICP-MS for the characterization of ENM and their analytical determination in food and biological specimens. **Results:** Using innovative approaches, the analytical challenges associated to the determination of SiO2 and TiO2 in biological samples were overcome. For SiO2 (E551) we provided novel methodologies for the quantitative characterization of NPs (1-2), for detecting Si deposition in studies on the mode of action of SiO2 (3) and to assess biodistribution and toxicokinetics in rats, which enabled to carry out a preliminary yet comprehensive risk assessment of E551 use in food (4). For TiO2 (E171), we proved deposition in spleen (a target organ for bioaccumulation) after oral, short-term exposure to low doses of NPs, eliciting endocrine-related effects in the absence of general toxicity (5). **Conclusions:** Detection of unlabelled oxide ENM in food and biological samples is problematic and complicates their safety assessment, despite their importance related to the widespread use. In order to generate new data to support risk assessment of ENM in food we successfully applied novel approaches using state-of-the-art analytical platforms. (1) J Anal Atom Spectr 27:1540 (2)10.1039/C4JA00478G Nanotoxicology (3)10.3109/17435390.2014.969791 (4)10.3109/17435390.2014.940408 (5) 8:654

120. Occurrence of tropane alkaloids in cereal based food products

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ABSTRACT

Objectives: Human risk linked to the assumption of Tropane alkaloids (TAs), secondary metabolites naturally produced by different plant species as Brassicaceae and Solanaceae (i.e. *Datura stramonium* L.), is associated with several diseases. In fact, TAs can influence the human central nervous system and bring to alteration of movement and of heart rate, secretion of salivary glands, dilatation of pupils and gastro-intestinal problems. These compounds, among them the most representative and studied are Atropine

and Scopolamine, could naturally occur in wheat, maize, soybean, millet, linseed, sunflower and buckwheat, as in the derived products. The presence of TAs in cereals is due to a sort of indirect contamination, in fact TAs plant producer (*Datura stramonium L.*) could infest crops and be harvested together with the other plants, so some seeds containing alkaloids could occur among the other seeds and go undetected through the processing of a food. Since data about the occurrence of these particular compounds are still limited, EFSA (European Food Safety Authority) presented a scientific opinion about human exposure linked to the consumption of food containing TAs. Thus, more data concerning the occurrence of TAs, in particular of Atropine and Scopolamine, in cereal as in cereal based food are needed in order to better evaluate the actual human exposure. At this aim an UHPLC-MS/MS method was developed and applied to several cereal based food products collected from different markets. **Materials and Methods:** All the collected samples (cereal based products, some of which containing buckwheat) were extracted with aqueous methanol and then analysed on a UHPLC-ESI-MS/MS apparatus. **Results and Conclusions:** Obtained results allowed us to have a more precise idea of the actual occurrence of Tropane alkaloids in cereal based food, with a particular focus on those products containing buckwheat. Furthermore, different samples of *Datura stramonium L.* seeds were analysed in order to identify unknown Tropane alkaloids and then to check their presence in food.

121. A 'wet & dry' joint effort to investigate the xenoestrogenic activity of zearalenone

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ABSTRACT

Objectives: Zearalenone (ZEN) is a secondary metabolite produced by fungi and molds belonging to *Fusarium* spp., which is found as a contaminant in various grains – especially maize – and grain-based products worldwide. ZEN is responsible for various toxicological effects in humans and animals, among them the most critical are due to the estrogenic activity triggered by the activation of estrogen receptors. The toxicokinetic of ZEN in mammals has been widely investigated, leading to the identification of the major and minor phase I metabolites. In addition, phase II glucuronidated and sulfated forms have been described as well. Beside the longstanding investigation of xenoestrogenicity of reduced forms, the response mediated by oxidized metabolites and phase-II conjugated forms is largely overlooked. Similarly, although the existence of phase-II plant metabolites has been largely documented, their role in the overall xenoestrogenic dietary load is actually understudied. All these modified forms should be investigated to promote a whole scenario for risk assessment, but the lack of commercial standards actually makes such study still challenging. **Material and Methods:** In this context, the integration of in silico and in vitro analysis smartly improves the research by giving the opportunity to focus experimental efforts solely on noteworthy candidates. The in silico modeling – based on docking simulations and rescoring procedures – provides indeed structural insight to better understand the triggering of xenoestrogenic response. **Results:** Within this framework, a series of oxidized ZEN metabolites and conjugated forms (including glucuronide, glucoside and sulphate) have been investigated for their capacity to bind and activate the estrogen receptor alpha. Notably, a properly validated and effective in silico model has made affordable the analysis of a wide range of modified forms. **Conclusion:** Overall, our findings outlined the xenoestrogenic activity of ZEN modified forms not yet included in risk assessment studies. Interestingly, the conjugation seemed to be the only metabolic modification truly capable to quench the xenoestrogenic activity of ZEN.

122. ECsafeSEAFOOD: Development of strategies for evaluating the incidence of ciguatera in the EU

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ABSTRACT

Cases of ciguatera fish poisoning (CFP) have been documented in Macaronesia from consumption of amberjack (Seriola spp.), where ciguatoxins (CTXs) were confirmed by liquid chromatography coupled to mass spectrometry (LC-MS). Cases of CFP have also been reported in European hospitals from consumption of imported fish. Additionally, EU citizens travelling abroad to CFP endemic areas face the possibility of illness through consumption of ciguatoxic fish where symptoms characteristic of CFP may persist upon return. A framework to evaluate the risk of CFP to EU citizens is proposed based on approaches applied in the French Antilles island of Guadeloupe. In Guadeloupe, samples of fish implicated in several episodes of CFP and associated epidemiological data were collected. Composite toxin levels and structural confirmation of CTXs in the samples were determined by cell-based assay and LC-MS/MS, respectively. The lowest adverse effect level (LOAEL) for CTXs was determined from these toxin measurements and epidemiological records. In another study, the content of CTXs in lionfish from different islands in the Guadeloupe archipelago was determined using the same methodologies. The evaluation of CTXs in fish samples allowed us to define regions where these toxins occur and the proportion that contained CTXs at levels considered hazardous for consumption. A first approach using this framework has been adopted for the evaluation of *Gambierdiscus spp.* in Madeira and Canary Islands, and CTXs in fish from Canary Islands, Madeira (Atlantic Ocean) and La Réunion (Indian Ocean). Therefore, based on our results and risk management strategies (adopted in areas where ciguatera is endemic), we are presenting a plan to determine the incidence of CFP in the EU. Acknowledgements: Part of the research was conducted in the frame of the ECsafeSEAFOOD project funded by the European Union's Seventh Framework Programme for Research and Technological development (FP7/2007-2013) under Grant Agreement n°311820.

123. The essential micronutrient vanadium – Effects of viability and proliferation of chicken and rat virus-transformed cancer cells

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ABSTRACT

Objectives: The interest in biological activity of vanadium and its compounds has increased significantly during the recent years. This fact could be explained by at least three reasons: i) Vanadium is included in the list of 40 essential micronutrients that are required in small amounts for normal metabolism; 2) There are many data confirming the antitumor and anti-diabetic properties of vanadium; ii) The daily human exposure to vanadium compounds due to the wide application of this metal in current industry. Up to now the influence of this metal on growth of virus-transformed tumor cells is not fully clarified. Aim. The aim of our study was to evaluate the effect of ammonium vanadate (NH4VO3) on viability and proliferation of cultured virus-transformed chicken and rat cancer cells. **Materials and Methods:** Permanent cell lines LSCC-SF-Mc29 (chicken liver cancer induced by the myelocytomatosis virus Mc29)

and LSR-SF-SR (sarcoma in rat induced by Rous sarcoma virus strain Schmidt Ruppin) were used as model systems in experiments. The investigations were performed by thiazolyl blue tetrazolium bromide (MTT) test, neutral red uptake cytotoxicity assay (NR), double staining with acridine orange and propidium iodide, method of Pappenheim and colony forming method. **Results:** The results obtained revealed that applied at a concentration range of 0.1–20 µg/ml NH4VO3 expresses significant cytotoxic and/or cytostatic effects that are time- and concentration dependent. The compound completely inhibited colony-forming ability of tumor cells in semisolid medium at concentrations ≥ 5 µg/ml. Positive correlations between the data coming from short-term tests (NR and MTT, 24-72 h) in monolayer cultures and long-term colony-forming assay (16 days, 3D colonies in semi-solid medium) as well as between MTT (which reflects damage to mitochondria) and NR (indicates damage to lysosomes and Golgi apparatus) methods were observed. **Conclusions:** NH4VO3 decreases significantly viability and proliferation of LSCC-SF-Mc29 and LSR-SF-SR cells that express oncogenes v-myc (chicken liver cancer) and v-src (rat sarcoma). The cellular analogues of these genes are involved in signaling pathways responsible for cell proliferation, differentiation, survival, migration, invasion, and angiogenesis and disturbed balance of their expression has been suggested to be connected with pathogenesis of > 80% of human cancers. **Acknowledgements:** Supportet by Grant DFNI Б-02/30 from 12.12.2014 from National Scientific Fund in Bulgaria.

124. Predicting the mutagenic and carcinogenic potential of heat-induced food contaminants using open source QSAR software models

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ABSTRACT

Introduction: Numerous chemical compounds are formed in the course of the thermal treatment of foods. Some of these heat-induced food contaminants such as acrylamide or furan are toxicologically well-characterised; however, there are hardly any toxicological data available for most of these substances. At least more than 800 reaction products of the Maillard reaction and lipid oxidation products are known to be present in foods such as heated cereals, roasted meat, refined oils, coffee, juices, and many others. **Objectives:** Due to the lack of experimental toxicological data for most of these compounds, an in silico approach was conducted in order to predict the hazard potential of these substances. Freely available software tools for the prediction of toxicological endpoints such as mutagenicity and carcinogenicity on the basis of the structure of the respective compound were employed to examine (quantitative) structure-activity relationships (QSAR) for these more than 800 substances. **Methods:** The T.E.S.T. software package (version 4.1; US EPA) and three models of the VEGA platform (version 1.0.6; Mario Negri Institute, Milan) were used for the prediction of mutagenicity (CEASAR, Benigni-Bossa, and SarPy) and two models to predict the carcinogenic potential (CEASAR and Benigni-Bossa) of the compounds. The lazy structure-activity relationship (lazar) framework contributed to both the mutagenic and the carcinogenic endpoint. Finally, the results of the five different mutagenicity models as well as the results of the three carcinogenicity models were combined in order to rank the 800 compounds for their predicted mutagenic and carcinogenic potential, respectively. **Results:** Based on this in silico analysis, the class of vinylic aldehydes as well as furan derivatives and chloropropanols were predicted to be the most harmful compounds and are therefore highly recommended to be characterised toxicologically in detail in the future in order to generate the needed experimental data for a prospective risk assessment of these heat-induced food contaminants. **Conclusion:** A combination of different computer models can be used to predict the mutagenicity and the carcinogenicity of new compounds for prioritization purposes.

125. The ILSI Europe expert group on the re-evaluation of the cancer potency database

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ABSTRACT

Objectives: Based on evaluation of the Carcinogen Potency Database (CPDB) and structural prediction of potential genotoxicity, it has been concluded that there is a very low probability of harm to human health when exposure to certain compounds from the diet is below 0.15 µg/day, a threshold of toxicological concern (TTC) (see Kroes et al., 2004). With the expansion of applications for the TTC approach, it is important to provide a robust, scientific, and transparent basis for this TTC value. ILSI Europe has therefore recently started re-evaluating the CPDB supporting the TTC for potential genotoxic carcinogens, with a focus on accepted approaches to cancer risk assessment. The CPDB will be refined and reanalysed, to establish whether this TTC value is still appropriate, based on the most up-to-date information. **Materials and methods:** - Clean-up the CPDB with respect to data relevance and analysis of studies; - Re-analyse the 'clean' dataset and re-evaluate the TTC based on identified modes of action and the results of genotoxicity tests; - Recommend an appropriate strategy for the stratification of compounds that are potentially genotoxic and/or carcinogenic, with respective TTC values, based on structural alerts; - Confirm whether human exposure thresholds for Cramer classes I, II and III are appropriate for non-genotoxic carcinogens. **Expected results:** This work will provide state-of-the-science support for one or more TTC values for potential genotoxic carcinogens, based on chemical class and/or structural prediction of mode of action. Additionally, analyses of the impact of data on non-genotoxic carcinogens on the thresholds and on the applicability of the Munro/Cramer non-cancer thresholds to such compounds should increase confidence in the approach, or identify areas requiring further work. **Conclusions:** The results will be published as peer-reviewed publications. In addition to strengthening the scientific basis of the cancer-based TTC tier(s), this activity will also provide a publically-available database, allowing separate analyses by independent parties. In addition to carcinogenic potency, the database will include information on: - Chemical class; - Presence/absence of structural alerts for DNA reactivity; - Genotoxicity mechanism and data; - Existing regulatory classifications, such that one could choose to evaluate only those chemicals currently recognized as known/probable carcinogens and/or mutagens.

126. In silico preliminary hazard evaluation of androgen receptor-mediated endocrine-disrupting effects of thioxanthone photoinitiators and their metabolites

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ABSTRACT

Objectives: The purpose of this work is to show how in silico tools can (i) predict toxicological behavior exerted by food contaminants and (ii) contribute to reveal their sub-molecular implications. In 2005, EFSA reported the case of infant formulas contamination by thioxanthone photoinitiators. In this regard, we have (i) built a computational model to correctly predict endocrine disrupting effects of these photoinitiators and (ii) analyzed the potential effects of their in silico predicted phase I metabolites. **Materials and Methods:** The applied protocol can be summarized as follows: validation, metabolites prediction, docking, rescoring and binding-affinity prediction. Previously reported data on 2-ITX metabolism and anti-androgen activity was used as a validation set respectively for making metabolic and androgen receptor binding-affinity predictions. Metaprint2D (http://www-metaprint2d.ch.cam.ac.uk) was used for metabolic predictions whereas GOLD docking software and HINT scoring function were used respectively for protein-ligand pose generation and energetic evaluations. **Results:** Twelve phase I metabolites were predicted starting from the five photoinitiators reported by Reitsma and co-workers. All these structures were docked into the androgen

receptor ligand binding pocket (AR-LBP) and energetically rescored with HINT function. Competitive ability towards the endogenous ligand (testosterone) was measured as affinity ratio by dividing the interaction score of each ligand for that of testosterone. At least six of the twelve analyzed compounds revealed to be suitable to interact with AR-LBP: in particular, hydroxylation of thioxanthone isopropyl moiety enhances protein-ligand interaction thanks to H-bond generation. Conversely, sulfoxidation is poorly tolerated because of negative hydrophobic-polar interactions. **Conclusions:** Our results suggest that an hazard evaluation focused not only on unaltered thioxanthones but also on their metabolites could be necessary to avoid a potential underestimation of the toxicological risk for this class of chemicals. The case study reported above, is only one example of the wide possible applications of computational tools so it shows that this approach could be a good support for a more complete risk assessment in food safety.

127. Serum protein profiling as a tool to identify dioxin contaminated cattle

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ABSTRACT

Objectives: Animal productions represent the major source of exposure to dioxin-like (DL) compounds (PCDD, PCDF and PCB) for humans. As analytical techniques for surveillance purposes are time-consuming and expensive, the development of faster and cost effective screening methods able to identify the contaminated animals is of growing interest. In this respect, proteomics represents a promising approach to detect the modifications of protein expression in response to different biological events, such the exposure to toxic agents. The aim of this study was to investigate the changes in serum protein profile from heifers contaminated by DL-PCBs, in order to identify reliable biomarkers of exposure in easily collectable samples from living animals. **Materials and methods:** One-year old DL-PCBs accidentally exposed heifers (n=8) were reared for 6 months in an uncontaminated experimental facility. From each animal perirenal fat biopsies and blood samples were collected 4 times at two-month intervals (A-B-C-D). Fat DL-PCB content was measured by GC-HRMS. Processed serum samples were analyzed in the 2-20 kDa range using a Bruker Microflex LT mass spectrometer. Mass spectra were subjected to statistical analysis by the ClinProTools software through the Wilcoxon and Kruskal-Wallis test. After isolation by liquid chromatography, statistically relevant peaks were identified by MALDI-TOF/TOF-MS and/or nanoLC-ESI-linear ion trap-MS/MS, and by protein database searching using the MASCOT engine. **Results and Conclusions:** Initial DL-PCB level of each animal was higher than 20 pg/g fat, and decreased rapidly in sampling B (7.71± 0.32 pg/g fat), complying with legal limits in samplings C and D (below 5 pg/g fat). Protein profiling revealed 48 statistically significant peaks ($P<0.05$), which were selected for identification according to their increasing or decreasing intensity among the 4 samplings. Fibrinogen β-chain, apolipoprotein A-II, and apolipoprotein C-III raised throughout the decontamination, whereas apolipoprotein C-II short-form, serpin A3-7, serum amyloid A-4 protein, and hemoglobin subunit-α declined. Interestingly, similar changes in serum apolipoproteins have been reported in TCDD-exposed humans, while amyloid A, a known inflammatory marker, is associated with PAH exposure. Although further research is needed, serum proteomics seems to be a promising technology to identify biomarkers of exposure to DL-compounds.

128. The substitution principle in the LIFE-EDESIA animal free approach: an in silico project update

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ABSTRACT

The main goal of the LIFE-EDESIA project is to apply the substitution principle to Endocrine Disrupting Chemicals (EDCs), such as phthalates and bisphenols, in order to find chemical with a lower environmental and human health impact. Within REACH, on case-by-case, EDCs are considered of 'equivalent concern' to Substances of Very High Concern (SVHC): some phthalates and bisphenols, indeed, are classified as toxic for reproduction because of their endocrine disruption effects and to the potential exposure of the whole population. The project involves the use a new, robust and cost-effective in silico/in vitro approach to evaluate suitable chemicals for EDC replacement that combines both computational screening methods and in vitro functional assays based on endocrine-dependent human clinical biomarkers of effect (endocrine-activity). To identify potential substitutive chemicals, the first selection criteria were based both on the state of the art and applying available computational in silico methods (e.g., QSAR, virtual screning, molecular docking,). Using in silico methods, endocrine properties of a list of potential alternatives were analyzed to evaluate their potential toxicity (carcinogenicity, mutagenicity and development toxicity) or detrimental environmental properties (persistence and bioaccumulation). Different in silico approaches were used addressing primarily estrogen receptor (ER) and androgen receptor (AR) binding but including also models for more global effects derived on the basis of EC priority lists. Moreover, using a 'consensus scoring' approach, two different docking techniques have been used to predict the interactions of the list of potential alternatives to ER and AR and, hence, to classify the chemical set as positive interactors, negative or may be. Only molecules predicted as negative interactors should be considered for in vitro test as no ED-like chemicals. Both techniques converged to the same results allwing to drive a reduced set of in vitro tests.

129. Simultaneous determination by liquid chromatography/mass spectrometry of five low calorie intense sweeteners in urine: a potential biomarker approach for assessing intakes

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ABSTRACT

Background: Low calorie intense sweeteners (LCS) are food additives which are commonly used as sugar substitutes in a variety of products in order to provide a sweet taste without increasing the energy content. Methods of monitoring exposure to these compounds can be prone to error and given that several LCS are excreted via the urine largely un-metabolised(1-3), an opportunity exists to implement a biomarker approach for investigating levels of exposure more objectively. Simultaneous analysis of various LCS has been carried out in various foodstuffs(4) and waste water(5). No such method, however, has been published for urine. **Objectives:** This project aimed to develop a method for the simultaneous determination of four LCS (acesulfame-K, saccharin, cyclamate, sucralose) and the excretory product of a relatively new sweetener, steviol glycosides (steviol glucuronide), in human urine. **Materials and Methods:** Liquid chromatography with mass spectrometry was utilised in order to separate and quantify the five compounds of interest. A simple sample preparation procedure, consisting of a 10-fold dilution followed by filtration, was used. Method validation involved the assessment of linearity & range, limits of detection (LOD) and quantification (LOQ), as well as accuracy and precision (intra-batch and inter-day) which were assessed at three concentrations across the dynamic range (12.5, 550 and 930ng/ml). **Results:** Very good linearity was observed for all five LCSs across a large dynamic range of 10-1000ng/ml (R2 0.997-0.999) with

LOD and LOQ ranging from 0.01-0.4ng/ml and 0.05-1ng/ml respectively. At low, medium and high concentrations, accuracy ranged from 92.7-104.4% while %CVs were below 8% for both intra-batch and inter-day precision. **Conclusion:** A simple and reliable method has been developed representing an opportunity to implement a novel biomarker approach suitable for objectively assessing dietary exposure to five LCS. Further work is currently underway to investigate the dose response relationship between intakes and excretion and which will help determine the feasibility of implementing such an approach. 1. Renwick AG (1986) Xenobiotica 16, 1057-1071 2. Roberts A, Renwick AG, Sims J et al (2000) Food Chem Toxicol 38 (Suppl 2), S31-S41 3. Guens JMC, Buyse J, Vankeirsbilck et al (2007) Exp Biol Med 232, 164-173 4. Zygler A, Wasik A, Namiesnik J (2009) Trends Anal Chem 28, 1082-1102 5. Ordonez EY, Quintana JB, Rodil R et al (2012) J Chromatogr A 1256

130. Simultaneous determination of azoxystrobin, acetamiprid and metalaxyl pesticide residues in tomato by solid-phase extraction and rapid resolution liquid chromatography

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ABSTRACT

Tomato is affected by many pests, hence farmers applied excessive amounts of different types of pesticides to protect their crops. It is important to know about the presence of pesticide residues in tomato which will help to minimize the potential human's health risk. This study presents a new method for simultaneous determination of three pesticides (azoxystrobin, acetamiprid and metalaxyl) frequently used in tomato production in Macedonia. Pesticide residues were extracted with acetone and purified by both liquid-liquid extraction (LLE) and solid-phase extraction (SPE). The separation and quantification of investigated pesticides was performed by reverse-phase rapid resolution liquid chromatography (RP-RRLC) with diode-array detection (DAD). The best results were obtained using acetonitrile:water (50:50, V/V) as the mobile phase with flow rate of 1 mL/min, and UV detection at 220 nm and 250 nm. The optimal analytical separation was achieved with a LiChrospher 60 RP-select B (250 x 4 mm, 5 µm) column. The method had a good linear relationship in the range of 1.05 - 3.59 mg/kg for azoxystrobin, 0.07 - 0.24 mg/kg for acetamiprid and 0.07 - 0.24 mg/kg for metalaxyl. The precision was evaluated for the retention times, peak areas and peak heights of investigated pesticides. Under the established condition, the recovery of azoxystrobin, acetamiprid and metalaxyl was 96.03 % - 102.95 %, 95.56 % - 104.68 %, and 96.62 % - 106.11 %, with relative standard deviations below 2.53%. The limits of detection obtained for the developed method were 0.35 mg/kg for azoxystrobin, 0.023 mg/kg for acetamiprid and 0.023 mg/kg for metalaxyl. The quantification limits were 1.05 mg/kg, 0.07 mg/kg and 0.07 mg/kg for azoxystrobin, acetamiprid and metalaxyl, respectively. The proposed method was successfully applied to the monitoring of investigated pesticides in tomatoes grown in different regions in Macedonia in 2013. The obtained results showed that analyzed tomato samples did not contain detectable pesticide residues.

131. Threshold of Toxicological Concern: application in the assessment of chemically complex food matrices

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ABSTRACT

Food products are usually chemically complex food matrices (CCFM) containing a high percentage of unknown substances. The safety assessment of CCFM may therefore be time consuming and expensive since it may involve animal studies, and may require the full composition of the food to be determined. Furthermore, each of the chemical constituents known to be present require a safety assessment whereas the intake of those constituents is often relatively low, making the necessity for a full safety assessment of these substances questionable. The Threshold of Toxicological Concern (TTC) concept provides a generic safety assessment strategy relating human exposure threshold values to chemical structures. We drafted a framework in which the TTC concept and its thresholds are used for the assessment of unknown structures in CCFM (Rennen et al., 2011). The framework consists of a step-by-step multidisciplinary strategy combining analytical techniques and bioassays with the TTC principle. The major advantage of this complex mixture safety assessment strategy (CoMSAS) is that it enables to distinguish toxicologically relevant substances from toxicologically less relevant substances, when related to their respective levels of exposure, and allows one to focus on substances of potential health relevance. This reduces the amount of work needed for identification, characterization and evaluation of each unknown substance present without jeopardizing consumer safety. This includes the relevance of cumulative effects of substances present at low exposure levels, which also has been investigated (Leeman et al., 2013). The effective applicability of CoMSAS in the assessment of both food packaging materials and of food products has been demonstrated with case studies (Koster et al., 2014 and Koster et al., 2015). Over the past years, the thresholds used in the TTC principle, and consequently in CoMSAS, have further evolved. For substances in Cramer class III it has been concluded that for organophosphates and organohalogens a separate threshold may apply. However, the overall Cramer class III threshold has not been adjusted after exclusion of these chemicals. We have reevaluated the original TTC database based on current insights and were able to elaborate new thresholds (Leeman et al., 2014). The research performed on TTC and CoMSAS will be presented in an integrated way to demonstrate possibilities and limitations of TTC in the safety assessment of CCFM.

132. B-13 Progenitor-derived hepatocytes (B-13/H cells) as a model for lipid dysregulation

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ABSTRACT

Obesity and type II diabetes is a growing epidemic globally and the use of food additives could potentially have a role in these diseases. Steatosis is the accumulation of lipid droplets; this can occur in hepatocytes due to an imbalance of storage and metabolism of triglycerides and can develop into non-alcoholic steatohepatitis, fibrosis and cirrhosis. An effective in vitro model for steatosis could be used to study the mechanism of how this occurs and how steatotic cells respond to other stresses. We hypothesised that B-13 progenitor-derived hepatocytes (B-13/H cells) can provide a simple, cost-effective in vitro model for lipid accumulation in liver cells in response to exposure to high concentrations of fatty acids. B-13/H cells were treated with medium containing oleic and linoleic acid for 3 days, after treatment the cells were fixed and stained for lipids using Oil Red O staining. B-13/H cells accumulated lipid droplets in a dose- and time-dependent manner when they are exposed to fatty acids. This effect was not reversible when the fatty acid media was replaced with normal growth media. Lower concentrations of fatty acids (0.25mM) resulted in microsteatosis, whereas at higher concentrations (2mM) resulted in macrosteatosis. B-13/H cells also expressed the liver X receptor α (LXRα) that is activated by oxysterols in response to increases in cholesterol levels. However LXR also activates sterol regulatory element-binding protein-1c (SREBP-1c) and carbohydrate response element binding protein (ChREBP) proteins in the lipogenic pathway, resulting in de novo fatty acid synthesis. Therefore activators of the LXR cause steatosis. When treated with the synthetic LXR agonist T0901317, B-13/H cells showed a dose-depended increase in lipid accumulation. These results demonstrate that B-13/H cells provide a simple and cost-effective model for lipid accumulation. B-13/H cells could be used to investigate the mechanism underpinning steatosis and the effects that exposure of food chemicals have on the response. This model could also be used screen drugs and food chemicals for LXR activation and steatosis.

133. E171 as ingredient for cake decorations: physico-chemical characterisation of titania (nano)particles

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ABSTRACT

Since the last few years titanium dioxide (TiO2) has become a common additive in food stuff and personal care products. Even though its use as food colorant (E171) is regulated by the European Directive 94/36/EC, the daily human exposure is raising, causing lots of concerns about its safety. Being aware that E171 is spreadly used as white pigment and thickening agent, TiO2-based food colours for cake decoration were chosen for this case study. The main objective was to physico-chemically characterise different E171-based colorant powders, as a fundamental step to predict their biological and environmental risks. Scanning Electron Microscopy, Particle Size Distribution Analysis and X-ray Diffractometry were applied to study the particle morphology, size and size distribution, as well as the chemical and phase composition of the dry powders. Measurements of zeta potential in water and in physiological solution disclosed the surface charge of the particles in different pH conditions. The analyses revealed that the powders were made of titanium dioxide, as declared. The particles appeared rounded, with an average size ≈120 nm and quite spread size distribution. The majority of particles were micron-sized, but a fraction up to 30% fitted within the nano-range. As a semiconductor, nano-sized TiO2 undergoes photocatalysis, if activated by UV light; thus, our purpose was to study the photocatalytic activity of the E171-based powders under UV light, following the degradation of the other components which constitute the food colorants (champagne, petal blue and cream colours). Pure TiO2 food grade white pigments from different brands were also used to assess the organic degradation of caffeine (used as model molecule) upon UV irradiation. The results showed that the TiO2-based food colorant powders were able to photocatalitically destroy organic molecules, even though in different extents. Their photocatalytic activity was related to their physico-chemical properties. Due to the large amount of nanoparticles, which can interact with the gastrointestinal tract once ingested, and their dispersivity in liquids, the outcomes of this study contribute to warn the scientific community and the authorities about the possible toxic effect of such commercially available products. Future studies will concern in vitro toxicological tests of food grade colorants containing TiO2 (nano)particles.

134. DHT-induced PSA secretion to screen plant bioactives with an anti-androgen-like activity

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ABSTRACT

Introduction: Prostate, a critical but yet overlooked target for endocrine disruptors (EDCs), has received limited attention in male reproductive toxicity screening. Prostatic fluid contains proteins (e.g., the Prostate-Specific Antigen, PSA), trace elements (e.g., zinc) and other molecules (e.g., citrate) essential to sperm cell activation and capacitation. PSA is also used as a specific and reliable toxicological biomarker to monitor prostate epithelial human cells upon treatment with EDCs, including plant bioactives. The soy isoflane genistein /GEN, five common Mediterranean diet plant bioactives (apigenin/API, luteolin/LUT, naringenin/NRG, quercetin/QRC, resveratrol/RESV) were tested in comparison to the sex steroids DHT and E2 to set up the in vitro model system (human LNCaP cells secreting PSA). Finally, the model system was tested also with a Bulgarian plant extract from Verbascum xanthophoeniceum in comparison to the worldwide used natural anti-inflammatory agent Serenoa repens. **Methods:** LNCaP were treated in a dose-dependent manner (1pM-100μM range) with plant bioactives (± sex steroids) to test i) cell viability (MTS assay), ii) DHT-induced PSA secretion and iii) intracellular distribution (differential centrifugation followed by chemical analysis by HPLC and/or GC-MS). **Results:** In the presence of DHT, plant bioactives partially reduced the DHT-induced PSA secretion to a different extent (generally between 20%-40%) and in a non-linear manner. The intracellular distribution showed that: i) plant bioactives enter within LNCaP similarly to E2, ii) intracellular plant bioactives distribute within N similarly to DHT, iii) plant bioactive 'nominal concentration' is different from the 'real' amount effectively entering within the cell, iv) DHT or E2 influence their ability to enter within the cell and to distribute in the different cellular compartments. Moreover, V. xanthophoeniceum and S. repens extracts were for the first time to have, in the absence of cytotoxicity, an inhibitory effect on DHT-induced PSA secretion. **Conclusions:** In an in vitro model of human prostate epithelium, using the cell-specific DHT-induced PSA secretion assay, plant bioactives or extracts potentially interfering with the prostate function can be successfully screened for their (anti)androgenic ability.

