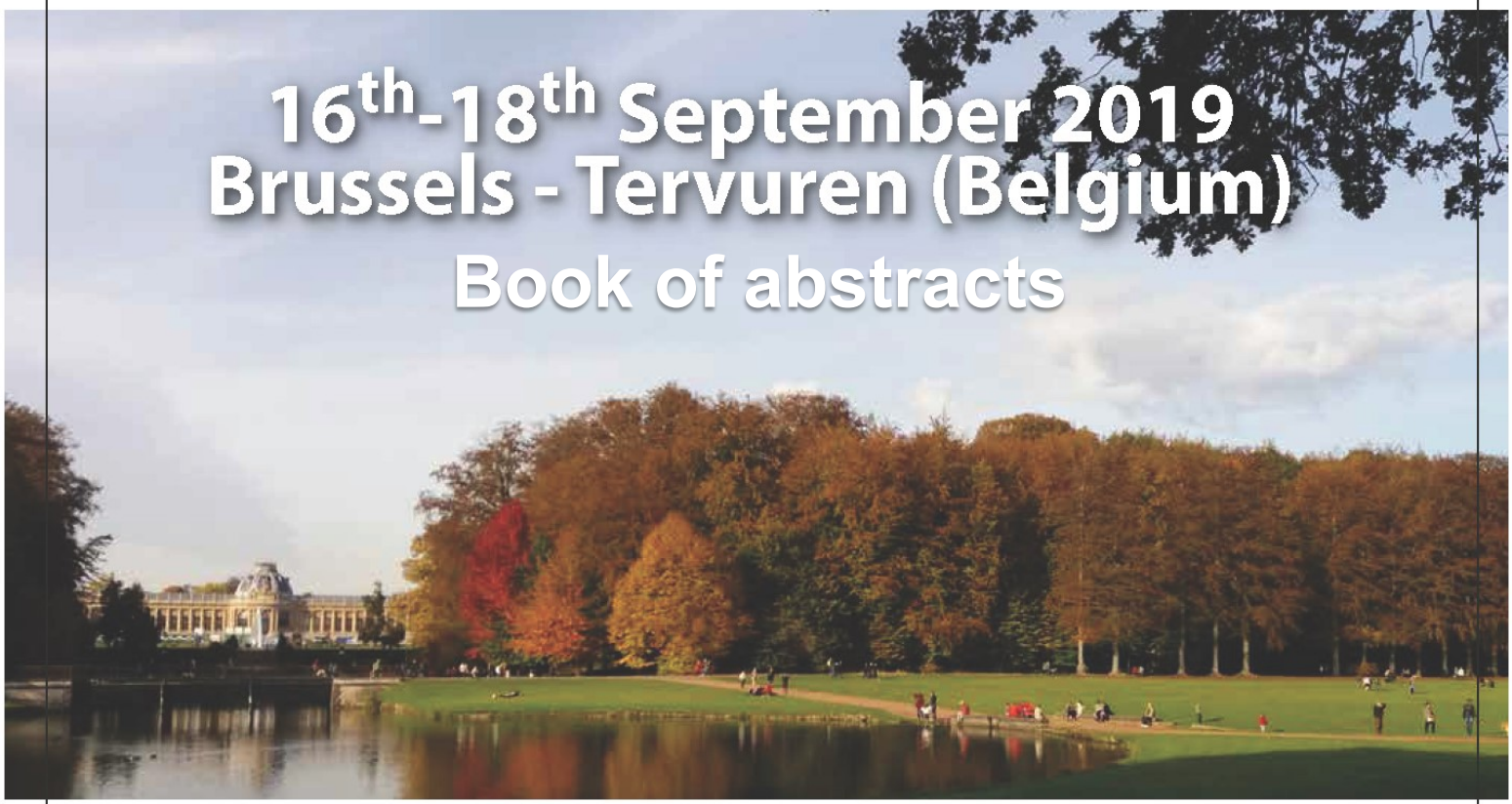
INTERNATIONAL MEASUREMENT CONFEDERATION
TC23 "Metrology in Food and Nutrition"

4th IMEKO FOODS

4th International Conference on Metrology in Food and Nutrition

Metrology supporting emerging food topics

http://www.imekofoods4.be



sciensano

TITLE
Book of Abstracts of the 4th International Conference on Metrology in Food and Nutrition
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EDITORS
J. Alexandre, S. Janssens, J. Van Loco
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WELCOME MESSAGE

We cannot imagine a modern society without metrology. Industry, science and legal instances rely on the correctness of results and data they are using. Novel technologies and emerging concerns put new challenges for metrologists in terms of standardisation and harmonisation and to assess the uncertainties of their data. Furthermore, there is an increasing need of data and knowledge sharing, strengthening the application of the FAIR data principle. The fourth Imekofoods conference will address metrological issues in measurement and data in food quality, food integrity, food safety and nutrition, with emphasis on new technologies.

On behalf of Sciensano and Imeko TC 23 we cordially invite you to take part in the fourth international conference IMEKOFOODS. The symposium will be organised at the Sciensano site in Tervuren, which is located in the green long around Brussels. Sciensano is a new born federal research centre in Belgium which finds its foundation in the concept of 'One Health'.

The IMEKOFOODS 4 conference brings together scientists from academia, laboratories and industry, control authorities and representatives of national and international agencies to exchange scientific results and to discuss the latest findings in food quality, -integrity, -safety, -traceability and nutrition. The conference will offer keynote lectures, oral and poster sessions, workshops, poster awards, vendor seminars and exhibition. A social program with get-2-gether party, visit of the Africa museum, gala dinner and Farewell BBQ will be offered in the splendid premises of the Africa Museum and in the park of Tervuren.

Tervuren is the ideal place for delegates to enjoy the beautiful green surroundings of Brussels with a walking or a biking trip, to visit the Africa Museum or to visit Brussels or Leuven. Due to his central location the historical cities Ghent, Bruges, Antwerp, and Liège… can be easily visited.

Welcome!

Joris Van Loco
Chair of the organizing committee

TABLE OF CONTENTS

COMMITTEES

Scientific Committee

Name		*Institute | company*
Mariana	Arce Osuna	CENAM - Mexico
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Name		*Institute | company*
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PLENARY AND KEYNOTE SPEAKERS

DR. URSKA VRHOVSEK

Metabolomic Unit, Food Quality and Nutrition Department
Research and Innovation Centre, Edmund Mach Foundation
San Michele all'Adige, Italy

Session: Plenary Lecture | Monday 16th of September 9:30

Title of lecture: Foodomics: a milestone in food and nutritional studies

Biography

Urska Vrhovsek got her Bachelor degree at Food sciences at Biotechnical faculty of University of Ljubljana, Slovenia, and a Ph.D. in Food sciences at Universität für Bodenkultur, Vienna, Austria. At present she is a senior researcher and currently leads the metabolomic unit at Edmund Mach Foundation. She was a visiting scientist at the Department of viticulture and enology at University of California Davis, Metabolomics Fiehn laboratory at the University of California Davis and at the Plant Products and Food Quality Department of The James Hutton Institute.

Her scientific fields of work are food chemistry and human nutrition. She is especially interested in studies of food composition especially berries, apples, grape and wine. The second part of her research activity is devoted to the studies of the mechanistic approaches of polyphenols absorption in different models, cell cultures, rats and humans. Her current primary scientific interest is metabolomics. She is a supervisor of graduate and postgraduate students in the fields of food chemistry and nutrition and author of more than 160 ISI papers.

Senior Scientist Jens J. Sloth

National Food Institute, Technical University of Denmark
Lyngby, Denmark

Session: Food Safety, Trace Elements, Contaminants | Monday 16th of September 11h10-h11h40

Title of lecture: Determination of trace elements in food - recent developments from research, reference lab and standardization activities

Biography

Jens Sloth is a MSc graduate from the Technical University in Denmark (1995) and later finalized his phd studies at the University of Bergen in Norway (2005). He has extensive experience within various research projects on toxic and essential trace elements in food and feed with special emphasis on the development of methods for trace element speciation (e.g. selenium, arsenic, iodine, mercury) using HPLC-ICPMS based methods and has been the project leader of several research projects with activities in this area. He is a lecturer in analytical food chemistry at DTU and has been supervisor for 25+ thesis works at PhD, MSc and BSc level. He has for more than a decade been an expert member of the CEN standardization committees for trace elements in food and feed and contributed to the development of new standardized methods of analysis for the determination of trace elements and their species. Recently, he was appointed as director of the European Reference Laboratory for metals and nitrogenous compounds in feed and food (EURL-MN) and also as convenor of the CEN expert group for Elements and their chemical species in animal feed (CEN TC327/WG4).

Prof. Jana Hajslova
University of Chemistry and Technology,
Prague, Czech Republic

Session: Food Integrity and Quality | Monday 16th of September 13:45

Title of lecture: Well established analytical methods for food quality and safety control: any risk of biased results?

Biography

Prof Jana Hajslova is the Head of Accredited (ISO17025) Food Quality and Safety Laboratory of the Department of Food Analysis and Nutrition at the University of Chemistry and Technology, Prague. She is a widely recognized expert in the field of food/natural products chemistry and analysis, having published more than 300 original papers on the development of advanced analytical strategies of contaminants, residues, natural toxins and other biologically active compounds, authentication and metabolomics. In 2016, Prof Hajslova obtained from Association of Official Analytical Chemists (AOAC Int.) prestigious scientific Harvey W. Wiley Award for her excellent scientific work. Her research team has participated in many international and national projects at both research, management and coordination levels, including the EC 5-7th Framework Programme, H2020, COSTs and EEA grants. Jana's team has been also involved in various bilateral international research activities. Under her supervision, close collaboration has taken place with many world-renowned institutions, such as WHO, FAO, USDA and the European Commission's Joint Research Centre. Currently, she is the Czech Republic delegate in HORIZON 2020 SOCIETAL CHALLENGE 2: "Food Security, Sustainable Agriculture and Forestry, Marine and Maritime and Inland Water Research and the Bioeconomy" committee. In her capacity as Chairwoman, she had a key input into establishing a series of reputable international symposia, the 'Recent Advances in Food Analysis' series, from 2003 till 2019

Dr. Frans verstraete

DG Health and Food Safety, European Commission
Brussels, Belgium

Session: Food Safety, Contaminants & Pesticides | Monday 16th of September 16h00-16h30

Title of lecture: EU policy on contaminants in food: recent developments and outlook

Biography

Frans Verstraete graduated in 1985 as agricultural engineer at the University of Ghent (Belgium). After his studies he held positions at the University of Ghent and thereafter at the Belgian Ministry of Agriculture and he was for a period technical adviser of the Belgian Minister of Agriculture. He is working for the European Commission since 1997. In the European Commission he has had various functions but since 2000 he is working at the Directorate General Health and Food Safety. He is responsible for the elaboration, development and management of the EU-legislation on contaminants in feed and food.

Prof. Sarah De Saeger

Centre of Excellence in Mycotoxicology and Public Health, Department of Bioanalysis, Faculty of Pharmaceutical Sciences, Ghent University
Ghent, Belgium

Session: Plenary Lecture | Tuesday 17th of September 9h00-9h40

Title of lecture: Recent technologies in the (bio)analysis of mycotoxins.

Biography

Prof. Dr. Sarah De Saeger is head of the Centre of Excellence in Mycotoxicology and Public Health at Ghent University, Belgium. She is coordinator of the international thematic network MYTOX-SOUTH.

As a full professor she is teaching all food-related courses in the Faculty of Pharmaceutical Sciences (Bromatology, Bioanalytical Practical, Food Safety, Special Nutrition).
The laboratory focuses on 4 research lines: mycotoxins and human health, detection methods, metabolomics and untargeted analysis, and mycotoxin occurrence. Many research proposals are running and funded by the EU H2020 programme, HERCULES, FWO, FOD, BELSPO, BOF, VLIR-UOS.

Research results are published in more than 300 A1 peer reviewed papers (h-index 41).
She was an expert in EFSA CONTAM working groups in the period 2011-2018 and she is a member of the Scientific Committee (SciCom) of the Belgian Federal Agency for Food Chain Safety since 2015. In June 2015 she established the Joint Laboratory of Mycotoxin Research of the Ghent University-Shanghai Jiao Tong University-Chinese Academy of Sciences (Shanghai Institutes of Biological Sciences). In 2015 she was awarded the Ghent University Prometheus Award for research.

, confirmatory LC-MS/MS, high resolution MS/MS, biomonitoring, human metabolism, MYTOX-SOUTH.

Prof. Lynn Vanhaecke

Ghent University, Lab of Chemical Analysis
Merelbeke, Belgium

Session: Food Omics | Tuesday 17th of September 9h50-10h40

Title of lecture: Nutrimetabolomics: integrative action for metabolomic analyses in human nutritional research as proposed by the Foodball consortium

Biography

Lynn Vanhaecke is a Master in Bioscience Engineering and was from 2004-2008 affiliated with the Lab of Microbial Ecology and Technology, Faculty of Bioscience Engineering at Ghent University as a PhD student. During this period her research focused on the impact of the human intestinal microbiota on the metabolism and biological activity of meat and environmental contaminants. In 2008 she graduated as a PhD in Bioscience Engineering and shifted to the Laboratory of Chemical Analysis within the Department of Veterinary Public Health and Food Safety at Ghent University, where she obtained a postdoctoral fellowship from the FWO-Vlaanderen. Since October 2011 she is appointed as Associate Professor and head of the Lab of Chemical Analysis. The chemical analyses of food, biofluids and environmental matrices, the metabolism and biological activity of food and contaminants, and the holistic analysis of small molecules using metabolomics using advanced high-resolution mass spectrometry in relation to human health belong to her major research objectives now. She is author and co-author of 184 peer-reviewed international publications and presented her work in many national and international conferences. Prof. Vanhaecke is also one of the 5 promotors of MSsmall (Mass Spectrometry for Small Organic Molecules: 14 UGent partners from 5 faculties). Prof. Vanhaecke is member of the Ghent University Research Council, and of several scientific committees of international symposia.