135. Systematic literature review on in vitro and alternative Developmental Neurotoxicity (DNT) testing methods

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ABSTRACT

The goal of this systematic review performed under a contract with EFSA was a comprehensive literature search and the evaluation of information on alternative testing methods in the field of developmental neurotoxicity (DNT) that could support the EFSA Pesticides Unit with respect to the peer review of active Substances under Reg. 1107/2009. The review question herein was summarized as: 'Which test methods or approaches are available to evaluate developmental neurotoxic effects of chemical exposure?' Thus a strategy was developed to search for all available published literature, relevant grey literature and website information (in English language) from 1990 to mid of April 2014. This should result into the state of the art of in vivo DNT testing methods including novel and alternative non-mammalian models, in vitro test methods, in silico-methods, read across approaches and combinations of testing methods in test batteries. Inclusion and exclusion criteria had to be defined to select appropriate papers for further critical evaluation of methods and strategies for DNT testing. This systematic review retrieved a variety of methods suitable to identify adverse effects of chemicals during different stages of neurodevelopment. In general the published data across all method types is very heterogeneous, with a range of different cell types from different brain regions, different exposure paradigms and various compounds tested. From the alternative methods a testing strategy covering early and later neurodevelopmental stages (from stem cell to zebrafish larvae motor behavior) can be assembled. For ultimately gaining regulatory acceptance, definition of biological application domains of human NS/PC-based methods by performing in vitro - in vivo validation is needed. Moreover, protocols for cell-based and zebrafish assays need international standardization. With such standardized protocols, a test battery needs to be tested for its performance parameters (sensitivity and specificity) by testing concentration-responses of known DNT positive and negative compounds across different in vitro and zebra fish assays. Such data might in the end be used to support international regulatory assessments or compound prioritization, both in favor of a reduction of rodent DNT in vivo testing.

136. Investigating hepatic and gastrointestinal effects of tartrazine in an experimental mouse model

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ABSTRACT

Tartrazine (E 102) is a water soluble food additive (ADI 7.5 mg/kg bw/d) with a known history of causing intolerance reactions in sensitive individuals (EFSA, 2009). Tartrazine has also been shown to be a human estrogen receptor (ER) activator in vitro. Since (xeno)estrogens are known to be cholestatic, we hypothesized that tartrazine will cause a cholestatic (periportal) injury if significant absorption occurs. To test this hypothesis, mouse ERs were

cloned and their activation by tartrazine and its gut-derived metabolites was examined. To test for mouse ERα activation in vivo, 19 day old female mice were dosed i.p. with up to 50mg tartrazine/kg bw/d for 4 days prior to determination of relative uterine wet weight on day 5. Tartrazine was also administered either i.p. (male adult mice, 50mg/kg bw, 10 doses over 14 days) or by oral gavage (male adult NF-kB-luciferase (Tg) or wild type mice, 50mg/kg bw twice daily for 10 weeks (+/- 6g/kg bw/d ethanol to modulate gut permeability). In reporter gene assays, estrogen – but not tartrazine nor its metabolites – activated the ERα and ERβv1. In vivo, tartrazine also failed to increase relative uterine wet weight. Exposure to tartrazine by i.p. injection resulted in mild cholestatic liver injury (increases in serum ALP and pathology). Live imaging of Tg mice demonstrated that oral tartrazine caused increases in inflammation after 8 weeks in the gut and by 10 weeks in liver, whereas the combined treatment with ethanol had no effect. However, serum ALP and histopathological examinations did not identify any hepatic changes of toxicological relevance. These data indicate that tartrazine is not an activator of the mouse ERs and that mild hepatic injury through direct (i.p. administration) exposure to tartrazine is independent of the ER in mouse. However, oral exposure to tartrazine failed to re-capitulate these effects, despite evidence of an inflammatory response in the gut and liver. Increasing gut permeability through alcohol exposure reduced the apparent inflammatory effects of tartrazine. Tartrazine is therefore not a mouse ER activator and not a good model with respect to potential estrogenic effects of tartrazine in man. Although tartrazine caused a mild cholestatic injury when injected i.p., oral exposure showed minimal effects likely due to minimal absorption via this route. Reference: EFSA. Scientific Opinion on the re-evaluation tartrazine (E 102). EFSA Journal 2009; 7(11):1331

137. Subchronic toxicity study with a GM Maize MON810 Pioneer

Ondrejkova J, Alacova R, Ambrusova K, Bartusova M, Kebis A, Kovriznych J, Krivosikova Z, Kuricova M, Liskova A, Rollerova E, Spustova V, Szabova E, Tulinska J, Zeljenkova D

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ABSTRACT

Objectives: One of the aims of the GRACE (GMO Risk Assessment and Communication of Evidence) project is to conduct 90-day animal feeding trials and animal studies with an extended time frame on genetically modified (GM) maize in order to comparatively evaluate their use in GM plant risk assessment. We present results of a 90-day feeding study with a GM maize MON810 Pioneer variety, its near-isogenic non-GM variety and two additional conventional maize varieties. **Methods:** The rat feeding study on MON810 Pioneer was performed by taking into account the EFSA Guidance on conducting repeated-dose 90-day oral toxicity study in rodents on whole food/feed (EFSA Scientific Committee, 2011) and the OECD TG 408. Three dietary treatments represent the groups of near-isogenic control, 11% GMO and 33% GMO and two additional groups consisted of two conventional maize varieties. **Results:** No statistically significant differences in feed consumption were observed between the male and female rats in the five experimental groups. Male rats fed the conventional 1 diet had a significantly lower percentage of monocytes than the male animals fed the control diet while no significant differences regarding the haematology parameters were observed between the female rats fed the conventional 1 and the control diet. The alanine transferase (ALT) and aspartate aminotranferase (AST) activities were significantly increased in the serum of female rats being fed the 33% GMO diet if compared to the animals receiving the control diet. However, the ALT and AST activities measured in the serum of 33% GMO-fed female rats are in the same range as the historical ALT and AST data collected by the breeder company for control animals of the same strain, age and gender. Furthermore, the ALP activity was similar to that of the control diet-fed animals and no sign of liver injury was observed in the gross necropsy as well as the histopathological analyses. **Conclusion:** The results obtained when feeding rats with the near-isogenic non-GM maize and the conventional maize varieties were mostly similar. In those cases, in which statistically significant differences between the control group and the groups fed the conventional maize varieties were observed, the measured values were within the range described for the individual parameters in control animals of the same strain and age. The research study was supported by the European Commission: 7th Framework Programme for the project GRACE 311957.

138. Adverse Outcome Pathway (AOP) describing key events linked to motor deficit of Parkinson's disease caused by exposure to rotenone

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ABSTRACT

The Adverse Outcome Pathway (AOP) concept facilitates understanding of complex biological systems and the pathways of toxicity that result in adverse outcomes (AOs). It has been developed as a tool for a knowledge-based safety assessment that relies on understanding mechanisms of toxicity. Using the AOP framework we described the Key Events (KEs) that are triggered by the Molecular Initiating Event (MIE) when an inhibitor binds to the Complex I of the mitochondria and finally leads to motor deficit of Parkinson's disease (PD). Indeed, chronic systemic complex I inhibition caused by a pesticide such as rotenone induces features of PD in rats including selective nigrostriatal dopaminergic lesions and development of alfa-synuclein-positive cytoplasmic inclusions in nigral neurons resulting in hypokinesia and rigidity. In the brain, mitochondria play a pivotal role in neuronal and glial cell survival and cell death because they are regulators of both energy metabolism and apoptotic/necrotic pathways. Due to their structural and functional complexity, they present multiple targets for neurotoxic compounds and mitochondrial dysfunction contributes to the pathology of various neurodegenerative disorders. Based on the systematic literature reviewing the following KEs of this AOP have been identified: MIE: Binding of inhibitor to subunit of NADH- ubiquinone oxidoreductas; KE1: Partial or total Inhibition of Complex I; KE2: Mitochondrial dysfunction; KE3: Impaired protein degradation and damaged intracellular transport; KE4: Degeneration of Dopaminergic (DA) terminals in striatum; KE5: Cell death of DA neurons in substantia nigra (SN, pars compacta) and AO: Motor deficit symptoms of PD. This AOP can be applied to chemicals targeting the brain and inhibiting mitochondrial Complex I, such as rotenone and MPP+, increasing oxidative stress, inducing selective degeneration of the nigrostriatal pathway and cell death of dopaminergic neurons in the substantia nigra, pars compacta. As a result, dopamine release in the striatum is reduced affecting processes related to motor control and various cognitive functions, suggesting that these toxicants may be associated with increased risk of PD. The proposed AOP will be further developed in a qualitative and where possible quantitative manner to support systematic use of mechanistic knowledge for regulatory safety assessment of human health.

139. Identification of transcriptome signatures and biomarkers specific for migration-inhibiting potential developmental toxicants in human neural crest cells

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ABSTRACT

Disturbance of the neural crest cell (NCC) migration process by toxicants is known to lead to severe malformations in model organisms, and several factors (e.g. genetics and chemicals) have already been identified as causes for neural crest- related developmental defects. The Migration of Neural Crest cell (MINC) assay was set up as first human stem cell-based method that is able to detect the functional effects of chemicals on one of the key events of nervous system development, the neural crest cell migration (Zimmer et al., 2012). Its performance characteristics allowed the MINC assay to be included in the screening test battery of the European project ESNATS (Embryonic Stem cell-based Novel Alternative Testing Strategies). This project allowed the testing of a wide array of substances comprising environmental pollutants and modern pharmaceutical substances and it lead to the identification of several migration-impairing toxicants (Zimmer et al., 2014). In this study, we compared transcriptome profiles of different MINC-positive compounds, including drugs (geldanamycin), environmental chemicals (triadimefon, arsenic trioxide and the flame retardant PBDE-99) and known developmental toxicants (valproic acid

and trichostatin A), in order to investigate which processes are mainly targeted by the different migration inhibitory drugs. We used an unbiased system-wide transcriptome analysis by micro-array to gain insight into altered gene expression patterns; transcriptome profiles were compared and enriched GOs and KEGG pathways were classified and contrasted, showing the presence of different processes targeted by the MINC-positive compounds. Furthermore, we used different approaches to identify suitable biomarker candidates. The first method is based on the comparison and the overlap of the transcriptome profiles among the different toxicants; the second approach is based on a novel system of scoring, which assigns different weight to each gene, according to its characteristics of 'measurability', 'statistical power' and 'biological relevance'. The found candidate biomarker genes will contribute to optimize the biomarker identification strategy for new developmental toxicant identification and characterization. References: Zimmer B. et al., Environ Health Perspect. 2012 Aug;120(8):1116-22 Zimmer B. et al., Arch Toxicol. 2014 May;88(5):1109-26.

140. Neurodevelopmental toxicity of perinatal exposure to a food matrix contaminated with the six indicator non-dioxin-like polychlorinated biphenyls in a mouse model

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ABSTRACT

Polychlorinated biphenyls (PCBs) are a class of man-made organic compounds, hazardous for human health, biomagnified in the food chain despite a regulatory stopping of their production in the 80's. Among them, the toxicity of the six non-dioxin-like PCBs (6 NDL-PCBs; PCBs: 28, 52, 101, 138, 153 and 180), considered as indicators because of their significant occurrence in food and feed, are less studied. Since no data were available on their impact in a realistic context of exposure, this study enrolling in the NeuroDeveTox project (ANR-CESA-2011) focused on the effects of a naturally contaminated food matrix. Fishery products being for the 6 NDL-PCBs the major source of human exposure, different regimes based on eel, the most accumulative specie, was tested in an animal model. For this, Swiss albino female mice were daily subjected from gestational day 7 until postnatal day (PND) 21 to the 6 NDL-PCBs at 0, 85, 200 and 400 ng/kg/day via a food paste containing lyophilisate eel. From parturition to PND 11, reproductive and physiological parameters (sex ratio, litter size, mortality and body weight), neuromotor development (surface righting and negative geotaxis tests) and ultra-vocalizations in response to maternal separation were recorded in offspring as well as maternal behavior (nest-building and retrieving tests) in dams. Results showed significant disturbances depending on the level of contamination, gender and age, in motor reflexes and in the number of vocalizations in exposed pups, whereas no effects on reproductive parameters and in maternal care have been observed. Concentrations of the 6 NDL-PCBs being measured in maternal milk and in the pups' brain at PND14 confirm the mother-young transfer of contaminants. This study demonstrated that the exposure to the 6 NDL-PCBs via a food product naturally contaminated at low levels is able to impact neurodevelopmental endpoints in offspring indirectly exposed by the maternal organism. Additional functional and mechanistic analysis conducted as part of the NeuroDeveTox project will be compare to these results to determine the persistence of cognitive troubles and suggest ways of action underlying the early defects.

141. Cross-omics profiling reveals novel adverse outcome pathways in brains of juvenile female mice following dietary exposure to CB-153, BDE-47, HBCD or TCDD

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ABSTRACT

Introduction: Little data exist on the molecular effects in brain of juvenile mice following dietary exposure to the dioxin 2,3,7,8-tetrachlorodibenzodioxin (TCDD) or the non-dioxin-like (NDL) chemicals hexabromocyclododecane (HBCD), 2,2'4,4' - tetrabromodiphenylether (BDE-47). The present study assessed if eating a diet, spiked with any of these persistent organic pollutants (POPs), affects gene and protein expression in the maturing mouse brain. **Methods:** Juvenile female Balb/c mice (22 days of age) were exposed for 28 days to fish-based diets spiked with TCDD, CB-153, BDE-47 or HBCD at doses approximating their respective lowest observed adverse effect levels (LOAEL). To generate an unbiased view on cellular pathways and functions affected in the brain, we measured global gene and protein expression profiles. Biological network analysis was used to concomitantly interpret the transcriptomics and proteomics data obtained. **Results:** It was found that all POPs elicited changes in neural gene and protein expression profiles. Bioinformatics analysis of gene expression data highlighted the importance of the aryl hydrocarbon receptor (AHR) in dioxin toxicity and revealed that zinc regulation in the brain is targeted by TCDD through the AHR. Calcium homeostasis was affected by both TCDD and the NDL chemicals. In contrast to the transcriptomics analysis, the proteomics data did not allow for a clear distinction between dioxin and NDL responses in the juvenile brain but indicated that proteins associated with excitotoxicity were affected in all exposure groups. **Conclusions:** Integrated interpretation of data lead to the conclusion that the POPs investigated in the present study cross the blood brain barrier (BBB) and accumulate in the juvenile brain where they might induce excitotoxic insults by dysregulation of the otherwise tightly controlled homeostasis of calcium and zinc.

142. Assessment of the short-term neurobehavioral toxicity of a perinatal exposure to the HexaBromoCycloDoDecane (HBCDD) alpha-isomer in rats

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ABSTRACT

The present study aimed to investigate the developmental neurotoxicity of an early exposure to alpha-HBCDD through the ingestion of contaminated hen's egg in pregnant and lactating Wistar female rats. Eggs were obtained from hens after 3 months of daily feeding with alpha-HBCDD-contaminated feed at two levels of exposure, resulting in a content of alpha-HBCDD within the eggs of 30 and 100 ng/g of lipids, respectively. Female rats were daily administered p.o. with an appropriate volume of the whole egg calculated from the daily quantity ingested in humans from the day of fertilization (GD0) to the weaning day for pups (PND21). Thus, fetuses and pups were continuously exposed to HBCDD through the dam at levels of 22 and 66 ng/kg body weight/day over a whole 42-day period including both gestation and lactation. Neurobehavioral development of pups was investigated from PND3 to PND 25 using various tasks including the righting reflex (PND3-5), the grasping reflex (PND4-6), the negative geotaxis (PND8-10) and the locomotor coordination test (PND19-21). Ultrasonic vocalizations of pups were also daily recorded from PND4 to PND16. After weaning, spontaneous motor activity and anxiety-related behavior were examined at PND25 in the open-field and in the elevated-plus maze, respectively. Present results showed a significant decrease in body weight of both pups exposed to the lower HBCDD level from PND3 to PND25, whereas the weight of rat pups exposed to 66 ng/kg/day of HBCDD was not different from controls. During the first 3 weeks of life, impairments in the motor maturation of pups were observed in a dose-dependent manner depending on the test, whereas no significant differences were reported between male and female pups. At PND25, the anxiety level of female rats exposed to the lowest dose of HBCDD (22 ng/kg/day) was significantly reduced whereas it remained unchanged in males. No significant variations were measured in rats exposed to the higher level of HBCD (66 ng/kg/day). These results suggest the potent developmental neurotoxicity of an early chronic exposure to the HBCDD alpha-isomer through the ingestion of hen's eggs contaminated with this pollutant and question the long-lasting consequences of this exposure on behavior abilities and brain functioning in adulthood.

143. Safety evaluation of nanomaterials by using in vitro digestive process coupled with intestinal-like epithelium model

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ABSTRACT

Objectives: Food industry is showing interest on nanotechnologies to improve food properties and nutritional value. According to EFSA guidelines, digestive process simulation is an important step to evaluate nanotechnology physico-chemical modifications occurring after exposure to gastro-intestinal fluids. Moreover, reliable in vitro models for nanotechnology hazard identification are also required. Aim of this work is to evaluate absorption efficacy and safety of silver nanoparticles (AgNPs) and zinc oxide nanoparticles (ZnO NPs) as the most used nanomaterials in food sector following EFSA approach. To this end we set up an in vitro digestive process coupled with an intestinal-like epithelium model characterized by polarized enterocytes covered with a mucous layer on the apical side, and follicle-associated epithelial cells as immune component. **Materials and Methods:** AgNPs (35 nm) and ZnO NPs (20 nm) were characterized for size distribution by transmission electron microscopy; chemical purity by inductively coupled plasma mass spectrometry; and stability in biological fluids with dynamic light scattering. Nanomaterials were incubated on reconstructed saliva, gastric fluid and duodenal juice with bile. Size distribution, stability and ion release were evaluated before and after in vitro digestive process. Their toxicity was tested on intestinal-like epithelium model composed of Caco-2, HT-29 and M cells co-culture, before and after in vitro digestion. The impact on trans-epithelial electric resistance, apparent permeability, tight junction integrity was also evaluated. **Results:** AgNPs and ZnO NPs are characterized by high chemical purity and stability in biological fluids. Exposure of AgNPs and ZnO NPs to simulated in vitro digestive process causes dispersion instability and promotes dissolution. Digested nanoparticles are more toxic than pristine ones. AgNPs are more toxic than ZnO NPs and affect trans-epithelial electric resistance, apparent permeability and viability irreversibly. **Conclusions:** Digested nanomaterials show a different hazard profile compared to pristine ones, with digested AgNPs and ZnO NPs are more toxic than undigested particles. The use of appropriate in vitro model coupling digestive process and intestinal-like epithelium provides more realistic data on efficacy (e.g. supplements) and safety of nanomaterials for oral exposure.

144. Reproductive disruption: The impact of bisphenol S on oocyte development

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ABSTRACT

Bisphenols are an essential part of most plastics. The most widespread bisphenol is bisphenol A. BPA mimics the effects of natural estrogens, disrupts hormonal balance and negatively affects female reproduction. These negative changes result in fault cell signalling, cytoskeletal abnormalities and aneuploidy during oogenesis. Therefore, BPA has been replaced by its analogue, bisphenol S (BPS). Only few studies have described the effects of BPS on cell cycle and chromatin organisation, and reported negative results. However, the effects of BPS in mammalian oogenesis, including oocyte maturation, has not been revealed. **Objectives:** Based on these facts we hypothesised the negative effects of BPS on oocyte maturation. The objective of our study was to evaluate BPS treatment of porcine oocytes maturing in vitro, estrogenous receptors (ER) expression and microtubular organisation. **Materials and methods:** Porcine oocytes were matured in vitro for 48 hrs at 39°C. Oocytes were treated with 0.003, 0.3 and 30 µM BPS. Immunocytochemical analysis of alpha/beta-ER and alpha-tubulin was performed. Confocal laser scanning microscopy and image analysis were used for BPS evaluation. The data were statistically analysed by ANOVA and t-test with SAS 9.0 software. **Results:** We demonstrated the negative effects of BPS on oocyte maturation as well as a significant disturbance of this important process. Our results indicated ER expression changes in a dose dependent manner. The negative effects of BPS on maturing oocytes with decreased of ER expression and microtubular arrangement were also detected. **Conclusions:** These findings showed that BPS caused a time and dose-dependent-delay in cell cycle progession by interfering mainly with centrosomal proteins, and to a lesser extent, with microtubules, which ultimately lead to the formation of abnormal spindle and chromosome non-disjunction. Futher studies are needed for better understanding of BPS impact on animal and human reproduction. This project was supported by NAZV QJ1510138 and CIGA 20132035.

145. From farm animals to human's food: effects on cell viability/proliferation and DNA integrity of the antibiotics Monensin, Salinomycin and their metal complexes

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ABSTRACT

Objectives: Due to their anticoccidial and antibacterial activity the polyether ionophore antibiotics Monensin and Salinomycin are widely used in veterinary medicine. Thus, Monensin has been applied for more than 40 years in poultry farming, as well as in the control of coccidiosis in game birds, sheep, and cattle. Profound information on the impact of these compounds and their complexes with trace metals on various aspects of cell viability and behavior will allow us to assess better potential health risk to humans and animals and to avoid the unwanted adverse consequences. Aim. The purpose of our study was to evaluate the influence of these antibiotics and their metal complexes on viability/proliferation and DNA integrity of cultured cells. **Materials and Methods:** The following cell cultures were used as model systems in our study: Permanent cell lines established from cancers in human (A549, MCF-7, HepG2, HeLa, 8MGBA), rat (LSR-SF-SR) and chicken (LSCC-SF-Mc29); Non-tumor human (Lep-3), bovine (MDBK) and murine (BALB/c 3T3) cell lines and primary cultures from healthy chicken embryos, transplantable hepatoma of Zajdela and bone-marrow cells from the same tumor bearing rats and healthy animals. The total of 12 complexes of Monensin (Mg, Ca, Mn, Co, Ni, Zn) and Salinomycin (Ni, Mn, Na, Zn, Co, Cu, K) were tested. The investigations were performed by MTT test, neutral red uptake cytotoxicity assay, crystal violet staining, trypan blue dye exclusion technique, double staining with acridine orange and propidium iodide, single gel electrophoresis at neutral pH, colony-forming method. **Results:** The results obtained have revealed that applied at concentrations of 0.3 – 35 µM for Monensin and its metal complexes and 2.5 nM -25 µM for Salinomycin and its metal complexes for 24, 48 and 72 h the compounds investigated decreased significantly the viability and proliferation of the treated cells as well as their ability to grow in semisolid medium in a time- and concentration-dependent manner. Cytopathological changes (membrane blebbing, nuclear fragmentation, total cell shrinkage) and double stranded DNA damages in the treated cells were also found.

146. Processing of almonds and apricot kernels: new insights on evolution of aflatoxins

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ABSTRACT

Objectives: Almonds and apricot kernels have several health benefits. They are a good source of proteins, fiber, unsaturated fats, vitamins and minerals. They are consumed as raw nuts or as ingredients of derived products. However they can be contaminated by aflatoxins (AFs), well known carcinogenic mycotoxins. We evaluated the evolution of AFs during transformation processes of almonds into traditional Italian products, such as pastries, nougat and syrup which involves

blanching, peeling, cooking, roasting and water infusion. The efficacy of sorting to segregate AFs contaminated apricot kernels was also assessed. **Materials and Methods:** Experiments were conducted on naturally contaminated apricot kernels and almonds inoculated with a toxigenic strain of Aspergillus flavus. A robust and horizontal HPLC-FLD method was optimized and used for AFs determination in all the matrices considered in this study1. **Results:** Blanching processes (by steaming or boiling in water) did not reduce AFs levels in peeled almonds and apricot kernels. Standard roasting conditions produced up to 50% reduction of AFs. During nougat preparation it was observed a 54-70% AFs reduction due to the caramelisation of sugar on almond surface. AFs were instead stable during cooking of pastries. During almond syrup preparation 18-25% of AFs passed in the final syrup and the whole process produced a marked increase of total AFs. This increase was probably due to the water infusion of ground peeled almonds that activates endogenous almond enzymes that release free AFs from masked AFs1. To reduce AFs contamination in apricot kernels we applied the manual colour sorting to peeled kernels and obtained excellent results: 97-99% AFs reduction by removing only discoloured kernels. **Conclusions:** Roasting, caramelisation and manual sorting of peeled nuts were identified as effective processes to reduce AFs in nuts. We observed, for the first time, a marked increase of AFs during almond syrup preparation which suggests the existence of masked AFs never reported before. The separation of discolored apricot kernels is a feasible strategy to remove almost all AFs in contaminated peeled kernels. These information are extremely useful and can be exploited by food producers to improve the safety of nuts and derived products. 1 Zivoli et al. 2014. Effect of Almond Processing on Levels and Distribution of Aflatoxins in Finished Products and By-products. J. Agric. Food Chem. 62: 5707-5715.

Microbiological risk assessment

147. Risks of *Cronobacter* spp. in neonatal intensive care units of selected Malaysian hospitals

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ABSTRACT

The risks of exposure to *Cronobacter* spp. in neonatal intensive care units (NICU), particularly in infant milk preparation lines in three hospitals in Malaysia were determined. The risk of infection in the NICU was determined. Risk Ranger software was used to generate risk estimates and model different processing scenarios. Should good hygiene and sanitation systems during milk preparation be ineffective, the probability of contracting *Cronobacter* spp. infection per day per neonate was 4.29 x 10-1 and the total number of predicted illnesses per year for the target population was 2.96 x 104. The risk rank was 77. It is recommended that good hygiene practices in terms of milk preparation be strictly and consistently followed and monitored by milk handlers in order to ensure that milk prepared in NICUs are safe from harmful pathogens.

148. Vaccination of pigs and lambs against *Toxoplasma gondii* reduces tissue cyst formation; safer meat for human consumption

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ABSTRACT

The protozoan parasite *Toxoplasma gondii (T. gondii)* is a zoonotic pathogen that has the ability to infect all warm blooded animals including humans (Tenter et al 2000). Toxoplasmosis is a major opportunistic disease of immunocompromised patients. It also represents a serious threat during pregnancy, causing severe foetal abnormalities or potentially leads to problems in childhood or later adult life (Koppe et al 1986). Undercooked or raw meat containing infective tissue cysts are a significant source of human infection (Cook et al 2000). The production of *T. gondii* tissue cyst free meat could reduce the risk of human exposure to *T. gondii*. In two different animal studies a group of 23 pigs and 32 lambs were used to determine the efficacy of a commercially available vaccine that protects sheep from abortions caused by *T. gondii*, with an aim to reduce tissue cyst formation. Following vaccination, animals were challenged with oocysts. Subsequently a mouse bioassay, using a variety of porcine tissues, resulted in a 100% survival of mice that received tissues from vaccinated/challenged pigs. While bioassays of tissues from non-vaccinated pigs resulted in a survival rate of 51%. Parasite DNA was also identified in the homogenate used in bioassays from the non-vaccinated/challenged group but not in the vaccinated/challenged pigs. In a similar experiment, *T. gondii* DNA was tested for in the tissues of lambs. Following vaccination and challenge with 100,000 oocysts of the Moredun M4 strain, the parasite was detected at significantly lower levels in heart and skeletal muscle samples from the vaccinated/challenged group (0% and 5.9% respectively), when compared to the non-vaccinated/challenged animals (75% heart, 87.9% skeletal muscle). The results demonstrate that vaccination of pigs and lambs with the S48 attenuated *T. gondii* strain can reduce the formation of tissue cysts, resulting in potentially safer meat for human consumption.

149. A microbial subtyping approach for the identification of food potentially involved in listeriosis in Northern Italy

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ABSTRACT

Objectives: Listeriosis is a relatively rare but life threatening foodborne disease caused by *Listeria monocytogenes*. Molecular microbial subtyping allows to identify genotypes recurring in clinical cases and compare them with food subtypes. The objective of this work was to investigate the existence of dominant genotypes that could be found both in human cases and in food, in order to identify and prioritize appropriate food safety interventions aimed at controlling listeriosis in Lombardy region. **Materials and Methods:** All *L. monocytogenes* isolates collected in the period 2012-2014 from clinical cases (n=73) and food (n=232), were subtyped by Pulsed-Field Gel Electrophoresis (PFGE) according to the PulseNet protocol with *Asc*I enzyme. Subsequently, all human isolates (n=30) that showed a pulsotype correlating with food isolates were analyzed with Multi-locus Sequence Typing (MLST). Sequence Types (STs) were assigned in accordance with the Listeria MLST database (Pasteur Institute, France). **Results:** PFGE analysis of human and food isolates identified a total of 153 AscI pulsotypes, among which were identified 14 AscI clusters of 3 to 77 isolates. MLST showed that the 14 AscI clusters belonged to 13 different STs: ST1, ST5, ST7, ST8, ST9, ST37, ST38, ST101, ST121, ST218, ST288, ST325 and ST398. *L. monocytogenes* were isolated from meat products and preparations (43.2%), fishery products (24.9%), other ready-to-eat products (26.6%) and cheeses (5.3%). Analyzing subtyping data it was possible to observe that genotypes identified most frequently in human cases (ST8, n = 12 and ST37, n = 3) are distributed evenly in different food categories, while rare genotypes seem to be related to specific ecological niches, being identified respectively in meat products (ST7, ST101, ST121 and ST218), fishery products (ST1 and ST288) and cheeses (ST325). **Conclusions:** This preliminary study identified the main genotypes found in the food supply chain, as well as in human cases. It was possible to highlight the existence of genetic profiles recurring in different food categories, probably less adapted to specific ecological niches but mainly responsible in human cases. Conversely, genotypes less frequently identified in clinical cases are observed in specific food categories, suggesting a higher adaptation to peculiar production environments. These findings could be helpful in the epidemiological investigations of listeriosis, which are often challenging.

150. Prevalence and antimicrobial resistance of thermophylic Campylobacters at broilers

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ABSTRACT

Objectives: *Campylobacter* spp. is a leading cause of foodborne diarrhea. The most common source of infection is contaminated food, mainly poultry meat. The uncontrolled use of veterinary drugs in animal breeding has been suggested as main reason for antimicrobial resistance in human isolates of this foodborne pathogen. **Materials and methods:** In this study 136 samples of cloacal swabs from two broiler farms were tested for *Campylobacter* spp. prevalence, for the first time in Republic of Macedonia. The antimicrobial resistance of all *Campylobacter* isolates was determined for the following antimicrobial agents: erythromycin, ciprofloxacin, tetracycline, streptomycin, gentamicin and nalidixic acid. **Results:** *Campylobacter* was present in 77.9% of chickens. A total of 106 isolates were recovered, with *C. jejuni* being the predominant species with prevalence of 52.2%. *C. coli* showed prevalence of 21.3%. *C. lari* 3.7% and *C. upsaliensis* 0.7%. Antibiotic resistance tests performed by disk diffusion assay indicated that most C. *jejuni* isolates were resistant to nalidixic acid (70.4%), followed by ciprofloxacin (42.2%) and tetracycline (42.2%). Considerable lower antimicrobial resistance was detected towards erythromycin, streptomycin and gentamicin. Comparable results for antimicrobial resistance were recovered from the C. coli isolates. Highest resistance was detected towards nalidixic acid (72.4%), ciprofloxacin (58.6%) and tetracycline (58.6%). Only a small number of isolates of C. coli showed resistance to streptomycin and erythromycin. No resistance to gentamicin was identified in all *C. coli* isolates. Multidrug resistance to at least three antibiotics was seen in 8.4% of *C. jejuni* and 37.7% of *C. coli* isolates. **Conclusions:** Overall, higher antimicrobial resistance was detected to quinolones and tetracyclines and lower resistance towards macrolides and amino glycosides. Observing the results it can be noticed that *C. jejuni* and *C. coli* showed low resistance to erythromycin and gentamicin. The results emphasize the higher resistance of *C. coli* versus *C. jejuni* to ciprofloxacin, tetracycline, streptomycin and erythromycin.