Prof. Dr. Cristina Nerín

University of Zaragoza, I3A, Dept. Analytical Chemistry
Zaragoza, Spain

Session: Food Contact Materials | Tuesday 17th of September 13h45-15h15

Title of lecture: Analysis of food contact materials: IAS and NIAS

Biography

Professor at the University of Zaragoza (Spain), PhD in Analytical Chemistry, Master in Science and Degree in Chemistry at the University of Zaragoza (Spain). Director of the Research Group GUIA. Director of the Master in Environmental Engineering (1990-2012). Author of > 340 Scientific Publications, Director of 37 PhD Thesis (+5 in process). Principal Investigator of > 200 Research Projects in Competitive calls and R&D&i with Industry. Inventor in 7 International Patents on active packaging and one International Patent on Intelligent Packaging, all in exploitation. 6 Research Awards received. Participant in >140 International Conferences and Professor of several International Courses in Europe, Asia and South-America. Organizing Committee and Chairwomen of several International Symposiums and > 50 Plenary Lectures in International Conferences.

Member of WG Recycling in EFSA from 2010 to end 2018. Member of Scientific Panel in AESAN in 2010-2015. Board of Directors in ILSI Europe (2019-2021). Editor of Packaging Technology and Science journal.

Evaluator of R&D&i Projects (EU-VII Frame Program, H2020, Chile, Brasil, The Netherlands, Austria, Belgium, Portugal, Argentina, France, Saudi Arabia, USA, Israel, Spain). Evaluator of PhD Thesis in Spain, Belgium, France, Sweden, India, Ireland, South Africa, New Zealand, Australia. Referee of many Scientific Journals in Analytical Chemistry, Materials Science, Food Chemistry, Food Technology, Food Packaging, Food Science, Environmental…

Prof. Michael Rychlik
TUM, Analytical Food Chemistry
Munich, Germany

Session: Workshop Toxins | Tuesday 17th of September 13h45-15h15

Title of lecture: Alternaria toxins: Analysis and risk assessment of emerging and modified mycotoxins

Biography

Prof. Dr. Michael Rychlik is the Head of the Chair of Analytical Food Chemistry at the Technical University of Munich, Germany (TUM).
He graduated in food chemistry at the University of Kaiserslautern in 1988.
His PhD studies on the flavour of bread were completed in 1996 and he was appointed full professor at the TUM in 2010.
In 2015 he served as a Visiting Professor at the University of Queensland (UQ), Australia and in 2016 he was appointed an Honorary Professor at the latter University.
In the last years he was also active as a Visiting Professor in 2016 at the National University of Singapore (NUS) and i salso teaching since 2018 at the University of Hong Kong.
At the TUM School of Life Sciences Weihenstephan, Michael Rychlik is engaged as the Director of the Research Department Nutrition and Food Sciences since 2016.

His group has been working for 15 years in the field of developing analytical methods for bioactive food components, in particular for vitamins, mycotoxins, odorants and lipids. For these compounds, he developed stable isotope dilution assays that reveal superior accuracy. Moreover, his research is focused on the application of these methods to recent areas in food chemistry, technology, toxicology and nutrition. Since 2014 he serves as the Head of the “Committee on Contaminants in the Food Chain” at the Federal Institute for Risk Assessment, Berlin, Germany.

Dr. Johanna Noireaux
LNE, Biomedical and inorganic chemistry department
Paris, France

Session: Nanomaterials and Microplastics | Wednesday 18th of September 9h00-10h30

Title of lecture: Perspective in nanoparticle analysis in food with single particle ICPMS

Biography

I received a PhD in geochemistry at IPGP (Institute for earth's physics) in Paris, with a specialization on boron isotope ratio measurements and application. During that time I became an expert on ICPMS measurements, mainly trace analysis and isotope ratios.

I joined Paola Fisicaro's group at LNE in 2017 where I continued my work on trace element analysis and developed an interest on nanoparticles. I started training myself on single particle ICPMS analysis and my work now focuses on sample preparation and data treatment with this technique and others available at the nanoparticle characterization platform at LNE.

LNE is partner of the Research Infrastructure METROFOOD, where it is recognized for its role of National Metrology Institute. LNE brings its competencies in the development of metrological references such as reference materials and methods, in particular in the field of inorganic and organic contaminants and NP characterization.

Dr. Ralf Kaegi
Eawag, Swiss Federal Institute for Water Science and Technology,
Department of Process Engineering
Duebendorf, Switzerland

Session: Nanomaterials & Microplastics | Wednesday 18th of September 11h00-11h30

Title of lecture: Quantification of (Engineered) Nanoparticles in Complex Matrices: More than a Silver Lining on the Horizon?

Biography

Ralf Kaegi studied Earth sciences at University of Basel and did his PhD in the field of magmatic petrology at ETH Zürich. He then spent 5 years as a scientist at the Swiss Federal Institute for Materials Science and Technology (Empa) in the field of aerosol science. During that time, the research of Ralf Kaegi was focused on the representative collection, detection and quantification of ultrafine particles in the atmosphere using electron microscopy techniques. In 2006, Ralf Kaegi joined Eawag as a scientist and leader of the particle laboratory. The main research interests of Ralf Kaegi include the fate and transformation of engineered nanoparticles in urban (waste)water systems. For that purpose his group has established a suite of pilot scale facilities, including a wastewater treatment plant simulating the activated sludge process, an anaerobic digester and a fluidize bed incinerator. This unique infrastructure allows investigating the behavior of engineered nanomaterials under most realistic conditions. In addition and related to these activities, the group of Ralf Kaegi is involved in the development of new analytical methods to detect and quantify engineered nanoparticles in aqueous suspensions and complex matrices, in general.

SCIENTIFIC PROGRAM

Monday 16th

8h00	**Registration**		
9h00	**Opening** *J. Van Loco | Welcome word of the symposium Chair* *I. Castanheira | Welcome word of IMEKO*		
	Chair: I. Castanheira (INSA,PT)		
9h30	**Plenary presentation** **Prof U. Vrhovsek (FMACH, IT)** **Foodomics: a milestone in food and nutritional studies**		
10h10	**Coffee Break & Posters Exhibition**		
Auditorium	**Session**: Food Safety, Trace Elements, Contaminants	L333	**Session:** Food Integrity / Quality
	Chairs: N. Ogrinc (IJS, SI) and K. Cheyns (Sciensano, BE)		**Chairs:** N. Belc (IBA, RO) and S. Malysheva (Sciensano, BE)
11h10	**Key note presentation**	11h10	The effect of temperature on the nutritional quality of edible mealworm Tenebrio molitor
	J. J. Sloth (DTU) | Determination of trace elements in food - recent developments from research, reference laboratory and standardization activities		*L. Kouřimská, M. Kulma, A. Nevrzalová, D. Homolková*
11h40	Nickel in feed and food – Results of proficiency tests by the EURL-MN	11h30	Direct elemental analysis of cereal and rice flour using total reflection X-ray fluorescence: new challenges
	H. Amlund, H. Fodnæss, A. Landin and J.J. Sloth		*F. Bilo, L. Borgese, C. Zoani, G. Zappa, R. Dalipi, L. E. Depero*
12h00	Occurrence of perfluoroalkylated substances (PFAS) in drinking water in Czech Republic	11h50	Development of the analytical method for 87Sr/86Sr determination in olive oil
	J. Pulkrabova, M. Buresova, D. Lankova and J. Hajslova		*M. Furdek Turk, E. Epova, J. Barre, S. Berail, O. X. Donard and T. Zuliani*
12h20	Is there too much lead in Belgian big game meat? *'Short communication'*	12h10	Nutrients, secondary metabolites and anti-oxidant activity Moringa oleifera leaves and Moringa-based commercial products -
	A. Ruttens, J. Casaer, C. Marien, A. Rutten and N. Waegeneers		*N.S. Mokgalaka, M.Y. Aphane, V.J. Tembu and L.M. Cele*
12h30	**Lunch**		
Auditorium	**Session:** Food Safety, Allergens	*L333*	**Session:** Food Integrity / Quality
	Chairs: N. Belc (IBA, RO) and S. De Vos (Sciensano, BE)		**Chairs:** C. Zoani (ENEA, IT) and K. Vandermeiren (Sciensano, BE)
13h45	Flaws and hurdles concerning the harmonization of detecting allergens in food	13h45	**Key note presentation**
	K. Van Vlierberghe, M. Gavage, M. Dieu, P. Renard, T. Arnould, N. Gillard, I. Taverniers, M. De Loose, K. Gevaert, C. Van Poucke		*J. Hajslova (UCT)* | Well established analytical methods for food quality and safety control: any risk of biased results?

14h05	Quantification of SO2 in wines by Surface Enhanced Raman Spectroscopy and the comparison with the official OIV method *I. Cagnasso, L. Berta, L. Mandrile, A.M. Giovannozzi, M. Petrozziello, A.M. Rossi, F. Durbiano*	14h15	Enhanced capability of a purge-and-trap, thermal desorption and GCxGC-MS methodology for aroma profiling *F. A. Franchina, D. Zanella, E. Lazzari, P-H. Stefanuto, and J-F. Focant*
14h25	Development of a DNA barcoding-like approach to detect mustard allergens in wheat flours *J. Frigerio, R. Pellesi, V. Mezzasalma , F. De Mattia , A. Galimberti , F. Lambertini , M. Suman , S. Zanardi , A. Leporati and M. Labra.*	14h35	Implementing sodium reduction in food regulations in South Africa – the analytical measurement challenges *M. Fernandes-Whaley, M. Linsky and D. Prevoo-Franzsen*
14h45	**Coffee Break & Exhibition**		
Auditorium 15:10-15:50	**Vendor Presentations** **Waters** Advances in Ion Mobility Mass Spectrometry for Food Analysis *J. Claereboudt*	*L308* 15:10-17:00	**QualiT - A new Quality Control Toolbox for Mycotoxin and Allergen Analysis** (offered by r-Biopharm AG)
	chair: S. Goscinny (Sciensano, BE)		**chair:** B. Huybrechts (Sciensano, BE)
Auditorium	**Session:** Food Safety, Contaminants & Pesticides	*L333*	**Session:** Food Safety / Food Hygiene
	Chairs: J.M. Rodrigues (INMETRO, BRA) and L. Joly (Sciensano, BE)		**Chairs:** F. Limonier (Sciensano, BE) and M. C. Garcia-Graels (Sciensano, BE)
16h00	**Key note presentation** *F. Verstraete (DG Santé)* | EU policy on contaminants in food: recent developments and outlook	16h00	Monitoring on hygiene in institutional kitchens in Belgium *E. Duthoo, S. Krings, G. Daube, B. Taminiau, M. Heyndrickx, and K. De Reu*
16h30	Multi-approach determination of dithiocarbamate fungicides and of their degradation products in fruits and vegetables *A. C. Dirtu, G. Lavison-Bompard, A. Ducrocq, C. Inthavong, T. Guérin, P. Jitaru*	16h20	Detection and quantification of biogenic amines in Cambodian smoked freshwater fish *C. Douny, H. Mith, F. Brose, A. Igout, M-L. Scippo*
16h50	New developments in integrated 'sample to results' workflows for the multi-residue analysis of polar anionic pesticides and their metabolites *R.J. Fussell, F. Pigozzo, Q. Guo, Y. Li and T. Bo, E. George*	16h40	Proposal for a European Metrology Network on food safety – EMN-FS *A. M. Rossi and F. Durbiano*
17h00-19h30	**Get together party**		

Tuesday 17th

Auditorium

9h00	**Plenary lecture** **Prof. S. De Saeger (UGent)** **Recent technologies in the (bio)analysis of mycotoxins**		
	chair: C. Zoani (ENEA, IT)		
Auditorium	**Session**: Food Omics	*L333*	**Session:** Proficiency Testing & Reference Materials
	Chairs: K. Presser (PREMOTEC, CH) and S. De Keersmaecker (Sciensano, BE)		**Chairs:** N. Ogrinc (IJS, SI) and E. Tangni (Sciensano, BE)
9h50	**Key note presentation** *Prof. L. Vanhaecke (UGent)* | Nutrimetabolomics: integrative action for metabolomic analyses in human nutritional research as proposed by the Foodball consortium	9h50	Certified reference material of nitrofuran metabolites in chicken breast muscle from incurred samples *F. G. M. Violante, B. C. Garrido, E. C. P. Rego, E. F. Guimarães; N. O. C. Zúniga, W. Wollinger; J. M. Rodrigues; F. R. Aquino Neto.*
10h20	Avoiding the culture step in outbreak investigations: parameters for optimised metagenomics of contaminated food *F. Buytaers, A. Saltykova, S. Denayer, B. Verhaegen, N. Roosens, K. Vanneste, D. Piérard, K. Marchal, S. C. J. De Keersmaecker*	10h10	Proficiency Testing Scheme for Benzoic acid in Banana-based Condiment to Support the Traceability of Chemical Measurements to SI units *B. S. Ebarvia, A. C. Dacuya, A. R. C. Veranga, J. A. C. Valdueza*
10h30	**Coffee Break & Poster Exhibition**		
Auditorium	**Session:** Food Genomics	*L333*	**Session:** Proficiency Testing, Reference Materials and accreditation
	Chairs: M. Heyndrickx (ILVO, BE) and N. Roosens (Sciensano, BE)		**Chairs:** M. Fernandes-Whaley (NMISA, ZA) and K. Cheyns (Sciensano, BE)
11h30	Detection of antibiotic resistance genes in microbial fermentation products *M-A. Fraiture, M. Deckers, N. HC Roosens*	11h30	Development of new stable isotope reference materials for food authentication and traceability *N. Ogrinc, A. Schimmelmann, F. Camin, D. Potočnik, H. Qi and S. Kelly*
11h50	Reconstruction of plasmids carrying antimicrobial resistance genes in food, feed and human bacterial isolates using short and long read sequencing reads *B. Berbers, A. Saltykova, P.J. Ceyssens, C. Garcia-Graells, K. Vanneste, N. H. Roosens, K. Marchal, S. C.J. De Keersmaecker*	11h50	ICAR proficiency testing scheme and a novel calculation model to compare proficiency testing schemes *S. Orlandini*
12h10	Rationalizing the GMO analytical detection procedure: optimization of subsampling, homogenization and milling steps	12h10	International co governance of food safety Based on quality infrastructure

	I. Taverniers, S. Liévrard, J. Baert, M Dhondt, A. Staelens, M. De Loose		*L. Jun , X. Xuelin, F. Xiang*			
12:30-13:45	**Lunch**					
Auditorium	**Session:** Food Contact Materials			*L333*	**Workshop Toxins**	
	Chairs: I. Castanheira (INSA, PT) and E. Van Hoeck (Sciensano, BE)				**Chair:** A. Troupiotis-Tsaïlaki (Sciensano, BE)	
13h45	**Key note presentation** *Prof. C. Nerin (UNIZAR)* | Analysis of individual (MOAH) by APGC- QTOF-MS and comparison to the conventional method LC-GC-MS			*13h45*	**Key note presentation** *Prof. Dr. M. Rychlik (TUM)* | Alternaria toxins: Analysis and Risk assessment of Emerging and Modified Mycotoxins	
14h15	Mineral oil in food: How is the situation in Belgium and what are the risks? *A. Van Heyst, S. Goscinny, B. Mertens, M. Vanlancker, J. Vercammen, S. Bel, S. Vandevijvere, M. Elskens, E. Van Hoeck*			14h15	MYCOSUGAR: Investigation on mycotoxins and their producing fungi in sugarcane and its by-products *M.F. Abdallah, C. Bereket, V. Kagot, L. Matumba, S. Okoth, M. De Boevre, G. Haesaert, K. Audenaert and S. De Saeger*	
14h35	Effect of food composition on the migration of surrogate contaminants from paperboard packaging *M. Baele, A. Vermeulen, P. Ragaert, B. De Meulenaer*			14h35	Occurrence, toxicokinetics and toxicity of citrinin and risk assessment *C. Meerpoel, A. Vidal, B. Huybrechts, E.K. Tangni, M. Devreese, S. Croubels and S. De Saeger*	
14h55	Investigation of migrating substances from food fabrics *K. Van Den Houwe, B. Dewilde, J. Van Loco, S. Goscinny and E. Van Hoeck*			14h55	Analytical methods for mycotoxin determination *B. Huybrechts*	
15:15-16:20	**Coffee break** **Exhibition**					
Auditorium 15:30-16:10	**Vendor presentations** **ABSciex** Ensuring the authenticity & safety of food with new advances in LC-MS/MS workflows *D. McMillan* **chair:** S. Goscinny (Sciensano, BE)			L308 15:30-17:20	**QualiT - A new Quality Control Toolbox for Mycotoxin and Allergen Analysis (offered by r-Biopharm AG)** **chair:** B. Huybrechts (Sciensano, BE)	
Auditorium	**Session:** Nutrition	*L333*	**Session:** Metrofood-RI			
	Chairs: L. Kouřimská (CZU, CZ) and M. Andelkovic (Sciensano, BE)		**Chair**: Claudia Zoani (ENEA, IT)			
16h20	Calcium and vitamin D intake from foods and supplements in the Dutch population *MC Ocké, HAM Brants, CS Dinnissen, J Verkaik-Kloosterman, CTM van Rossum*	16h00 16h15	The Eurachem contribute to quality of measurements (analysis/sampling) in the agrifood sector | *I. Vercruysse (Eurachem)* ISO/IDF - role of standardisation in the dairy sector | *S. Orlandini (ISO/IDF)*			
16h40	School meals in light of the regulation – Assessment of the public catering decree in primary schools in Hungary *A. Varga, M. Bakacs, A. Zentai, B. Nagy, Z. Nagy-Lőrincz, G. Erdei, É. Illés, V. Varga-Nagy, K. Miháldy, E. Sarkadi Nagy, C. Kaposvári*	16h25 16h35	Food Metrology - Food safety laboratory at METAS | *S. Mallia (METAS)* Characterization of nanomaterials in food by Transmission Electron Microscopy | *K. Vandermeiren (Sciensano)*			

17h00	Probiotic properties of lactic acid bacteria isolated from household fermented Sorghum slurries	16h45	Improvement of food quality and safety during food processing: pilot plant experiments | *N. Belc (IBA)*
	M.L. Thaoge and S. Rapoo	16h55	Data standards, data exchange and the electronic part of METROFOOD-RI | *K. Presser (PREMOTEC)*
		17h05	Circular Economy for Food - Partnership for Sustainable Coastal Cities in the Western Mediterranean | *C. Chiavetta (ENEA)*
17:30-19:00	**TC 23 Meeting (invitation only)**		
20:00-23:30	**Gala Diner Koloniënpaleis, Paleizenlaan, 3080 Tervuren**		
	Wednesday 18th		
	Auditorium		
	Session: Nanomaterials & Microplastics		
	Chairs: K. Smeets (Uhasselt, BE), J. Mast (Sciensano, BE)		
9h00	**Key note presentation**		
	J. Noireaux | Perspective in nanoparticle analysis in food with single particle ICPMS		
9h30	Method validation for determination of microplastics in food		
	M. Dekimpe, D. Deloof, J. Robbens, K. Hostens, B. De Witte		
9h50	Characterization of the nano-sized fraction of silver particles in food additive E174 by EM and SP-ICP-MS		
	S. De Vos, E. Verleysen, M. Ledecq, N. Waegeneers and J. Mast		
10h10	Inhibition of pathogenic bacteria in duck meat using nanoclay encapsulated oregano essential oil		
	P. Klouček, J. Táborský, N. West, A. Fraňková and M. Božik		
10h30	**Coffee break**		
	Auditorium		
	Session: Nanomaterials & Microplastics		
	Chairs: K. Smeets (Uhasselt, BE), J. Mast (Sciensano, BE)		
11h00	**Key note presentation**		
	R. Kaegi | Quantification of (Engineered) Nanoparticles in Complex Matrices: More than a Silver Lining on the Horizon?		
11h30	Physicochemical characterisation of several types of the E171 food additive		
	E. Verleysen, M. Ledecq, S. De Vos, I. Ojea Jimenez, F. Brassinne, N. Waegeneers and J. Mast		
11h50	Towards a routine method for the characterisation of TiO2 nanoparticles in food by single particle-ICP-MS		
	L. Givelet, P. Jitaru, D. Truffier-Boutry, J.-F. Damlencourt and T. Guérin		
12h10	*Validation of single particle ICP-MS for routine sizing and quantification of the fraction of silver nanoparticles in E174 food additives and confectionery products.*		
	N. Waegeneers, L. Delfosse, S. De Vos, E. Verleysen, J. Mast		
12:30-13:15	**Best poster award** **Announcement Imekofoods 5** **Closing ceremony**		
13:30-15:30	**Farewell BBQ**		

LIST OF PLENARY AND KEYNOTE PRESENTATIONS

PL1 - Foodomics: a milestone in food and nutritional studies
U. Vrhovsek
Monday 16th of September 9h30-10h10 - Auditorium

K01 - Determination of trace elements in food - recent developments from research, reference laboratory and standardization activities
J.J. Sloth
Monday 16th of September 11h10-11h40 - Auditorium

K02 - Well established analytical methods for food quality and safety control: any risk of biased results?
J. Hajslova, M.Stupak, Z. Dzuman, V. Kocourek and J. Pulkrabova
Monday 16th of September 13h45-14h15 – L333

K03 - EU policy on contaminants in food: recent developments and outlook
F. Verstraete
Monday 16th of September 16h00-16h30 - Auditorium

PL2 - Recent technologies in the (bio)analysis of mycotoxins
S. De Saeger
Tuesday 17th of September | 9h00-9h40 - Auditorium

K04 - Nutrimetabolomics: integrative action for metabolomics analyses in human nutritional research as proposed by the Foodball consortium
L. Vanhaecke, M. M. Ulaszewska, C. Weinert, A. Trimigno, R. Portmann, G. Vergères
Tuesday 17th of September 9h50-10h20 - Auditorium

K05 - Analysis of individual (MOAH) by AGPC-QTOF-MS and comparison to the conventional method LC-GC-MS
J. Jaén, C. Domeño, P. Alfaro, C. Nerín
Tuesday 17th of September 13h45-14h15 - Auditorium

K06 - Alternaria toxins: Analysis and Risk assessment of Emerging and Modified Mycotoxins
M. Rychlik
Tuesday 17th of September 13h45-14h15 – L333

K07 - Perspective in nanoparticle analysis in food with single particle ICPMS
J. Noireaux, P. Fisicaro
Wednesday 18th of September 9h00-9h30 - Auditorium

K08 - Quantification of (Engineered) Nanoparticles in Complex Matrices: More than a Silver Lining on the Horizon?
R. Kaegi, T. Uusimäki
Wednesday 18th of September 11h00-11h30 - Auditorium

LIST OF ORAL COMMUNICATIONS

Session Food safety, Trace Elements and Contaminants - Auditorium

OC01 - Nickel in feed and food – Results of proficiency tests by the EURL-MN
H. Amlund, H. Fodnæss, A. Landin and J.J. Sloth
Monday 16th of September 11h40-12h00

OC02 - Occurrence of perfluoroalkylated substances (PFAS) in drinking water in Czech Republic
J. Pulkrabova, M. Buresova, D. Lankova and J. Hajslova
Monday 16th of September 12h00-12h20

OC03 - Is there too much lead in Belgian big game meat?
A.Ruttens
Monday 16th of September 12h20-12h30

Session Food Integrity / Quality – L333

OC04 - The effect of temperature on the nutritional quality of edible mealworm Tenebrio molitor
L. Kouřimská, M. Kulma, A. Nevrzalová, D. Homolková
Monday 16th of September 11h10-11h30

OC05 - Direct elemental analysis of cereal and rice flour using total reflection X-ray fluorescence: new challenges
F. Bilo, L. Borgese, C. Zoani, G. Zappa, R. Dalipi, L. E. Depero
Monday 16th of September 11h30-11h50

OC06 - Development of the analytical method for 87Sr/86Sr determination in olive oil
M. Furdek Turk, E. Epova, J. Barre, S. Berail, O. X. Donard and T. Zuliani
Monday 16th of September 11h50-12h10

OC07 - Nutrients, secondary metabolites and anti-oxidant activity Moringa oleifera leaves and Moringa-based commercial products
N.S. Mokgalaka, M.Y. Aphane, V.J. Tembu and L.M. Cele
Monday 16th of September 12h10-12h30

OC08 - Enhanced capability of a purge-and-trap, thermal desorption and GCxGC-MS methodology for aroma profiling
F. A. Franchina, D. Zanella, E. Lazzari, P-H. Stefanuto, and J-F. Focant
Monday 16th of September 14h15-14h35

OC09 - Implementing sodium reduction in food regulations in South Africa – The analytical measurement challenges
M. Fernandes-Whaley, M. Linsky and D. Prevoo-Franzsen
Monday 16th of September 14h35-14h55

Session Food Safety, Allergens – Auditorium

OC10 - Flaws and hurdles concerning the harmonization of detecting allergens in food
K. Van Vlierberghe, M. Gavage, M. Dieu, P. Renard, T. Arnould, N. Gillard, I. Taverniers, M. De Loose, K. Gevaert, C. Van Poucke
Monday 16th of September 13h45-14h05

OC11 - Quantification of SO2 in wines by Surface Enhanced Raman Spectroscopy and the comparison with the official OIV method
I. Cagnasso, L. Berta, L. Mandrile, A.M. Giovannozzi, M. Petrozziello, A.M. Rossi, F. Durbiano
Monday 16th of September 14h05-14h25

OC12 - Development of a DNA barcoding-like approach to detect mustard allergens in wheat flours
J. Frigerio, R. Pellesi, V. Mezzasalma , F. De Mattia , A. Galimberti , F. Lambertini , M. Suman , S. Zanardi , A. Leporati and M. Labra.
Monday 16th of September 14h25-14h45

Session Food Safety, Contaminants and Pesticides - Auditorium

OC13 - Multi-approach determination of dithiocarbamate fungicides and of their degradation products in fruits and vegetables
A.C. Dirtu, G. Lavison-Bompard, A. Ducrocq, C. Inthavong, T. Guérin, P. Jitaru
Monday 16th of September 16h30-16h50

OC14 - New developments in integrated 'sample to results' workflows for the multi-residue analysis of polar anionic pesticides and their metabolites
R.J. Fussell, F. Pigozzo, Q. Guo, Y. Li and T. Bo, E. George
Monday 16th of September 16h50-17h10

Session Food Safety / Food Hygiene –L333

OC15 - Monitoring on hygiene in institutional kitchens in Belgium
E. Duthoo, S. Krings, G. Daube, B. Taminiau, M. Heyndrickx, and K. De Reu
Monday 16th of September 16h00-16h20

OC16 - Detection and quantification of biogenic amines in Cambodian smoked freshwater fish
C. Douny, H. Mith, F. Brose, A. Igout, M-L. Scippo
Monday 16th of September 16h20-16h40

OC17 - Proposal for a European Metrology Network on food safety – EMN-FS
A. M. Rossi and F. Durbiano
Monday 16th of September 16h40-17h00

Session Food Omics - Auditorium

OC18 - Avoiding the culture step in outbreak investigations: parameters for optimised metagenomics of contaminated food
F. Buytaers, A. Saltykova, S. Denayer, B. Verhaegen, N. Roosens, K. Vanneste, D. Piérard, K. Marchal, S. C. J. De Keersmaecker
Tuesday 17th of September 10h20-10h40