151. The epidemiology and control of *Salmonella* in the pork production chain: the approach in a high prevalence country (Spain)

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ABSTRACT

Objectives: The aim of the study was to obtain relevant insights of the epidemiology of the infection through the pig production chain (breeders, finishers and post-farm stages), in order to approach the control of *Salmonella* in a highly prevalent country such as Spain. **Materials and Methods:** -Breeders: Cross-sectional study to determine the bacteriological and serological prevalence as well as the risk factors associated to the infection in sows. - Finishers: Considering the high prevalence determined by previous studies, we evaluated the efficacy of two control strategies on highly infected farms. First the administration of organic acids ((1) a mixture of lactic-propionic-formic-acetic 0.035% in drinking water and (2) potassium-diformate 0.5% in feed) and second the usefulness inactivated-vaccine (*S.*Typhimurium). - Post-farm: Study of the epidemiology of the infection in transport, lairage, and slaughtering. **Results:** - Breeders: The study performed in 309 herds evidenced, by serology and bacteriology, the presence of *Salmonella* in 60% of the herds. Pelleted feed and individual housing were linked to *Salmonella*-infection, while slatted floor was pointed out as protective factor. - Finishers: The use of organic-acids during the last 6 weeks of finishing reduced the risk of finding seropositive pigs and faecal shedding at the end of the treatment, regardless of the acid used or the administration (water/feed). The protection conferred by a *S.* Typhimurium-inactivated vaccine was demonstrated by the reduction of shedders (six times compared to control animals), when a homologous infection was established; while no protection was achieved in herds infected by serotypes different from *S.* Typhimurium. - Post-farm: The post-farm studies revealed the high burden of *Salmonella* and the risk of new infections in transport and lairage as well as the significance of manual activities on carcass contamination (the estimation of cross-contamination by molecular typing achieved the 75% of the total *Salmonella* contaminated carcasses). The studies stressed the importance of including these stages in a potential control programme, by the inclusion of improved cleaning protocols and establishment of CCP and GMP within HACCPs. **Conclusions:** An exhaustive evaluation of all the production stages is required to establish the strategy, targets, and actions to control Salmonella. The present summary reflects potential actions across the pig production in a high prevalence country.

152. Monitoring individual cell death using time-lapse microscopy: Application to stochastic modelling of microbial inactivation

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ABSTRACT

Objectives: Individual cell inactivation behavior was assessed and characterized through a direct microscopic method and single cell heterogeneity was evaluated as a source of variability in microbial inactivation. **Materials and Methods:** Single cell inactivation behavior of *Salmonella enterica* ser. Agona was studied using a confocal laser scanning microscope. Direct monitoring of cell death was achieved with the time-lapse method and the use of Propidium Iodide (PI), a fluorescent dye that is permeant only in cells with damaged membrane thus, enabling the discrimination between dead and alive cells. The cells of the pathogen were surface plated on solid laboratory medium, where micro-colonies were formed, and then covered with the inactivation solution (26%w/w NaCl) containing PI. A sequence of frames for the selected micro-colony with time was obtained allowing the monitoring of each cell inactivation. **Results:** An image analysis program was used in order to estimate the percentage of cell surface that is covered with PI for a total of 250 cells. The coverage data of each cell with time were fitted to the modified Gompertz equation and lag phase duration was obtained as individual cell time of inactivation. Individual cell time of death in a micro-colony was found to be highly heterogeneous, indicating single cell inactivation behavior as a source of biological variability in microbial inactivation. Individual cells' inactivation times were fitted to a variety of continuous distributions, using @ RISK 6.1 software. The best fitted distribution (LogLogistic) was further used to predict the inactivation of small *S.* Agona populations with the aid of Monte Carlo simulation, with the number of iterations being equal to initial population and the number of simulations representing the variability of the population inactivation behavior. For small populations, the D-value used in deterministic inactivation models was found to be better characterized by a probability distribution rather than a uniform value. **Conclusions:** Individual cell time of inactivation is highly heterogeneous. Single cell inactivation behavior is a source of biological variability in microbial inactivation. We acknowledge the action THALIS: 'Biological Investigation Of the Forces that Influence the Life of pathogens having as Mission to Survive in various Lifestyles; BIOFILM

153. Raw milk sold by vending machines in Piedmont, Italy: microbiological evaluation over 4-year period (2011–2014)

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ABSTRACT

Objectives: The sale of raw milk for human consumption through vending machines is permitted in some EU Countries and Italy has the largest number (1,066 vending machines in 2013). Although the Italian legislation enforces the consumers to boil raw milk before consumption, in Piedmont a monitoring plan on vending machines is defined, according to the Italian legislation which set out microbiological criteria on raw milk. The aim of this study is to verify the microbiological compliance to the Italian legislation in raw milk sold by vending machines in Piedmont from 2011 to 2014. **Materials and methods:** Raw milk was collected by the Local Veterinary Service and delivered to the Food Control Laboratory of the Istituto Zooprofilattico Sperimentale del Piemonte, Liguria e Valle d'Aosta. Each sample was tested to detect *Salmonella* spp., *L. monocytogenes*, *E. coli* O:157, *Campylobacter spp.*, and to enumerate Coagulase-Positive *Staphylococci* (CPS), using accredited procedures; identification of *S. aureus* was performed in case of CPS positivity. **Results:** In 2011, 222 samples from 145 vending machines were analysed; in 2012, 254 from 147; in 2013, 246 from 132; in 2014, 204 from 113. In 2011, 12 samples (5.41%) were not compliant (4 samples for *L. monocytogenes*, 4 for *Campylobacter jejuni*, 4 for *S. aureus*); 7 (2.76%) in 2012 (6 for S. aureus and 1 for *Salmonella* spp.); 5 (2.03%) in 2013 (1 for L. monocytogenes, 2 for *Campylobacter jejuni*, 2 for *S. aureus*); 4 (1.96%) in 2014 (1 for *L. monocytogenes* and 3 for *S. aureus*). **Conclusions:** Results showed that the number of positive samples decreased during the period, as well as the number of vending machines; that is probably due to the producer awareness relative to the complexity of the management and the high level of safety required for this matrix. *Campylobacter jejuni* was one of the most frequently pathogenic microorganism detected, in accordance to the human data from the EFSA report on zoonoses and foodborne outbreaks (EFSA Journal 2015, 13:3991 [162 pp.]). Considering the presence of *S. aureus*, the number of positive samples remains constant during the considered period, which suggests the need of more attention in herd management. Data on consumers' habits confirm that a lot of people do not boil raw milk before consumption and use inadequate bottle to transport it. Thus, considering the microbiological hazard related to this practise, improved risk communication to consumers is recommended.

154. A structured path for the foodborne outbreak investigation

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ABSTRACT

Objectives: Define a structured path for coordinating the different national and regional agencies involved in the foodborne disease outbreak management. **Materials and Methods:** Through the integration of knowledge of different partners, the research group was able to define an investigation standard protocol to apply in case of suspected foodborne disease. The operating guideline, produced by the Foodborne Disease Working Group, together with the epidemiologic and diagnostic survey protocol studied by the research group have been integrated in the, standard investigation protocol. The research tools developed have been applied when a suspected foodborne outbreak have been served during the research period. Traditional diagnostic methods were integrated with molecular biology-based methods, like Duplex PCR end-point, PCR end-point, Pulsed field gel electrophoresis (PFGE) and Spa-typing. **Results:** The comparison between the two genetic typing methods showed both their agreement and their usefulness for defining the origin of the infection. However the Spa-typing method performed better for rapidity of execution and reproducibility. The revision of the Informatic System (IS) of the Istituto Zooprofilattico Sperimentale del Piemonte Liguria e Valle d'Aosta (IZSPLV) highlighted some gaps in the recording process. The performance of the IS was improved increasing the number and the quality of the information uploaded. The results of the research project were disseminated trough the organization of different national courses. The guidelines that have been produced are a tool to be used whenever there is a suspicion of an outbreak of MTA. **Conclusions:** More detailed surveys are necessary nowadays to take into account the increased number of factors that influence the complex pathway from farm to fork. Using the foodborne outbreak investigation guidelines epidemiologist can most likely to reveal the food carrier of infection. In addition, using techniques of molecular epidemiology, investigators are able to go back with a very high level of confidence to the strain responsible, and if there were, the eventual bearers and infectives.

155. Bivalve molluscs: a link between environment and food safety

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ABSTRACT

Objectives: The Blue Growth initiative launched by FAO to promote aquaculture in a responsible and sustainable manner must be in line with the WHO's twelfth programme of work outcome on food safety, to ensure that all countries must control production to prevent and mitigate risks to food safety. Bivalve molluscs, as filter-feeding organisms, can concentrate many contaminants: for this reason the bivalve molluscs safety control can be a model on how it is necessary to consider environmental problems to produce safely in a global food security view. Aim of this work was to address the link between environmental microbiological risk factors and the safety of shellfish. **Materials and methods:** Clams (*Chamelea gallina*) production areas located on the Marche region coast (Italy) were studied. For the spatial analysis and statistical evaluation, we used the 2007-2012 microbiological results of *E. coli*. On the basis of the bibliography we investigated the following risk factors of *E.coli* contamination: waste water treatment plant characteristic, the potential trophic load, the presence of rivers, the rainfall and the production data about animal farms. By using GIS we linked each production area with its own catchment area, to obtain the corresponding risk factors and to create a dataset available for statistic evaluation. For descriptive analysis and statistical models, non parametric statistics and mixed effects models were used. All analysis were conducted by using Stata®11.1. statistical software package. Maps were done by using Map Info Professional Version 7.5®G.I.S statistical software package. **Results:** Thematic maps show higher level of *E.coli* contamination in correspondence with higher level of animal and human waste. By statistical analysis we found that considering the potential trophic load as a categorical variable and taking as reference the first quartile of the distribution there is an increment of the fourth quartile (0.40 95% CI: 0.08- 0.71). Furthermore the presence of rivers (0.06; 95% CI: 0.002 – 0.13) is also a risk factor and there is a positive linear trend of rainfall (0.10; 95% CI: 0.07- 0.13). Considering served treated load as a continuous variable, there is a negative linear trend (- .051 95% CI: -. 097 -.006). **Conclusion:** This study confirms the necessity to consider environment factors in a global view based on risk assessment for a food safety spatial planning Blue Growth purpose.

156. Genomes analysis as a tool for risk assessment of bacteria used in the food chain

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ABSTRACT

Objectives: Recent improvements in the quality, efficiency and cost of next generation sequencing (NGS) technologies have led to a revolution in the study of a great number of bacterial genomes. Aim of this work is to highlight how NGS might provide a tool for deeper understanding of bacteria intentionally introduced in the food chain and that require an authorization by risk managers. To reach this goal, whole genome sequencing together with the sequence comparison among different strains was applied in the risk assessment process, highlighting the presence of putative genes involved in

virulence and antimicrobial resistances. **Materials and Methods:** A total of 22 strains belonging to different species of lactic acid bacteria (LAB) used as human and animal probiotics, silage and food starter cultures, belonging to the species *Lactobacillus rhamnosus, L. plantarum, L. helveticus, L. casei, Streptococcus thermophilus, Lactococcus lactis, Staphylococcus epidermidis and Enterococcus faecium*, were subjected to genome analysis. Sequencing was made using an Illumina Genome Analyzer Hiseq1000. Quality reader filter and assembly was performed using Velvet 1.2.08. Functional annotations were performed with Manatee and RAST. KEGG was used to reconstruct biochemical pathways. Antimicrobial resistance genes and genes coding for putative virulence factors were detected using ResFinder 2.0, PatogenFinder, VirulenceFinder, VFDB and MvirDB databases. Phylogenomic approaches were used to assign enterococcal strain to the commensal or clinical clade. **Results:** Strains of LAB species belonging or not to the EFSA QPS list were studied. NGS technique provided relevant information on these bacterial strains. Data deriving from genome annotation showed a great variability in the genome sizes and number of genes among the analyzed species. The comparative sequence analysis added new insights in themetabolic pathways and safety features. Genes coding for antibiotic resistances were detected in strains of *Lactobacillus* and *Enterococcus* and virulence factors in strain of *E. faecium* and *Staphylococcus epidermidis*. **Conclusions:** The interpretation of the sequence information constitutes an essential approach to perform the risk assessment of strains intentionally introduced in the food chain. Moreover it allows the study bacterial biodiversity, genetic drift, to reconstruct the biochemical pathways and to analyze the phylogenetic relationship among strains and species.

157. Microbiological risk of farmed edible insects: a conceptual framework

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ABSTRACT

Objectives: Edible insects are currently classified as Novel Food in the European legislation. The interest for this alternative protein source is growing worldwide as also in Europe. In the scientific literature several aspects linked to insect consumption have been widely explored, whereas food safety aspects have been scarcely considered. This lead to a paradoxical situation as legal approval is tied to the availability of scientific data, which are largely missing in published literature. For this reason risk assessors are 'waiting for evidence' and not, unfortunately, 'weighting existing evidence'. Aim of this work is to identify data gaps in the context of a conceptual microbiological risk assessment model. **Materials and methods:** A conceptual framework was drawn, based on a hypothetical sequence of stages. Data gaps regarding the microbiological safety of edible insects with a 'farm to fork approach' have been highlighted through an exercise aimed at identifying and qualitatively weighting the amount of available evidence for each specific stage. **Results:** The conceptual framework highlights data gaps in the field of microbiological risk assessment of edible insects. Knowledge about the pathogenicity of intrinsic bacteria of insects and about the vertical transmission of pathogens in insect farms has been judged as almost completely lacking. Moreover knowledge about persistence of extrinsic pathogens and horizontal transmission within insect farms should be implemented. Other relevant data regard the study of potential transfer of foodborne pathogens between farming and processing stages **Conclusions:** Published data on the microbiological hazards of edible insects are few and generally not belonging to ad hoc studies. For this reason evidence based risk assessment models are affected by a lack of relevant data and strongly rely on expert opinion. Reasons behind the scarcity of data could be either the safe and easily controlled production chain or the limited attention of the scientific community to this topic, as absence of evidence not necessarily indicates evidence of absence. The situation gets complicated by the existence of a high number of insect species candidate for human consumption. Our conceptual framework suggests a potential way to identify data gaps. Research should take advantage of the existing insect producing facilities and stimulate a collaborative effort to produce the data needed to satisfy risk analysis requirements.

158. Relative risk of *E. coli* O157:H7 illness using visual inspection to determine doneness of hamburgers packed in Modified atmosphere

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ABSTRACT

Objectives: Risks of illness associated with different consumer preferences were evaluated for Modified Atmosphere Packaging (MAP) hamburgers using two different scores to evaluate doneness based on visual inspection. The increased relative risk of illness due to premature browning (PMB) of MAP hamburgers was estimated. **Materials and methods:** At the last consume-by-day, hamburgers made from minced meat packaged in 80/20 CO2/O2 (MAP hamburger) and from meat minced at retail (control hamburger) were inoculated with a gfp-tagged strain of *E. coli* O157:H7. Hamburgers were heated for different times and cut in halves after heating. Doneness was evaluated based on visual judgement of the internal colour using a colour score chart (C-score) from 'uncooked' (score 1) to 'well done' (score 5). An alternative score chart (TCC-score) including texture of the meat, clarity of meat juice and internal colour was also used. Enumeration of viable *E. coli* O157:H7 in heated hamburgers were based on fluorescent colonies recovered from plates. **Results:** Based on internal colour (C-score) MAP hamburgers developed PMB when compared with controls (P=0.0003). The use of TCC-score instead of C-score reduced the difference between MAP and control hamburgers. The predicted absolute risks for illness were highest for MAP hamburgers for all C- and TCC–scores, and the relative risk of PMB increased with doneness. For a C-score of 4 (slightly pink) the predicted relative risks for illness was 300 times higher for MAP hamburger than for controls. The TCC-score reduced the relative risk for illness between the MAP and control hamburgers. The relative risk for illness in MAP hamburgers was 14 times higher for a C-score of 5 (well done) compared with the same TCC score. There was no difference in relative risk for illness when comparing the controls. **Conclusions:** Efforts to inform consumers about PMB in minced meat packaged in high oxygen packages are needed with the aim to make consumers use thermometers correctly or at least not determine doneness based only on meat colour.

159. Association of *Campylobacter* colonisation and welfare parameters in different broiler systems

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ABSTRACT

Campylobacter, the most prevalent foodborne human bacterial pathogen, is not an essential component of the normal avian intestinal microbiota. Infection of chicken with *Campylobacter* does not result in gross pathologic lesions despite the high colonisation levels. However, the fact that broiler chicken may mount an inflammatory immune response to infection indicates something other than commensalism. In this study the association between *Campylobacter* colonisation and the level of house mortality, rejections, pododermatitis and hock marks was examined in four different commercial production systems (with different welfare standards and bird breed). We report the results of the univariate analysis of indicators of poor welfare and colonisation status. Among the 76 batches sampled 46 (61%) were colonised by *Campylobacter*. *Campylobacter* infection was associated with an increased prevalence of mortality (2.29 v 1.64%), carcass rejections (0.49 v 0.38%), pododermatitis (20.20 v 9.68%) and hock burns (8.40 v 6.64%) ($p<0.01$). The presence of *Campylobacter* was significantly associated with the prevalence of pododermatitis and hock burns in both faster and slower growing breeds ($p<0.001$). However, it was noted that *Campylobacter* infection had a stronger association with pododermatitis and hock burns in the faster growing breed. These results show *Campylobacter* colonisation is positively associated with welfare associated pathologies. Further research is needed to investigate the mechanisms that link *Campylobacter* infection with the health and welfare of the birds including gut health and bird immune status under production stress. Our results indicate

that there is a combined benefit for public health and animal welfare in the control of *Campylobacter* infection.

160. Comparing different molecular typing methods of *Listeria monocytogenes* isolates from human and food circulating in Italy

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ABSTRACT

Objectives: Listeriosis is a severe food-borne disease causing every year over 1600 cases in European Union. Attribution to sources of human food-borne illness is crucial in order to establish preventive strategies, and it can be achieved using different laboratory methods. Pulsed-field gel electrophoresis (PFGE) is considered the 'gold standard' for source attribution of *Listeria monocytogenes,* but Multi Locus Sequence Typing (MLST) and Whole Genome Sequencing (WGS) are considered more discriminant methods. The aim of this work is to compare molecular typing results from different methods applied on human and food isolates, in order to highlight homologies for source attribution. **Materials and methods:** In this study 115 isolates from clinical cases in Northern Italy hospitals and isolates from foodstuffs and food processing environments sampled in Italy in the framework of official controls and surveillance programs from 2003 to 2014 have been collected and characterized by PFGE, MLST and WGS. Preliminary results related to 3 isolates of *Listeria monocytogenes* from clinical cases and 5 from foodstuffs have been obtained so far. The isolates were characterized by PFGE (CDC protocol 2013) and 7-locus MLST, while WGS of the isolates was performed using an Ion Torrent PGM platform. The Genome-wide Single Nucleotide Polymorphism (SNP) analysis was carried on using the FDA snp-pipeline program, a SNP reference-based matrix was produced and a UPGMA phylogenetic tree was created. **Results:** The isolates from clinical cases were characterized as belonging to 3 different serotypes, pulsotypes and sequence types. Three sequenced isolates from foodstuff and food processing areas shared the same PFGE profile and MLST sequence type (ST3) with one isolate from clinical case. Phylogenetic relationships showed that the selected isolates are similar but according to SNP analysis only one food isolate is very close to the human one. **Conclusions:** This study has confirmed the high discriminatory power of WGS in comparison to other molecular typing methods. More results will be obtained from this study analysing other isolates currently available. In the future, large scale sampling and WGS analysis of food and human isolates could increase our capability to identify *L. monocytogenes* clusters, improving surveillance systems.

161. Improving knowledge of human norovirus in the marine environment to reduce the incidence of shellfish-related illness

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ABSTRACT

Human norovirus (NoV) is an important cause of infectious intestinal disease linked to consumption of raw shellfish, particularly oysters. The UK Food Standards Agency Foodborne Disease Strategy 2012/15 identified that further evidence on the fate of NoV in the marine environment is required before a risk management programme for this pathogen can be developed. This evidence is critical since it has been emphasised in EFSA Scientific Opinions that effective risk management must focus on prevention of contamination in shellfish production areas (SPA) because post-harvest purification treatments have not prevented the occurrence of NoV outbreaks. To address this knowledge gap, field experiments have been conducted to investigate the characteristics of NoV contamination in two SPA and adjacent hydrological catchments in England. The experiments have provided information on the relationships between NoV contamination in three species of shellfish (Pacific oysters, native oysters, mussels) and environmental variables (rainfall, degree of sewage dilution and dispersion) and typical concentrations of NoV in sewage effluents subject to different treatment processes. This evidence has been used to develop risk management options such as prohibition of shellfish harvesting near inputs of human contamination, management of harvesting to avoid periods of high risk of contamination and development of monitoring programmes for NoV. Modelling of the relationships between NoV and the current legislative faecal pollution indicator (*E. coli*) has also been used to develop regulatory controls to better mitigate the NoV risk. This research will help inform recommendations for applying the NoV risk management measures across the UK.

162. Hazard identification of *Arcobacter* spp. in bivalve molluscs collected from the central Adriatic Sea, Italy

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ABSTRACT

Arcobacter spp. belong to the family *Campylobacteraceae* and are present in the intestine of livestock animals which may act as reservoirs of these bacteria. *Arcobacter* spp. have been associated with cases of human gastrointestinal disease. Contaminated food and water may represent possible routes of transmission. *A. butzleri* is the most commonly recognised species for human disease, followed by *A. cryaerophilus*. This work aimed to investigate the presence of *Arcobacter* spp. in bivalve molluscs collected from harvesting areas of the central Adriatic Sea and study the correlation with levels of *Escherichia coli*, the bacterial indicator of faecal contamination of molluscan shellfish (Reg. (EC) N. 854/2004, 2073/2005). 123 samples of bivalve molluscs (35 mussels and 88 clams) were collected between Dec 2012 and Jul 2014 and analyzed by a culturing method for detection of Arcobacter spp. 71 and 52 samples were from class B harvesting areas (post-harvest treatment required) and A (postharvest treatment not required), respectively. Presumptive colonies were tested by PCR for the rpoB gene of *Arcobacter* spp. and species identification was performed by DNA sequencing analysis of the rpoB gene. 47 (38.2%) of the 123 samples were positive for *Arcobacter* spp.: 37/88 (42%) of clams; 10/35 (28.6%) of mussels. Sequencing analysis identified *A. butzleri*, *A. cryaerophilus* and *A. skirrowii* in 32 (68%), 14 (29.8%) and 1 (2.1%) of the 47 positive samples. 38 (80.9%) of the 47 positive samples for *Arcobacter* spp. were from class B areas or A areas closed for the level of *E. coli* >230 MPN/100g. *A. butzleri* was significantly more common among clams of class B areas with *E. coli* > 230 MPN/100g than in clams with *E. coli* < 230 MPN/100g ($\chi 2 = 5.22$; $P = 0.0224$). In agreement with other studies, *A. butzleri* was the most prevalent species identified in shellfish. A significant correlation between the indicator of faecal contamination *E. coli* above 230 MPN/100g and A. *butzleri* presence was found. For shellfish from class B areas post-harvest treatment as purification is required, until *E. coli* concentration decreases to levels below 230 MPN/100g. Previous experimental studies performed in our laboratory demonstrated that *A. butzleri* had a similar behaviour to *E. coli* in bioaccumulation experiments in mussels, providing evidence on *E. coli* as indicator of the hazard posed by A. butzleri.

163. Microbiological evaluation of cantaloupe packinghouses in Florida, USA, over two years

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ABSTRACT

In 2011 and 2012, US outbreaks of listeriosis and salmonellosis linked to consumption of cantaloupe melons were traced to unsanitary conditions in cantaloupe packinghouses. The purpose of this study was to evaluate the sanitary conditions of cantaloupe packinghouses in Florida, USA, over two years. In 2013, 5 cantaloupe packinghouses (facility A-D) were visited; in 2014, 2 cantaloupe packinghouses (B and C) were visited. Up to 60 swabs per facility were collected from food contact and non-contact surfaces, and water. Swabs were enumerated for total plate counts (TPC), generic *E. coli*, and coliforms, and enriched for *Listeria* spp. and *L. monocytogenes* by standard methods.

Presumptive *L. monocytogenes* colonies were confirmed by amplification of the sigB gene by PCR and analyzed by PFGE. In 2013, 270 swabs and 1 water sample (172 zone 1 food contact swabs) were collected; in 2014, 93 swabs (61 zone 1) were collected. In 2013, average zone 1 TPC for all facilities was 5.54±1.40 log CFU/swab; TPC's ranged from 5.21±1.22 (facility E) to 6.05±1.46 (facility C) log CFU/swab. In 2014, the TPC for zone 1 in facilities B and C were 4.31±0.76 and 5.33±1.04 log CFU/swab, respectively. Coliforms and *Listeria* spp. were recovered from 36.6% and 73.2% of zone 1 swabs from all facilities samples in 2013, ranging between 27.8% (facility A)-47.1% (facility D) and 50.0% (facility D)-86.5.4% (facility A), respectively. In 2014, coliforms were detected in 14.8% of zone 1 swabs in facility B and 32.4% of zone 1 swabs in facility C; *Listeria* spp. were recovered from 74.1% and 82.4% of zone 1 swabs in facilities B and C respectively. In 2013, *E. coli* was not recovered from any facility (limit of detection 50 CFU/swab); in 2014 was recovered from 5.3% and 3.6% of swabs from facilities B and C, respectively. Sanitation managers were advised to clean and sanitize their packing lines following the detection of *E. coli. Listeria monocytogenes* was recovered in 2013 from 2/270 swabs (0.7%); both isolates were recovered from facility B on food contact surfaces. PFGE patterns of the two isolates were identical. After recovery of *L. monocytogenes*, the sanitation manager was advised to clean and sanitize their packing line; *L. monocytogenes* was not detected upon retesting of the facility following sanitation. The prevalence of *L. monocytogenes* in Florida cantaloupe packinghouses was low; TPC, coliforms, *E. coli* and *Listeria* spp. were not good indicators of L. *monocytogenes*.

164. Performance Objectives for *Bacillus cereus* in RTE salads: a whole food chain approach

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ABSTRACT

The Performance Objective (PO) is a risk management concept we should became familiar with in the next future. The aim of this study was to derive POs for *Bacillus cereus* (BC) in spelt and cheese to be added as ingredients in RTE mixed spelt salads, packaged under air or modified atmosphere, with a shelf life of 12 days. This RTE product was selected as model product because the majority of reported BC emetic intoxications are linked to the consumption of heat treated dishes containing farinaceous ingredients. To derive the POs enumeration results and censored data were considered. Moreover, the most probable distribution of censored data was determined and Monte Carlo simulations performed to assess the uncertainty. In the simulation the authors assumed no cross contamination and that spelt salad contamination may arise only from spelt and cheese. The PO values were calculated in order to meet a Food Safety Objective (FSO) fixed, according to the literature, at 4 Log cfu of BC for each g of spelt salads at the time of consumption. The values obtained using the simulated initial contaminations, in which as input there was a single lognormal distribution, were close to the values estimated by ComBase predictors (inactivation and growth) and used as parameters in the simulation, due to absence of uncertainty effects. The POs were derived by interpolating the proposed FSO in the regression obtained by representing the simulated contamination levels versus the contamination level at time of consumption, which was the output of the model. The PO values for BC in spelt to be added in spelt salads stored refrigerated under air or MAP for 12 days were -0.64 and 0.22 Log cfu/g, respectively. The PO values for BC in cheese to be added in the same product stored refrigerated under air or MAP for 12 days were -2.22 and -1.36 Log cfu/g, respectively. The verification of the calculated POs should be done using a detection method including an enrichment step or molecular methods with extremely low detection and quantification levels. The approach presented in this study can be easily adapted to different FSOs and changing assumptions. The use of spelt and cheese compliant with the suggested POs might impact the incidence of foodborne intoxications due to BC and the proportion of food recalls, causing huge economic losses to food companies commercializing RTE products. This study was funded grant agreement 289262, STARTEC.

165. Prevalence and genetic characterization of *Toxoplasma gondii* in pigs from Northern Italy and its implication for consumers

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ABSTRACT

Objectives: Toxoplasmosis is a worldwide zoonosis. The consumption of raw or undercooked pork and products thereof such as fresh sausages containing *T. gondii* cysts may be a source of toxoplasmosis in humans. In Nord America and Europe, *T. gondii* isolates can be classified into 3 main clonal Types (I, II, III). Type I is highly virulent to out-bred mice and Type II and III strains are significantly less virulent. Currently, there are few data on both the occurrence of *T. gondii* in pigs destined to be transformed into high value Italian food products and its genotypes circulating in Italy. The aim of this study was to evaluate the occurrence of *T. gondii* and genetically characterize the isolates in industrial pigs slaughtered in Northern Italy. **Materials and Methods:** From January to June 2014, blood, diaphragm and heart samples were collected from 505 slaughtered pigs (large white or hybrids, 9-month-old, ≈160 Kg of weight) from 73 farms located in Lombardy and Emilia Romagna. Sera were examined for IgG antibodies to *T. gondii* by modified agglutination test and titers ≥1:10 were considered positive; tissue samples (≈25 gr) from seropositive pigs were processed for the presence of *T. gondii* DNA (B1 locus) by EvaGreen® RealTime PCR and HRM analysis. **Results:** Anti-*Toxoplasma* antibodies were detected in 1.6% of pigs with titers from 1:10 to 1:320. *T. gondii* DNA was found in all seropositive animals and in 12/18 muscular samples. Types III, II, or both were identified in 5, 2, and 1 subjects, respectively. **Conclusions:** This is the largest epidemiological study on *T. gondii* in pigs in Italy; less pathogenic *Toxoplasma* types (Type II and III) were found by high sensitive and rapid molecular techniques (RealTime + HRM). The low prevalence combined with the finding of less virulent strains show that industrial pigs play a limited role in the epidemiology of *T. gondii*. The extent of the infection in pigs depends on the management system. Industrial pigs are unlikely to ingest rodents harboring tissue cysts and food or water contaminated with sporulated oocysts. Our results also suggest that the consumption of cured products prepared from these animals represents a very low risk to consumers of acquiring toxoplasmosis. Indeed, when pork is processed for curing (salting, drying, smoking), it is unlikely that *T. gondii* survives the processing procedures.

166. The discriminative power of phenotypic and genotypic typing methods on *Campylobacter*

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ABSTRACT

Objectives: The aim is to compare phenotypic and genotypic typing methods, individually and combined, using *Campylobacter* isolates collected from broiler carcasses to gather information on the typing methods' discriminatory power and congruence. **Material & Methods:** *C. jejuni* (n=94) and *C. coli* (n=52) isolated from different broiler carcasses sampled in Belgium slaughterhouses (2011-2013) were characterized using different typing methods: multi-locus sequence typing (MLST) using the Miller et al. primers (2005); antibiotic microbiological resistance (AMR) determination for 7 antibiotics via a micro-broth dilution method (EUCAMP-Sensititre Trek); presence/absence of 5 putative virulence genes; and exclusively for *C. jejuni*, determination of lipooligosaccharides (LOS) class and the presence/absence of the LOS locus. Discriminatory power was calculated by the Simpson's index and the congruence measurement by the Wallace coefficient. **Results:** The predominant MLST clonal complex (CC) for *C. coli* was CC-828 (84%), which also contained the predominant sequence type (ST) ST-854 with 13% of the isolates. For *C. jejuni*, 21% of the isolates could

not be assigned to a CC; the most frequent CCs were CC-21 and CC-45, with 20 and 11% of the isolates, respectively. However, the most frequent STs, ST-464 and ST-5970, each representing 6% of the isolates, belonged respectively to CC-464 and to an unassigned CC. The presence of all 5 putative virulence genes was the most frequent virulence profile (*C. jejuni* 95% and *C. coli* 65%). The combined resistance to ciprofloxacin, nalidixic acid and tetracycline was the most frequent profile on the two tested species: 26% for *C. jejuni* and 30% for *C. coli*. LOS class A was the only LOS class absent from the tested *C. jejuni* isolates. MLST ST was the most discriminative typing method in both *C. jejuni* and *C. coli* (0.981 and 0.957). For MLST CC, the discrimination was higher for *C. jejuni* than C.coli (0.890 vs 0.265). MLST ST combined with AMR profiling was the most discriminative combination in both tested species: 0.988 for C. jejuni and 0.957 for *C. coli*. In general, lower congruence coefficients were obtained when MLST ST typing was the 2nd method. On the other hand, when MLST ST was the 1st method, the 2nd one with a lower Wallace coefficient that gave more additional information was the AMR (0.506). **Conclusions:** Individually, the typing by MLST ST showed the highest discrimination. This discrimination increased when the AMR was combined.