Session Proficiency Testing & Reference Materials – L333

OC19 - Certified reference material of nitrofuran metabolites in chicken breast muscle from incurred samples
F.G.M. Violante, B. C. Garrido, E. C. P. Rego, E. F. Guimarães; N. O. C. Zúniga, W. Wollinger; J.M. Rodrigues, F. R. Aquino Neto.
Tuesday 17th of September 9h50-10h10

OC20 - Proficiency testing scheme for benzoic acid in banana-based condiment to support the traceability of chemical measurements to SI units
B. S. Ebarvia, A. C. Dacuya, A. R. C. Veranga, J. A. C. Valdueza
Tuesday 17th of September 10h10-10h30

Session Food Genomics - Auditorium

OC21 - Detection of antibiotic resistance genes in microbial fermentation products
M-A. Fraiture, M. Deckers, N. HC Roosens
Tuesday 17th of September 11h30-11h50

OC22 - Reconstruction of plasmids carrying antimicrobial resistance genes in food, feed and human bacterial isolates using short and long read sequencing reads
B. Berbers, A. Saltykova, P.J. Ceyssens, C. Garcia-Graells, K. Vanneste, N. H. Roosens, K. Marchal, S. C.J. De Keersmaecker
Tuesday 17th of September 11h50-12h10

OC23 - Rationalizing the GMO analytical detection procedure: optimization of subsampling, homogenization and milling steps
I. Taverniers, S. Liévrard, J. Baert, M Dhondt, A. Staelens, M. De Loose
Tuesday 17th of September 12h10-12h30

Session Proficiency Testing, Reference Materials and accreditation – L333

OC24 - Development of new stable isotope reference materials for food authentication and traceability
N. Ogrinc, A. Schimmelmann, F. Camin, D. Potočnik, H. Qi and S. Kelly
Tuesday 17th of September 11h30-11h50

OC25 - ICAR proficiency testing scheme and a novel calculation model to compare proficiency testing schemes
S. Orlandini
Tuesday 17th of September 11h50-12h10

OC26 - International co governance of food safety based on quality infrastructure
L. Jun , X. Xuelin, F. Xiang
Tuesday 17th of September 12h10-12h30

Session Food Contact Materials - Auditorium

OC27 - Mineral oil in food: How is the situation in Belgium and what are the risks?
A. Van Heyst, S. Goscinny, B. Mertens, M. Vanlancker, J. Vercammen, S. Bel, S. Vandevijvere, M. Elskens, E. Van Hoeck
Tuesday 17th of September 14h15-14h35

OC28 - Effect of food composition on the migration of surrogate contaminants from paperboard packaging
M. Baele, A. Vermeulen, P. Ragaert, B. De Meulenaer
Tuesday 17th of September 14h35-14h55

OC29 - Investigation of migrating substances from food fabrics
K. Van Den Houwe, B. Dewilde, J. Van Loco, S. Goscinny and E. Van Hoeck
Tuesday 17th of September 14h55-15h15

Workshop Toxins – L333

OC30 - MYCOSUGAR: Investigation on mycotoxins and their producing fungi in sugarcane and its by-products
M.F. Abdallah, C. Bereket, V. Kagot, L. Matumba, S. Okoth, M. De Boevre, G. Haesaert, K. Audenaert and S. De Saeger
Tuesday 17th of September 14h15-14h35

OC31 - Occurrence, toxicokinetics and toxicity of citrinin and risk assessment
C. Meerpoel, A. Vidal, B. Huybrechts, E.K. Tangni, M. Devreese, S. Croubels and S. De Saeger
Tuesday 17th of September 14h35-14h55

OC32 - Analytical methods for mycotoxin determination
B. Huybrechts
Tuesday 17th of September 14h55-15h15

Session Nutrition - Auditorium

OC33 - Calcium and vitamin D intake from foods and supplements in the Dutch population
MC Ocké, HAM Brants, CS Dinnissen, J Verkaik-Kloosterman, CTM van Rossum
Tuesday 17th of September 16h20-16h40

OC34 - School meals in light of the regulation – Assessment of the public catering decree in primary schools in Hungary
A. Varga, M. Bakacs, A. Zentai, B. Nagy, Z. Nagy-Lőrincz, G. Erdei, É. Illés, V. Varga-Nagy, K. Miháldy, E. Sarkadi Nagy, C. Kaposvári
Tuesday 17th of September 16h40-17h00

OC35 - Probiotic properties of lactic acid bacteria isolated from household fermented Sorghum slurries
M.L. Thaoge and S. Rapoo
Tuesday 17th of September 17h00-17h20

Session Nanomaterials and Microplastics - Auditorium

OC36 - Method validation for determination of microplastics in food
M. Dekimpe, D. Deloof, J. Robbens, K. Hostens, B. De Witte
Wednesday 18th of September 9h30-9h50

OC37 - Characterization of the nano-sized fraction of silver particles in food additive E174 by EM and sp-ICP-MS
S. De Vos, E. Verleysen, M. Ledecq, N. Waegeneers and J. Mast
Wednesday 18th of September 9h50-10h10

OC38 - Inhibition of pathogenic bacteria in duck meat using nanoclay encapsulated oregano essential oil

P. Klouček, J. Táborský, N. West, A. Fraňková and M. Božik

Wednesday 18th of September 10h10-10h30

OC39 - Physicochemical characterisation of several types of the E171 food additive

E. Verleysen, M. Ledecq, S. De Vos, I. Ojea Jimenez, F. Brassinne, N. Waegeneers and J. Mast

Wednesday 18th of September 11h30-11h50

OC40 - Towards a routine method for the characterisation of TiO2 nanoparticles in food by single particle-ICP-MS

L. Givelet, P. Jitaru, D. Truffier-Boutry, J.-F. Damlencourt and T. Guérin

Wednesday 18th of September 11h50-12h10.

OC41 - Validation of single particle ICP-MS for routine sizing and quantification of the fraction of silver nanoparticles in E174 food additives and confectionery products.

N. Waegeneers, L. Delfosse, S. De Vos, E. Verleysen, J. Mast

Wednesday 18th of September 12h10-12h30

LIST OF POSTER COMMUNICATIONS

Session A | Food Safety

1. **Isolation and identification of microorganisms from processed milkfish products for the development of matrix-based PT material for salmonella sp. in milkfish**
   M.S.A. Aguinaldo, A.P. de Asis, N.M.L. Dela Cruz and S.M. Estrada

2. **Antioxidant and antimicrobial activities of leaves extracts of urtica dioica L. from Algeria: application in apple fruits**
   A. Ouelhadj, L. Khati , D. Tadjenant

3. **Occurrence of polyether ionophores residues in Brazilian Minas cheese**
   F. R. N. Silva, P. A. de C. Braga, F. G. R. Reyes, M. F. Capristo, A. P. Arisseto-Bragotto

4. **Use of additives in Brazilian dairy products and compliance with current legislation**
   A. V. Botaro, A. P. Arisseto Bragotto

5. **Ultra-low level quantification of pesticides in baby foods using an advanced triple quadrupole GC-MS/MS**
   R. J. Fussell, R. Law, A. Lamb, P. Silcock, T. Anderson, J. Cole and C. Cojocariu

6. **Fluorimetric method and trends in aflatoxins determination**
   A.Najdenkoska, P.Misevski, B.Georgievski

7. **Optimization and validation of HS-SPME GC/MS to measure furan and alkylfurans in babyfood**
   Z. Alsafra, G. Scholl, B. De Meulenaer, G. Eppe.

8. **Investigation of the Polycyclic Aromatic Hydrocarbons contamination in spices and dried herbs available on the Belgian market**
   Ph. Szternfeld

9. **Exposure to pesticides and metals following tea consumption in Belgium**
   K. Cheyns, Ph. Szternfeld

10. **Development and application of a novel analytical methodology for simultaneous speciation analysis of Cr(III) and Cr(VI) in foodstuffs by HPLC-ICP-MS using species-specific isotope dilution**
    M. Saraiva, A. Leufroy, R. Chekri, T. Guérin, J.J. Sloth and P. Jitaru

11. **SIMBA: Design, formulation and optimization of plant growth-promoting microbes for their use as microbial consortia inoculants**
    A. Bevivino, C. Cantale, S. Tabacchioni, P. Ambrosino, S. Passato, C. Nobili, A. Fiore, A. Del Fiore, O. Presenti, G. Giovannetti, D. Neuhoff, M. Sudau, E. Maestri, M. Caldara, N. Marmiroli, S.J. Sørensen, J. Nesme, T. Evison, A. Sczyrba, A. Schlüter, A. Brunori, A. Pihlanto

12. **Toxic element mobility in soil-wheat system: comparison between 6 wheat varieties**
    E. Pucci, G. Zappa, C. Zoani, L. Gazza, C. Manetti

13. **Accomplishment of the EU regulations 2017/644-771 for PCDD/Fs and PCBs in food by using a novel triplequadrupole MS generation**
    E. Lazzari, F. A. Franchina, G. Scholl and J-F. Focant

14. **Aluminium in food**
N. Møller Iversen, H. Amlund, B. Koch Herbst and J.J. Sloth

16. **Pilot study for developing an interactive platform to facilitate communication between main actors in the food packaging chain**
G. Mustatea, E. L. Ungureanu, D. E. Duta and N. Belc

17. **Fast Analysis of a Multi-class Pesticides Panel in Wine and Olive Oil Extracts by LC-MS/MS**
L. De Keyzer, B. Miserez, C. Brodie, A. Hilkert, O. Kracht, D. Juchelka and J. Radke

18. **Food and Beverage Fraud Prevention Using Isotope Fingerprints**
B. Miserez, I. Paolini, S. Bani, D. D'Addona, . T. Yang and D. Ghosh

Session B | Food Data

1. **Smart Store-Keep your sample and data safe forever.**
F. Bilo, L. Borgese, A. Zacco, E. Bontempi, L. E. Depero

2. **The use of Biotechnology to improve the use of indigenous complementary food**
M.L. Thaoge and M.M. Mosele

Session C | Food Integrity

1. **Comparison between ELISA and qPCR kits for detection of hazelnut incurred in rice, cookies and chocolate samples**
I. Taverniers, S. Maes, M. Dhondt, N. Victoor, A-C. Huet, A. Lamote, M. Paulus, G. Janssens, N. Gillard and M. De Loose

Session D | Food Quality

1. **Assessment of physicochemical, antioxidant and antimicrobial characteristics of Algerian honeys**
A. Ouelhadj, R. Nakib, C. Seijo

2. **Estimation of flavonoid and chlorophyll changes in several micro-green species due to different LED light regimes with multiparametric fluorescence indexes**
C. Nugnes, G. Metelli, L. Nardi, E. Benvenuto

3. **Validation of HPLC method with pre-column derivitization for the determination of histamine in dried fish**
B.S. Ebarvia, A.C. Dacuya, R.R.L. Rondilla

4. **Comparison of ascorbic acid and nitrates content in fruit and vegetables from farmers´ market and supermarket**
M. Sabolová, T. Dupalová, L. Kouřimská

5. **The use of a biofixators for the removal of AFM1 from milk**
J. Bošnir, M. Ivešić, Ž. Pavlek, Ž. Kuharić, S. Serdar, S. Šikić, K. Markov, J. Frece, Ž. Jakopović

6. **Could the antioxidant profile be related to the presence of toxic elements in Olea Europea L. leaves and in drupes? A study on 11 Italian cultivars**
S. Mastrantonio, C. Nobili, C. Zoani, S. Procacci, D. Palumbo, A.M. Giusti, E. Pucci, L. Bacchetta

7. Sustainability in the food industry: managerial and technological actions to reduce the environmental footprint of Tinaia wine and Taleggio
A. Del Fiore, M. Valerio, C. Chiavetta, S. Cortesi, V. Fantin, C. Rinaldi, O. Presenti and N. Colonna

8. Evaluation of antifungal activity of essential oils against Penicillium spp. in in vitro and in vivo conditions by integrated approaches
A. Del Fiore, P. De Rossi, D. Palumbo, L. Quercia, C. Dalmastri, S. Sarrocco, G. Zappa and A. Bevivino

9. METROFOOD-CZ: state-of-the-art national research infrastructure in the fields of food quality & nutrition
L. Pacek, L. Kouřimská , P. Klouček , T. Míčka, M. Božík, K. Jíralov, J. Hajšlová and M. Urban

10. Researches on the impact of minimal processing on the antioxidant potential of cabbage varieties
M. Mulțescu, M. Zachia, N. Belc, T. Manasia, F. Burnichi and F. Israel-Roming

Session E | Food Omics

1. Development of a bioinformatics pipeline for routine analysis of whole genome sequencing data of Escherichia coli isolates
B. Bogaerts, S. Nouws, R. Winand, Q. Fu, J. Van Braekel, S. Denayer, B. Verhaegen, S. C. J. De Keersmaecker, N. Roosens, K. Marchal and K. Vanneste

2. Detection of food enzyme-producing microorganisms in food enzyme preparations
M. Deckers, K. Vanneste, R. Winand, S. C.J. De Keersmacker, S. Denayer, M. Heyndrickx, D. Deforce, M-A. Fraiture, N. H.C. Roosens

3. An integrated strategy combining DNA walking and NGS to detect GMOs
M-A. Fraiture, N. H.C. Roosens

4. The added value of WGS for foodborne outbreak investigation and surveillance of STEC and Staphylococcal enterotoxins
S. Nouws, B. Bogaerts, S. Denayer, B. Verhaegen, D. Piérard, N. Roosens, K. Vanneste, K. Marchal, and S. C. J. De Keersmaecker

Session F | Proficiency Testing & Reference Materials

1. Proficiency test round for the determination of deoxynivalenol, ochratoxin A and zearalenone in cereals
E.K. Tangni, Z. Han, B. Huybrechts, J. Mesquelier, E. Van Hoeck, W. Guo , Du. Ying, Z. Zhao

2. Certified reference materials and quality control materials …… Valuable tools for laboratory management
C. Maune, R. Malone, J. Rodgers and R. Niemeijer

3. The role of scientific research in increasing the performance of agro-food testing/analysis
F. Șerbancea, N. Belc, C. Uțoiu, V. Ionescu, A. Culețu, F. Manolache

4. Primary reference material for somatic cell counting in milk
S. Orlandini and H. van den Bijgaart

5. Establishment of the international joint research center of reference material for mycotoxins: contexts and fields of activities

E.K. Tangni, Z. Han, B. Huybrechts, J. Mesquelier, E. Van Hoeck, W. Guo, K. Jiang, J. Van Loco, Z. Zhao

6. Preparation of dimethylarsinic acid reference material

Q.Y. Sun, G. Bo, L. Feng-li, W. Yun, Q. Dai-jun, H. Qing-bo, Y. Zhong-yuan, Z. Jian-qiang

Session G | Food Contact Materials

1. Mineral oil migration from cardboard food contact materials: assessing the endocrine activity of mineral oils (PAHs) using the DRE-, ERE- and PPARγ CALUX bioassays

I. Boonen, A. Van Heyst, K. Van Langenhove, E. Van Hoeck, B. Mertens, M. Elskens, H. Demaegdt

2. Analysis of individual (MOAH) by APGC- QTOF-MS and comparison to the conventional method LC-GC-MS

J. Jaén, C. Domeño, P. Alfaro, C. Nerín

3. Release of trace elements from porcelain enamelware

H. Demaegdt, K. Cheyns

Session I | Nutrition

1. Level of minerals and inorganic contaminants in meat, fish and mixed dishes: an exploratory analysis developed in Portugal

M. Mendes, I. Coelho, I. Castanheira, I. Cabral, A.S. Matos

2. Influence of fat types on the fatty acids and trans fatty acids composition of biscuits

A.L. Mihai, M. Negoiță, G.A. Horneț and N. Belc

3. Soy protein hydrolysates in bakery products

A. Culetu, D.E. Duta, Z. Knežević-Jugović, J. Jovanović, N. Šekuljica, D.L. Comaniciu, L.V. Ordodi

4. Analysis of the amylose content of starch from different gluten-free flours

A. Culetu, D.E. Duta, M. Schimbator, I. Susman, G. Stamatie, N. Belc

5. Characterization of Jerusalem artichoke as ingredient in bakery products

L. Apostole, N. Belc, I. Susman, M. Schimbator

6. HPLC/DAD Method for Determination of Flavonoids Rutin and Quercetin in Herbal Supplements

T. Janeva, A. Najdenkoska, Z. Arsova-Sarafinovska

Session J | Nanomaterials

1. Optimization and validation of quantitative TEM analysis of pristine titanium dioxide powders in a regulatory context

F. Brassinne, E. Verleysen, M. Ledecq and J. Mast

2. Towards a routine method for the characterisation of TiO2 nanoparticles in food by single particle-ICP-MS

L. Givelet, P. Jitaru, D. Truffier-Boutry, J.-F. Damlencourt and T. Guérin

Session K | Accreditation

1. **Shortening the traceability chain for Africa: purity assignments of organic calibrators**
   M. Fernandes-Whaley, N. Nhlapo, L. Quinn, D. Prevoo-Franzsen

ABSTRACTS: PLENARY AND KEYNOTE PRESENTATIONS

PL1 - Foodomics: a milestone in food and nutritional studies

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Metabolomics is applied to a variety of biological fields from medical science to agriculture. Most of human beneficial properties of plants, be they foods, medical resources, or industrial raw materials, are ascribed to plant metabolites. One of the most important class of compounds are the polyphenols, due to compelling evidences of their beneficial health properties and to their impact on food quality. The complexity and remarkable diversity of polyphenols has challenged the analytical performances of the separation and detection methods in terms of resolving power, selectivity and sensitivity required for the identification and quantification of these compounds in different matrices. Targeted metabolomics is a strategy based on the use of predefined metabolite-specific signals, such as MRM transitions, that can be used to accurately and selectively determine the concentrations of a wide range of known metabolites. A targeted metabolomics method using UPLC/MS/MS system has been developed for the quantification >150 polyphenols and for the quantification of some polyphenols catabolites associated with the consumption of fruits. The validated method was found to be particularly flexible, since it can be easily expanded and adapted to the needs of different experiments. It was successfully applied to the analysis of various fruits and wine, as well as in nutritional studies, providing a valuable tool for the metabolite profiling of both the native compounds present in food and some nutritionally important bioactive catabolites in biofluids and organs.
For a mechanistic understanding of the action of polyphenols in living organisms it is fundamental in nutritional studies to include also untargeted metabolomics, a powerful tool to study processes in organisms and to detect new biologically important biomarkers. In our study, a un-targeted high resolution mass spectrometry-based investigation was chosen to monitor the metabolic effects induced by administration of different dietary polyphenols, at a physiologically relevant dose.

K01 - Determination of trace elements in food - recent developments from research, reference laboratory and standardization activities

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Confidence in the quality and safety of food is a high priority worldwide. The presence of undesired chemicals as well as the lack of essential chemical substances to fulfill the dietary requirement can potentially lead to serious consequences for human health. The trace elements have their own place in this context. When assessing the quality and/or safety of foods there is a demand for reliable experimental information, which in turn is based on the availability of fit-for-purpose analytical methodologies.

Trace element speciation analysis has been among the most important research topics within the field of trace element analysis over the last decades. Food samples are comprised of high variety of chemical compounds from which many can interact with metals and metalloids forming complex elemental species with various influence on the human body. In order to achieve the full picture it is important not only to determine the total amount of a certain trace element present in the food sample but also to identify the chemical form in which given element occurs in given sample (i.e. its speciation). Selected examples on trace element speciation will be presented.

The increasing World population has lead to an increased demand for food and research initiatives in exploitation of novel food ressources. In the western part of the world several projects have been initiated on exploration of increased use of e.g. insects and seaweed as ingredients in food production. The use of novel bioressources demands that these matrices are characterised for the their content of essential and harmful chemicals, incl trace elements. Examples from the determination of trace elements in seaweed and how data is used to evaluate food safety will be provided.

Recently, DTU FOOD were appointed as hosts of the European Reference Laboratory for metals and nitrogenous compounds in feed and food (EURL-MN). The EURL-MN collaborates closely with the network of NRLs (National Reference Laboratories) in the EU members states and organises proficiency tests, workshops and training for the NRL with the aim of harmonising and increasing the analytical competences of the laboratories involved in official food control of trace elements. An important player here is also the European Standardisation Committee (CEN) and the Working group 10 on Elements and their chemical species in Food, which develops standardised methods and procedures for analysis of trace elements in food. An update on the recent activities and future plans within the EURL-MN and CEN standardisation work will be presented.

K02 - Well established analytical methods for food quality and safety control: any risk of biased results?

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In line with increasing demands for food quality and safety, the need for reliable analytical methods for determination compliance with national regulations as well as international requirements is continuously growing. The reliability of a method should be determined by validation. Herewith we present two case studies document that special attention should be paid to trueness of generated data even in case of well established methods.

Case study no. 1 was concerned with analysis of spirit drinks. According to the Regulation (EU) No.1169/2011on the provision of food information to consumers, alcoholic beverages containing more than 1.2% (v/v) of ethanol must be labelled with alcoholic strength. To control this parameter we developed a rapid GC -MS method employing for ethanol sampling in the sample headspace SPME technique. When comparing the results obtained by this new approach with those obtained by pycnometry, a Comunity ´reference ´ method laid down in The Regulation (EC) No. 2870/2000, significant differences were found in case of some spirits such as fruit brandies, the determined alcohol content was higher when using the latter analytical approach. The cause of this discrepancy was co-distillation of other volatile sample components including fuel alcohols.

Case study no. 2 documents resolving the problem encountered within the external quality control, proficiency inter-laboratory testing focused on mycotoxins in maize. Routine ISO 17025 accredited method HPLC-MS/MS (contrary to other laboratories performing calibration by standard in a net solvent, we employed matrix-matched calibration for quantification. While a very good compliance of our results with other laboratories was achieved, the reported concentration of deoxynivalenol (DON) by our laboratory was fairly higher. The cause of the problem were unexpected interferences at the masses belonging to the 13C isotopically labelled deoxynivalenol fragment ions originating from the sample matrix. It should be noted, that the interfering matrix ions were present at the quantitative, as well as at both confirmatory ion transitions detected by the triple quadrupole unit resolution mass spectrometry.

Under these conditions, increased signal of isotopically labelled DON resulted in a significant overestimation of native DON content. The problem was satisfactorily elucidated by using high-resolution tandem mass spectrometry, HRMS (Q-orbitrap mass analyser), which enabled resolution of ions with very close m/z values belonging to 13C-DON and those of interferences. This practical troubleshooting example just underlined the necessity of confirmation of results by combination of different mass spectrometric approaches. In this particular case, the unique selectivity of HRMS technique helped to avoid the quantification error.

Keywords: spirits, ethanol content, maize, deoxynivalenol quantification, trueness of results.

K03 - EU policy on contaminants in food: recent developments and outlook

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The EU legislation on contaminants Council Regulation (EEC) No 315/93 of 8 February 1993 provides that food containing a contaminant in an amount which is unacceptable from the public health viewpoint shall not be placed on the market (food can only be placed on the market when it is safe). Furthermore, it is foreseen that contaminant levels shall be kept as low as can reasonably be achieved by following good practices at all stages of the production chain and in order to protect public health, maximum levels for specific contaminants shall be established where necessary. Also, the consultation of EFSA for all provisions which may have an effect upon public health is mandatory.

Following requests of the European Commission, the Panel on Contaminants in the Food Chain (CONTAM) from the European Food Safety Authority (EFSA) has completed in recent years several scientific opinions on contaminants, including on mycotoxins, processing contaminants, plant toxins and environmental contaminants, reviewing the possible risks for human health due to the presence of these substances in food.

The presentation shall provide up to date information and background on the recent and ongoing discussions on several contaminants such as

- the mycotoxins citrinin, ergot alkaloids, T-2 and HT-2 toxin, deoxynivalenol and modified forms, ochratoxin A, Alternaria toxins and enniatins
- the processing contaminants glycidyl esters, 3-MCPD esters, acrylamide, furan and alkylfurans.
- the plant toxins erucic acid, pyrrolizidine alkaloids, tropane alkaloids and opium alkaloids
- the environmental contaminants perchlorate, cadmium, dioxins and PCBs, perfluoroalkylated substances (PFAS) and brominated flame retardants (BFR)

PL2 - Recent technologies in the (bio)analysis of mycotoxins.

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Mycotoxins, toxic fungal secondary metabolites, are one of the main food safety threats worldwide. Co-occurrence of multiple mycotoxins in one crop as well as effects of climate change complicate this research field. The mycotoxin problem needs to be tackled in a multi-disciplinary way primarily focusing on prevention measures, but also mycotoxin analysis for monitoring and control purposes is definitely needed.

There are plenty of analytical techniques available for mycotoxin analyses in food, feed as well as in biological samples. Mycotoxin analytical methods include rapid screening as well as confirmation techniques. Sensitive *on-site* detection of mycotoxins in various matrices is highly needed. Therefore, both quantitative (ELISA, biosensors) and qualitative (lateral flow) systems for (multi)mycotoxin detection are being developed. Specific recognition elements as well as ultra-sensitive labels are under development in order to design more reliable rapid test-systems. HPLC and LC-MS/MS methods are mainly used as confirmatory methods and can be adopted to multi-mycotoxin analysis including modified mycotoxins. Moreover, high resolution mass spectrometry is gaining interest as it is an invaluable tool to discover unknown secondary fungal metabolites; to study degradation products and modified forms of mycotoxins as a result of processing techniques and use of detoxifying enzymes or microbes; and to unravel the human and animal mycotoxin metabolism.

Human mycotoxin exposure can be determined both indirectly (based on the combination of chemical analysis of foodstuffs and food consumption data) as well as directly by the determination of exposure biomarkers, mycotoxin biotransformation products, in biological fluids, such as urine or blood. In recent years many efforts have been put in the development of ultra-sensitive multi-mycotoxin LC-MS/MS for analysis of mycotoxin exposure biomarkers in urine and blood. However, still many gaps in our understanding of the human mycotoxin metabolism exist.

An overview of recent developments in the (bio)analysis will be given during this presentation. Also, the need for further international collaborations and how this can be practically achieved through the MYTOX-SOUTH network for training and education will be highlighted.

Keywords: rapid screening.

K04 - Nutrimetabolomics: integrative action for metabolomic analyses in human nutritional research as proposed by the Foodball consortium

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Life sciences are currently being transformed by an unprecedented wave of developments in molecular 'omics' analysis, which include important advances in instrumental analysis as well as chemometrics. In light of the central role played by metabolism in nutrition, metabolomics is rapidly being established as a key analytical tool in nutritional research. Consequently, an increasing number of researchers integrate metabolomics into their workflow. Within this dynamic landscape, the potential of nutritional metabolomics (nutrimetabolomics) to be translated into a science, which can impact on health policies, is gaining momentum.

A key element to reach this goal is the ability of the research community to join, to collectively make the best use of the potential offered by nutritional metabolomics. In this talk, a methodological basis for nutritional metabolomics will be offered that reflects on the state-of-the-art techniques used in the laboratories of the Food Biomarker Alliance (FoodBAll) (funded by the European Joint Programming Initiative a Healthy Diet for a Healthy Life (JPI HDHL)) as well as points of reflections to harmonize this field. This will be illustrated using practical examples from the Lab of Chemical Analysis.

Keywords: Mass spectrometry, GC, LC, metabolomics, biofluids.