167. Antibiotic resistance of lactobacilli isolated from rainbow trout (*Oncorhynchus mykkis*, Walbaum)

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ABSTRACT

Objectives: To isolate and identify lactic acid bacterial (LAB) strains from rainbow trout (Oncorhynchus mykkis, Walbaum) and subsequently perform antibiotic susceptibility test for the LAB strains on the basis of Guidance on the assessment of bacterial susceptibility to antimicrobials of human and veterinary importance by the EFSA panel on additives and products or substances used in animal feed (FEEDAP). **Materials and methods:** Sixty samples of rainbow trout were collected from a commercial fish farm in the Slovak Republic. A total of 6 lactic acid bacteria isolates from intestines of rainbow trout belonging to the genera *Lactobacillus* were included in this study and encompassed the following species: *L. plantarum* (n=3), *L. paraplantarum* (n=1), *L. fermentum* (n=1), *L. brevis* (n=1). The identification of these strains were performed by matrix-assisted laser desorption ionization-time-of-flight mass spektrometry (MALDI-TOF MS) (Ultraflex III, Bruker Daltonics). Antimicrobial sensitivity has been tested to 9 antibiotics listed by EFSA (ampicillin, vancomycin, gentamicin, kanamycin, streptomycin, erythromycin, clindamycin, tetracycline and chloramphenicol) and was evaluated using the disc diffusion method and E-test. **Results:** Distribution of resistance was found in different species. All isolates were susceptible to chloramphenicol, tetracycline, ampicillin and erythromycin. In addition, isolates resistant to streptomycin, gentamicin and clindamycin were detected, although the incidence of resistance to these antibiotics was relatively low. In contrast, all the strains were resistant to kanamycin and vancomycin. **Conclusions:** The results show that lactobacilli may be resistant to antimicrobial agents and resistance is a key indicator for selection of suitable probiotic bacteria that will be used in aquaculture. The safety of these probiotic strains is essential, as is the impossibility to transfer their potential resistance to antimicrobial agents to other bacteria, since antibiotic resistance is an emerging issue.

168. Human Sapovirus in mussels from Galicia, Spain

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ABSTRACT

Sapovirus (SaV), from the Caliciviridae family, is an etiological agent of human gastroenteritis classified by the U.S. Environmental Protection Agency as an emergent human pathogen with risk for public health in aquatic environments. Spain is the main aquaculture producer of the European Union, and Galicia the main producer region and one of the most important in the world, especially for bivalve molluscs. Bivalve, due to their filtration feeding, concentrate viral particles presented in seawater that can resist depuration treatments to which bivalves are submitted. In addition, the traditional way of bivalve consumption increases the probability of infection, making them a high risk food. The main objective of this work was the quantification and genetic characterization of SaV in bivalves from Galicia, Spain. A total of 81 mussels samples (Mytilus galloprovincialis) were analyzed from October 2010 to March 2012 from Ría do Burgo, an estuary located close to the city of A Coruña. Sampling points were located in Class B harvesting areas according to the EU legislation. All bivalve samples consisted of 10 mussels which were dissected and processed under aseptic conditions to obtain their digestive tissue. After the viral RNA extraction, detection and quantification of human SaV using RT-qPCR (Reverse Transcriptase - real time Polimerase Chain Reaction) were performed according to the recently developed standard method ISO/TS 15216:2013. Primers SaV124F and SaV124R, and probe SaV124TP designed to amplify human genogroups I, II and IV were used. SaV was detected in 30 of total 81 mussel samples (37.0 %). Most of the positive samples were detected within the cold months of the study (November 2010 – March 2011 and November 2011 – February 2012). The average quantification ranged from 2.2 x 103 copies of viral RNA per gram of digestive tissue (copies/g) to 2.1 x 105 copies/g, with an average 9.4 x 103copies/g. Genotyping studies are currently in progress to characterize genetically the positive samples. It will allow us to compare these results with previously ones obtained in Galicia from other bivalve and clinical samples, and with those from other areas in the world. Finally we will be able to achieve a global vision of the prevalence of SaV in Galicia as an important global exporter of bivalve molluscs, and to determine the risks for food safety.

169. Monitoring virulence potential of *Salmonella enterica* from animal food products

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ABSTRACT

Introduction: *Salmonella* infection is a major foodborne illness worldwide. *Salmonella* Pathogenicity Islands (SPI) are necessary for invasion and infection of host cells. *S.* Typhi produces a cytolethal distending toxin (CDT: CdtB, the active form, PltA and PltB). CdtB arrests cell cycle induces apoptosis and extends persistence of pathogenic bacteria in the host. We assess the virulence potential of infection by *Salmonella* isolated from food products of animal origin. **Methods:** A virulence DNA microarray was developed, and used to assess the virulence potential of *Salmonella* from animal products. Adhesion/invasion assays were performed to *S.* Infantis Sal147 missing several SPI-1 genes. RT-PCR and toxin assays for *S.* Typhimurium Sal199 with cdtB gene were performed in HeLa cells; and flow cytometry was used to check the transition G2/M phase arrest. Whole genome sequencing performed to both strains. The virulence potential was evaluated in vivo with *G. mellonella* model. Representative results were obtained from three independent experiments. Data were expressed as percentage of survival and analyzed by using the statistical package GraphPad Prism 5. **Results:** *S.* Infantis Sal147 missing thirty-eight SPI-1 genes; and *S.* Typhimurium Sal199 expressed cdtB gene and others Typhi-related genes (pltA, pltB, envF, pagC, pagD. taiA, tcfA, and hlyE). cdtB gene together with pltA and pltB genes were inserted in transposase IS911. Tissue cultures showed non-invasion of human-colon cell by Sal147. HeLa cells infected with Sal199 became distended with prominent cell cycle arrest in G2/M phase compared with the control *S.* Typhimurium LT2. Sal199 showed a mean of survival of 10 % (range 0-20 %), while the control LT2 presented a survival rate of 16.7 % (range 10-20 %). **Conclusion:** Microarray results predict atypical phenotype. *S.* Infantis missing several SPI-1 genes was able to adhere but not to invade human cells. *S.* Typhimurium was able to cause cytoplasm and nuclear enlargement, increased invasion, cytotoxicity and mortality of caterpillars. CDT might have been horizontally transferred by a prophage that integrated into the bacterial DNA chromosome. To our knowledge, this is the first report showing that *S.* Typhimurium strains from food sources may contain active CDT genes similar to *S.* Typhi. This study highlights the importance of monitoring food products of animal origin, avoiding worst *Salmonella* infection.

170. Human Listeriosis in Piedmont, Northern Italy (2010-2013): an insight on surveillance efficacy and strain diffusion

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ABSTRACT

Objectives: Listeriosis is a notifiable foodborne disease, with high hospitalization (up to 90%) and mortality rates (20-50%). Still, due to unspecific symptoms and long incubation time, the epidemiology of this disease is often unclear. In 2011, the incidence of listeriosis was 0.31 cases/100.000 inhabitants in Europe, and 0.17 cases/100.000 in Italy, with some Italian regions regularly reporting no cases and others with an incidence of 0.88 cases/100.000. We aimed to assess if the number of reported listeriosis cases in Piedmont is accurate and type the isolates circulating in the same area through genomic subtyping. **Materials and Methods:** Notification rate was assessed conducting a retrospective comparison of notification data with Hospital Dismissal Records (HDRs) listing listeriosis as diagnosis, for the period 2010-2014. Regional hospitals were asked to send *Listeria monocytogenes* clinical isolates for characterization with Multi-Virulence-Locus-Sequence-Typing (MVLST). MVLST results were compared with reference database and a neighbor-joining tree based on the number of nucleotide differences in the gene fragments analyzed was constructed. **Results:** Our analysis showed that on average 40% of cases that listed listeriosis as a diagnosis in the HDRs had not been notified. When adjusted according to HDRs, yearly incidence in Piedmont increased from 0.24 cases/100.000 inhabitants (notification data) to 0.42 cases/100.000 inhabitants. Only 2% of reported cases were pregnancy related. In total, 17 *L. monocytogenes* clinical strains were collected, 41% of them had a genetic profile matching strains involved in previous outbreaks and interestingly, 2 cases had a genetic profile previously recovered only in Piedmont in a cheese production plant. **Conclusions:** Currently there is no systematic clinical *Listeria* strain collection in Piedmont and thus, the collection of even relatively few human isolates is important to start implementing listeriosis surveillance strategies. Overall, the percentage of pregnancy related cases is lower compared to other countries (reporting 10-20%), probably because causes of miscarriage are seldom investigated. Moreover, 40% of human listeriosis cases diagnosed in Piedmont every year are not notified, highlighting the need to raise awareness of notification procedures among medical staff. Stressing the importance of collecting and typing clinical strains, starting at regional level, might be an efficient way to reach this goal.

171. Whole genome analysis as a tool for the safety assessment of antibiotic resistance in food-processing bacteria

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ABSTRACT

Acquisition of antibiotic resistances (AR) by pathogens leads ultimately to a failure of antibiotic therapy. The food chain is considered a key player in the transmission of AR determinants to pathogens from reservoirs in commensal and beneficial bacteria. Therefore, the absence of transmissible AR genes in bacteria used as starter and adjunct cultures for food and feed processing is considered to be critical (EFSA, 2012; EFSA Journal, 10:2740). Objetives: Genome sequencing allows the inspection of the whole genetic makeup of bacteria in the search for the basis of desirable and undesirable traits, including that of AR. **Material and methods:** In silico sequence analysis and comparison against databases can be used as a tool for the safety assessment of microorganisms intended to be used in food systems. **Results:** This communication reports on the genome analysis of three *Leuconostoc mesenteroides* strains of dairy origin showing atypical resistances to tetracycline (LbT16), erythromycin and clindamycin (LbE15), and kanamycin, streptomycin, tetracycline and virginiamycin (LbE16). Genes encoding for erythromycin [*erm*(B)] and tetracycline [*tet*(S)] resistance had already been detected by PCR. Genome analysis confirmed the presence of these genes and identified others which encode uncommon AR in lactic acid bacteria. Analysis of the genes and their flanking regions revealed a potential of some to be horizontally transferred to other bacteria. **Conclusions:** This study demonstrates the effectiveness of combining genome sequencing and bioinformatics analysis as an affordable tool for the safety assessment of food bacteria. This innovative approach could become a novel paradigm in the selection programs of starters for the food industry.

172. Characteristics and survival of *Bacillus cereus* group members in herbs

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ABSTRACT

Introduction: According to EFSA (2013) *Bacillus* spp. and dried herbs and spices are on the top four of the ranking groups of food/pathogen combinations in food of non-animal origin. The aim of our study was to characterize and investigate the survival of *B. cereus* group members (*B. weihenstephanensis*, *B. mycoides*, *B. pseudomycoides*, *B. anthracis*, *B. thuringiensis* and, *B. cereus*) in herbs. **Material and Methods**: The occurrence of presumptive *B. cereus* in oregano, basil and parsley was investigated in accordance with ISO 7932. Species identification and detection of toxin genes was conducted by multiplex real-time PCR, toxin producing capabilities of strains were investigated using the BCET-RPLA Kit and the Duopath GLISA assay, resp. The survival of spores of a *B. thuringiensis* strain was investigated in the three herbs (air dried spore suspension on 0.5 g sand + 4.5 g herb or sand (positive control), final concentration 106 cfu/g). **Results:** Different species of the *B. cereus* group were detectable in the three analysed herbs. Toxin genes known to be associated with diarrhoea in humans (nheA, hblD and cytK) were identified in strains in different combinations. Studies on the toxin production capabilities of these strains are ongoing. First results of the survival study indicate that *B. thuringiensis* spores remain viable in high numbers for several months. **Conclusion:** The microbial flora of herbs can contain potentially toxin producing members of the *B. cereus* group. On the model of *B. thuringiensis* spores it can be assumed that spores have high survival capacities in herbs. More data on the characteristics and pathogenic potential of *B. cereus* group members are needed when assessing microbiological risks related to consumption and processing of herbs. Amendment This research was executed in the framework of the EU-project SPICED (Grant Agreement: 312631) with the financial support from the 7th Framework Programme of the European Union. This publication reflects the views only of the authors, and the European Commission cannot be held responsible for any use which may be made of the information contained therein. For further information on the project please see www.spiced.eu. References EFSA (2013) Scientific Opinion on the risk posed by pathogens in food of non-animal origin. Part 1 EFSA Journal 2013;11(1):3025

173. Antibiotic resistant bacteria in Ready to Eat Foods

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ABSTRACT

Objectives: The Antimicrobial resistance (AMR) is a serious threat to public health worldwide. The surveillance programmes indicate that there is a general world-wide increase of antimicrobial resistance. The wide use of antibiotics in the farm environments acts as selective pressure for associated bacteria and can influence the bacteria growing in fermented food of animal origin. Aim of this work was to assess the risk of antimicrobial resistant bacteria in ready to eat food (RTE), including fermented foods where, due to high counts of viable bacterial cells at the consumption stage, the exposure of the consumer might be the highest. Moreover the horizontal gene exchange among foodborne bacteria was studied. **Materials and Methods:** Food of animal origin, such as raw and pasteurised milk, cheese from raw and pasteurised milk, meat, dry fermented sausages, and cured meat, collected form retail were included in

the study. Microbiological analyses were performed selecting bacteria resistant to clinically and veterinary relevant antibiotics. Resistant strains were identified and the genotype of resistance investigated by molecular approaches. The whole genome sequence of multidrug resistant strains was achieved. The rate of AMR gene exchange in complex microbial communities was assessed in experiments in vitro and in food **Results:** Resistant strains of *Bacillus, Enterococcus, Escherichia, Klebsiella, Lactobacillus, Shigella* and *Staphylococcus* were isolated from RTE foods. Tetracycline, erythromycin and ampicillin resistance were the most spread antimicrobial resistance among Gram- and Gram+ isolated bacteria. The most present determinant found were, *tet*M and *tet*K for tetracycline; *erm*B and *erm*C for erythromycin; *blaZ*, and the resistant variant of *pbp5* gene for ampicillin resistance. Multidrug resistant strains were found. Genome analysis of the these bacteria, showed the presence of some of the AMR determinant on mobile genetic elements. Mating experiments, demonstrated the intra and intergeneric horizontal gene transfer of *tet*M and *erm*B. **Conclusions:** These results indicate that RTE foods are a source of AMR bacteria and that the gene transfer can occur at high frequency during food fermentation. Moreover these data underline the need of deeper understanding of the role of foodborne bacteria in the spreading of AMR.

174. Detection of antibodies anti-*Toxoplasma gondii* in tank bulk milk of caprine dairy herds in Lombardy (Northern Italy)

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ABSTRACT

T. gondii is a major cause of reproductive failure in goats; besides, these species plays an important role in human infection and plans of surveillance are mandatory (Kijlstra and Jongert 2008 Int J Parasitol 38:1359-1370). In the screening of infections within a herd, the analysis on tank bulk milk, easy to obtain and representative of all milking animals, has been investigated and validated for many pathogens (Sekiya et al. 2013 Irish Veterinary Journal 66). Aim of the present survey was to collect epidemiological data on *T. gondii* infection in caprine herd in Northern Italy through the analysis on tank bulk milk. With the collaboration of the Regional Breeders Association of Lombardy (ARAL), 100 samples of tank bulk milk of dairy goats' farms in Lombardy were collected within the control quality program. A commercial ELISA kit (ID Screen® Toxoplasmosis Indirect Multi-Species, IDVET) was validated for analysis on milk using a panel of 30 sera-milk pairs previously tested for *T. gondii* by IFAT. A linear regression analysis on ELISA results of sera-milk samples was performed to obtain the optimal dilution for milk; the agreement between results on sera and milk, analyzed at optimal conditions, was further evaluated (k=0.873). Tank bulk milk samples were therefore analyzed; on ELISA results, a general linear model (GLM) was performed to determine factors that could be considered predictors of the presence of antibodies to *T. gondii* in milk, using as independent variables 'province', 'farm size', 'n° of milking in a day'. Antibodies were detected in 58 out of 100 (58%) of tank bulk milk samples. Small farms resulted at low risk of infection, increasing the risk of infection with the increase of the number of animals in farm (OR=1.001; p=0.014). On the contrary, the increasing of milking in a day correspond to a decreasing of the risk of resulting infected (OR=0.902; p=0.028). Any difference concerning the provenience resulted statistically significant. The present survey confirms *T. gondii* infection as very common among dairy goats' farms in Northern Italy. Analysis on tank bulk milk revealed an affordable tool in the context of a health monitoring program as a first screening, allowing epidemiological surveys on large scale and planning monitoring controls and selection within positive farms.

175. Multidrug resistant *Escherichia coli* from raw chicken meat: when your chicken dinner is hiding a superbug

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ABSTRACT

Objectives: Multidrug resistant (MDR) bacteria, such as extended spectrum β-lactamase (ESBL) producing *Enterobacteriaceae* pose a serious challenge to the human health. In recent years, these MDR bacteria have been detected increasingly outside the hospital setting. Also the contamination of food with MDR bacteria, particularly of meat and meat products, is a concern. Moreover, antibiotic resistance in *Escherichia coli* is especially worrying because it is the most common Gram-negative pathogen in human gut. The aim of this study was to evaluate the occurrence of MDR *E. coli* in chicken meat on sale in Palermo, Italy. **Materials and Methods:** In a prospective survey conducted in Palermo, Italy, during 2013–2014, selective cultures, disk diffusion drug susceptibility assays and PCR–based tests to define antibiotic resistance traits were performed on 250 raw retail chicken meat samples. **Results:** A total of 237 (94.8%) biochemically confirmed isolates of *E. coli* were isolated from a total of 250 raw retail chicken meat. Resistance to at least one antimicrobial agents class was found in all isolates. The predominant resistances were to ciprofloxacin and tetracycline (92.4% and 91.6%, respectively), followed by co-trimoxazole (81%), cefotaxime (78.5%), ceftazidime (74.3%), amoxyclav (62.9%), cefepime (36.3%), gentamicin (17.7%) and cefoxitin (11.8%). None isolate was resistant to imipenem. ESBLs and/or AmpC β-lactamases were detected in 94% (223/237) of isolates. Plasmid-mediated quinolone resistance (PMQR) genes were detected in 95.3% (226/237) isolates. Moreover, both ESBL and PMQR genes were detected in 92.4% (219/237) of isolates. **Conclusions:** The results of this study show an alarming high prevalence of co-resistance to third generation cephalosporins and quinolones in broilers' meat which suggest a possible role of the food chain in the MDR *E. coli* spread. According to previous reports, the ongoing use of antimicrobial drugs in mass therapy and prophylaxis in zootechny should be urgently revised and stopped, particularly in the poultry sector.

176. A risk assessment approach evaluating the spoilage of yogurt with respect to moulds

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ABSTRACT

Objectives: As the matter of yogurt spoilage from moulds poses important challenges to the dairy industry (e.g., leading to recalls and claims), and due to the limited documentation on Quantitative Microbial Risk Assessment with respect to quality, the objective of the current study was to provide a basis of risk assessment for mould spoilage. **Materials and Methods:** A stochastic modelling approach was applied based on an *Aspergillus niger* mycelium growth model by 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. The ability of *A. niger* spores to germinate and initiate mycelium growth in the yogurt environment was assessed with a growth-no growth boundary model. Input parameters were fitted to the appropriate distributions and *A. niger* colony's diameter distributions for each stage of chill chain were derived using Monte Carlo simulation. **Results:** By combining the output of the model with the mould prevalence, which is being estimated by the industry using challenge tests, the probability distribution of the number of cups in which a visible mycelium of *A. niger* is being formed at the time of consumption was determined. The model output showed that for a batch of 100,000 cups in which the percentage of contaminated cups with *A. niger* is 1% (that is 1000 yogurt cups) the predicted numbers (median (5th, 95th percentiles)) of the spoiled cups at consumption time were 8 (5, 14). For higher percentages of 3, 5 and 10% the predicted numbers (median (5th, 95th percentiles)) of the spoiled cups at consumption time were estimated to be 24 (16, 35), 39 (29, 52) and 80 (64, 94), respectively. **Conclusions:** Beside the current value of developed model, which can lead to a more effective risk-based quality

management of yogurt and support the decision making in yogurt production, the general concept of this study constitutes the first study in risk assessment of mould spoilage**.**

177. *Toxoplasma gondii* in Caribbean livestock

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ABSTRACT

Objectives: *Toxoplasma gondii* is a ubiquitous protozoan parasite capable of infecting all warm-blooded animals including livestock. In these animals, the parasite forms cysts in the tissues which may pose a risk to public health if infected meat is consumed undercooked or raw. The aim of this study was to determine the exposure of livestock to *T. gondii* in the Caribbean. **Materials and Methods:** Initially, sera from 305 sheep and 442 goats from Dominica, Grenada, Montserrat and St. Kitts and Nevis were examined for *T. gondii* antibodies using an in house ELISA. Secondly, sera and/or heart tissue and meat juice were collected from 124 pigs, 116 sheep and 66 goats at the St. Kitts and Nevis Abattoir. Sera and meat juice were screened for reactive antibodies to T. gondii using an in-house ELISA. Heart tissue was screened for *T. gondii* DNA using quantitative PCR and positive samples were genotyped using RFLP. **Results:** Antibodies to *T. gondii* were detected in sera from over 40% of animals collected in the initial study of 4 Caribbean islands. In samples from St Kitts and Nevis abattoir, *T. gondii* antibodies were detected in 48% of pigs, 26% of sheep and 34% of goats tested. Antibodies were also detected in the meat juice from 55% of pig hearts, 22% of sheep hearts and 31% of goat hearts tested. There was a significant positive correlation between serology and meat juice results. T. gondii DNA was detected in heart tissue of 21% of pigs, 16% of sheep and 23% of goats tested. Preliminary PCR-RFLP analysis identified a predominance of the Type III genotype of T. gondii although work is currently underway to investigate this further. **Conclusions:** These results suggest widespread environmental contamination with *T. gondii* oocysts and that livestock could be a potentially important source of T. gondii infection if their infected meat is consumed (or handled) undercooked.

178. A Comparison of the Active and Passive *Salmonella* Surveillance Systems for Antimicrobial Resistance in the French Poultry Agro-Food Chain

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ABSTRACT

Objectives: Two surveillance systems are established in France for the monitoring of antimicrobial resistance (AMR) in *Salmonella enterica* subsp. enterica isolated from the agro-food chain: 1) the passive event-based *Salmonella* Network (PS) and 2) the mandatory active surveillance system (AS) regulated by the European Commission. This parallel monitoring provides an excellent opportunity to gain new knowledge in AMR surveillance through the exploration of trends in diversity and resistance profiles, as well as an understanding of the emergence of new phenotypes using historical data collected by passive and active surveillance systems. **Materials and Methods:** Three food animal populations were investigated in this study: laying hens, broilers and fattening turkeys, which have been continuously monitored through PS and AS since 2008. The AS system requires antimicrobial susceptibility testing against a panel of 15 antimicrobials for a minimum of 170 Salmonella isolates per poultry sector each year. All bacterial isolates (e.g. representing animal health, food, or feed) collected through the PS surveillance system are tested for antimicrobial susceptibility for a similar panel of 15 antimicrobials. To perform a retrospective analysis, 2015 Clinical Laboratory Standards Institute clinical breakpoints were used to harmonize and interpret the antimicrobial minimum inhibitory concentrations of all isolates. **Results and Conclusions:** This study was performed to determine if the two surveillance systems produced similar findings in poultry production over time. Direct comparison of PS and AS in terms of total resistant phenotypes, the prevalence of resistance to certain antimicrobials, and measures such as yearly trends in AMR among regulated *Salmonella* serovars in poultry will be evaluated and presented. Assessment of the PS and AS systems will be beneficial to inform stakeholders for future designs of surveillance programs to provide a cost effective means to make the most informed regulatory decisions regarding AMR in the agro-food chain.

179. A predictive model for *Alicyclobacillus acidoterrestris* growth as a tool to assess risk of fruit juice spoilage

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ABSTRACT

Objectives: The objective of the present study was to develop a predictive model for the effect of temperature and pH on *Alicyclobacillus acidoterrestris* growth and validate it in predicting spoilage of fruit juices at dynamic temperature conditions simulating distribution and storage of the product. A product specific model, like this, can be used for the development of a risk assessment approach for assuring fruit juice quality. **Materials and methods:** Aiming at modeling the behavior of *A. acidoterrestris* its growth was studied i) in K broth (2.5g/l yeast extract; 5.0g/l peptone; 1.0g/l glucose; 1.0g/l tween 80) adjusted to pH=4.5 with filtered 25% (w/v) citric acid at temperatures (T) ranging from 25 to 55°C and ii) in K broth of pH ranging from 3.03 to 5.53, adjusted with filtered 25% (w/v) citric acid, at the optimum growth temperature of 48°C using the turbidimetric system BioscreenC. In both cases, the growth rates were modeled using a Cardinal Model with Inflection (CMI). To adjust the model to the product, the kinetic behavior of *A. acidoterrestris* was assessed in fruit juices at 48°C and the estimated rates was used in the CMI. The model was further applied to assess the risk of fruit juice spoilage in the market of Greece based on the average daily temperature (WunderSearch® database) during a shelf life of a semester. **Results:** The estimated values for the cardinal parameters Tmin, Tmax, Topt and the optimum maximum specific growth rate (μ_(max_opt)) of *A. acidoterrestris* were found to be Tmin=19.90°C, Tmax=55.71°C, Topt=48.29°C and μ_(max_opt)=1.001/h, respectively. Simultaneously, the values for the cardinal parameters pHmin, pHmax, pHopt and μ_(max_opt) were equal to pHmin=2.90, pHmax=5.93, pHopt=4.23 and μ_(max_opt)=1.077/h, respectively. The experiments with fruit juices, in agreement with the literature data, showed that spoilage was observed at population level of 105-6CFU/ml (spoilage level). Validation at various dynamic temperature profiles showed that the model can accurately predict both microbial growth and spoilage time. **Conclusions:** We demonstrated that the risk assessment model can be used by the industry as a decision making tool for adjusting the shelf life during a semester in order to achieve an accepted risk of spoilage. This study has been co-financed by the European Regional Development Fund and Greek national funds, Project MOIKOM-09SYN-22-977.

180. Risk factors for broiler flocks contamination with *Campylobacter* spp. in Lithuania

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ABSTRACT

Campylobacteriosis is a common human bacterial infection that is spread through the food. Broilers are frequent carriers of *Campylobacter* spp. During the processing, if intestines are damaged, *Campylobacter* may cross-contaminate the carcasses which act as the primary source of *Campylobacter* infection to humans. The objective of our study was to determine risk factors for *Campylobacter* infection in broiler flocks in Lithuania. Over one-year period 81 broiler flocks slaughtered in two abattoirs were included in this study. Each broiler flock was tested for contamination with *Campylobacter* spp., and for the presence of various pathologies at the abbatoir. To isolate *Campylobacter* spp. we were using multiplex-PCR and microbiological methods. Several statistical criteria such as Pearson χ2 compatibility criterium, Mann-Whitney Z criterium, Spearman's rank correlation coefficient and the binary logistic regression

(method ENTER) were employed to identify risk factors at the flock, the farm, and the abatoir levels. Study revealed that 59.3% of the examined broiler flocks were contaminated with *Campylobacter* spp. The most prevalent detected species was *C. jejuni* (44.4%) followed by *C. coli* (2.5%). Following a single factor statistical analysis, we revealed that the risk of broiler flock contamination with *Campylobacter* was significantly associated with the sampled farm, the slaughterhouse, the number of broiler houses at the farm, the slaughter weight, and the type of the ventilation system. At the abattoir, broiler flock contamination with *Campylobacter* increased with the increase of runt and peritonitis pathologies in broilers of the slaughtered flock. The statistical analysis of multiple risk factor demonstrated that broiler age and the average weight per bird at slaughter, assigned to health at slaughter risk factor group, had highest effect on the increased prevalence of *Campylobacter* spp. in broilers. The risk factors determined in present study may have a decisive influence on the transmission of *Campylobacter* in broiler farms. Improving health and welfare could also reduce *Campylobacter* levels in broilers.

181. *Campylobacter* prevalence, counts and resistance profiles in poultry meat of Estonian, Latvian and Lithuanian origin at Estonian retail level and from patients with severe enteric infections in Estonia

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ABSTRACT

Objectives: The main aim of the study was to determine prevalence, counts, seasonality and antimicrobial susceptibility patterns of *Campylobacter* isolates obtained from Estonian retail level. **Material and methods:** A total of 606 fresh poultry meat samples were collected monthly within one year period to determine the *Campylobacter* prevalence. Counts were determined for 220 fresh broiler chicken meat samples. From Estonian hospitals 28 *Campylobacter* isolates were obtained during 2011-2013. The detection, enumeration and identification was carried out according to ISO 10272-1:2006, ISO 10272-2:2006 and multiplex PCR assay. Antimicrobial susceptibility of randomly selected 126 isolates was determined using minimal inhibitory concentration test. Statistical analyses were performed with Excel 2010, Statistical Package R and Statistical Package for Social Sciences 13.0. **Results:** The overall *Campylobacter* prevalence in fresh poultry meat was 20.8% at Estonian retail level. The prevalence among 220 fresh broiler chicken meat samples was 35.0%: 20.3% in Estonian, 60.0% in Latvian and 50.0% in Lithuanian origin products with mean contamination loads 2.8; 3.4; 3.2 log10CFU/g, respectively. There were lower ($p<0.001$) *Campylobacter* prevalence and counts in Estonian compared to Latvian and Lithuanian origin samples. Distinct seasonal peak of *Campylobacter* positive samples was during warm summer months. Species identification resulted in estimation of 89.0% isolates harbouring *C. jejuni*, 8.0% *C. coli* and 3.0% *Campylobacter* spp. specific genes. Resistance to ≥1 antimicrobials was detected 63.3% of *Campylobacter* broiler chicken meat and 71.4% of human origin isolates. Multidrug resistance was observed in 5.1% and 7.1%, respectively. Among of broiler chicken meat isolates 60.2% were resistant to ciprofloxacin and 71.4% of human isolates to fluoroquinolones. Antimicrobial resistance to ≥1 antimicrobials, to ciprofloxacin and nalidixic acid was less frequent ($p<0.05$) in Estonian compared to Latvian and Lithuanian isolates. **Conclusions:** The greater prevalence, counts and antimicrobial resistance of the *Campylobacter* isolated from the fresh broiler chicken meat of Latvian and Lithuanian origin might pose greater *Campylobacter* exposure risks to the Estonian population. Currently the focus is on analyzing WGS data of 53 isolates. **Acknowledgements:** Veterinary and Food Board Health Board Estonian Scientific Council, grant 9315 Ministry of Agriculture of Estonia, project T13057VLTH

182. *Salmonella* Napoli in fresh vegetables: a multidisciplinary approach to risk assessment and management

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ABSTRACT

Human infections caused by *Salmonella enterica* serovar Napoli are relatively rare in Europe. In 1982 a large outbreak due to *S.* Napoli was reported in EU linked to consumption food of Italian origin. In particular, the outbreak occurred in England and Wales and was associated with the consumption of chocolate bars manufactured in Northern Italy. In Italy, S. Napoli represents 2- 4% of all *Salmonella* serovars isolated from human infections each year. No animal reservoirs have been recognized for this serovar which is isolated very rarely from food animal. Since 2004 the European Rapid Alert System for Food and Feed (RASFF) has notified different alerts involving salad produced in Campania contaminated by *S.* Napoli. A cluster of cases associated to the consumption of rucola produced in Italy was also detected in Sweden in 2009. Since 2009 the Italian Ministry of Health has organized a multidisciplinary task force with the aim in defining strategies to reduce or eliminate the risk of contamination of fresh vegetables and manage the risk for human beings. The initial work of the taskforce was the surveillance of the farms located in Campania region and involved in the RASFF alerts. In order to evaluate the presence of *Salmonella* fresh vegetables and irrigation waters, were sampled. The results showed that *S.* Napoli was the prevalent serovar accounting for 6% of the samples. At light of these finding, in 2012 a pilot study was initiated with the aim to identify a possible reservoir of *S.* Napoli among wild animals. During the study *S.* Napoli was isolated from nightingales and this represent the first isolation of this serovar from birds. Furthermore, the environmental sources and fresh and leafy vegetables should be considered important for the transmission of this serovar to humans. This work represents an example of how a multidisciplinary approach may contribute to improve the knowledge of epidemiology of *S.* Napoli. This emerging serovar is a relevant public health concern in Italy. In the light of this, the networks operating in the framework of food safety, animal and human health should cooperate and take in to account of environmental compartment in the risk assessment and management.