K05 - Analysis of individual (MOAH) by APGC- QTOF-MS and comparison to the conventional method LC-GC-MS

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Mineral oil hydrocarbons (MOH) consist of a mixture of chemical isomers of saturated hydrocarbons (MOSH) and aromatic hydrocarbons (MOAH). Their analysis is very complex, as there are hundreds of isomers both linear, branched and with multiple aromatic rings, with or without sulphur. The contamination of food from food contact materials by MOH, mainly by MOAH, represents a risk to human health as MOAH are mutagenic agents and genotoxic carcinogens [1].

The contamination of food can come from different stages of food processing or packaging. Recycled materials, printing inks or some adhesives are common sources of MOAH.

Conventional methods to determine mineral oils involves the separation of MOSH and MOAH fractions either by silica column with AgNO3 (0.33%) and subsequent quantification by GC-FID or by LC-GC-FID or LC-GC-MS. A hump of unresolved peaks, which is quantified by GC-FID using an internal standard, gives the final result [2].

However, MOSH and MOAH contain a large number of compounds, which have not been identified and require a deeper evaluation before being quantified, to avoid overestimating or underestimating them. The analysis of mineral oil samples treated with solid phase extraction cartridges (SPE) followed by GC-MS and APGC–QTOF revealed the presence of different aromatic and polyaromatic compounds, such as alkylated benzenes, alkylated naphthalenes, benzothiophenes, compounds up to four aromatic rings, and dinaphthothiophenes.

GC-MS(EI) is commonly used for identification of unknown substances, since the large number of fragments generated by this technique and its high reproducibility allow identification of unknown substances through the comparison of mass spectra to those from the scientific libraries. APGC is a soft ionization that generates less fragmentation and allows obtaining the exact mass of molecular ion. APGC coupled to QTOF-MS allows the structural elucidation and identification of a large number of unknown substances.

This research explores these two techniques as complementary tools for identification and quantification of mineral oil components. Qualitative and quantitative analysis of individual MOAH present in several mineral oils and in a series of recycled materials were carried out by APGC-QTOF-MS and the results obtained are shown.

Topics: Food safety, minerals oils.

Keywords: Food safety, mineral oils, migration, analysis, APGC-QTOF-MS.

References:

[1] EFSA. Scientific opinion on mineral oil hydrocarbons in food. EFSA Journal. 2012, 10(6):2704

[2] S. Bratinova, Guidance on sampling , analysis and data reporting for the monitoring of mineral oil hydrocarbons in food and food contact materials, 2019.

K06 - Alternaria toxins: Analysis and risk assessment of emerging and modified mycotoxins

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According to a WHO estimation about 25% of agricultural commodities are contaminated with mycotoxins world-wide. During the last years so called “modified” [1] or “emerging” mycotoxins have been discovered, which are either plant metabolites of the fungal toxins or produced by other ubiquitous fungi such as Alternaria species, respectively.

Targeted approaches have been developed to accurately quantitate „emerging“ and „modified“ mycotoxins along with multi-analyte approaches based on stable isotope dilution assays (SIDAs) [2] for efficient mycotoxin control.

In this regard, major Alternaria toxins were analyzed and risk assessments along with management actions for infant foods were initiated. Moreover, non-targeted approaches were initiated to screen for other fungal metabolites.

Keywords: modified mycotoxins, Alternaria toxins, stable isotope dilution assay, metabolomicsRYCHLIK

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K07 - Perspective in nanoparticle analysis in food with single particle ICPMS

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Nanoparticles can be found in a variety of food products due to additives used for various properties such as colouring or texturing or due to the food production process. Quite often the nanoparticle content of these additives are known neither from the additive manufacturer nor from the food producer. The EU obligation of labeling food than contain nanomaterials in the list of ingredient has led to a large amount of studies to determine the best suitable methodology to sort engineered particles between non-nano and nanomaterial. Among the techniques available for nanoparticle counting and sizing, sp-ICPMS has the advantage of being element-specific, and adaptable to complex matrices. Metrological challenges however remain to be solved in order to achieve robust and reproducible measurements in all laboratories. An overview of the analytical and metrological difficulties posed by the analysis by sp-ICPMS will be presented. Then, some examples of nanoparticle characterization in food product performed at LNE will be discussed.

K08 - Quantification of (engineered) nanoparticles in complex matrices: more than a silver lining on the horizon?

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Engineered nanoparticles (ENP) are generally defined by their size - referring to any kind of equivalent diameter - and include particles with diameters between 1 and 100 nm. Nanotechnology is the art and science of manipulating and engineering objects at the 1 - 100 nm scale and opens up a tremendous field of new applications beneficial to mankind and the environment. Amongst others, the food and cosmetics industries have started using ENP to increase the quality of their products. However, despite the beneficial use of ENP in many products, their interaction with biological systems and ultimately their fate in the environment is still poorly understood. This triggered the development of novel or refurbished analytical techniques, rapidly appearing on the market, to detect and quantify emerging pollutants of this new class of materials. However, after more than a decade of intensive research and development with to some extent disillusioning results, it is obvious that the analytical Swiss Army knife has not been invented, yet. The quantitative measurement of ENP in complex matrices, therefore, remains very challenging and to some extent, wishful thinking. However, recent advances in the field of single particle inductively coupled plasma mass spectrometry, especially when combined with a time of flight system, and electron microscopy represent a huge step towards a reliable and robust quantification of ENPs, also in complex matrices. With its enormous resolving power well beyond the nm scale, transmission electron microscopy (TEM) is the only technique that covers the whole nano-size range and delivers true number based particle size distributions. In addition to size, auxiliary techniques, such as energy dispersive X-ray analyses and selected area electron diffraction allow collecting information of the elemental composition and the structure on an individual particle level. All this wealth of information comes at the prize of manpower, apparently making TEM too expensive to be competitive with other well-established but less powerful techniques. However, the combination of automated image analyses algorithms with automated TEM operations opens new possibilities for the detailed characterization of ENP at considerably reduced manpower and, thus, costs. To benefit from the immense potential of automated TEM, a (well defined) minimal image quality is required, which directly transfers into the development and tailoring of sample preparation methods. With the basic TEM automation being realized and elaborate image processing software being available, the next challenge will be on optimizing and standardizing sample preparation protocols resulting in samples suitable for automated TEM analyses. The bottleneck for TEM will therefore likely shift from the actual time spent on the microscope to the time required for suitable sample preparation.

Keywords: Electron microscopy, nanoparticles, sample preparation, automation.

ABSTRACTS: ORAL COMMUNICATIONS

OC1 - Nickel in feed and food – Results of proficiency tests by the EURL-MN

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The European Food Safety Authority (EFSA) published their Scientific Opinion on Nickel in feed in 2015. EFSA concluded that any adverse effect after dietary exposure to nickel (Ni) in farmed animals is unlikely, and that chronic dietary exposure to Ni considering only foods of animal origin may be of concern in the young population. For some food groups, occurrence data were limited, and for some sample types, e.g. feed, data originated mainly from one country. Following the European Commission recommends the monitoring of Ni in feed (Commission recommendation (EU) 2016/1110) and food (Commission regulation (EU) 2016/1111).

The European Union reference laboratory for metals and nitrogenous compound in feed and food (EURL-MN) included nickel as an analyte in their proficiency tests (PTs) in 2018 in order to evaluate the capability of national reference laboratories (NRLs) to analyse Ni.

The PTs materials were a mixed corm poultry feed and a chili powder, and the scope was to assess the performance of the NRLs in determining the mass fraction of cadmium, lead, mercury, arsenic and Ni in feed and food. The PTs were conducted according to ISO 13528:2015. The assigned values are consensus values based on the results of the participants, and the performance of the NRLs were evaluated using z and ζ scores.

Overall, the performance of the NRLs were very satisfactory. For Ni, the performance was good. The performance of the participants was satisfactory, but not a good as for the other elements, due to higher number of questionable and unacceptable results. The poorer performance may be due to lack of a CEN standard method for the analysis of Ni in feed, and that the laboratories analyse this parameter less frequently than the other analytes.

The results of the PTs will be discussed in detail, with focus on the analysis of Ni.

Keywords: Analysis, elements, EURL-MN, metals, performance, z score.

OC2 - Occurrence of perfluoroalkylated substances (PFAS) in drinking water in the Czech Republic

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Per- and polyfluoroalkylated substances (PFASs) as a group of potentially harmful environmental pollutants are currently in the center of interest of health and safety authorities. In 2018, the European Food and Safety Authority (EFSA) issued new values for a tolerable weekly intake (TWI) for perfluorooctane sulfonate (PFOS) – 13 ng/kg bw per week and perfluorooctanoic acid (PFOA) – 6 ng/kg bw per week (1). Based on the EFSA opinion, together with fish and seafood products a drinking water were classified as one of the most important contributor to the mean chronic exposure to some PFASs. The main aim of our study is to validate of a sample preparation procedure for the determination of PFASs in water based on solid phase extraction (SPE) followed by UHPLC-MS/MS analysis. Then to use this method for the investigation of a large set of tap water samples originated from various localities in the Czech Republic.

For the analysis of 24 PFASs, the extraction method based on SPE using sorbent WAX was used. The target analysis was performed using a 1290 Infinity II LC system coupled to a Triple Quadrupole G6495 (Agilent Technologies, USA) with the negative electrospray ionization.

The method was successfully validated; the recoveries were in the range of 61-119% with the repeatability expressed as the relative standard deviation lower than 24%. The limits of quantification were in the range of 0.05-0.25 ng/L. Using this method 55 samples of tap water collected during 2018 and 2019 were investigated. The most abundant contaminants were PFOS (both linear and branched isomers) and PFOA in the concentration range of 0.1-16 ng/L. Nevertheless, other representatives from both groups of perfluorosulfonates (PFBS, PFHxS, and PFHpS) and perfluorocarboxylic acids (PFHxA, PFNA, and PFDA) were detected, often at comparable or even higher concentration to PFOS and PFOA. The amount of PFASs in tap water samples collected in various places (e.g. households) but originated from the same water supply was almost identical. Regarding the comparison of results in water from various supply area (e.g. town, countryside, mountains, wells) the occurrence of PFASs differs significantly. Regarding the estimations of daily intake for the average consumer (70 kg body weight, 2 liters) the TWI can be filled 10-20% by the consumption of the tap waters with the highest PFOA and PFOA concentrations.

Keywords: PFOS, PFOA, LC-MS, drinking water, TWI.

OC3 - Is there too much lead in Belgian big game meat?

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Lead (Pb) is a widely occurring, hazardous contaminant. Human exposure to Pb is associated with a wide range of effects, including neurotoxicity, nephrotoxicity and increased systolic blood pressure (Nawrot & Staessen, 2006). Although Pb has been banned from several applications (e.g. gasoline, plumbing,…), it is still widely used in hunting ammunition. Several studies have shown that hunting big game with Pb-containing bullets can result in increased Pb levels in meat for consumption (Knott et al; 2010; Lindboe et al., 2012). The presence of small bullet fragments in venison was demonstrated by radiographic studies even at large distance of obviously injured tissue (Hunt et al., 2009).

The present study aims to collect data of Pb in edible meat of 250 different animals hunted in Belgium (work package WP1), comprising roe deer, wild boar and red deer, and to evaluate human dietary exposure and risk. Work package 2 focuses on potential differences in Pb concentration in meat as a function of the distance to the wound channel. Information about bullet type, age class, sex and geographic origin of all samples, is collected based on the official ID numbers complemented with questionnaire data, and will be included during data processing. Samples for WP1 are bought from hunters, game processing centres or supermarkets, while for WP2 entire animals are bought from the hunters.

Because Pb fragments may continue to exist in traditionally blended meat samples, leading to heterogeneous -and hence non representative- sample results, an alternative sample treatment and homogenisation step is applied as described by Lindboe et al. (2012). According to this method each meat sample (±200 g in WP1; ±100 g in WP2) is -after the first grinding step- blended with nitric acid (15% v/v) and shaken ±20h at room temperature. This allows potential Pb fragments to dissolve (Kollander et al., 2014). The obtained slurry is further digested in a microwave and analysed by ICP-MS.

The first results of WP1 (currently ±65 samples of roe deer and wild boar) indicate that maximum Pb concentrations were similar in both species (min: <0.003 mg kg-1; max: ±0.800 mg kg-1), while P50 and P90 concentrations were higher in wild boar (P50 = 0.029 µg kg-1; P90 = 0.190 µg kg-1) compared to roe deer (P50 = 0.006 mg kg-1; P90 = 0.134 mg kg-1). In both species respectively 12% and 13% of the samples exceeded the European maximum limit of 0.1 mg kg-1 Pb for meat of various farm animals (EU 1881/2006). None of the samples exceeded the Belgian action limit of 1 mg kg-1 for game meat as applied by the Federal Agency for the Safety of the Food Chain. In WP2, roe deer samples taken close to the wound channel (5-20 cm) showed higher P50 and P90 Pb concentrations compared to samples taken at larger distance (40-70 cm), while this trend was not observed in wild boar. Data treatment will be finalized in the coming months and the results obtained will be discussed in relation to literature data.

Keywords: Lead, game meat, risk, hunting, bullets.

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OC4 - The effect of temperature on the nutritional quality of edible mealworm Tenebrio molitor

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Recently, the insects are becoming more and more popular as food. They are a good source of protein, lipids, essential amino acids and fatty acids, and many beneficial substances such as minerals and vitamins. Since January 2018, insects have been recognized as novel foods in the EU. Their nutritional value is known to be not constant and factors affecting their composition are debated. As rearing conditions belong among such factors, the effect of temperature on the nutritional value of mealworm (Tenebrio molitor L.) was studied.

Mealworms were reared in the incubator at the temperatures 22 and 25°C. The amount of dry matter and ash were determined gravimetrically, lipids by the Soxhlet extraction, and crude proteins by the Kjeldahl method. Moreover, their life cycle duration, length and weight were monitored as well.
Weight and length of mealworms reared at 25°C were significantly higher than in another group. Concerning nutritional parameters, contents of proteins, lipids and ash were found to be influenced by rearing temperature as well. While the levels of ash and crude protein were higher in worms reared at 22°C, the amount of lipids was higher at 25°C. On the contrary, the dry matter content was proved to be not depending on the temperature.

The results of this work will contribute to research on seeking for the optimization of mealworm rearing technology in order to achieve the desired nutritional values.

Keywords: Edible insects, nutritional value, rearing, temperature.

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OC5 - Direct elemental analysis of cereal and rice flour using total reflection X-Ray fluorescence: new challenges

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Food is the main source of essential, major, and trace elements for human and animal nutrition. The toxic metal content of foods is influenced by many factors ranging from environmental conditions during growth to post-harvest handling, processing etc. Of particular concern is the presence of toxic or potentially toxic elements such as arsenic, cadmium, lead, and mercury. These metals can disturb human metabolomics, contributing to morbidity and even mortality. Development of more robust, efficient, sensitive, and cost-effective analytical methodologies that guarantee the safety, quality, and traceability of foods in compliance with legislation and consumers' demands is the main issue for food analysis. Elemental chemical characterization of foods results complicated due to samples' complexity. Foodstuff analysis is generally performed by Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-AES) and Inductively Coupled Plasma Mass Spectrometry (ICP-MS) techniques, which require complete sample mineralization by acid digestion [1].

Although in Total reflection X-Ray Fluorescence (TXRF) spectrometry, wet acid digestion stands as the most used sample preparation method, other approaches are continuously studied for reliable elemental chemical analysis of solid samples [2]. Development of new reliable strategies for a faster and easier determination of potentially toxic elements – to be eventually confirmed or refined with more sensitive methods - could represent a promising improvement for foodstuff characterization enhancing food safety and consumer protection.

In this contribution, we propose a simple analytical methodology that consisted in suspending the powdered samples in MilliQ water for direct analysis. For the first time, the novel SMART STORETM method, explored previously for direct analysis of tree leaves [3] and elemental analysis of particulate matter collected on filters [4], is tested for foodstuff. The sample was deposited on acetate cellulose filters, which were sandwiched between two thin adhesive sheets of polypropylene and cut in a circle of 30 mm diameter. The obtained disc was placed onto a quartz reflector carrier for measurements. From an analytical point of view, results revealed that the developed TXRF methods are less sensitive and precise than sample mineralization. Notwithstanding this, preliminary data highlight the potentialities of these fast, simple and suitable strategies for screening purposes.

Keywords: cereal, rice flour, TXRF, suspension, SMART STORE.

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OC6 - Development of the analytical method for 87Sr/86Sr determination in olive oil

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Olive tree (Olea europaea L.) is considered as one of the most important crops in the Mediterranean basin. Namely, its fruit is used in the production of olive oil, which is a substantial part of the traditional diet. Virgin olive oil is a very appreciated natural food product due to its beneficial effects to human health known for centuries. They depend on oil composition, which varies widely depending on the fruit variety, degree of fruit ripeness, environmental conditions, production and storage process, as well as growing region i.e. its origin.

EU Member States agreed to obligatory origin labelling of extra virgin olive oils to prevent consumers being tricked about their true characteristics and origin (EC 182/2009). However, high commercial benefits in olive oil trade often lead to frauds and misleading statements about its origin. Fraud detection is a great challenge from an analytical point of view; it implies searching for region-specific indicators such as isotopic, chemical or element composition patterns. Up to now, many analytical tools were developed with the purpose of discrimination of the olive oils' geographic origin, such as analysis of volatile compounds, fatty acids and triacylglycerol composition, trace elements and stable isotopes of light elements. In addition, Sr isotope composition was also demonstrated as a reliable tool in the investigation of geographical origin of several food products, but has been only in one study applied to olive oil due to the lack of the adequate analytical method. The determination of elements in olive oils is a big analytical challenge due to the complex matrix with high viscosity and high organic content as well as very low concentrations of trace elements, including Sr.

In this work, a sensitive and accurate analytical method for the determination of 87Sr/86Sr isotope ratio in olive oil by multicollector inductively coupled plasma mass spectrometer (MC-ICP-MS) was set up. The method development consisted of: i) optimization of Sr extraction from the oil matrices and ii) optimization of the procedure for organic matter removal prior to Rb extraction on Sr-resin. Namely, the organic matter could hamper the Sr-resin extraction efficiency and influence the measurement of 87Sr/86Sr isotope ratio. Various extraction parameters such as different volume of extractant (water, 8 M HNO3 and 2% HNO3 + 0.1% HCl) and sample were tested, as well as the performance of three different extraction procedure (ultrasonic, microwave and mechanical stirring). For the organic matter removal, the efficiencies of microwave and hot plate assisted digestion with HNO3 and H2O2, and dry ashing were compared. Sr and Rb concentrations were measured to check for Sr mass balance and efficiency of Rb removal. The Sr isotope composition was determined by MC-ICP-MS. The developed method was successfully applied for the determination of 87Sr/86Sr isotope ratio in olive oil samples.

OC7 - Nutrients, secondary metabolites and anti-oxidant activity Moringa oleifera leaves and Moringa-based commercial products

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Moringa oleifera (family Moringaceae) is a fast-growing tree that is widely distributed in tropical and subtropical regions of the world. The plant has been reported widely for its nutritional, medicinal and industrial value. Malnutrition is endemic to most developing and underdeveloped countries and Moringa holds potential to alleviating the problem of under nourishment in South Africa. South African provinces engaged in production of Moringa (viz. Limpopo, KwaZulu-Natal and Mpumalanga are those that are also highly affected by poverty and malnutrition. Climatic conditions and farming practices vary significantly and may have an effect on the profile of nutrients, secondary metabolites of the leaves and possibly the antioxidant capacity of the leaf extracts of M. oleifera. The nutrient content, antioxidant activity and phytochemistry of M. oleifera leaves from different regions of South Africa (Limpopo, Mpumalanga and Gauteng) including Moringa-based commercial products (loose leaves, tablets, tea, chocolate, juice and porridge) were investigated. The leaves of M. oleifera from Limpopo had the highest nutrient content and highest antioxidant capacity (IC50 = 19.1 ± 0.26 µg/ml). Fourier Transform Infrared Spectroscopy was used for rapid profiling of the chemical composition of M. oleifera leaves and for authenticating Moringa-based commercial products. The spectra for all samples exhibited a similar profile, with strong absorbance bands associated with the presence of phenolic compounds appearing at 3287 cm-1, 2920 cm-1, 2850 cm-1, 1611 cm-1 and 1051 cm-1. Two marker bands at 2920 cm-1 and 2850 cm-1 representing the stretching vibration of the aromatic group (C-H) were observed in all Moringa-based commercial products except in Moringa juice. The absence of these marker bands could mean that some phytochemicals are destroyed during the processing of Moringa juice. Phytochemical investigation of the leaves extracts of M. oleifera was carried out through the sequential extraction method and analysed using ultra-performance liquid chromatography time-of-flight mass spectroscopy.

The results revealed 20 compounds, belonging to; flavonoids, steriods and phenolic acid derivatives being the most abundant from the leaves of M. oleifera. This study concluded that leaves of M. oleifera collected from different geographical regions of South Africa contain commercially viable secondary metabolites, which can be exploited for opportunities in the manufacture of drugs and nutraceuticals.

Keywords: Moringa oleifera, phenolics, flavonoids, antioxidant, DPPH, FTIR.

OC8 - Enhanced capability of a purge-and-trap, thermal desorption and GC×GC-MS methodology for aroma profiling

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Headspace gas chromatography has been frequently used for aroma profiling because of its ability to naturally exploit the volatility of aroma compounds, and also to provide information on the composition of the sample [1]. Its main practical advantages are its simplicity, no use of solvent, amenability to automation, and the cleanliness of the extract.

In the present contribution, the most effective sampling (dynamic sampling), separation (multidimensional gas chromatography) and detection (mass spectrometry) techniques are combined, showing their potential in unravelling aroma profiles in beverages.

In addition, a neat workflow for data analysis is discussed and used for the successful characterization and identification of different beer flavors, if the steps in the analytical process are properly controlled. From the technological viewpoint, this is the first time that a purge-and-trap (P&T), comprehensive 2D gas chromatography (GC×GC), and mass spectrometry (MS) are exploited in combination. A newly-thought flow modulation approach allowed for multidimensional 2D gas chromatography, with the full eluate transfer onto the second dimension and the MS detector with no need to divert the flow, making the overall method highly sensitive and selective [2-3].

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OC9 - Implementing sodium reduction in food regulations in South Africa – the analytical measurement challenges

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In March 2013, the South African National Department of Health published regulation R214 under the Foodstuffs, Cosmetics and Disinfectant Act 54 of 1972. This regulation is aimed at reducing the sodium content in selected food products, to minimize the increasing number of non-communicable lifestyle diseases such as high blood pressure, heart disease and stroke in the South African population. South Africa is currently the only country in the world to have implemented mandatory sodium reduction regulations in several food categories, in addition to bread.

Food manufacturers were allowed three years in which to comply with the new regulation which became effective on 30 June 2016, with a further reduction in sodium levels that will become effective in June 2019. Since implementation, some disputes have arisen from discrepancies between analytical testing laboratories' results for sodium content in foodstuffs. NMISA was requested to assist with a customized proficiency testing scheme (PTS) that targets the various food categories to provide an independent assessment of measurements performed between the private and public-sector institutions. The foodstuff categories included: stock powder, bread, noodles, fat spread, savoury snacks and canned meat. NMISA has homogenized and packaged these materials, together with commercial food producers, and has provided the traceable reference value by ICP-OES or ICP-MS for sodium in each round of the PTS. The regulated tolerance of 20% on the sodium result was used as the standard deviation of proficiency assessment. An overview of the challenges encountered with each completed round of the PTS, initiated in September 2018, will be highlighted with suggested improvements.

Evaluation of PTS performance has allowed the regulator and participants to identify the possible sources of measurement result discrepancy, which will support improvements in the implementation of national health promotion strategies.

Keywords: Mandatory sodium reduction, proficiency testing, analytical challenges.

OC10 - Flaws and hurdles concerning the harmonisation of detecting allergens in food

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Food allergies form a growing health problem worldwide. With awareness and incidence rising, the need for accurate food allergen detection methods is obvious. Although a lot of research in this field is being done, some important hurdles still remain, especially in terms of worldwide harmonization. This harmonization is required for the development and the validation of detection methods, but is also crucial in terms of legislation.

Various techniques exist for detecting food allergens. Depending on the purpose of the detection, one technique can be more suitable than another. DNA-based techniques (PCR, qPCR and ddPCR) are rapid and offer the possibility of multiplexing, but the outcome is less informative for patients suffering from food allergies since DNA is not the elicitor of the allergic reaction. In the case of the protein-based detection technique ELISA, a lack of knowledge on the antibody used in the kit can result in misinterpretation. The exact epitope sequence recognized by the antibody in the kit is often not known, as well as if the kit uses poly- or monoclonal antibodies. Such information is however crucial to evaluate test results since food processing can influence the recognized epitope.

In case of mass spectrometry-based detection techniques, where the analyte is a peptide with known sequence unique and specific to the ingredient, there is still work to do with respect to harmonization. Some aspects such as the criteria to determine whether a peptide is detected in the LC-MS/MS method and how to quantify this peptide, should be discussed on an international level, in function of a consensus.

Regardless of the technique applied, there is a need of international reference materials and internal standards for the uniformity of testing and the interpretation of test results. Different techniques apply different calibration methods, and even within the same technique, these calibration methods can vary. Reference materials and standards are thus necessary to obtain accurately assigned values and correctly interpretated results.

At last, there should be a general consensus on how to report results. There is general agreement that results are reported in mg total protein per kg food material. However, when using peptides or DNA as analytes, correct conversion factors should be established and applied to be able to report values in the suggested unit.

Keywords: food allergens, detection, harmonization, reporting unit, reference materials.

OC11 - Quantification of SO2 in wines by Surface Enhanced Raman Spectroscopy and the comparison with the official OIV method

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Sulfur dioxide (SO2), is commonly added as a preserving and antioxidant agent during the entire winemaking process. The content of sulfur dioxide in wine is limited by specific laws in force in every country. The EU has set a legal limit for total SO2 of 150 mg/L in red wines and 200 mg/L in white wines. Moreover, SO2 and sulphites are potential allergens, consequently, rigid analytic controls across the entire winemaking process are imposed. Total SO2 is defined as the sum of all its forms, either in the free state or combined with other molecules in wine. The official method by the International Organisation of Vine and Wine, OIV-ma-as323-04a, for SO2 quantification is based on the distillation and ri-dissolution of SO2 in the form of sulfuric acid. Then, this latter is determined by titration with a sodium hydroxide solution. However, this method is time-consuming and subject to many possible sources of error. For these reasons, it is important to have alternative fast and reliable methods for the quantification of SO2. In this study, Surface Enhanced Raman Scattering (SERS) in a liquid suspension of silver nanoparticles (AgNPs) is presented as a possible rapid method for total SO2 quantification in white and red wine. AgNPs are used as a plasmonic substrate to enhance the intensity of Raman signals, increasing the sensitivity of the technique. Thanks to the chemical affinity between S and Ag, SO2 is bounded to the active surface of AgNPs providing intense and specific SERS signals around 920 cm-1 and 620 cm-1, associated with S=O stretching and deformation, respectively. The intensity of those signals can be correlated with the amount of SO2 in wine. Calibration curves were tested in the concentration range 0-100 mg/L using simil-wine solutions of 12 % EtOH/H2O. However, the matrix effect, strongly dependent on the type of wine, was observed when real wine samples were analyzed. For this reason, the standard addition method was then used to determine the amount of SO2 in wines. In this case, the only pretreatment of the samples consisted of a rapid solid phase extraction of main interfering molecules with C18 cartridges. Moreover, the measurement uncertainty of the method was evaluated. The results obtained by SERS method were compared with the ones achieved by the official OIV method and the consistency both for red and white wines was confirmed.