183. How to move to risk based metrics to improve food safety in the European Union

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ABSTRACT

The aim of this poster is to present selected outputs of the BASELINE project concerning the definition of risk based metrics, such as the Performance Objectives (POs), for *Campylobacter* in broiler carcasses after chilling and *Listeria monocytogenes* as well as *Salmonella* in pork cuts intended to be eaten cooked. The POs for *Campylobacter*, calculated using the EFSA data of the monitoring study performed in 2008, correspond to the *Campylobacter* concentrations after chilling resulting in a final concentration on carcasses at the time of consumption equal or below to a FSO set at -1.2 log10 cfu/g. The estimated mean concentrations of *Campylobacter* on carcasses were 1.05 and 2.38 Log10 cfu/g for non thinned and thinned flocks, respectively. Further, for carcasses starting with a high *Campylobacter* contamination (i.e., > 2.5 log10 cfu/g) a reduction in PO values higher than 1.5 log10 cfu/g was calculated as necessary to meet the FSO. The POs for *L. monocytogenes* and *Salmonella* in pork cuts were calculated on results of ten lots of pork cuts tested for presence and concentration of both pathogens under different scenarios through the shelf life. For *L. monocytogenes* the median values of prevalence distributions ranged between 0.41 and 0.68. The number of samples to test to verify lot compliance to these values ranged between six, for samples tested immediately after packaging, to three, for

samples tested at the end of the shelf life. The concentration values ranged between 2.02 log10 CFU/g, for samples tested immediately after packaging, up to 3.14 log10 CFU/g for samples tested after final storage at 14°C. Basing on log normal distributions, the maximum contamination level of the lots to achieve the suggested POs, were calculated. It was obtained that mean concentration estimated as PO should be between -0.43 and 0.48 log10 CFU/g. The POs for *Salmonella* were calculated at the 50th percentile of prevalence distributions under selected stages, from pork cuts packaging up to the consumption. The results indicated that values increased between 26.10 % of positives after final storage at 14°C to 46.70 % of positives after storage at retail shops. The number of samples to test in order to verify the compliance to the estimated POs ranged between five, for samples after storage at retail, and ten, for samples stored at 14°C before the expiration date. For risk management purposes, percentiles different from the 50th can be selected in order to derive the POs.

184. Serosurvey based on meat juice analysis on *T. gondii* infection in slaughtered small ruminants

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ABSTRACT

Toxoplasma gondii infection still represents worldwide a public health concern, with one third of human population affected (Cook et al., 2000 British Medical Journal 321: 142-147). A major way of infection is represented by the consumption of raw or undercooked meat, particularly of certain species including small ruminants (EFSA 2007). Considered the high seroprevalence values encountered in sheep and goats bred in Northern Italy, a serosurvey based on the detection of antibodies in meat juice obtained from muscle samples collected at three slaughterhouses in Lombardy (Northern Italy) was planned. During a period comprised between April 2013 and January 2014, hearth or diaphragm (basing on convenience) were collected from 223 sheep and goats. From samples, meat juice was obtained and tested with a commercial ELISA kit (ID Screen® Toxoplasmosis Indirect Multi-Species, IDVET) according to manufacturer's instruction. Antibodies anti-*T. gondii* were found in 46 (20.6%) of 223 examined animals (sheep: 19.9%; goats: 25.9%). Prevalence resulted related to age (p=0.0001), with adult animals more at risk of infection (OR=1.045) than kids or lambs; adult animals are indeed more exposed to parasite in environment, as suggested by several Authors (Tenter et al., 2000, Int J Parasitol 30: 1217-1258; Halos et al., 2010 Int J Parasitol 40:193-200). Moreover, concerning lambs, the provenience of the animals resulted a risk factor for the infection, with animals proceeding from Italian herds more at risk of infection (OR=10.392) in comparison to those imported from abroad, probably due to differences in the age at which lambs are slaughtered in each country. Although the detection of circulating antibodies does not necessary mean infectivity of meats, the detection of antibodies reflect the proportion of infected animals that potentially represent a risk for consumers. Meat juice, easier to collect at slaughterhouses than serum, has been proven suitable for the screening of *T. gondii* infection in different meat-producing animals destined to human consumption (Berger-Schoch et al., 2011 Zoonoses and Public Health 58: 472-478) and its use should be implemented in *T. gondii* surveillance and reporting system.

185. Understanding the factors leading to *Salmonella* proliferation in post-harvested tomato. Can a 'perfect storm' scenario lead to outbreaks?

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ABSTRACT

Vegetables are increasingly recognized as vehicles of human pathogens. Outbreaks of gastroenteritis caused by strains of non-typhoidal *Salmonella* and enterovirulent *Escherichia coli* linked to the consumption of fresh fruit and vegetables show that these human pathogens can contaminate produce at any stage of the production cycle, farm to fork. The risk of occurrence of these human pathogens has increased as a consequence of global changes in the way food is produced and consumed: intensive production, longer shelf life and increased consumption of raw vegetables. Our goal was to test how different agronomic practices (irrigation and fertilization), tomato cultivar selection, pathogen and host genotypes affect the ability of *Salmonella* to multiply in tomatoes post-harvest. Our methodology was based on the implementation of such variables in field or in tomatoes post-harvest. A multifactorial statistical analysis on about 6000 harvested and infected tomatoes was carried out. Our results show that in terms of fertilization regimes, differences in nitrogen concentrations in the plant tissues were correlated with the susceptibility of partially-ripe Solar Fire tomatoes to *Salmonella*. The irrigation regimes per se did not affected susceptibility of the crop to post-harvest proliferation of *Salmonella*. However, *Salmonella* grew significantly better in water-congested tissues of green tomatoes. In addition, tomato maturity, cultivar, *Salmonella* genotype, and inter-seasonal differences were the strongest factors affecting proliferation. Our research also showed that *Salmonella* proliferation was strongly reduced in tomato mutants with defects in ethylene synthesis, perception and signal transduction, highlighting the ethylene signaling as possible key-role able to sustain *Salmonella* proliferation. These results contribute in understanding the factors that orchestrated together can lead a 'perfect storm' scenario increasing the risk of outbreaks of *Salmonella*.

186. Growth, colonisation and internalisation of VTEC in fresh produce: the potential impact on food security

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ABSTRACT

Objectives: Fresh produce accounts for a significant proportion of food-borne 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 Hazard Analysis and Critical Control Point (HACCP) framework. However, pre-harvest factors, such as internalisation, 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. 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 food-borne outbreaks. **Materials and Methods:** Colonisation 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 green fluorescent protein (GFP)-tagged *E. coli*. The metabolite complement of plant extracts was also determined by HPLC and GC/MS. **Results:** E. coli shows marked differences in growth and internalisation in plant tissues that is dependent on the plant species; tissue type and bacteria strain type. Biochemical analysis revealed major differences in metabolites between the host species, which can be used to relate growth rate variances to plant species-specific differentiation. **Conclusions:** 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 the RA to show the basis for important differences in bacterial growth potential on plant hosts.

187. Food Safety of Dairy Products and TSEs Control Through Genetic Selection In Small Ruminants: Prion Gene Analysis In Sicilian Goat Breeds, Italy

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ABSTRACT

Objectives: The full eradication of prion strains circulation in sheep and goats is important for food safety concern related to risks over exposure of Transmissible Spongiform Encephalopathies (TSEs) through dairy products. Breeding for genetic resistance in sheep is accomplished in several countries in EU. In goats, recent studies provided a potential association of some PrP gene (PRNP) polymorphisms with resistance to TSEs. A potential protective allele 222 (Q222K) showed promising results by in vitro study and in vivo challenges. Small ruminants are often bred in rural and economic marginal areas in developing but also in developed countries. The feasibility of breeding program for TSEs resistance could increase the interest in autochthon breeds with a good impact on the preservation of livestock biodiversity and valorisation of typical food productions. PRNP genetic variability in autochthon Sicilian goat breeds was analysed in this study. **Materials and Methods:** The sequence of caprine PRNP was determined in 568 goats of five breeds commonly reared in Sicily: Girgentana (n = 160), Derivata di Siria (n =153), Maltese (n = 143), Argentata 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:** 12 polymorphic sites were identified, G34V, V125I, G127S, M137I, I142T, H143R, R151H, R154H, P168Q, R211Q, Q222K e S240P. We found 222K variant in all breeds, among them the Girgentana, Derivata di Siria and Argentata dell'Etna breeds showed a highest frequency: 18,7%, 14,6% and 16% respectively. The results showed that the realization of breeding programs for TSE resistance in some Sicilian breeds might be easier to accomplish compared to other European breeds in which K222 is not present at such high level. **Conclusion:** Breeding for TSEs resistance to eliminate the prion strains from small ruminant flocks can assure a higher biosafety of milk and dairy products with added benefits for consumers. A major quality and safety of dairy products are also important for fair competition among dairy industries on the global market. These results might be important to increase interest in the breeding of Sicilian goat breeds preserving biodiversity and typical food productions. This study was supported by EMIDA project: 'GOAT-TSE-FREE and RF-2010-2318525 from Italian Ministry of Health.

188. Risk assessment of meat inspection on *Cysticercus* in beef supply chain in Pakistan

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ABSTRACT

Objectives: Food safety and quality are main steps towards food security. Health problems due to unsafe food are known from the start of history. Beside other hazards, biological hazards including bacteria, virus and parasites are major part of food borne disease problems globally. Current information on the prevalence of *Cysticercus* in beef in Pakistan is based on the visual meat inspection of beef carcass in export-slaughterhouses. Data at national level is lacking due, as no inspections are made in the informal meat chain, which is the dominant route of national beef supply. Present work is an effort is to assess the *Cysticercus* risk in beef and human population in Pakistan. **Material and Method:** The assessment was based on an excel spreadsheet template generously provided by B.R. Berends (IRAS, Utrecht University). Prevalence data at slaughter were estimated from previous studies at Lahore slaughterhouse. Sensitivity of meat inspection was divided into average, worse and best inspection conditions (20, 10 and 30%). It is also assumed that survival rate of *Cysticercus* after cooking was 20-40% (average 30%) and probability of human infection is on an average 29% (10-50%). Calculation was made with Pakistan population which was 182.1 million in 2014. Calculations were done for a 'meat inspection in place' on 'no meat inspection at all' scenario. **Results:** Based on above mentioned assumptions, the incidence of human taeniasis acquired via beef consumption in Pakistan would range from 8 to 6305/100,000/yr. without meat inspection compared to 5-5674/100,000/yr. with meat inspection implemented. Corresponding average values are 331 and 265/100,000. **Conclusion:** Results indicate the limited effect of meat inspection in the control of human taeniasis, unless inspection results are communicated to the farmer and corrective actions can be taken at primary production level. Although this finding is not novel, it indicates that a focus should be put on biosecurity at primary production level (i.e. to avoid infection of grazing cattle) and on the safety of beef processing before consumption. This does not out rule inspection of slaughter animals and subsequent meat inspection, as animal welfare issues, sampling for zoonosis monitoring and examination for pathological alterations specific for certain bacteria can be done at these stages.

189. The microbiology of beef chilling: do the observations fit the predictions?

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ABSTRACT

This study investigated the microbiology of beef carcasses and vacuum packed primals during chilling and compared the observed data with model predictions (Combase and Food safety Spoilage Predictor) for carcasses. On three separate occasions 10 carcasses were swabbed daily at the neck, brisket, flank and rump during 4 days chilling in a commercial beef export plant and tested for TVC mesophiles, TVC psychrophiles, Total *Enterobacteriacae* Count (TEC), *Pseudomonas* spp., Clostridial spp., lactic acid bacteria (LAB) and *Brochotrix thermospachta* using ISO or internationally accepted methods. The chilling processes were also characterised in terms of ambient, carcass surface and core temperature, relative humidity, carcass surface pH and water activity (aw) for both lean and fat tissue. Vacuum packed beef primals were then prepared from the carcasses and stored at 0°C for 6 weeks. These samples were subjected to the same microbial, temperature, pH and water activity testing as described above. Carcass TVC mesophiles, TVC psychrophiles, TEC, *Pseudomonas* spp., Clostridial spp., and *Br. Thermospachta* counts increased by, 0.62, 0.81, 0.51, 0.72, 0.27 and 1.42 log10 CFU/cm2, respectively, over the 4 day chill. LAB counts decreased by 0.16 log10 CFU/cm2. The corresponding figures on vacuum packaged beef primals were 4.27, 4.06, 0.58, 2.29, 4.55 and 3.67 log10 CFU/cm2, respectively, over 6 weeks storage at 0°C. LAB increased by 4.45 log10 CFU/cm2. Mean pH and aw as well as the slowest and fastest chilling curves were used in the modeling studies for the carcasses. Observed growth was compared with predicted growth from models in Combase (*Br. thermosphacta* and *Pseudomonas* spp.) and the Food Safety & Spoilage Predictor (LAB). The study provides data on the microbiology of beef carcasses during chilling and the resultant primals after deboning. Combase accurately predicted the growth for both *Pseudomonas* spp. and *Br. Thermosphacta* on carcasses. However, the FSSP over-estimated LAB growth on the beef carcasses during chilling. Future work in this study will include comparing the microbiology of vacuum packaged beef primals to Combase and the Food Safety Spoilage Predictor to investigate if these models can predict similar counts to the observed growth.

190. Study of the presence of Aichi Virus in Shellfish and clinical samples

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ABSTRACT

Bivalve molluscs obtain their food by filtering small particles suspended in water. Often these waters are contaminated and in the process of filtering, molluscs concentrate and retain pathogens including enteric viruses. Although Norovirus and Hepatitis A virus, causing gastroenteritis and hepatitis, are considered the most important viral agents transmitted by shellfish, in recent years other viruses have emerged such as Aichi virus (AiV), as responsible for outbreaks associated with this type of food. AiV belongs to the genus Kobuvirus of the family *Picornaviridae*. It is a virus with icosahedral morphology that

present a single stranded RNA virus positive sense (8,280 nucleotides) and a poly (A) chain. AiV was detected from clinical samples, and is considered today one of the main causes of nonbacterial gastroenteritis in some geographic areas. Furthermore, several studies conducted in Japan, Germany, France and Tunisia showed a high prevalence of AiV antibodies in adults (between 80 and 99%) which is indicative of a large exposure to this virus. The aim of this study is the detection and quantification of AiV in bivalve molluscs from the Galician rias and in molluscs imported from developing countries (Vietnam, Peru, South Korea and Morocco). Samples consisted in 10 or 20 individuals, depending on the species, that were dissected under sterile conditions to obtain the digestive gland of the animal. The viral RNA was extracted using Nucleospin RNA Virus Kit (Macherey-Nagel) from digestive tissue homogenates. Viral detection and quantification was performed by real time RT-PCR using the kits Platinum Quantitative RT-PCR Thermoscript One-Step System (Invitrogen) and aichi@ ceeramTools®.Health (Ceeram) with AIV-AB-F and AiV-AB-R primers and AB-TP probe to amplify different genogroups. Internal controls were included to avoid false positive or negative results, and to determine the presence of inhibitors. Until now 45% of samples samples were analyzed, resulting all negative for AiV. This could indicate that the prevalence of this virus in Galicia is lower than that described for other geographical areas. It will be necessary to complete the analysis to confirm this hypothesis and to know the real importance of AiV in our region.

191. Risk-based control options to mitigate microbial risks associated to traditional fermented Italian salami and soppresse consumption

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ABSTRACT

Traditionally fermented pork-meat products are becoming increasingly popular among European consumers and a matter of concern is the potential exposure to food-borne pathogens via these products. Therefore, with the aim of identifying control strategies, that can be easily implemented by the producers themselves, the peculiar traditional production system of Italian salami and soppresse was investigated with a risk-based approach. From farm to fork samples were collected during 2008-2009 in a selection of productive units in order to obtain data on microbiological contamination along all the production chain, moreover physical-chemical parameters (pH, aw) of the products were recorded. The collected data were used as input for a model based on that one designed by Hwang and coll. (2009) to estimate the quantitative variation of *Salmonella* spp., *E. coli* O157 and *L. monocytogenes* during time according to the recorded pH and aw values. Moreover a microbiological predictive modelling was applied to estimate the probability of pathogens survival at retail time. In 2009-2010 an intensive sampling plan was carried out in order to fill in the data gaps and collect detailed information on the ripening conditions of all the producers so that to define targeted control measures. Data collected during 2008-2009, suggested that the production process was not standardized even within each producer. Moreover, the risk matrix implemented with qualitative and quantitative data and with microbiological predictive models outputs, highlighted that these products may pose a moderate risk to human health in relation to *E. coli* O157 and *L. monocytogenes* in particular and that the aw trend influences greatly the pathogens level. Data from 2009-2010 sampling allowed to identify a targeted control strategy, based on the fact that the salami and soppresse safety depends not only on the microbiological characteristics of the raw meat but also on the ripening conditions, which trend may be measured by the aw fluctuations, thus a correlation was estimated between the aw decrease and the products weight loss. Finally a risk mitigation strategy was defined as follows: salami and soppresse weight decrease must be controlled by producers during ripening and products are allowed to be marketed when no pathogens are detected in the batter (sampled and analysed by the Competent Authority) and a weight decrease of at least 25% is reached.

192. Risk assessment of salmonella in feed and animal production

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ABSTRACT

Food items of animal origin, such as pork products, have been suggested as the main source of zoonotic salmonella infections in Europe. Contaminated feed can potentially introduce the pathogen into the animal-derived food chains, especially in countries with low salmonella prevalence. In Finland, the low level of salmonella infections is maintained by monitoring the pathogens prevalence not only on food products and animals but also already in the animal feed. The objective of the study was to assess the impact of this feed control as well as the feed manufacturing and feeding practices on the salmonella risk in Finland, focusing on the pork-derived food chain. Information on feed hygiene practices of commercial feed mills, mobile mixers, and pig farms was collected from reports of the Finnish Food Safety Authority, the customs office, and the Finnish Farm Registry, as well as from a questionnaire carried out during the study. Data from 2013 was chosen as the starting point for the risk assessment. A simplified mathematical model of feed flow was built based on national average proportions of feed raw materials and feed types used for pig production. The model can be used to evaluate the exposure risk of pig production types (as a whole) as a mixture of risks in the feed types used, which again are mixtures of feed raw materials, in given proportions. Control sample information was attached to each component within the feed flow, if available, to assess contamination probability. There were 1600 pig farms and around 1.3 million pigs in Finland in 2013. More than 186 million kilograms of pork was produced during that year. Around 80 % of the farms fed pigs with commercial feeds, either complete feeds or supplement feeds, whereas only 0,7 % of the farms produced all their feed components by themselves and less than 8 % of the farms were the clients of a mobile mixer. According to questionnaire, none of the pig farms had heating process to ensure feed hygiene. At the same time, none of the farms resold the feed produced on the farm. The early findings of the study suggest that pig farms are vulnerable over feed-acquired salmonella contamination. Next, feed control is evaluated by comparing the present monitoring effort to scenarios where there would be no monitoring for salmonella. The impact of certain feed ingredients to salmonella risk is also exploited.

193. Presence of *Listeria monocytogenes* in Chilean food matrices

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ABSTRACT

Objectives: To compare *Listeria monocytogenes* strains obtained from food matrices with those from environmental samples in the same food processing plant. **Methods and Results:** Between 2008 and 2012 the presence of *L. monocytogenes* was evaluated in 2647 food samples. A total of 448 work surfaces and 92 equipment's were also evaluated from 6 plants which produce ready-to-eat (RTE) foods in Santiago, Chile. An additional selected sample of hand and nails samples was also obtained from 13 food handlers working in a sausage elaboration plant. As a whole *L. monocytogenes* was present in 265 (10%) food samples and 22 (4%) environmental samples. The foods with highest recovery were red meats 14/60 (23%), poultry 223/1196 (19%), the remaining samples accounted a total of 27/1391 (2%). The environmental samples positive for *L. monocytogenes* were obtained from two food plants both the cheese 8/8 (100%) and from a fresh peaches exporter 3/3 (100%). Finally *L. monocytogenes* was isolated from 5/13 (38%) food handlers studied. **Conclusions:** The study confirms the presence of *L. monocytogenes* in different matrices, especially in meat and RTE products. Analyses conducted on work surfaces revealed that contamination comes mostly from both raw materials and surfaces in indirect contact with foods. Significance and impact of study: The study reinforces the need for companies to apply regulations related to food quality and safety systems (HACCP, Hazard Analysis & Critical Control Points) to prevent *L. monocytogenes* contamination from food processing plants.

194. A comparison of available microbiological standards for dried culinary herbs and spices

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ABSTRACT

Dried culinary herbs and spices (DCHS) are minor food components that are added to most of our foods. Due to their low water activity they usually not enable microbial growth. Nevertheless, some microorganisms, including pathogenic ones, can survive in DCHS. The addition of microbial contaminated DCHS to ready-to-eat food in combination with improper food storage can cause a serious health risk for the consumer. Thus, DCHS in combination with *Bacillus* spp. and *Salmonella* spp. were listed in the top four ranked group of food/pathogen combinations linked to outbreaks originating from food of non-animal origin in the EU (EFSA Panel on Biological Hazards, 2013). Our study aimed to identify and compare available microbial standards for DCHS addressing European Union (EU) member and non-member states. To receive an overview on current microbiological standards for DCHS, a literature study and internet research was performed. The standards considered comprise facultative and obligate standards of private and public bodies on global, EU and national level. In the EU, no microbiological limits for DCHS are set by EU law. However, the Commission Recommendation of 19 December 2003 concerning a coordinated programme for the official control of foodstuffs for 2004 [document number C(2003) 4878] includes values for satisfactory, acceptable and unsatisfactory levels of *Salmonella* spp., *Bacillus cereus, C. perfringens* and *Enterobacteriaceae* to interpret the results and to assess the biological safety of spices and herbs on retail and non-retail level in frame of this official control programme. The available national standards, including those of non-EU member states, often address ready-to-eat DCHS. In summary, microbiological criteria are, except of few food categories, less defined for food of non-animal origin compared to food of animal origin and data for DCHS are limited. In general, most of the microbiological standards available are facultative, even if set by public bodies. In addition to those, the spice/herb industry and the retail usually use individual buyer–seller agreements.
Acknowledgements: This research was performed in the framework of the EU project SPICED (Grant Agreement: 312631; www.spiced.eu) with the financial support from the 7th Framework Programme of the European Union. Reference EFSA Panel on Biological Hazards (BIOHAZ), 2013. Scientific Opinion on the risk posed by pathogens in food of non-animal origin. Part 1. EFSA Journal 11(1): 3025

195. A risk assessment model for *Escherichia coli* in lymph nodes of bovine carcasses

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ABSTRACT

Objectives: This research arises from the background of a progressive streamlining of food controls on abattoir on behalf of visual inspection and limitation of palpation and cuts. For a comprehensive risk assessment, this approach has to consider the presence of lymph nodes which adhere to the carcass, therefore accidentally destined to consumption. In fact, a potential source of pathogenic bacteria in ground beef is the lymphatic system, specifically the lymph nodes. The objective of the current study was to determine the prevalence of *Escherichia coli* in bovine lymph nodes. **Materials and methods:** Internal and external iliac bovine lymph nodes (n = 652) were collected from 326 carcasses at a commercial slaughterhouse. 638 lymph nodes sampled were obtained from regular slaughter, and the remainder 14 were obtained from 7 emergency slaughters. **Results:** *E. coli* prevalence in the lymph node samples was high, with an overall prevalence of 38.65%. Lymph nodes from emergency slaughter carcasses had a higher prevalence (85.71%) of *E. coli* than did those from regular slaughter carcasses (37.61%). *E.coli* were isolated according to ISO 16654:2001. 200 lymph nodes were analysed for hemolysin (*hlyA*) gene but it was never detected. Further exams are being conducted for the detection of genes encoding Shiga toxin 1 and 2 (*stx1* and *stx2*), intimin (*eaeA*) and hemolysin (*hlyA*) by Multiplex PCR. **Conclusions:** This research underlines the role of lymph nodes as a source of pathogens' contaminations, mostly for ground meat, and focuses the attention on the importance of accurate and detailed inspection of the carcasses despite modern trends and revisions of procedures. Results from Fisher's exact test; Regular slaughter: 120 +ve (36,80%), 199 -ve (61,05%), Total 319 (97.85%); Emergency slaughter: 6 +ve (1,85%), 1 –ve (0.30%), Total 7 (2.15%); Total: +ve 126 (38,65%), -ve 200 (61.35%). The two-tailed P value equals 0.0147. The association between groups and outcomes is considered to be statistically significant.

196. Effect of absorbent pads containing pinosylvin inclusion complexes on *Campylobacter* spp. control: in vitro and in vivo studies

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ABSTRACT

Objectives: To evaluate the *in vitro* and *in vivo* efficacy of hydroxypropylated cyclodextrins-pinosylvin inclusion complexes (HP-CD-PS ICs) coating on absorbent pads for *Campylobacter* control in chicken meat packages. **Materials and Methods:** The absorbent pads were coated with hydroxypropyl-β-cyclodextrin (HP-β-CD) or hydroxypropyl-γ-cyclodextrin (HP-γ-CD) inclusion complexes with pinosylvin (PS ICs) [1] at 20% (v/v) final concentration in a cellulose solution to achieve a more uniform coating of the pads. *In vitro* testing of PS ICs containing pads (0.08 to 0.8 mg/cm^2) was performed in solid and liquid media inoculated with *C. jejuni*. *In vivo* testing of PS ICs absorbent pads was performed, firstly, in chicken exudates and, secondly, in non-sterile chicken breast fillets, all artificially contaminated with *C. jejuni*. Furthermore, the antimicrobial action of the pads on common chicken bacteria such as lactic acid and psychrotrophs, Pseudomonads and total viable cell counts was also evaluated. **Results:** *In vitro* testing in solid media showed that the inhibition halos produced with HP-β-CD PS ICs ranged from 11 to 26 mm; whereas for HP-γ-CD PS ICs, they ranged from 12 to 26 mm. In liquid medium, the pads caused a 4 log CFU/mL reduction in *C. jejuni* growth, thus exhibiting a bactericidal activity, as previously demonstrated [2]. PS ICs pads were also effective in reducing *C. jejuni* contamination in chicken exudates, with 100% of inhibition at concentrations of 0.4 mg/cm^2. In chicken meat stored at 4 ºC, the pads caused up to 80% of microbial reductions in all common chicken bacteria after 3 days of incubation, while also effectively reduced *C. jejuni* load, with a more pronounced efficacy of HP-γ-CD PS ICs pads in these assays. **Conclusions:** This study describes a natural coating for absorbent pads commonly used in chicken packages with good antimicrobial activity not only against *C. jejuni* but also against other bacteria usually present in chicken meat, deeply encouraging the use of this approach as a possible solution for the efficient microbial control of chicken meat. **References:** [1]. F. Silva et al. (2014) Food Chem 145:115-125. [2]. F. Silva et al. (2015) Food Control 54:66-73.

197. Plasmid dynamics between *Clostridium botulinum, Clostridium novyi* and *Clostridium haemolyticum* convert strains into different pathogens

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ABSTRACT

Objectives: *Clostridium botulinum* is a strictly anaerobic spore-forming bacterium found in soil and water sediments. It produces the extremely potent botulinum neurotoxin (BoNT), which is responsible for causing the paralytic disease botulism. The majority of botulism cases are food- or feedborne and poses a great risk to humans and animals across the world. A close genetic relationship exists between *C. botulinum* group III, *Clostridium haemolyticum* and *Clostridium novyi*, which carry most of their toxin genes on plasmids or bacteriophages. They are traditionally organized into these three species due to their ability to cause different diseases. In this study we have explored the genetic relationship between them and we have especially looked for plasmid interactions and the exchange of pathogenic traits. **Materials and Methods:** Strains of *C. botulinum* group III, *C. novyi* and *C. haemolyticum* from different origins were subjected to whole genome sequencing using Roche 454,

Illumina HiSeq and MiSeq technology. Genomes were de novo assembled using GS assembler or MIRA and plasmids were further assembled using consed. **Results:** The genomes included in this study could be divided into four different lineages on the basis of the overall genomic sequence similarity. Three lineages contained strains of more than one species, and one lineage contained strains of all three species. An unusually large plasmidome was discovered (61 plasmids in 24 genomes), which could be organized into 13 different plasmid groups on the basis of their similarity and conservation of plasmid replication or partitioning genes. However, there were patterns of interactions within and between plasmid groups. This dynamic process appears to be primarily driven by phages. Some plasmids or bacteriophages were present in more than one species and lineage, for example the botulinum neurotoxin phage, the noyvi alpha-toxin phage and the botulinum C2-toxin plasmid. **Conclusions:** This study shows that plasmid interactions have taken place between species and across lineages, sometimes unlinking pathogenicity from its genomic background. This makes the pathogenesis of *C. botulinum* more unpredictable and poses an increased risk for escaping detection.

198. Antimicrobial resistance in indicator *Escherichia coli* isolated from broilers and turkeys slaughtered in Poland, 2008 – 2014

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ABSTRACT

Objectives: Monitoring of resistance in indicator *E. coli* acc. 2013/652/EU requirements was implemented in 2014 in broilers and turkeys. The aim of current analysis was to assess the obtained results and compare with 2009 – 2013 data. **Materials and Methods:** Veterinary Inspection randomly sampled 350 caecal contents from broilers (N=180) and turkeys (N=170) in slaughterhouses (respectively, 90 and 23 abattoirs) contributing to majority of annual country production. MacConkey culture followed by PCR identification (uspA gene) provided 349 *E. coli*. Minimal Inhibitory Concentration was determined with Sensititre® EUVSEC (Trek D.S.) and interpreted according to epidemiological criteria. Cephalosporin resistant isolates were further tested with EUVSEC2. The background data were obtained from similar setup, different only by sample type (cloacal swabs), biochemical identification of *E. coli*, and previously EFSA-recommended MIC panels. The approach provided 854 broiler and 863 turkey isolates of *E. coli* (approx. 170 isolates by source and year). The differences in prevalence of microbiological resistance was assessed with 95% CI. **Results:** Official monitoring of broiler *E. coli* proved the highest prevalence of resistance against ciprofloxacin (87,7%), nalidixic acid (79,3%), ampicillin (72,6%), tetracycline (62,0%), sulfamethoxazole (57,5%), and less frequently to trimethoprim (38,6%) and chloramphenicol (19,0%). Few isolates were resistant to gentamycin (3,9%), cephalosporins (2,2%), and colistin (0,6%). Resistance level was similar in turkeys with exception of quinolones: the prevalence was lower than in broilers reaching 70,0% for ciprofloxacin and 58,8% for nalidixic acid. Cephalosporin resistance was identified as ampC-type cephalosporinases (N=6) and ESBL (N=1). No resistance to meropenem and tigecycline was observed, and azithromycin MICs were not interpreted. Similar to previous study (Front Microbiol, 2013, 4: 221) current comparison showed differences in only three out of 22 occasions: less frequent cephalosporin resistance in broilers than in 2009 – 2013 study (7,4%) and ciprofloxacin resistance in broilers and turkeys higher than previously (77,7% and 59,0%, respectively). **Conclusions:** The study confirmed substantial resistance levels of *E. coli* from slaughtered poultry, with few temporal changes. It shows also that caecal samples might be easily replaced by more convenient cloacal swabs.

199. Reduction of *Campylobacter jejuni* in broiler chickens: role of caprylic acid

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ABSTRACT

Campylobacteriosis has been currently recognised as the most frequently reported food-borne ilness within European Union. Since there is a very high prevalence of *Campylobacter* spp. in poultry, caecal carriage can often result in carcass contamination during processing. Reducing the number of campylobacters in chickens, as well as the reduction of carcass contamination in the processing plant, can decrease the risk of campylobacteriosis in humans. Therefore, the effect of caprylic acid (CA) on *Campylobacter jejuni* in chickens was evaluated using two approaches: dietary supplementation or surface treatment of chilled chicken carcasses. To analyze the dietary effect of CA, individually housed broiler chickens (n = 48) were artificially infected with *C. jejuni* VFU612 (106 CFU/bird) on the 21st and 35th days of life. Dietary CA (2.5 and 5 g/kg of feed) significantly decreased *C. jejuni* shedding ($p < 0.05$). However, the effect only lasted for 3-7 days after infection. The numbers of *Campylobacter* shed by the positive control birds reached its maximum on the 37th day of life, while on that same day, both Treatment I and Treatment II groups shed significantly lower ($p < 0.05$) numbers of *Campylobacter* (by 0.8 and 1.8 orders of magnitude, respectively). Also, peak shedding was delayed by one day in both treated groups. After euthanasia of each chicken on the 42nd day of life, no differences in *campylobacter* counts in the crop, gizzard, ileum and cecum were found between the positive control and the treated groups ($p < 0.05$). Surface contamination of the chilled chicken halves was performed with *C. jejuni* VFU612 and CCM6214. Surface treatment with CA at 1.25 and 2.5 mg/mL for 1 min significantly reduced *C. jejuni* VFU612 contamination of chicken skin ($p < 0.05$) by 0.29 – 0.53 and 1.14 – 1.58 orders of magnitude (log10 CFU/g of skin), respectively. Counts of *C. jejuni* CCM6214 were reduced by 0.72 – 1.65 orders of magnitude (log10 CFU/g of skin). In conclusion, dietary CA affected numbers of *C. jejuni* in the gastrointestinal contents of chickens, whereas surface-treatment reduced or eliminated *C. jejuni* contamination in processed chicken carcass. Supported by the projects of Ministry of Agriculture of the Czech Republic, No. MZeRO07014 and Czech University of Life Sciences in Prague, No. CIGA 20142014.

200. Faulty fridges and mixed-ingredient salads – a dangerous combination

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ABSTRACT

Background: There is a trend towards healthier eating habits with increased consumption of ready-to-eat (RTE) salads. At the same time there has been an increased number of foodborne outbreaks associated with consumption of RTE salad. Foodborne pathogens may contaminate at several steps during the production of RTE salad, e.g. via contaminated water or cross-contamination from human handling. If this occurs, there is no process in the following production chain that inactivates pathogens. Consequently, the safety of RTE salad depends on good hygiene practices and a well-functioning cold chain. **Objectives:** The study aim was to investigate the growth of *Escherichia coli* O157:H7 in RTE salad, mimicking recommended fridge temperature (8 °C) and temperature abuse (15 °C) to evaluate the risk for the consumer. **Materials and Methods:** Two different matrices were studied in this experiment; leafy greens (baby spinach) and mixed-ingredient salad (baby spinach and grilled chicken). A green fluorescent protein (GFP)–labeled strain of *E. coli* O157:H7 were inoculated in low numbers and enumerated after 3 and 7 days of storage at 8 or 15 °C. The results together with a dose response model for VTEC were used to estimate consumer´s risk for infection when eating RTE salad contaminated with *E. coli* O157:H7. The relative risk was estimated using a @Risk Monte Carlo simulation with an inoculation level of 1 cfu/gram. **Results:** Storage of mixed-ingredient salad at 15 °C strongly supported growth of *E. coli* O157:H7. The risk of EHEC-infection after eating a 100 gram portion stored for 3 days, varied with

temperature. At 15 °C the risk was 30-120 times higher than at 8°C. **Conclusion:** Cold storage of RTE salad is essential to reduce the risk of foodborne disease from *E. coli* O157:H7.

201. Application of whole genome sequencing in investigations of *Campylobacter* outbreaks

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ABSTRACT

Objectives: Infection with *Campylobacter* is characterised as an acute gastroenteritis. In the EU, *Campylobacter* is the most notified zoonosis, with more than 200,000 annually reported human cases. In the EU summary reports, *Campylobacter* is the second most common causative agent of the notified food- and waterborne outbreaks. Investigations of *Campylobacter* outbreaks are complicated due to several reasons. A simple and robust typing method is lacking. *Campylobacter* isolates from human cases are seldom collected and saved. Also, *Campylobacter* can be isolated from multiple sources. We aimed to assess the use of the typing methods of PFGE (pulsed-field gel electrophoresis) and WGS (whole-genome sequencing) in outbreak investigations. **Materials and Methods:** *Campylobacter* isolates from human cases and potential sources of three *Campylobacter* outbreaks were analysed using PFGE (Pulse-Net) and WGS. Outbreak 1 was a family outbreak associated with consumption of unpasteurised milk. Outbreak 2 was associated with eating undercooked liver pâté at a wedding. Outbreak 3 occurred after a kindergarten visit to a dairy farm. **Results:** Outbreak 1: *C. jejuni* from humans and cattle shared indistinguishable PFGE profiles which was verified by WGS. Outbreak 2: *C. jejuni* from humans and one chicken had indistinguishable PFGE profiles. The human isolates were similar in WGS whereas the chicken isolate differed slightly. *C. coli* was isolated from one human case and a similar profile was identified in one chicken isolate. Outbreak 3: *C. jejuni* isolates from cases were indistinguishable in PFGE as well as three cattle isolates. In WGS, the human isolates and one cattle isolate were similar. **Conclusions:** PFGE proved to be a useful tool for point-source outbreaks which was verified by WGS.

202. Searching measures to protect the elderly against listeriosis

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ABSTRACT

Objectives: Finland is one of the leading countries among the EU member states regarding the number of human listeriosis. The symptoms of this foodborne infection vary from mild or nonexistent to fatal due to personal health status of the consumer. Large outbreaks in Finland are rare but sporadic cases appear. Young children, elderly people and immunocompromised individuals are regarded as risk groups, whereas fish, meat, raw milk and vegetables are considered main sources. However, most often the worst cases have arisen among the elderly, who are also heavy users of fish and fish products. The number of retired people is increasing as is the average lifetime all over the EU. In Finland the elderly are urged to lead independent life at home as long as possible. The target of an ongoing project is to assess the *Listeria monocytogenes* (LMO) exposure caused to the elderly by fish products and to consider risk management measures needed for LMO prevention especially for the elderly living at home. **Materials and Methods:** Hygiene grading was exploited in order to weigh the possibility of microbial endurance and cross contamination during processing of fish products. Data on LMO prevalence and concentration in certain foods collected in different projects and national statistics were used. The Food Spoilage and Safety Predictor (FSSP) was used for LMO growth prediction. Concentration of samples with detected LMO but concentration below the limit of determination was estimated by Bayesian modeling. **Results:** LMO is able to survive in temperatures used for some fish products. Depending on the process and layout of the establishment, cross-contamination is possible before packing. At home product handling, refrigerator temperatures and the way of using fish products may favour LMO growth. Low LMO concentration may increase onto a level that poses a person's health to risk. **Conclusions:** Even a few number of LMO colonies (below detection limit) may be enough to affect a person's health and thus account for sporadic cases, especially if he or she belongs to a risk group. Novel aids are needed, and special knowledge should be utilized in order to develop efficient protection measures.

203. Risk assessment of human pathogenic *Yersinia enterocolitica* in minced meat in Belgium

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ABSTRACT

Yersiniosis is the third most common zoonosis in Europe, and is mainly caused by *Yersinia enterocolitica*, particularly bioserotype 4/O:3. The main reservoirs of these pathogens are domestic pigs and pork is considered the main source of human infections. More specifically, the consumption of raw minced meat has been shown an important risk factor for human infections. A modular process risk model (MPRM) was used to perform an exposure assessment of human pathogenic *Y. enterocolitica* in minced meat produced by industrial meat processing plants in Belgium. The model described the production of minced pork starting from the contamination of pigs with *Y. enterocolitica* at time of slaughter. Data obtained from previous studies about the prevalence and contamination level of incoming pigs and their carcasses during slaughter were used as input in the model. Data from the literature were used when observational data were not available. For *Y. enterocolitica,* no dose response model is available. Therefore, the end point of the exposure assessment was the prevalence of *Y. enterocolitica* in minced meat packages (just before consumption) and the number of colony forming units in these contaminated products. The entire model was simulated with Monte Carlo techniques using @Risk software. A reduction of the level of carcass contamination would reduce the proportion of *Y. enterocolitica* positive minced meat packages. Storage of minced meat by consumers has an important effect on the concentration of *Y. enterocolitica* in minced meat at the time of consumption.

204. Microbiological risks and benefits of the consumption of raw milk and raw dairy products

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ABSTRACT

The Scientific Committee of the Belgian FASFC has published several opinions where the objective was to assess the risks and benefits of the consumption of raw milk and raw dairy products (from multiple species), based on an elaborate literature study and expert opinion. In Belgium, the most relevant microbiological hazards related to the consumption of raw cow, sheep and goat milk are *Campylobacter*, *Salmonella* and human pathogenic verocytotoxin producing *E. coli* (VTEC). Raw donkey and horse milk generally has a high microbial quality. A risk assessment at an European level identified the same hazards and included also *Brucella* spp. in sheep milk, *Mycobacterium bovis* in cow milk and tick-borne encephalitis virus in milk from several species. As potential emerging hazards, *Coxiella burnetii* and *Mycobacterium avium* subsp. *paratuberculosis* (MAP) were identified. The risks of raw dairy products (especially (semi-)soft cheeses) in Belgium are mainly linked to *Listeria monocytogenes*, VTEC, *Staphylococcus aureus, Salmonella* and *Campylobacter*. Dairy products from cows with subclinical mastitis may contain high numbers of *L. monocytogenes* and *S. aureus*. *L. monocytogenes*, VTEC and *S. aureus* have been identified as microbiological hazards in raw milk butter and cream albeit to a lesser extent because of a reduced growth potential of these pathogens compared to cheese. In endemic areas in Belgium or abroad, raw dairy products may also be contaminated with *Brucella* spp., *Mycobacterium bovis*, the tick-borne encephalitis virus, *C. burnetii* and MAP. Based on the health threat due to the possible presence of human pathogens, it is stated that heat treatment of milk before consumption and before the manufacturing of dairy products is important to insure their safety. Concerning so-called beneficial (nutritional and

health) effects attributed to raw milk consumption, it was concluded that there is no scientific evidence that, with the exception of an altered organoleptic profile, heating raw milk would substantially change its nutritional value or other hypothesized benefits. The benefits of probiotic and lactic acid bacteria are not relevant due to low numbers encountered in raw milk.

205. Prevalence of foodborne bacterial pathogens in irrigation water used by produce farms in the Lower Mainland of British Columbia, Canada

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ABSTRACT

Objectives: Foodborne outbreaks associated with fresh produce have become increasingly common worldwide. Many of these outbreaks are caused by bacterial pathogens including *Salmonella enterica* (Se), *Listeria monocytogenes* (Lm), and verotoxin-producing *Escherichia coli* (VTEC). A potential etiological factor is contaminated water, which has been shown to effectively transmit bacteria through crop irrigation. The main objectives are to (i) investigate the prevalence of VTEC, Se, and Lm in irrigation water in the lower mainland of British Columbia (BC), Canada; (ii) evaluate the effectiveness of coliform/E. coli count as an indicator for the presence of foodborne pathogens; and (ii) identify environmental factors affecting the prevalence of bacterial pathogens. **Materials and Methods:** Water samples were collected monthly at October to March and semi-monthly at April to September from six produce farms in two BC watersheds (i.e., Serpentine [S1] and Sumas [S2] respectively). S1 is adjacent to urban developing areas, while S2 is surrounded by a number of cattle farms. An immunoblot assay was used for detection and isolation of VTEC using antibodies to directly target verotoxin; allowing for the detection of all VTEC strains, including non-O157 serotypes. The virulotype of VTEC was determined by multiplex PCR. Detection and isolation of Se and Lm were conducted following Health Canada methods, MFHPB-20 and MFHPB-30 respectively. The water samples were analyzed for (i) pH; (ii) temperature; (iii) turbidity using a microplate reader; and (iv) total coliform/E. coli count using 3M Petrifilm. **Results:** Up to date, eleven VTEC isolates and twelve Lm isolates were recovered from the two sites. All of the VTEC isolated were recovered from S2, with three distinct virulotypes observed. Eight Lm isolates were recovered from S1and four from S2. Overall, the pH of water in S1 (7.56+/-0.04) was significantly higher than that in S2 (6.52+/-0.23) ($p<0.05$). The coliform/*E. coli* counts vary across the farms, with the highest being 4/33 per ml and the lowest being 0/0 per ml. No correlation was observed between the coliform/*E. coli* count and the presence of VTEC or Lm. **Conclusions:** These data suggested that: (i) the coliform/*E. coli* count may not be an effective indicator for the presence of foodborne bacterial pathogens in irrigation water; (ii) the prevalence of foodborne pathogens may be affected by landscape factors and physicochemical characteristics of the water.

206. Tracking of *Campylobacter* genetic diversity and contamination of broilers carcasses in Poland

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ABSTRACT

Objectives: Chicken meat and its products are commonly consumed in Poland, but campylobaceriosis is rarely reported. The objective of this study was to investigate a molecular characteristic and a clonal relationships as well as antimicrobial resistance of *Campylobacter* isolates of poultry origin in Poland. **Materials and Methods:** A total of 254 *Campylobacter* isolates from broiler's caeca and corresponding carcasses were used. The microbroth dilution method to establish MIC (minimal inhibitory concentration) for six antimicrobials: gentamicin, streptomycin, erythromycin, nalidixic acid, ciprofloxacin and tetracyclines was used for determination of AMR profiles. PCR was applied for identification of 11 *Campylobacter* virulence genes (VG): flaA, flhA, cadF, docA, cdtA, cdtB, cdtC, ciaB, iam, wlaN, and virB11. Pulsed-filed gel electrophoresis (PFGE) with *Smal* restriction enzyme was used. The composite cluster analysis covering PFGE, AMR and VG profiles was performed using the UPMGA method and the average from the experiments as coefficient for similarity and correction for internal weights. **Results:** The composite analysis separated the *Campylobacter* isolates into 178 distinct types. The vast majority of the composite types (97.2%), including 95 *C. jejuni* and 78 *C. coli*, covered only one to three isolates but five composite profiles comprised from four to ten strains. It was also found that 11 pairs of *C. jejuni* and 10 pairs of *C. coli* isolated from caeca and the corresponding carcasses, at the same geographical place and sampling time possessed identical PFGE, AMR and VG patterns which may indicate a cross-contamination of chicken carcasses during the slaughter process. Furthermore, the identical composite patterns were found in *Campylobacter* isolates originating from different geographical regions of Poland or sampled at different time. No correlation between particular AMR or VG types and PFGE patterns was observed. **Conclusions:** *Campylobacter* poultry population in Poland was characterized as highly genetic diverse. Moreover, the evidences for cross-contaminations during the chicken slaughter process were found. Such epidemiological studies provide information necessary for assessment of *Campylobacter* transmission along food chain. The results of the study indicate the need for further monitoring of poultry that will provide information necessary for human *Campylobacter* infection control.

207. Microbial inactivation by novel food technologies – Overcoming challenges for the Implementation in the food industry of the future

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ABSTRACT

Novel preservation technologies enable to maintain excellent food quality. To handle the risk originating from food-borne pathogens and spoilage organisms, research and development is challenged by several questions: underlying inactivation mechanisms, kinetics, adaption and tolerance formation, reproducible technological process parameters and matrix effects. Pulsed light (PL) represents a fast, tailored and residue free technology that – via high frequency, high intensity pulses of broad-spectrum light rich in the UV fraction – is capable of inactivating microbial cells and spores. In the present work main factors related to an effectively PL-treated food have been elaborated by inoculation trials with *Listeria monocytogenes* strains. The influence of a complex food matrix alone or in combination with different process parameters has been tested. Further, the impact of prior sublethal injury representing typical food manufacture stresses on bacteria has been assessed. The tolerance formation potential against long-term application of PL was examined. Electron Microscopy elucidated an insight into the in-situ morphology of PL-treated cells. The findings showed that strain specific differences among tested *L. monocytogenes* strains influence the sensitivity to PL as well as the treatment parameters distance and time. Further, strong evidence of homologous tolerance formation in both strains was found when applying PL over an extended period of time. On closer examination of the survival curves, the Weibull model was found to be best suited for describing the process of PL activation on *L. monocytogenes* inoculated at lab scale. Further, results obtained for the complex food matrix have shown a significantly reduced inactivation degree but comparable survival curves. Tolerance formation after application of sublethal stresses could not be observed in combination with PL. This work provides increased knowledge regarding the antimicrobial effects of PL on a representative set of *L. monocytogenes* strains as well as the impact of repeated and consecutive stress factors on the tolerance development against PL. Hence future research should focus on the effect of tolerance formation related novel technologies of pathogenic microorganisms and its relevance for future risk assessments. This should include demonstration and quantification of this phenomenon in-vitro as well as under real processing conditions in order to facilitate approval of these technologies.

Drivers for emerging issues in animal and plant health

208. Detecting plant pest and stress symptoms in trees using remote sensing

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ABSTRACT

Tree pests increasingly threaten forests and tree crops, because pest species and their vectors cope more quickly with climate change than their tree hosts do, and because increasing trade facilitates the introduction and spread of new pests. Remote sensing is often used to map the damage that pest outbreaks cause to forests. Yet, usually these applications only detect extensive forest areas with advanced levels of damage, limiting their usefulness in combating pest spreads. For remote sensing to contribute to an early warning system for pest spread, it needs to detect small forest areas, or better, individual trees that display stress symptoms. Furthermore, these trees need detection early enough for forest managers to disrupt the reproduction or dispersal cycles of the pest. We developed remote sensing based methods for the early detection of symptoms of tree crown decline in a pine forest in Extremadura, Spain, an area at increasing risk of Pine Wilt Disease caused by *Bursaphelencus xylophilus* and currently spreading through Portugal. We collected narrowband hyperspectral, colour-inrared, and standard colour imagery from aircraft over a 3000 ha Pinus sylvestris forest at spatial resolutions ranging from 11 to 70 cm. Furthermore, we collected satellite imagery from the Pleiades, Worldview 2 and Skybox satellites at 2 m spatial resolution for the same area. The latter represents a new generation of Earth orbiting satellite constellations that acquire frequent (ie sub-weekly) colour-infrared imagery of very high spatial resolution. We evaluated the remote sensing data, and a suite of spectral indices derived from them, for their ability to detect varying degrees of canopy discoloration, defoliation, and die-off in pine trees using field observations of ca. 300 pine trees. Our results demonstrate the ability of high resolution airborne imagery to not only advanced but also early stages of canopy decline, provided that imagery with a sub 25 cm resolution is available to delineate individual tree crowns, and case-appropriate spectral indices are used. Our results also show the potential of the data generated by new constellations of satellites that combine relatively high spatial resolutions (2 m) and frequent revisit times, such as Skybox, to monitor forests for pest symptoms. Nonetheless their spatial resolution, which is coarser than that obtainable from aerial surveys, prevents the detection of pest symptoms that don't affect the entire tree crown.

209. The impact of changes in green offal inspection on detection of public health, animal health and animal welfare hazards in cattle, small ruminants and pigs

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ABSTRACT

Objectives: Weaknesses of the current meat inspection system are well recognized in the European Union, where significant actions have been initiated in order to review and modernize this resource-intensive activity. The aim of this study was to qualitatively assess the changes in detection of public health, animal health and animal welfare hazards in cattle, small ruminants and pigs posed by down-scaling to a visual-only inspection of green offal or by removing green offal inspection completely. **Materials and Methods:** Three green offal inspection scenarios were considered: i) current inspection as laid down in legislation, ii) visual-only inspection, and iii) absence of green offal inspection. Hazards were identified based on their ability to cause detectable lesions in green offal. For assessment of changes in detection, qualitative likelihood categories were used: negligible, very low, low, moderate and high. Available scientific literature was used, and, where no literature was available, experts were consulted to inform the assessment. **Results:** With respect to public health and animal health, the conditional likelihood of detection with the current green offal inspection was found to be low for eleven out of the twenty-four selected hazard-species pairings and very low for the remaining thirteen pairings. A difference between current and visual-only inspection scenarios was observed only in three hazard-species pairings all of which are hazards only relevant to animal health. For foot-and-mouth disease in cattle, the switch from current to visual-only green offal inspection would reduce the likelihood of detection from low to very low. For tuberculosis in cattle and pigs, moving to visual-only green offal inspection would lead to a change from very low to negligible. **Conclusions:** The results strongly suggest that the contribution of current green offal inspection to risk mitigation is very limited for public and animal health hazards. The removal of green offal inspection would reduce the detection of some animal welfare conditions. For all selected public and animal health as well as welfare hazards, the reduced detection could be compensated with other pre-harvest, harvest and/or post-harvest control measures including existing meat inspection tasks.

210. A transitional model for the evaluation of Rift Valley fever virus transmission in North Africa and related surveillance approaches

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ABSTRACT

Objectives: Vmerge project (Emerging, Viral Vector-Borne Diseases, FP7-KBBE project) is in an international project involving 16 veterinary diagnostic and research Institutes located in 12 countries, including 5 countries from western and northern Africa (Senegal, Mauritania, Morocco, Tunisia, and Egypt). The project intends to address the risk of introduction and spread throughout northern Africa and Europe of some mosquito-borne and Culicoides-borne diseases and enhance epidemiological surveillance strategies for the early detection of infections. In this context, Rift Valley fever (RVF) is one of the most important mosquito-borne zoonoses, able to cross international borders and to cause devastating effect on animal health and food production systems. Climate changes and the presence of competent vectors in the most of actual RVF-free countries lead to consider RVF among the most significant emerging viral threats for public and animal health in Europe and in the Mediterranean basin. One of the objectives of Vmerge project is to model vector-population dynamics and RVFV transmission to assess its spread capacities, evaluate the efficacy of existing surveillance approaches and propose new surveillance strategies in countries more at risk of RVF introduction. **Material and methods:** Mathematical models are often used to explain the mechanisms of biological processes, providing effective tools to plan measures for public health protection. Within Vmerge project, an epidemiological RVFV transmission model was developed, coupling climate (temperature, rainfall), biology of Aedes and Culex mosquitoes, and hosts densities data. The model was calibrated on vectors and epidemic data as observed in Mauritania and applied to the North African countries to estimate areas and seasons more suitable for disease spread. **Results and discussion:** The modeling approach followed in our study allowed the production of detailed spatio-temporal maps on the risk of RVFV transmission in North Africa, thus providing the bases for the assessment of alternative active and passive surveillance strategies aiming at early detecting the RVF infection. The prompt recognition of RVFV introduction and spread in North African countries is of paramount importance for limiting the impact of this disease on the animal husbandry and food production system of these countries as well as for their crucial role as possible doors for RVF entry into southern European countries.

211. Drivers for Ebola virus spillover in West Africa

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ABSTRACT

Objectives: Ebola viruses can cause fatal disease in humans and other animals such as monkeys, chimpanzees and duikers. Human outbreaks in West Africa are thought to originate from contact with susceptible animals infected with Zaïre Ebola virus including bats, the presumed reservoir. Hunting, preparing and eating wildlife for bushmeat are believed to be the main drivers of these spillover events. However, drivers are unlikely to occur in isolation. Identifying wider contributing drivers and understanding how they are connected may help in determining the origins of these events and prevent future outbreaks. Our objectives were to identify drivers of spillover in West Africa and provide network diagrams highlighting the main drivers and their interactions. **Materials and Methods:** An initial list of possible drivers from published literature was used as a starting point. Supporting data were collected through a literature search in Web of Science using a pre-defined search string which identified 566 papers. Following expert screening, 36 were considered relevant and a subset of these was used for data extraction. Relevant arguments in the selected texts were converted into linked drivers based on the original list, with the list refined during the extraction process. Multiple diagrams were produced using data-visualisation and network analysis techniques. The diagrams showed the drivers and their interactions, any mutual implication of the linkages, geographical locations and the strength of and citations used in the original argument. **Results:** Although bushmeat-associated drivers such as hunting were strongly represented, they were found to be extensively linked to broader drivers operating at local, national and international levels. Main interconnected drivers included human food security and livelihood concerns, urbanisation, increasing populations, deforestation, agro-economic changes, infrastructure development and international market pressures. These were linked to ecological and environmental drivers such as wildlife ecology and demographics, ecosystem changes and seasonality. **Conclusions:** Understanding Ebola virus spillover in West Africa requires a broad, transdisciplinary perspective which looks beyond the presumed roles of bushmeat and hunting. This necessitates consideration of the multidimensional interactions between drivers with wider social, economic, political and ecological origins.

212. *M.bovis* Tb in NorthWestern Italy: a fifteen-years survey for comparison of strain profiles in human and animal populations

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ABSTRACT

Objectives: *M.bovis* is the causative agent of bovine tuberculosis (bTB) with wide host range among livestock, wildlife and man, that can became infected by contaminated foodstuff (mainly raw milk) or by direct contact with infected animals. Eradication plans in industrialized countries had permitted the decrease of disease in farms. In NorthWestern Italy (NWI) bTB is near to be eradicated. Nevertheless, a low risk of human contagious is still persisting due to residual outbreaks. Recently, EFSA has included *M.bovis* among transmissible hazards via raw milk. This works describes genetic features and correlation among strains isolated from cattle outbreaks, hospitalised patients and wildlife, between 2001-2014 in NWI. **Materials and Methods:** All isolated strains from animal and man were identified firstly as *M. tuberculosis* complex and then genotyped by spoligotyping and VNTR- typing analysis for ETR-A, B, C, D, E. Data were collected in a DataBase (Microsoft Access 2000) and analyzed with SAS . Results In NWI 2170 cattle strains from 594 outbreaks, 22 wildlife strains (mostly from wild boar) and 34 human strains were characterized from 2001 to 2014; 14 out of 34 strains among people were not found in animals species; genetic profiles of the remaining 20 strains have their counterpart in animal populations and 13 of these belong to the four most frequent *M.bovis* profile detected in cattle (three of which present in wildlife too). People born before 1960 were commonly found infected with strains often isolated in bTB outbreaks. This evidence is statistically significant (χ^2 =5.62, p <0.01; OR 7.44, IC 95%). **Conclusion:** On the basis of collected data, it can be pointed out people born after 1960 were in the main infected with strains other than found in the local animal population; we can suppose that they become infected outside NWI. Otherwise elderly people could have been infected before eradication plans, in a period when bTB prevalence was significantly high. Tuberculosis is a chronic disease that in often causes latency, evolving clinically in about 10% of cases also many years after the first infection. Eradication plans in livestock are still confirmed as the principal tool to reduce *M.bovis* infection in human population. Genetic strain characterization is significantly important to monitoring new entries in a territory, and it is functional to understand the origin of the infections targeted to assess the risk of humans and animals related infections.

213. *Xylella fastidiosa* and its vectors in Europe

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ABSTRACT

Introduction and objectives: Following a request from the European Commission, the European Food Safety Authority (EFSA) provided a complete pest risk assessment and evaluation of risk reduction options (published in the EFSA Journal in 2015) prepared by the EFSA Scientific Panel on Plant Health. EFSA created a database of known host plants of *Xylella fastidiosa* and also mentioned the spittlebug Philaenus spumarius as a confirmed transmission vector in Apulia, Italy. The EFSA pest risk assessment describes the European putative vectors of the bacterium as well, all belonging to the same insect guild-the xylem sap feeders. Among them, there are confirmed vectors in the Americas (in the Cicadellidae, Aphrophoridae and Cercopidaeare families) and other species identified as potential vectors in the EU. A research question is: what are the limiting factors for a bacterium with a broad host range of over 300 plant species? Should the 'cold' countries be worried of this pathogen as it does appear to be present also in North America up to Canada on elm and maple? It is important to stress that even if current climatic conditions in the Northern EU countries could be limiting for the pathogen, it is possible that with climate change this climatic conditions will shift. **Materials and Methods:** to answer those questions, researches and field surveys should be carried out to improve our knowledge about potential insect vectors species which appear commonly in all EU member states, like the commonest: *P. spumarius, Cercopis vulnerata, Cicadella viridis*. A good study area could be primeval Białowieża Forest which is located in the North-East Poland: this is one of the coolest region in Poland, where some studies have already been conducted about spittlebugs. It is important to review all known information by extensive literature search in bibliographic databases and to conduct field surveys to determine how exactly those insects behave in 'cold' climates, such as in Poland, and to compare these results with their described ecology in warmer Mediterranean countries, such as Italy. Surveys for *X. fastidiosa* putative have already started also in other Mediterranean countries, such as Spain. **Results and Conclusions:** we can expect many different things from the literature screening and also from conducted field surveys. On the base of collected data on vector distribution and biology, it could be possible to better estimate the risk of *X. fastidiosa* in the 'cold' Northern EU Countries. Is this pest somehow a threat for Northern Europe or the species or subspecies features are limited by some factors? This remains nowadays an open question.

214. A cohort study on cause-specific mortality in dairy cattle

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ABSTRACT

The health and welfare of cattle may be assessed by mortality studies: however, while all-causes mortality studies give a general indication of trends and extent, cause-specific mortality data are unavailable and may be particularly

informative. Aim of our study was to investigate the causes of mortality in a cohort of dairy cattle in a Region of northern Italy (Piedmont). **Materials and methods:** The 'typical farm' was firstly identified by means of a descriptive analysis (i.e. by breeder association registration, herd size) of dairy farms active in Piedmont during the year 2011. A cohort of farms, stratified by province, was enrolled following predetermined inclusion criteria (breeders associates, herd size over 19 heads, willing to be involved in the project). Every calf, veal or cow died or euthanized during the period 01/04/2012-31/03/2015 was sent to post-mortem examination and ancillary diagnostics to identify the cause of death. Finally crude and age specific mortality rates (3 classes: 0-6 months, 7-24 months, >24 months) was obtained and the proportion of cause of death was calculated for each age-class. Data on denominators were drawn from the National Cattle Registry that suffers from an intrinsic underestimation of less than 15 days calves. **Results:** During the study period 15,289 cows (18,051.8 cow-years), belonging to 44 farms, were enrolled in our study. Crude mortality rate was 3.4/100 cattle-years (CI95% 3.2-3.7). The highest mortality rate was seen in the first age class (17/100 cattle-years, IC95% 15-19), mostly due to digestive disorders (43%), the lowest mortality rate in the second age class (0.9/100 cow-years, IC 95% 0.7-1.2) in part due to respiratory (37%) and digestive disorders (37%) while in adult animals mortality (2.9/100 cattle-years, IC95% 2.6-3.3) was mainly caused by digestive disorders (43%). **Conclusions:** Our crude rates referred to all causes mortality are consistent with national data. Mortality in the first age class could be overestimated: our effort to obtain a complete numerator may have faced an underestimation of the denominator. The proportion of digestive disorders in adults is higher than reported in literature, most of our cases are due to *Cl. perfrigens*. Currently within an international group, we are developing a classification system of the cause-specific mortality to be shared in order to allow international comparisons of bovine mortality data.

215. Cystic echinococcosis: risk factors of infection in human and animals (Case study: Province of Sidi Kacem, Morocco)

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ABSTRACT

A part of a large research project on five major neglected zoonotic diseases, ICONZ (Integrated Control of Neglected Zoonoses), including Cystic echinococcosis (CE), this study was undertaken in the Province of Sidi Kacem (North/west of Morocco) during a period of four years (April 2009-Marsh 2013). The main objectives were to determine the importance of Echinococcus granulosus (Eg) sources (CE prevalence of infection in ruminants and Eg prevalence of infection dogs), to identify the Eg's main strains present in the region, to evaluate the community knowledge on the disease and to indentify the dynamic of transmission to ruminants, dogs and human. The prevalence of infection in ruminants determined by post-mortem examination, of a total of 2,090 farm animals (1,302 sheep, 652 cattle, and 136 goats), carried out in the 10 abattoirs of the Province, the prevalence of infection in dogs was determined using bromhydrate arecoline purgation, while the evaluation of the community knowledge on the disease was carried out using a large-scale questionnaire survey undertaken at the population level in a total of 27 communes of the Province. Results showed a CE prevalence of 11%, 43% and 1.5%, respectively in sheep, cattle and goats. In dogs, the Eg prevalence of infection was 35.5%. Eg strain typing of a sample of 116 cysts revealed the presence, for the first time in Morocco, of G1 and G3 stains. Otherwise, surveys showed that 88.7% of respondents ignore the dog's role in disease transmission, whereas 39% allow their guard or sheep dogs to access to the family home including sometimes kitchen, and 54.3% feed them infected and inappropriate for human consumption organs. Furthermore, 61.2% of children do not wash their hands after petting or playing with dogs, 88.2% consume raw vegetables, and 68.9% of households still use traditional water sources. Thereby, it appears that CE is highly endemic in Sidi Kacem region. This zoonosis remains a major public health issue in Morocco and hence, the necessity for increased monitoring and global control of CE in the country.

216. Effect of dietary supplementation of layer pullets with a botanical additive on the growth performance, mortality, uniformity and caecal microflora

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ABSTRACT

Hy-Line W-36 is well known as one of the most efficient egg producing strains all over the world. However, it is hard for poultry producers to achieve the desired performance and uniformity goals among flocks due to their low feed consumption in comparison to other strains. Botanical feed additives have gained much attention in the last decade due to their natural growth promoting properties. A field trial was conducted to investigate the influence of supplementing pullet diets with a botanical blend containing carvacrol, cinnamaldehyde, and capsicum oleoresin on the growth performance (average body weight (ABW), average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR)), mortality, uniformity, and caecal bacterial count. A total of 60000 Hy-Line W-36 pullets were used in this trial from 2-12 weeks of age. The trial involved two dietary treatments. The number of birds per treatment was 30000 chicks divided into three replicate each containing 10000 chicks. Pullets of the first treatment were fed on a basal diet and kept as a control, while those of the second treatment were fed on a basal diet supplemented by 75 ppm of a botanical additive (Xtract, Pancosma Company, Switzerland). The feeding program involved three phases (starters 1 (2 to 4 wk); starter 2 (5 to 8 wk), grower (9 to 12 wk)). The bird's body weight and feed consumption were recorded on a weekly basis to determine the ABW, ADG, ADFI, and FCR. A total of 300 pullets (selected to represent different locations of the barn) per treatment were individually weighed. Mortality and uniformity were also estimated on a weekly basis. At the trial end, the left caeca of 30 birds per treatment were collected and used for examination and counting of caecal coliform and lactobacilli bacteria. The results were statistically analyzed via one way ANOVA using Statisitx 9©. The obtained results revealed that dietary supplementation of Hy-Line pullets with Xtract didn't result in significant difference in the overall ABD, ADG, ADFI, or FCR. However, the FCR was significantly lowered by 9.6 and 4.4% during 7 to 8 and 9 to 10 wks of age respectively in pullets fed on the Xtract diets. Mortality %, uniformity %, and lactobacilli count were not significantly differed among treatments. The caecal coliform count was significantly lowered by 27% in pullets fed the Xtract supplemented diet. It could be recommended to utilize higher levels of Xtract additive for delivering a better response.

217. Seroprevalence of *Toxoplasma gondii* in game ungulates as indicator of foodborne zoonoses risk

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ABSTRACT

Objectives: The protozoan *Toxoplasma gondii* is the most spread among parasitic zoonoses, and the consumption of raw or undercooked meat have been shown to be one of the main risk factor for human infection. *T. gondii* can infect many animal species and, among intermediate hosts, several ungulates are reported and may be source of infection for consumers, hunters and slaughterers (manipulation and handling of carcasses). Therefore, we performed a serological analysis in chamois (*Rupicapra r. rupicapra*), roe deer (*Capreolus capreolus*), red deer (*Cervus elaphus*) and mouflon (*Ovis musimon*), which represent the most consumed game meat in the Alps, in order to evaluate the potential foodborne zoonosic risk. **Material and methods:** Sera of game ungulates were gathered from two areas of Central-West Italian Alps. Overall 91 chamois, 74 roe deer and 63 red deer sera were sampled from area 1 (VB) during three hunting seasons (2011-2013) while 66 chamois, 44 roe deer, 25 red deer and 13 mouflon were sampled in area 2 (VC) during two hunting seasons (2013-

2014). For each subject age, gender and the shooting localities were registered. Sera were tested by a commercial ELISA kit and data were analysed through Generalized Linear Models. **Results:** Prevalence of area 1 were 3.3% in chamois, 24.3% in roe deer and 17.4% in red deer. Deer resulted significantly more infected than chamois. No significant effects of gender, age class, shooting localities and year were recorded on the probability of being positive. Prevalence of area 2 were 4.5% in chamois, 13.6% in roe deer, 8% in red deer and 46% in mouflon. Mouflon resulted significantly more infested than chamois. No other significant effects were recorded. **Conclusions:** The emerged seropositivities proves the presence of *T. gondii* in both study areas, in all the host species. Thus a wide spread of the protozoan in the Alpine ecosystem appears. As the contamination of pastures by cats' oocysts is the more likely transmission route, even if the transplacental one can not be excluded, the remote habitat-use of chamois could explain its lower infection than both deer and mouflon. Concerning zoonosic risk, mouflon and deer appear a more likely source of *T. gondii* infection for humans than chamois, although the risk associated with the consumption of its raw or undercooked meat or handling of its infected carcasses can not be completely excluded.

218. Tackling chronic fear in sheep through genetic selection

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ABSTRACT

Chronic fear in sheep has been defined as a welfare consequence in the EFSA Journal 2014(12):3933. With previous investigations outlining the influence that behavioural reactivity has on productive and reproductive performance in sheep. Moreover, animals which are highly reactive to humans and handling exhibit poor adaptation to their environment and experience high levels of stress, reducing their level of welfare. The objectives of the current research were: i) to evaluate the effects that behavioural reactivity has on production performance of sheep reared for meat production; ii) to estimate the heritability of temperament in Dorper breed; iii) with the ultimate purpose, to test the feasibility and reliability of introducing new 'green' selection traits such as temperament into commercial breeding of sheep in order to improve animal welfare. A data-set from 161 Dorper sheep (72 ewes, 86 lambs and their sires) managed under semi-intensive production system, and more than 720 records was analysed for estimation of the genetic parameters for temperament and correlations of behavioral reactivity with production traits. Temperament of the animals was evaluated based on a subjective method, using a 5-points scoring system at weighting, while spending 30 s on the scale ('scale' test, described by Dodd et al. 2012 Appl. Anim. Behav. Sci. 140:1–15). The heritability (±SEM) of temperament in Dorper breed was found to be low (0.10±0.03). Significant negative phenotypic (rp = -0.40, p≤0.05) and genetic (rg = -0.44, p≤0.01) correlations were found between ewe temperament and body weight, thus, the more reactive the ewe the lower its body weight. Correlations between temperament and litter size in ewes were negative and negligible (p>0.05). Significant genetic correlations were found between lamb temperament and pre-weaning growth rates (rg = -0.44±0.07, p≤0.05) and post-weaning growth till the age of 120 days (rg = -0.52±0.08, p≤0.05). Thus, selection for calmer animals will have a positive impact on lambs' growth rates during first three months of life, and vice versa. Negative correlations between temperament and production traits, in both ewes and lambs, suggest that selection against animals that are highly reactive in order to improve animal welfare and ease of handling would not have detrimental impacts on productivity in sheep reared for meat.

219. A new pest in vineyards: Approaches to sustainable management to support indigenous people's ethics

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ABSTRACT

Global agriculture has been increasing the yields of several crops, by using agrochemicals. Amongst them, pesticides produced several side-effects such as, insect acquired resistance, environmental degradation and human health problems. To promote sustainable agriculture, important efforts have been made during the last 30 years to find alternatives to pest control, which include habitat manipulation, biological control and use of alternative products to broad-spectrum insecticides, such as plant extracts, natural oils and feeding deterrents. Adults of the grass grub *Costelytra zealandica* (Coleoptera: Melolonthinae) hover and land on vine plants, producing severe damage to vine leaves (http://bioprotection.org.nz/news/grass-grub-beetle-invasion-captured-camera). Kono Beverages has strong ethical concerns about nature conservation, rooted on their Maori (New Zealand's indigenous people) cultural heritage based on kaitiakitanga which is the spiritual consideration of protecting the land for descendants. This encourages the search for sustainable solutions for this pest problem without the use of synthetic pesticides. Here, we investigated the distribution of adult grass grub within vineyard blocks and the use of feeding deterrents such as kaolin particle films and diatomaceous earths in a Kono vineyard in the Awatere Valley, Marlborough, New Zealand. The feeding deterrents were applied at a rate of 400 L ha-1 (20 g L-1), sprayed onto the vine foliage in a randomized block design. A generalized linear model (GLM) was used to analyze the data (link=log; family=Quasi-binomial). The adults' distribution within vineyard blocks was assessed by counting and removing the *C. zealandica* adults after they landed on vine plants, and this was analyzed by a GLM (link=log; family=Quasi-Poisson). The feeding deterrents significantly reduced the damage on vine plants by 37% ($\chi2=6.25$; df=284; $p<0.001$) compared with control, with no statistical difference between the treatments. Furthermore, significantly higher numbers of *C. zealandica* adults were found at the edge of the vineyard block, when compared to the centre ($\chi2=2033.59$; df=198; $p<0.001$). The above suggests that these feeding deterrents should be applied only to the edge of the vineyard block. If this approach is applied, an overall reduction in types and volume of external inputs is achieved. In addition, this management is in agreement with the spiritual perception of kaitiakitanga.

220. Activities within the fight against antimicrobial resistance in food producing animals

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ABSTRACT

The objective of the study is to make reference to activities, which have recently been carried out by a number of international organizations engaged in the field of human and animal health protection. New legislative modifications concerning antimicrobial resistance are discussed as well. As far as antibiotic policy is concerned, we also described the situation in the Czech Republic with focus on the National antibiotic programme content. The sales of antimicrobials in the Czech Republic has been under surveillance since 2003, now in accordance with European Surveillance of Veterinary Antimicrobial Consumption programme. In 2013 the Czech authorities agreed to participation in the pilot testing project concerning the consumption of antimicrobials on pig farms. National antibiotic programme of the Ministry of Health in the Czech Republic was established in 2009. This programme is based on the valid Action Plan with 11 priority points. There is an active Working Group for antimicrobials of the Ministry of Agriculture which operates in the area of veterinary medicines including medicated feed for food producing animals. Antibiotics play irreplaceable role in medicine, nevertheless there is however a risk, that we might slowly enter post-antibiotic era. For further progress, international cooperation and consistent implementation of measures in regions are necessary.

221. Risk assessment of amoebic gill disease (AGD)

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ABSTRACT

The Norwegian Food Safety Authority (NFSA) asked the Norwegian Scientific Committee for Food Safety (Vitenskapskomiteen for mattrygghet, VKM) to assess the risk of amoebic gill disease (AGD) in order to evaluate whether the disease should be listed on the national list of aquatic animal diseases. The VKM Panel on Animal Health and Welfare, established a project group consisting of both VKM members and external experts to answer the questions from the NFSA. Amoebic gill disease in farmed Atlantic salmon (Salmo salar) was originally described in Tasmania, Australia, in the 1980s and has continued to cause severe economic losses in this region. Since the 1990s, AGD has occurred sporadically in different farmed fish species in the Mediterranean Sea and the North-East Atlantic. In Norway, AGD was observed for the first time in association with health problems in farmed Atlantic salmon in the autumn 2006. Since 2010, the occurrence of AGD in farmed Atlantic salmon has increased significantly in the North-East Atlantic. The disease affects Atlantic salmon in the seawater phase, in particular post-smolts during the first autumn in sea. In Norway, AGD has additionally been observed in farmed rainbow trout (*Oncorhynchus mykiss*), ballan wrasse (*Labrus bergylta*) and in wild caught corkwing wrasse (*Symphodus melops*) used as cleaner fish in salmonid farms. Amoebic gill disease caused by Paramoeba perurans is a serious health risk to farmed Atlantic salmon and rainbow trout along parts of the Norwegian coast. The amoeba can cause high mortality, poor fish welfare and reduced growth if not treated at an early stage. High temperature and high salinity are major risk factors. Paramoeba perurans is present along major parts the western coast of Norway, together with diagnosed gill disease. Infections have been difficult to control in farms with open operation. Sporadic detections have been made as far north as Troms. Given the suitable conditions, P. perurans might establish further north. Restriction on movement of fish from affected areas could delay the spread, but probably not prevent it. Much of the existing knowledge on P. perurans comes from Tasmania. The relevance for Norwegian conditions is uncertain. Knowledge on the infection reservoir and spreading dynamics is lacking. Traditional hygienic measures, such as coordinated treatment and fallowing, could reduce the severity of transmission and infection in enzootic areas.

222. Application of in silico biology in livestock vaccine development

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ABSTRACT

Parasitic diseases continue to cause huge agricultural loses, morbidity and mortality in humans, domestic livestock and wild animals. During transmission of vector-borne diseases like malaria, sleeping sickness, Theileriosis etc., expression of parasite proteins occur in both the host and vector life cycle stages. There are also interactions between proteins expressed by the vector, host and the parasite. The pathogenesis of such diseases can be unraveled by understanding protein-protein interaction at the molecular level. Studies on such protein-protein interactions are limited by lack of data on the protein domains that interact with each other as well as models that can describe such inter-species interactions. East coast fever (ECF) is a fatal, tick-borne disease caused by the protozoan parasite *Theileria parva*, which is transmitted by the *Rhipicephalus appendiculatus* ticks. The disease is endemic in eastern, central and southern Africa, where it causes great economic losses of up to 200 million dollars annually, and puts the lives of more than 25 million cattle at risk in 11 countries. An effective vaccine against the disease is still elusive and the Infection and Treatment method (ITM) currently in use has limited application due to the use of whole live organism. These limitations can be eliminated by use of subunit vaccines, which are antigenic proteins that can induce a protective immune response. This approach is being used to identify candidate vaccines but is limited low availability of protein sequences and also models to demonstrate such interactions. The aim of this work is to develop a model that can predict protein-protein interactions between *T. parva* and its tick vector, *R. appendiculatus*. This model can be used in the development of a vaccine for East Coast Fever. Predictions of the interactions will be done based on homology, gene ontology terms and protein domain data in addition to the interaction databases. Support Vector Machine (SVM) classifier, will be applied to identify interacting *T. parva* and *R. appendiculatus* proteins. The output of this project will be identification of protein subunits expressed during the tick vector infection by *T. parva* which have potential to be developed into a vaccine for ECF.

223. Parallel animal welfare regulations

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ABSTRACT

Animal welfare and animal health are important aspects of food safety, and official control is carried out to ensure compliance with governmental legislation. However, in many countries animal welfare is not regulated and controlled only by the authorities but also through private standards developed by the industry, retailers or non-governmental organisations. Hence, many farmers have more than one animal welfare regulation to comply with. Such private standards can be more or less voluntary for a farmer to affiliate to. The aim of this study was to investigate the relations, both in theory and in practice, within and between the official binding animal welfare legislation and private standards concerning the requirements and control of dairy cows at farms, using a Swedish example. The study was carried out by making text analysis of regulations and corresponding guidelines, and by collecting outcome results from animal welfare controls made by the authorities and by the food company Arla, which has a standard that all farmers delivering milk to their dairy plants have to comply with. We saw that requirements can be expressed similarly but result in different outcomes e.g. accepted welfare levels, and vice versa. This illustrates the importance of discussing the measurements applied, in order to be able to create transparency and predictability both within and between animal welfare regulations. Our study also showed that the legislation had more of an individual animal perspective, whereas the private standard to a larger extent was based on group evaluation. Both legislation and standard covered resource-, management- and animal-based requirements. The official animal welfare inspectors' controls were more detailed compared to the private standard. However, at the time of on-farm control, the official inspectors made more remarks on resource-based requirements, even if the main non-compliance was animal-based (dirty animals). The Arla auditors to a larger extent made remarks on the animal-based requirements, for example dirty animals and long claws, but the main non-compliance for Arla was about insufficient cleaning of stables/cowsheds. By illuminating differences and similarities between these regulations, the transparency of animal welfare control can increase for farmers, inspectors and consumers. This may help avoiding confusion and ensure compliance, resulting in an acceptable or improved animal welfare level.

224. Fungal contaminants of *Pirus communis* var. bambinella: identification and growth characterisation

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ABSTRACT

Bambinella is native to the Maltese Islands and is cultivated for horticultural and agricultural purposes. Fungal pathogens cause premature fruit spoilage, leading to the loss of fruit during the post harvest phase. Latent infections from fields result in mycelial growth and sporulation of a number of fungal fruit pathogens and potentially production of mycotoxins during storage. The primary objective of this research project was to characterise the fungal contaminants of the small Maltese June Pear, *Pirus communis* var. bambinella. Hereafter, the aggressiveness of these fungi was assessed at different temperatures in order to perform a comparative assessment of their growth kinetics. Fungal spores were recovered from diseased fruits following washing steps in sterile water and a mild detergent. Aliquots from the washings were used to isolate predominant fungi which were then sub-cultured on separate isolation plates. The isolated fungi were

identified to the species level by means of colonial and microscopical features compared to known isolates. For the aggressiveness study, a colony of fungus was used to prepare the conidial suspension which was then suspsnded in 10% Tween 80 solution. 10µL of diluted conidial suspension was used to inoculate the point of intersection of the culture medium. Four different temperatures per species were studied (10°C, 15°C, 25°C, 30°C). The mycelium size was measured using a ruler and an average of the four diameters drawn onto the plate was calculated. Fungi isolated from healthy and diseased Bambinella fruit included *Cladosporium* spp., *Penicillium* spp., *Alternaria* sp. and *Ulocladium* sp.. All isolates are known to be causative factors of fungal diseases in pears. However, all these isolates, especially *Cladosporium* spp. have been found to be quite common air contaminants especially in the Maltese islands. Based on the kinetic studies, the order of most to least aggressive is as follows; *Ulocladium* sp., *Alternaria* sp., *Penicillium* spp. and *Cladosporium* spp., The fastest growth seemed to lie at a temperature around 25°C. Growth at 30°C was still positive but slower while growth at lower temperatures of 10-15°C was much slower. This suggested that temperatures above or below the optimum are limiting the fungal germination and growth. Further studies will focus on assessing in vivo the impact of fungal contaminants on the quality of storage Bambinella fruits.

225. Food offences and organized Crime in Southern Italy

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ABSTRACT

Beyond the traditional mechanisms of criminal justice, the author seeks to describe principles and practices of mafia type associations involved in food offences. According to European Food Safety Authority, the study contends that most theoretical frameworks encompass values, aims and processes that have as their common factor attempts to repair the Harm caused by criminal and antisocial behaviours. We' ll present statistical data with a criminological approach in this Eco-food mafia perspective .

226. Innovative methods and tools for a multidisciplinary risk management in poultry slaughterhouses

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ABSTRACT

Objectives: The aims of controls in slaughterhouses are to ensure food safety and quality as well as to monitor animal health and welfare. Control efficiency can be guaranteed by a multidisciplinary approach. Indeed, thanks to their knowledge of process and product, the active participation of food business operators (FBOs) to poultry meat control has to be enhanced. But, in accordance with European regulations, determination of sanitary actions depending on the associated risk has to be under the responsibility and the supervision of official veterinary services (OVS). In this context, it is necessary to implement an adapted control system taking into account respective competences and organisational constraints of FBOs and OVS. **Material and methods:** A group of multidisciplinary experts developed an innovative system to manage efficiently risks in poultry slaughterhouses and provide decision-makers with useful methods and tools for a new approach of poultry meat inspection. They listed the inspection tasks to be performed, defined the articulation between FBOs and OVS to implement these tasks and set indicators of achievement. **Results:** Four tasks to be performed were selected: analysis of the Food Chain Information, ante and post mortem examinations and feedback to the farmer. For each task, a set of indicators was determined and a warning value was set for each of them. The indicators must be monitored by FBOs; if the threshold value was exceeded, they must alert OVS that managed the alert and implemented adapted actions. Record sheets were developed to register indicator values and actions taken by FBOs and OVS in order to provide evidence of the implementation of health inspection and to ensure data traceability. Standardised documents describing lesions leading to carcass condemnation and training programs were developed to provide reliable tools helping operators to detect post mortem abnormalities. **Conclusions:** An innovative control system was developed, based on the assessment of the risk presented by the flock and the implementation of methods adapted to the level of risk, involving all stakeholders in a sustainable interaction. The system was then tested in 11 voluntary slaughterhouses for 1 year. The perception of the method was also evaluated through sociological interviews with stakeholders in about 30 slaughterhouses. Results of these studies will enable to assess the efficacy and the feasibility of the system and if necessary, to adapt it.

227. Asymptomatic potted plants may carry potentially invasive soil-borne plant pathogens

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ABSTRACT

In the past decades, the most common pathway for the movement of plant pests (herbivores and microorganisms) across boundaries has been the trade of live plants, especially pot ornamentals. Microorganisms carried in pot plants, either in plant tissues or in the growing medium or both, have more chances to survive transportation, and may more easily become established once at destination. The number of alien and invasive soil-borne plant pathogens has in fact increased dramatically in Europe in the same time period. The EU has an open phytosanitary system, under which a commodity that is not specifically regulated can be imported. Inspections are concentrated on a small number of economically important plant pests, and limited to visual check of the crown. Inspections fail to detect other pests; especially if these are internal to the tissues or in the soil; if plants are asymptomatic or incipient symptoms are limited to the roots. Aim of this paper was to assess the level of infestation by Phytophthora, a genus of especially dreadful soil-borne pathogens, in nursery ornamentals largely traded to and within Europe as plants for planting. Since phytophthoras are not easily isolated, we developed a real-time PCR assay based on a genus-specific TaqMan probe in order to detect small quantities of DNA of the pathogen from plant tissues and soil before symptoms occurrence. Results revealed that pot plants in soil carry several *Phytophthora* species without showing any external symptom, and the level of infestation is shocking. The risk represented by trade of plants in soil is discussed. Since eradication of soil-borne organisms is difficult or impossible, prevention of introduction is the only advisable measure against spread of these pathogens.

228. Occurence of Honeybee viruses in experimental apiary

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ABSTRACT

Honey bee (*Apis mellifera*) play a vital role in agriculture producing wealth in terms of hive products (honey, royal jelly, pollen, wax and propolis) and by increasing the productivity of a wide variety of crops and flowers. So, for the role that honey bees have in nature and for their economical importance in agriculture, their diseases are of paramount importance. A broad spectrum of specific pathogens affects the honeybee colony including bacteria, viruses, microscopic fungi, and internal and external parasites. Some of these microorganisms and parasites are more harmful than others and infections/infestations may lead to colony collapse. Knowledge of the biology and epidemiology of these pathogens are needed for prevention of disease outbreak. To date, about 20 viruses infecting honeybees have been identified. The aim of this study was to assess the occurrence of viral disease in experimental apiary made up of nine colonies in the province of Bari, Italy. All beehives were infested by the mite Varroa destructor, which is currently endemic in honeybee colonies in Southern Italy. Nobody morphological alterations were reported. Between March and June 2014 for each family, 30 adults and 30 larvae were collected and analysed. Total viral RNA was extracted from each samples and the presence of bee viruses was demonstrated by employing RT-PCR methods. Our results highlight the prevalence of 100% of Deformed Wing Virus (DWV) and Kakugo Virus (KV) in each kind of sample; of 80% of Sacbrood Virus (SBV) and Black Queen Virus

(BCQV). Low prevalence was evaluated for Chronic and Acute Paralysis Virus (CBPV and ABPV). The survey demonstrates that honeybee RNA viruses have wide circulation in Italy. Accurate identification and continuing monitoring of infected bees and asymptomatic carries is essential for the understanding of viral epidemiology in apiaries. In fact, to date it's known that these viruses have a important role in the Colony collapse disorder in combination with another stressor like Varroa or Nosema. So it is essential to develop, as soon as possible, intervention strategies in order to safeguard the bees' health.

229. Early Signal Detection for Emerging Risks in Scientific and Technical Literature

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ABSTRACT

We have developed an information system to support the detection of early signals for emerging food safety risks in very large volumes of text. This supportive system is based on a generic conceptual food safety hazard model that we induced from the analysis of early signals preceding past unexpected and high impact food safety incidents, such as acrylamide (2002), Sudan dyes (2005) and melamine (2008) (Van de Brug et al, 2014). The conceptual model depicts the relationships between food, adverse health effects and substances. Our Emerging Risk Identification Support (ERIS) system is able to identify any linguistic combination between –for example- any hazardous chemical and any food product in document texts, including those combinations of chemicals and food products that one would not have expected beforehand. Also, relationships with adverse health effects can be detected and extracted from text. The ERIS information system dynamically processes and combines new incoming documents from scientific, technical and web sources. The system's output consists of relationships between concepts and terms structured in a tabular format, topped with an interactive information analysis dashboard, ready for expert evaluation. We applied this technology in the EU FP7 project Connect4Action to capture relationships between new food processing technologies and stakeholder perceptions from scientific literature. Also, the ERIS technology is applied for occupational chemical safety and in the domain of immune health. Key features of ERIS: - relationship mining in very large volumes of text; - food safety ontology, required to capture relevant relationships; - multidisciplinary expertise, needed for evaluation of results. With this poster presentation we will show how the system works, how expert evaluation may look like and how typical results are like. Ref: F.J. van de Brug, N.B. Lucas Luijckx, H.J. Cnossen, G.F. Houben (TNO; Utrechtseweg 48; 3704 HE Zeist; PO Box 360; The Netherlands). Early signals for emerging food safety risks: From past cases to future identification. Food Control, Volume 39, 2014, Pages 75-86.

230. Forecasting (re)emergence of infectious animal diseases: prospects for monitoring risk indicators and generic alert setting

Van Huffel X, Cardoen S, Dewulf J, Imberechts H, Berkvens D, van den Berg T, Saegerman C, Hooyberghs J, Vandecan M, Raemaekers M, Van der Stede Y, Tamignaux D, Desmecht D, Linden A, Thiry E

Belgian Food Safety Agency

ABSTRACT

Emerging infectious animal diseases and zoonoses may provoke significant losses to animal production and public health sectors and can be a threat for the safety and the security of the food chain. Traditional active and passive animal disease surveillance is focused on the monitoring and/or early detection of diseases already present on the territory, based on clinical signs or diagnostic testing. These systems are unable to predict the next emerging animal disease outbreak(s). The latter represents a major challenge for veterinary and public health authorities. The aim of this ongoing self-tasking mandate of the Scientific Committee of the Belgian Food Safety Agency is to identify reliable and measurable risk indicators enabling risk assessors to determine generic levels of risk status for infectious animal disease emergence in a country in order to timely alert risk managers and animal health professionals to take the appropriate preventive and disease surveillance measures. The hypothesis is that (re) emergence of (new, known or unknown) infectious animal diseases in a country is related to or caused by events or particular circumstances (risk drivers). Several studies have already listed drivers and subdrivers of emerging risks (i.e. VWA, 2005 and 2006; EFSA, 2014). It is postulated that risk drivers can be monitored by risk indicators. These can be specific or be a proxy for a particular infectious animal disease or rather be of a generic nature. The challenge is to identify reliable and measurable risk indicators, to follow their evolution over time and to identify threshold levels for alert setting. The workgroup of the Scientific Committee follows a pragmatic approach and identified examples of both specific and generic measurable risk indicators (i.e. increase of commercial exchange of animals, increase of animal or human (in case of zoonotic infections) mortality/morbidity statistics, geographical extension of infectious diseases) and their respective data sources. The feasibility of the measurement and follow-up of these risk indicators and their ability as early or forecasting signal for emergence of infectious animal diseases is under investigation by case studies involving scientific risk assessors and risk managers. Preliminary results will be shown.

231. miRNome profiling of Bt- and EPSPS-expressing transgenic maize

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ABSTRACT

Double stranded RNA molecules, such as miRNAs, are small non-coding regulatory RNAs that play important roles in the regulation of gene expression at the post-transcriptional level. Therefore, changing the nature, kind and quantity of particular regulatory-RNAs through genetic engineering can create biosafety risks. While some genetically modified organisms (GMOs) are intended to produce new regulatory-RNAs, these may also arise in other GMOs not intended to express them. The aim of our study was to compare the miRNome of stacked transgenic maize versus its single transgenic parental lines and conventional counterpart. The transgenic samples were not intended to contain any new regulatory RNA species; the intention was to produce two insecticidal Bt proteins (MON89034 event) as well as the CP4EPSPS protein (NK603 event, RR-expressing) conferring herbicide tolerance. We have performed an untargeted profiling approach to characterize the miRNome of maize leaf samples. Total RNA samples were enriched for miRNAs and normalized aliquots were used for next generation sequencing using Illumina HiSeq2000. Bioinformatics analysis allowed the identification of conserved miRNA families. Pairwise differential expression revealed 13 miRNAs that were accumulated at higher or lower levels in transgenic samples. Nine miRNAs were present at statistically different levels in Bt-expressing samples compared to a conventional counterpart. Three other miRNAs showed statistically different levels in stacked (Bt and RR-expressing samples) compared to conventional counterpart and also between RR-expressing samples and the stacked one. Enrichment analysis of all potential miRNA targets showed alterations in major metabolic processes, such as: nitrogen metabolism and RNA biosynthetic processes. The major altered molecular function was transcription factor activity. In-depth miRNAs target prediction analysis showed 136 potential endogenous gene targets, including splicing variants and gene expression inhibition by either mRNA cleavage or translational inhibition. Correlation between miRNA targets and our previous proteomic investigation indicates the proof-of-concept for how transgene insertions might affect endogenous gene expression by pleiotropy. To the best of our knowledge, this is the first study of miRNA profiling of transgenic versus conventional maize samples, and it demonstrates the occurrence of pleotropic effects in core metabolism in transgenic plants at the miRNA level.

232. Precision crop protection against sporadic diseases driven by climate change

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ABSTRACT

Objectives: To devise and assess methods to allow early detection or forecasting of sporadic plant disease epidemics to help safeguard food production and safety under future climate scenarios. **Methods:** Recent projects have developed and assessed precision agriculture methods including air sampling using automated devices with built-in diagnostic methods and wireless communications that link to infection models to provide a direct inoculum-based warning of imminent disease risk. Additionally optical sensing methods were reviewed for both proximal and remote sensors and for reflectance and chlorophyll fluorescence imaging to map positions of plant stress. The potential for using volatile biomarkers and hand-held diagnostic tests to identify problems is also being reviewed. **Results:** Automatic detection and wireless reporting of airborne spores of Sclerotinia sclerotiorum was demonstrated in the UK and Canada and validated using traditional spore traps and lab-based qPCR. Further refinements will enable detection of other pathogens. Simple LAMP and other isothermal DNA-based assays have also been applied manually to air samples to indicate arrival of inoculum. Volatile biomarkers have been used in detection of insect attack and also for deterring insect pests. In contrast, most remote sensing can only indicate disease issues usually too late for effective treatment, except for instances of some soil-borne diseases. Proximal reflectance and chlorophyll fluorescence imaging has greater potential for early detection and mapping of diseases in fields in time for application of crop protection products as part of integrated pest and disease management. **Conclusions:** Crop protection enhanced by these new developments in precision agriculture will be more important than ever before for intensive production systems to provide the lowest environmental impact per unit of produce harvested under more variable weather patterns.

Advancing environmental risk assessment

233. Environmental risk assessment of pesticides in the field of ecotoxicology: past, present and future

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ABSTRACT

The protection of non-target species is one of the goals of the environmental risk assessment (ERA). For the authorisation of pesticides, an evaluation of the risks should be performed for the following groups of non-target species: birds and other terrestrial vertebrates; aquatic organisms; bees and non-target arthropods; earthworms, other soil macro-organisms and micro-organisms; other non-target organisms (flora and fauna) and biological methods for sewage treatment. A tiered approach is followed. Tiers are characterized by an increasing complexity starting with simple conservative assessments (worst-case assumptions) towards more realistic evaluations (i.e. more realistic assumptions). In the last 20 years, since a harmonised risk assessment for pesticide is required at EU level, several scientific methodologies were developed and reflected in Guidance documents to address first and higher tier risk assessments (SANCO/4145/2000 for birds and mammals, SANCO/10329/2002 and SANCO/3268/2001 for terrestrial and aquatic organisms, respectively and ESCORT2 for non-target arthropods). However, the science behind the ERA is constantly evolving e.g. more structured data, metadata and availability of tools. In this context, EFSA's role is to revise and develop updated ERA approaches by taking into account the new scientific knowledge. EFSA developed several opinions and guidance documents for the ERA, including an overarching opinion on the definition of ERA specific protection goals based on the ecosystem service concept. Relevant milestones already achieved, were the scientific opinions and guidance on birds and mammals, bees, aquatic organisms, non-target terrestrial plants, non-target arthropods. Other activities are still on-going or to be initiated i.e. opinion for in-soil organisms, sediment organisms, amphibian and reptiles, population modelling for aquatic organisms.

234. Pesticides residue levels in the honey bees *Apis mellifera* foraging in agricultural areas

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ABSTRACT

The extensive use of pesticides, and neonicotinoids in particular, for crop protection has been suggested as one of the main causes of pollinator decline. However, whether the bees foraging on agricultural areas accumulate the insecticides to the levels which may cause their decline remains unclear. One of the widely produced crops in Europe is the oilseed rape (*Brassica napus*), which is the most important insect-pollinated oil plant. In 2014, due to the European Commission's implementation of restrictions on the use of neonicotinoids as a seed treatment, the oilseed rape from neonicotinoid treated seeds was grown in Poland for the last time. This gives a unique opportunity to compare pre-ban pesticide residues levels in bees, nectar and pollen collected from agriculture areas with the post-ban situation in the coming years. Due to the implementation of the restrictions on the use of neonicotinoids, it is expected that there will be a considerable increase in the use of other types of insecticides, especially the broad-spectrum organophosphate, such as chlorpyrifos. In this study, three fields have been chosen with oilseed rape crop grown from thiamethoxam treated seeds, on which chlorpyrifos was also used for foliar application. On each of the studied fields one bee hive was installed in April 2014. Foraging bees were collected during oilseed rape flowering at 3 time points: before the blooming, in full blooming and at the end of the blooming period. The bees were deprived of pollen sacs on hind legs and were killed by freezing at -20°C. The residue levels of insecticides in bees, nectar and pollen were determined using modified QUEChERS method followed by gas chromatography coupled with mass spectrometry in case of chlorpyrifos and gas chromatography with electron capture detector to determine thiamethoxam levels. Thiamethoxam concentrations in bees ranged from 0.01 to 0.20 ng/g, and chlorpyrifos levels ranged from 0.18 to 1.07 ng/g, with no differences between sampling time for both insecticides. The data on pesticide residues in collected nectar and pollen will be also presented and used further as a basis for toxicokinetic experiments to test how the selected insecticides are absorbed, metabolized and eliminated by honey bees. Those data will be compared later with the post-ban situation based on samples collected in 2015.

235. Searching for impacts of pesticides on earthworm diversity

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ABSTRACT

The integration of functional agro-biodiversity and the related ecosystem services into the Environmental Risk Assessment schemes of pesticides stresses the need for advancement in knowledge and research to meet challenging objectives associated with soil ecology. Earthworms are known organisms to sustain soil functions and qualities such as fertility in the agricultural landscape. Moreover they are sensitive to chemicals and can serve as bio-indicator organisms. There is no clear consensus in the literature, on which aspects and attributes of earthworm diversity research has focused to date. This literature review aims at investigating the research that has been conducted for the impacts of pesticides on earthworm diversity and the related functions in the field of ecotoxicology. The literature review was carried out by using keywords in all fields of the advanced research option 'Journals' of ScienceDirect with the formula 'earthworm diversity and pesticide' and 'ecotoxicology'. From 325 publications a thorough full text examination was conducted to include only

the publications with consistent information. Thus data from a corpus of 52 publications from 1996 to 2015 comprise the database of the current literature review. Thirteen earthworm species are mainly investigated worldwide as being affected by a variety of pesticides also including combined effects. Affected diversity attributes such as abundance, were also studied besides more functional attributes such as sperm count and DNA damage or cholinesterase inhibition. The review provides an insight for research already completed in physiological adaptation, behaviour, life-stage composition and included life-cycle parameters (survival, growth, and reproduction), variation in pollutant-induced biochemical and cellular biomarkers and the sensitivity levels of earthworms compared to other soil organisms.

236. Preliminary study on the interaction between bacterial enteric pathogens and acquatic macrophytes

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ABSTRACT

Some bacterial pathogens are able to penetrate into the plant tissues, they may further infect it endophytically, which will cause the plant to serve as an alternative host for the human pathogens. *Salmonella* might be considered an endophytic pathogen because it can cause plant diseases with symptoms such as wilting and chlorosis. *Salmonella* has been found to be resistant to environmental changes and its persistence in surface water may represent an important aspect of the interface between environment and food chain. In the respect few is known on the interaction between bacterial pathogens present in surface water and aquatic macrophytes, which could contribute to the persistence of a pathogen in the aquatic environment and to the risk of infection by the consumption of produce irrigated with contaminated water. Aim of this study was to evaluate the possible interaction between a bacterial pathogen such as *Salmonella* and an aquatic macrophyte such as *Phragmites australis*. *Phragmites* plants were contaminated by inoculating 6.5x106 cfu/ml of *S.* Napoli in the water used for hydroponic culture. An hydroponic system without plants was used as control and the persistence of *S.* Napoli in water was monitored in both systems by periodical culture of water, root and rhizome samples. The preliminary results seem to suggest a possible interaction between *Salmonella* and acquatic macrophytes. 24 hour after contamination the number of *Salmonella* decreased in water with *P. australis* and did not in control. After 72 hours the number of cfu/ml of *Salmonella* isolated from control water and from root and rizome samples was higher than that of *Salmonella* isolated from water samples from systems with plants. Further experiments are needed in order to assess if microbial internalization could happen either through surface contamination or uptake through the plant root system followed by dissemination to other parts of the plant during the growing stage.

237. Effect of the sediment contamination in the fish community structure of a temperate estuarine system (Ria de Aveiro, Portugal)

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ABSTRACT

Objectives: Estuaries and coastal lagoons are among the most productive and economically important ecosystems, and are generally close to sites of human activity. Most estuaries and coastal lagoons receive large amounts of nutrients and contaminants from urban, agricultural and industrial effluents. High metal contamination might result not only in reducing growth and condition of fish but also in a selective elimination of the most sensitive stages of vulnerable fish species. The objective of this study was to use the fish community structure to assess the effect of human impacts, particularly metallic contamination, on a coastal lagoon ecosystem. **Material and methods:** Sediment and biological samples were collected in winter (February 2012) and summer (August 2012) at 9 sampling sites located inside the Ria de Aveiro (Northwest Portugal). In the laboratory, a total of 6195 fishes were identified, counted and weighted. Sediment chemical characterization was conducted using thermal decomposition AAS (Hg) and ICP-OES (other metals). Multivariate approach, based on Bray-Curtis similarity matrices, were used to examine the relationship between fish community structure and both chemical and environmental variables. **Results:** Fish abundance, in terms of number and weight, and fish diversity were generally higher in summer than in winter. The maximum values of Shannon's index were found in Carregal and Saõ Jacinto during August whereas the minimum values were found in Areão, Laranjo and Rio Novo de Príncipe during February. Metal concentrations found in the sediments were mostly similar between winter and summer. In general, sediments showed low or residual levels of metal contamination. The exceptions were in Vagos for cadmium and in Laranjo for mercury, arsenic and zinc. Cadmium, combined with temperature and salinity, produced the best correlation with the biotic similarity matrix in terms of fish number and weight data. **Conclusions:** This study highlight the sediment composition as one of the important determinants to the structure of the fish community in ecotone systems.

238. Advanced environmental risk assessment for invasive alien species

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ABSTRACT

The risk invasive alien species (IAS) pose to the environment is usually assessed by extrapolating the impacts in the area where the pest is already present to the situation in the receiving area. However, this neither considers sufficiently the sequence of changes in the abundance/prevalence, distribution and characteristics of the invading population, nor that the impact is a context-dependent process based on the interaction among the IAS invading a new area, the recipient community and the abiotic environment. To address this lack, we have developed, tested and improved the following method. In the first phase of the development of the method, the impact of IAS on ecosystem services (ES) and biodiversity components (BC) was assessed prospectively under simplified scenario assumptions. This is demonstrated by an environmental risk assessment (ERA) that has been conducted on Anoplophora chinensis. In the second phase, the DPSIR framework was introduced into the method, in which the steps (Driver identity and Pressure, State of the system, Impact and Responses) are assessed subsequently in a causal chain. As an example, an ERA on Agrilus planipennis is presented. In a third phase a more detailed analysis of the causal relationship between the driver pressure and the ecological traits of the recipient community has been conducted. The level of resolution in the analysis allows a suitable integration of data with expert judgment, supporting a reliable and reproducible quantitative assessment of the impacts of an IAS on ecosystem traits (ET), ES and BC. This approach has been applied to Pomacea canaliculata. In a fourth phase the method has been improved with the introduction of two novelties – considering the impact on a continuous scale and the mapping of the impact the IAS has on ET, ES, and BC. The proposed approach allows an assessment of environmental risks by integrating the impacts on different components and levels of the environment as well as their probabilities of occurrence. Considering the impact on a continuous scale gives a major understanding of the series of processes leading from entry/introduction of the IAS to the impact it has on ET, ES and BC. Maps of risk improve the method and can be generated considering (i) functions relating the driver pressure with ecosystem traits, (ii) maps representing the heterogeneity in the driver pressure, and (iii) maps assessing the ecosystems and their services.

239. A user-friendly program of a TK-TD model to link variable exposure to effects on survival of aquatic organisms

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ABSTRACT

The use of plant protection products in agriculture may lead to concentrations in edge-of field waterbodies like ditches, ponds or streams which may have effects on aquatic organisms. To analyse these risk the concentrations in the waterbodies are predicted by exposure models and compared to effect threshold concentrations derived in ecotoxicological tests. However, the exposure can be variable over time and space (i. e. due to variable weather and soil conditions in Europe). One way to mechanistically link exposure and effects offer Toxicokinetic-Toxicodynamic (TK-TD) models which transfer the external concentration (e.g. in the water) to concentrations in the organism and the resulting hazard. For the General Unified Threshold model of Survival (GUTS) a few implementations are available which are not very user-friendly and which differ in their results due to differences in the parameter calibration. Therefore the objective of this project was to develop a user-friendly stand-alone program of GUTs with a special focus of optimized and well documented calibration and validation procedures. In the first step the model parameters are calibrated based on the experimental data, e.g. a standard acute toxicity tests with constant exposure over 4 days. The outcome of this calibration depends on several settings not clearly defined in the GUTS description yet, e.g. the selection of the start values and the optimization algorithm used. We have compared different options to suggest specifics settings. In the next step the fitted model should be tested on a data set not used for calibration, i.e. a toxicity test with different exposure profile (e.g. pulsed instead constant exposure). Suitable measures on the quality of the correspondence of data and predictions are provided. Finally, the validated model can be used to predict the effects of more complex profiles predicted by exposure models. The different steps are demonstrated using an example data set. The GUTS implementation including its documentation will allow a more routine use of the model as a tier 2 tool according to the EFSA aquatic guidance document to link dynamic exposure with lethal effects.

240. Environmental Risk Assessment of Genetically Modified Plants Field Trials using a A Fuzzy-based Decision Support System

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ABSTRACT

An harmonized approach in Environmental Risk Assessment (ERA) of GMOs represents an important goal to attain, in order to overcome potential controversial results. Taking into account the EU legal framework regulating the environmental release of GMOs, we developed a Decision Support System based on Fuzzy logic (FDSS), with the aim to help experts in making a thorough risk assessment. This tool helps describing the various relationships between potential receptors and hazards of a Genetically Modified Plant field trial, leading to the identification of potential impacts. Subsequently, the related risks are assessed by fuzzy concepts and a fuzzy reasoning system1. The FDSS is a web based Questionnaire (http://www.ogm-dss.isprambiente.it/index.xhtml) that gives the input to a fuzzy inference system, which has a knowledge-base consisting of 6,125 IF-THEN rules. It has been tested yielding 150 ERAs related to Bt-maize and Herbicide Tolerant oilseed rape releases. Furthermore, FDSS has been applied to seven different risk scenarios, on a case by cases basis, by the expert group of the LIFE+ MAN-GMP-ITA project (http://www.man-gmp-ita.sinanet.isprambiente.it/)2 using data related to the project areas. When the information requested by the questionnaire are missing, the rules have been set to drive the system to the worst-case scenario, according to the precautionary approach. The results have been considered consistent by the expert of the LIFE+ project. In conclusion, it was confirmed that the FDSS is a useful tool to carry out the various phases of the ERA, although it is a work in progress that deserves further research. The application of the FDSS, beside providing a list of potential effects and related receptors, allows the visualization of the paths from the source of risk up to the receptors, and eventually allows the assessment of the identified risks. On a sideline, it can also help identify those missing data needed for a thorough evaluation of the risks. 1. Camastra, F. et al. TÉRA: A tool for the environmental risk assessment of genetically modified plants. Ecol. Inform. 24, 186–193 (2014). 2. Lener, M. et al. Applying an operating model for the environmental risk assessment in Italian sites of community importance (SCI) of the European commission habitats directive (92/43/EEC). Bull. Insectology 66, 257–267 (2013).

241. Project ECsafeSEAFOOD – Priority environmental contaminants in seafood: safety assessment, impact and public perception

Marques A, Barranco A, Langerholc T, Verbeke W, Egaas E, Barceló D, Granby K, Robbens J, Fadini JD, Cunha S, Kotterman M, Domingo JL, Trevisan M, Reuver M, Turquet J, Schipper J, Mortensen A

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ABSTRACT

Objectives: This European project aims to assess food safety issues mainly related to non-regulated priority contaminants present in seafood as a result of environmental contamination and evaluate their impact on public health, in order to increase seafood safety and reduce human health risks. The aim of the presentation is to present the structure and relevance of the ECsafeSEAFOOD consortium for the European food safety. **Methodology:** The first step consisted on the creation of a database for environmental contaminants of emerging concern present in seafood, followed by the implementation of a monitoring scheme at European level and assessment of cooking effect on contaminant intake by consumers. Secondly, development of fast screening detection methods for these contaminants, seafood risk assessment and mitigation strategies are ongoing, as well as the use of innovative toxicological tools to test environmental contaminants in realistic conditions. Thirdly, links between the level of relevant priority contaminants in the environment and those in seafood are being studied combining the effect of climate change. Finally, the project will increase the consumer confidence through clear and practical communication and information spread in close collaboration with food safety authorities. **Results:** Outputs from contaminant monitoring for seafood in hotspot areas and European markets combined with database risk ranking of contaminants, as well as the information gathered from consumer surveys have been accomplished. Benefits and risks associated with processing and cooking seafood, based on contaminants bioaccessibility and bioavailability in marine organisms, as well as the effects of global warming on bioaccumulation and elimination of contaminants are being assessed. New methods for screening, detection and extraction of different toxins are being developed. Also, mitigation tools during processing, phycoremediation and an online consumer tool to guide consumers about the benefits and risks of consuming seafood are being developed. **Conclusions:** The interesting results and progresses obtained so far will enable to assess the impact of non-regulated contaminants of emerging concern on public health, leading to improved seafood risk management and to increase public awareness. **Acknowledgements:** The project is funded by the European Union's Seventh Framework Programme for Research and Technological development (FP7/2007-2013) under Grant Agreement n°311820

242. Tools for improving pesticide risk assessment on European amphibians and reptiles

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ABSTRACT

Environmental pollution is recognized as one of the major causes involved in amphibian and reptile declines, in spite of which these animals have not been traditionally considered in risk assessment of pesticides, being supposedly protected through data retrieved from other vertebrate taxa. The development of the Regulation (EC) 1107/2009 concerning the placing of Plant Protection Products on the market has introduced the consideration of amphibians or reptiles for pesticide risk assessment in terrestrial ecosystems. However, in

order to put this into practice, large information gaps still exist. For example, the permeable nature of amphibian skin or the strong links of reptiles with soils (especially during egg laying) where pesticides are applied make dermal assimilation a very relevant route of exposure of these animals. In this context, a recent report published by the EFSA concluded that information to estimate dermal exposure in amphibians is lacking. From a practical point of view, no guidelines exist to test chemical toxicity in these two taxa. With the purpose of providing tools for assessment, EFSA has planned to work during the next years in the elaboration of a Guidance Document for pesticide risk assessment on amphibians and reptiles. Over the last year, we have been developing the HerPesti project, with the aim of testing the degree of protection that EU legislation on pesticides confers to amphibians and reptiles and, if necessary, developing possible remediation measures. We have identified the major exposure routes to pesticide in both aquatic and terrestrial environments and quantified the impact of pesticides using mechanistic approaches, showing that the exposure ways considered in current risk assessment procedures are not always the most relevant ones. A field monitoring in areas of pesticide use is currently in process to characterize the exposure of native populations in realistic scenarios. The project strongly pursues the establishment of a fluent communication with industry and government sectors, providing the former with protocols to evaluate pesticide risks on amphibians and reptiles, and the latter with decision-making tools that we hope will contribute to improve the efficiency of the upcoming EFSA document. Study financed by the European Commission through MC-IEF actions of the 7th Framework Program

243. Linking exposure and effects towards an ecological risk assessment in Mediterranean Rice paddy ecosystem

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ABSTRACT

Objectives: Paddy rice ecosystems are important as hot spot of biodiversity, especially flooding paddies that provide safe sites to many aquatic species, being necessary to understand how they react/recover from exposure to stressors. Nevertheless, it has been questioned whether ecosystem structure should be the protection goal in rice fields or whether maintaining their function should be the main goal. However, the ecological effect chain following pesticides stress may evidently affect ecosystem function and even crop productivity. In order to set light on these issues the present study tested the community impacts of the systemic insecticide imidacloprid when applied at its recommended commercial rate in experimental rice paddies. **Material and Methods:** The following methodology was implemented: Water sampling for environmental parameters and chemical analysis; Sampling of zooplankton (ZP), phytoplankton (PT) and macroinvertebrates (MC); Taxonomic identification; Statistical analysis (e.g. PCR,-Principal Response curves); Compare and link exposure and effects data. The experiments were carried out in 3 experimental rice plots (C -control, C1, C2) set up according to standard agricultural practices, but no insecticides were used prior to the study. Imidacloprid was applied by aerial way on rice paddies C1 (outflow of water interrupted 7 days) and C2 (outflow not constrained). **Results:** Identification of MC assemblages and PCR analysis. Prior to imidacloprid application, the one-way ANOVA revealed significant differences between the MC of both plots with those from the C, but did not show significant differences between them. However at time 6 and 7 after application significant differences were observed only between C1 and C. Populations of 3 species decreased in abundance after imidacloprid application. Morrisey et al., 2015, indicate those species as the most sensitive species to imidacloprid. Till now a total of 90000 exemplars of ZP were identified. Preliminary analysis shows a decreasing of abundance of identified organisms. **Conclusions:** Statistical analysis shows a potential effect of imidacloprid on the abundance of the identified organisms. However a tendency for recovery of the main groups potentially affected along time was found. The obtained results of environmental influence over biological assemblages will be presented and discussed.

244. Imidacloprid in house dust: environmental risk assesment for intake

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ABSTRACT

Objectives: To evaluate the contribution of house dust to the intake of imidacloprid a neonicotinoide insecticide with a wide range of applications in agriculture, indoor environment and veterinary medicine. **Materials and Methods:** Within the Italian Ministry of Health granted 'EnviFood' project, house dust samples (N=282) were drawn from different Italian towns and analyzed for Imidacloprid by an accredited UPLC-MS/MS multi-residue method. Pesticides concentration was then transposed into intake estimates, accounting for grooming and mouthing activity in toddlers. The conservative upperbound approach (0.01mg/kg as LOQ) was adopted for left censored data. Average and conservative dust intakes estimates of 16 and 110 mg/day respectively, were derived from the 1995 German Exposure Factors Handbook (AUH report). Average body weight of 11.3 kg for Italian toddlers were adopted. Under a deterministic approach, risk characterization accounted for the following acute and chronic toxicity guidance values set on neurodevelopmental end-point: the ARfD and the ADI of 0.06 mg/kg/day proposed by EFSA (2014). **Results:** On 282 samples, 99 (35%) gave quantifiable results (>0.010mg/kg). Occurrence (mg/kg) descriptors were respectively; P75=0.06; P90=1.68; P95=18.8; Max=78.3. Accounting for the reported percentiles, under the 16 mg dust intake scenario, the estimated Imidacloprid assumption represented the 0.13%, 3.95%, 44.4% and 185% of the ADI and ARfD guidances value (0.06 mg/kg/day). Under the 110 mg conservative scenario, the estimates raised up to 0.89%(P75), 27.2%(P90), 305%(P95); and 1,270%(max). **Conclusions:** Apart the toxicological uncertainties related to the associated presence of other not related pesticides in house dust, as well to other chemicals sharing a neurotoxicological end-point such as PBDE congeners no. 47 and 99, the contribution of dust to the aggregate (dust+food) intake of imidacloprid may be relevant to approach/trespass the pertinent ADI starting from the 16 mg/P95 and 100 mg/P90 scenarios, on the basis of the EFSA food intake estimates in the range 30–65% of the ADI, in EU children. The recorded frequency and occurrence levels of Imidaclopid in houst dust, however,suggest a Consumers empowerment about a prudent use of related products in indoor environment (i.e.as anti-beetle/ant treatment, and/or as phytosanitary treatment of ornamental plants), especially in presence of children and puppies,also to be preventive towards potential acute intoxication.

245. Metabolism of pesticides in farmed fish – What can we learn from in vitro studies?

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ABSTRACT

Objectives: Owing to the increase in fish farming and the increase in use of plant commodities as a source of feed, there is a need to estimate the levels and nature of pesticide residues in edible products in fish. Therefore, the EU published new data requirements for fish as part of the approval process for pesticides. Fish metabolism data provide an estimate of total terminal residues and characterize the chemical nature of residues which may occur in edible commodities of fish exposed to pesticides. Currently, metabolism studies with fish are required when pesticides of logKow >3 are used in crops fed to farmed fish, which may lead to significant residues in fish feed considered to be >0.1 mg/kg of the total diet. Rainbow trout and common carp are suggested as test species. Fish metabolism studies using 14C labelled pesticide are carried out following the new working document on nature of pesticide residues in fish. It was the objective of this study to investigate the suitability of in vitro assays using isolated primary fish hepatocytes to provide information on the metabolism of pesticides in farmed fish. **Materials and Methods:** Fish metabolism studies on rainbow trout and common carp were carried out following the working document. Experimental diets fortified with a 14C labelled pesticide were applied. In parallel, siblings of the experimental animals were used to isolate

primary hepatocytes for in vitro biotransformation assays with the same test item. Cell preparations (primary hepatocytes) were extracted and analyzed by thin-layer chromatography for pesticide metabolite patterns which were compared with the results obtained for tissue samples (fillet and liver). **Results:** In vitro metabolism assays with primary trout and carp hepatocytes can be used to identify pesticide metabolite profiles. Species-specific differences in metabolite pattern found in the in vitro assays were in accordance to the results obtained for liver in the in vivo studies. The liver metabolite profile was partly reflected in the fillet samples. **Conclusion:** Information on the nature of pesticide residues in fish may be generated from hepatocyte assays. This makes the in vitro hepatocyte assay a valuable tool to support in vivo metabolism studies on fish carried out as part of the approval process for pesticides.

246. Sensitivity of bees to pesticides: a comparative approach

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ABSTRACT

Bees, including managed and wild bees, provide important ecological functions, sustaining basic ecosystem services and human food production. In Europe, many crops depend directly on insect pollination and the honey bee, *Apis mellifera*, is considered one of the most important pollinators. However, there are thousands of other bee species and their contribution in the pollination service has been recently identified. Following the recent honey bee and wild bee populations declining worldwide, concern has been growing about the risks posed by pesticides and, on the appropriateness of the current risk assessment scheme for the approval and authorisation of pesticides. The current risk assessment for pesticides focuses on *A. mellifera* and suggests to extrapolate data from honey bees to other bee species based on the assumption that they are the most sensitive species, although a quantitative approach for comparing the difference in sensitivity among bees has not yet been reported. In this study, a systematic review of the relevant literature on the topic followed by a meta-analysis has been performed. Both the contact and oral acute LD50 and the chronic LC50 reported in laboratory studies for as many substances as possible have been extracted from the papers in order to compare the sensitivity to pesticides of honey bees and other bee species (Apiformes). The sensitivity ratio between the endpoint for the species *A. mellifera* and the other species of bees was calculated considering 150 different combinations of bee species and pesticide, including 19 bee species and 53 pesticides in total. The results of the meta-analysis showed a high variability of sensitivity among bee species and 9 of the 19 species were more sensitive than honey bees to pesticides. In about 5% of cases, the sensitivity of other species was more than 10 times higher than honey bees. In conclusion, according to the results of this study, it needs to cover a greater range of bee species in the risk assessment of Plant Protection Products, in order to protect wild bees as well as honey bees. At the same time, exposure levels must also be considered because the effect of pesticides can vary among bees depending on the specific life cycle, nesting activity and foraging behaviour.

247. Risk assessment for GMOs in regulatory context – objectives, general principles, methodologies and current practices

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ABSTRACT

The overall policy of most countries toward modern biotechnology aims at 'maximising benefits and minimizing safety risks', an approach that has been outlined in many internationally agreed documents such as Agenda 21 (1992). For the purpose of maximising benefits and minimizing risks, many countries have established biotechnology research strategies as well as biosafety regulations, which offer tools for informed decision making in weighing benefits and risks. Biosafety regulations exist in many different forms, i.e. with different objectives, scopes and regulatory mechanisms (e.g. requirements to comply with general conditions, and requirements to obtain an authorisation for certain activities). Despite these differences, key in all regulatory systems are environmental risk assessment and – where it concerns a product for consumption – food/feed safety assessment. The Cartagena Protocol on Biosafety includes international agreement on the objective, general principles and methodology for environmental risk assessment. The Codex Alimentarius includes international agreement on the objective, general principles and methodology for food/feed safety assessment. The poster will discuss these internationally agreed objectives, general principles and methodologies and to what extent current practices are consistent with them.

248. MUST-B: A framework for the risk assessment of multiple stressors in honeybees

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ABSTRACT

Honeybees (*Apis mellifera* spp.) represent a small fraction of bee species diversity, but they are ranked as the third most economically farmed animal because of their role in the food chain via pollination. However, over recent years, an increasing trend of honeybee colony losses has been reported worldwide. Such losses have been attributed to multiple stressors (biological, chemical, nutritional and/or environmental). Nevertheless, a good knowledge of the potential interactions and effects of these multiple stressors at colony level and in field conditions is still lacking. To better reflect this complexity and reality, EFSA is deploying resources and efforts, in collaboration with relevant stakeholders, towards the development of a conceptual framework defined under the project 'MUST-B---: EU efforts towards the development of a holistic approach for the risk assessment on MUltiple STressors in Bees'. This project is supported by a multidisciplinary team of scientific officers from the Scientific Committee and Emerging Risk Unit, the Animal and Plant Health Unit, the Pesticides Unit, the Assistance and Methodological Support Unit, and officers from the Communications department. The team supports dedicated working groups which are providing guidance for data collection and identifying further data needs for the development of the conceptual framework. The intended framework consists of monitoring, experimental and modelling approaches. For the monitoring and experimental approaches, available tools and methods used for measuring predefined indicators of the health status of a honeybee colony will be identified. For the modelling approach, a conceptual model that takes into account current development made in this field, in particular from population dynamic models, and which include complexity linked to exposure from multiple stressors will be proposed for further implementation and possible calibration with proper field data. Regular communication and exchanges will take place with involved stakeholders throughout the duration of the project.

249. Reduction of uncertainty and variability in honeybee trials for the ecological risk assessment

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ABSTRACT

Since colony collapse disorder (CCD) has become a public issue in recent years, a number of new methods have been tested to obtain large amounts of reliable, quantitative data for risk assessment and to understand possible causes of colony losses. The use of digital photography and videography offers a potent method for large scale field trials, in particular to reduce variability and uncertainty. Variability and uncertainty are often a result of study design and small sample sizes. For example in brood trials according to OECD 75 often only a few hundred brood cells need to be monitored. This results in a large variability of results between hives, making the detection of effects difficult. Based on power analysis we show how this variation can be reduced using automated brood analyses, which makes it practical to analyse entire hives. This results in a smaller uncertainty of the outcome of a trial despite of small numbers of hives being used. I a second step we assess methods to measure forager losses based

on videography. While in conventional trials up to 100 bees are individually tracked, again resulting in large variability between hives, videography makes it possible to count all bees leaving and entering the hive without much practical effort. These new methods show examples how the uncertainty of the outcome of field trials can be reduced without necessarily increasing the effort and costs of trials.

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