Keywords: Sulfur dioxide, Surface Enhanced Raman Scattering, official OIV method, result comparison, wine.

OC12 - Development of a DNA barcoding-like approach to detect mustard allergens in wheat flours

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The spread of food allergens is a topic of global importance due to its impact on public health. National and International regulations ask food producers and manufacturers to declare product compositions on the label, especially in case of processed raw materials. Wheat flour (Triticum aestivum) can be contaminated by a wide range of species in the field or during grain harvests, storage, and processing. Among them, could be present different Brassicaceae species such as rapeseeds (Brassica napus), non-allergenic species, or mustards (Brassica nigra, Brassica juncea and Sinapis alba) which are well known as allergenic species. In the case of mustard species, the 2S albumin is a high heat-resistance protein, and it could remain in its intact form during high temperature industrial processes. Often, food quality laboratories adopt an ELISA approach to detect the presence of mustard species in raw materials or finished products; however, this approach shows cross-reactivity with other non-allergenic species such as rapeseed. This study aims to set up an easy and rapid DNA-based tool to detect only mustard, allergenic species, throughout DNA barcoding method. In the last few years, this approach was proposed as a valid identification method, and it is now commonly used in the authentication of food products. DNA barcoding (matK and ITS2) and chromosome markers (A6, B, C1 genome regions) were selected, and specific primers were validated on incurred reference food matrices. The developed test was proven to be able to distinguish mustard from rapeseed in common wheat matrixes, overcoming cross-reactivity with Brassica napus.

Keywords: allergen detection, DNA barcoding, Brassicaceae, Brassica napus, processed food, Triticum aestivum.

OC13 - Multi-approach determination of dithiocarbamate fungicides and of their degradation products in fruits and vegetables

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Dithiocarbamates (DTC) have been extensively used as fungicides for more than fifty years due to their cost-effectiveness and their antifungal properties against a large panel of plant diseases. In order to broaden their activity spectrum and further enhance plants resistance to various pathogens, DTCs are often used in combination with modern systemic fungicides leading consequently to elevate levels already reported for food samples collected from the European Union. Despite the significant environmental and food chain impact of DTCs, the current analytical approaches for their determination suffer from serious drawbacks. The European reference method for this purpose relies on non-selective quantification by indirect determination: after acidic hydrolysis, the sum DTCs is measured via the generated carbon disulfide by gas chromatography.

A multi-approach strategy was applied throughout this study in order to increase knowledge on the DTCs determination from various fruits and vegetable matrices from the following perspectives:
(i) determination of DTCs per class, depending on their chemical structure, as well as of their degradation products, like ethylene- and propylene-thiourea. The methods are based on hydrophilic interaction liquid chromatography (HILIC) or reverse phase (RP) HPLC hyphenated to either molecular or elemental mass spectrometry (MS) techniques through the detection of organic DTCs, metals and/or sulfur moieties;
(ii) selective and simultaneous determination of the three individually EU regulated DTCs (thiram, ziram and propineb) as well as of the DTCs degradation products by the use of HILIC-MS/MS.

The proposed analytical methodologies were in-house validated for the determination of the target analytes from several fruit and vegetable matrices over a wide concentration range. The results of the present study show that the analysis of DTCs by class as well as of their degradation products from fruits and vegetables can be achieved by a multi-approach methodology especially when employing soft surface extraction techniques and further separate and detect by the use of HILIC or RP-HPLC coupled to electrospray-MS/MS or to ICP-QQQ-MS.

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Keywords: dithiocarbamate fungicides, DTCs, ETU, PTU, HILIC-MS/MS, HPLC-ICP-QQQ-MS.

OC14 - New developments in integrated 'sample to result' workflows for the multi-residue analysis of polar anionic pesticides and their metabolites

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Laboratories are constantly challenged to analyze more classes of pesticides at lower concentrations in more different commodities, with faster turnaround times and little if any increase in costs. And with the expectation that residues will not go undetected and all results can be verified by associated analytical quality control data compliant with method performance guideline criteria.

One of the most challenging group of pesticides is the polar anionic pesticides, such as glyphosate, glufosinate, ethephon, perchlorate, chlorate and the like, which often occur as residues in food, but are not always included in pesticide monitoring programs. This presentation will highlight the development and validation of an IC-MS/MS based workflow for the robust, sensitive and reliable determination of polar anionic pesticides and metabolites at low µg/kg levels, all in a single chromatographic run. The integrated workflow from sample to results is based on the Thermo Scientific™ Dionex™ Integrion™ HPIC™ system, TSQ Altis™ Triple Quadrupole Mass Spectrometer System and all associated workflow components: IC column & suppressor, suitability check standard solutions, software system and comprehensive user guidelines for fast implementation, enablement of ongoing optimum performance. The workflow uses a modified QuPPe extraction with cartridge solid phase extraction clean-up and has been thoroughly tested and validated in two different laboratories. Results for wheat, leek and Baby food matrices are compliant with SANTE guidelines and EU MRLs. Quantification limits are 10ug/kg with % RSDs typically <10 %. Recoveries with and without internal standards, using using matrix-matched calibration, and matrix extracted calibrations (procedural standards) will be presented.

Keywords: Polar Pesticides, Glyphosate, IC-MS/MS, Ion Chromatography, QuPPE.

OC15 - Monitoring of hygiene in institutional kitchens in Belgium

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Introduction

Good hygiene in institutional kitchens is essential for prevention of foodborne illness. Respecting the basic rules of hygiene is the basis to guarantee safety in this sector where many steps are manual and different equipment is used.

Purpose

In this study, critical food contact surfaces and hand contact surfaces in institutional kitchens were monitored microbiologically .

Methods

From 40 Belgian institutional kitchens (10 schools, 10 day cares, 10 retirement homes and 10 hospitals), 598 surfaces were sampled. The surfaces contained hands, cutting boards, cutting knives, ladles, trays, workbenches and extras (e.g. whisks, strainers, cutting machines or blenders). Microbiology of the same surfaces were compared before and after cleaning. Additionally, sinks and aprons were sampled. Samples were taken by swabbing with a sterile moistened cotton pad and repeating the swabbing with a sterile dry cotton pad. Following microbiological enumerations were performed: total aerobic count (TAC), Enterobacteriaceae, Bacillus cereus, Staphylococcus aureus and Escherichia coli.

Results

The highest average TAC was found for sinks, used cutting boards and used cutting knives; 4.0±1.5, 4.0±1.6 and 3.9±1.7 log CFU/20 cm², respectively, with cutting boards and cutting knives also having the highest log CFU/20 cm² reductions after cleaning, 1.3 and 1.2, respectively. The highest average TAC of cleaned surface types was found for worktables with 3.1±1.6 log CFU/20 cm² with only achieving an average reduction of 0.3 log by cleaning. Cleaned cutting boards and worktables contained enumerations of Enterobacteriaceae in 33% and 45% of the samples, respectively, and of B. cereus in 23% and 25% of the samples, respectively. Washing of hands led to a slight reduction in TAC of 0.5 log CFU/20 cm².

Significance

This study shows the importance of good cleaning and disinfection of kitchen surfaces and utensils, using clean rags and towels, and frequently washing hands when preparing food to reduce cross-contamination.

Keywords: microbiological food safety, kitchen hygiene, microbiological food quality.

OC16 - Detection and quantification of biogenic amines in Cambodian smoked freshwater fish

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Freshwater fish, highly perishable food, has been traditionally processed and smoked to give special smoke flavor and crispiness to fish flesh and also preserve it for longer shelf-life. The quality of fish can be downgraded due to improper storage conditions and manufacturing practices following the fishing and prior to smoking production process. Biogenic amines could be produced consequently in fish and fish products by bacteria through decarboxylation of free amino acids. These could affect the quality of finished smoke fish products. Particularly, histamine and tyramine have been considered as the most toxic among the biogenic amines causing allergic reactions including difficulty in breathing, itching, rash, vomiting, fever and hypertension to consumers.

Therefore, a preliminary study was carried out to assess the quality of smoked freshwater fish in terms of contaminated biogenic amines in Phnom Penh local markets. Out of nine markets, Oreussey market was a main target for diversification of smoked fish as it possessed the highest number of stores (24) commercializing these products as retailers and wholesalers. Six types of smoked fish including Trey Andoeng (Clarias spp.), Trey Kaes (Micronema spp.), Trey Riel (Cirrhinus spp.), Trey Slek Reussey (Paralaubuca typus), Trey Ta Oan (Ompok bimaculatus) and Trey Changvachnot (Osteochilus spp.) were commonly available in the market and collected for further investigation. The samples were subjected to analysis by Ultra High Performance Liquid Chromatography coupled to fluorescence detection.

The biogenic amines analysis revealed that cadaverine was detected in all samples but one at the highest concentration, with values ranging from <1 to 172 mg/kg. Putrescine, spermine, spermidine serotonine, methylamine and tryptamine were also detected in nearly all samples at lower concentrations. Concerning histamine, tyramine and 2-phenylethylamine, they were present in 3 samples. For 7 biogenic amines out of 10 analysed, Ompok bimaculatus was the fish species containing the highest concentration.

The biogenic amines index was calculated for each sample. In this study, one sample can be considered as fresh with a value lower than 5 mg/kg, four samples were considered as acceptable (5-20 mg/kg), four samples were considered as low quality (20-50 mg/kg) and five samples were considered as spoiled (>50 mg/kg).

OC17 - Proposal for a European Metrology Network on food safety - EMN-FS

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Safe and high-quality food is a fundamental prerequisite for human health. Food production has evolved considerably during the past decades in many aspects. The objective of the EU food policy is to preserve the safety of food and to verify alteration and contamination in compliance with the precautionary principle of the Sanitary and Phytosanitary Measures agreement of WTO. By combining the metrology approach with the needs identified by the European Reference Laboratories (EURLs) and National Reference Laboratories (NRLs), a common vision can emerge to face the food-related challenges. A European Metrology Network on Food Safety – EMN-FS is needed to improve the reliability of measurement results along the food chain and to support the efforts in ensuring traceability to SI units in European food control. The overall objective of the proposal is to develop a European network among the metrology community, the standardization and the normative bodies and the EURLs/NRLs who have to deal with the Regulation (EC) 178/2002 laying down the general principles and requirements of food law of food safety and with Regulation (EC) 2017/625 on official controls and other official activities performed to ensure the application of food and feed law. This metrological network will deal with the harmonization of measurements and standard reference procedures, the conduction of measurement comparisons and the development of reference materials, used to control the safety of provided food in order to increase trust among consumers. The EMN-FS will focus not only on the food for human consumption but also on animal feed, thus enhancing animal health and welfare.

The network will further support the standardisation organisations, such as CEN and CENELEC. Solid coordination among all involved bodies can improve the current state of the art in food-related measurements and support the constant improvement of the legislation at the European level. Moreover, it will contribute to a reduction of costs deriving from the dispute and unnecessarily repeated measurements.

Keywords: EU food policy, food-related challenges, networking activity, measurements harmonization, reference material development.

OC18 - Avoiding the culture step in outbreak investigations: parameters for optimised metagenomics analysis of contaminated food

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Introduction

The management of an outbreak depends on the rapid and accurate characterization of the contamination source. In recent years, a reflection has begun on the use of next-generation sequencing (NGS) methods to improve the study of outbreaks. Indeed, it allows to obtain the complete DNA sequence and to draw useful information such as the subtype, the presence of virulence genes or antibiotic resistance factors, as well as making a link with the origin of the contamination. Until now, the focus has been on the use of whole genome sequencing (WGS) of pathogenic isolates. However, obtaining an isolate from a food sample is often time-consuming and not straightforward. We aim to improve this process by developing a method to characterize the pathogens present in a sample without prior isolation, i.e. a shotgun metagenomics-based approach. It would provide all the necessary information to investigate an outbreak in a single test, in a minimal time frame.

Materials and Methods

We worked with the pathogenic Shiga toxin-producing Escherichia coli (STEC) spiked at the minimal infectious dose on raw minced beef meat and enriched with different methods. The DNA was extracted with two commercial kits and after library preparation, the samples were sequenced with Illumina Miseq. This was done in parallel with the classical method (ISO 13136:2012) and WGS on the isolate. The data analysis consisted of a taxonomy classification of the reads, gene detection (virulence, serotyping), reference-based assembly and phylogeny.

Discussion

The results showed the potential of a metagenomics method on artificially contaminated food samples after enrichment with a limit of detection corresponding to the minimal infectious dose of STEC. This was studied in regard to the performances and limits of the classical method (qPCR, selective media, PFGE) and WGS on isolates. This method will be validated on real-life samples.

Acknowledgements: The research that yielded these results, was funded by the Belgian Federal Public Service of Health, Food Chain Safety and Environment through the contract RF 17/6316 StEQIDEMIC.be and by Sciensano through contract RP .Be READY.

OC19 - Certified reference material of nitrofuran metabolites in chicken breast muscle from incurred samples

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Certified Reference Materials (CRM) provide measurement results with quality and traceability to the International System of Units (SI). It is also important to highlight the need of the CRM usage in the measurements related to food safety, due to the commercial restrictions of some countries in order to control the veterinary drugs residues in the food products of animal origin. The results of the studies for the development and certification of a CRM candidate for the four nitrofuran metabolites mass fractions of approximately 1 µg.kg-1 in chicken breast muscle are presented in the present work. A batch of material was produced from chicken breast muscles containing incurred residues of the nitrofuran metabolites AOZ, AMOZ, AHD and SEM, obtained from the controlled administration of the precursor antibiotics in vivo to the animals. The chicken breasts of the treated animals were manually cut into small pieces, blended and freeze-dried separately for 24 h. The freeze-dried chicken breasts were milled in a knife mill, sieved at 420 µm and stored separately. Specific amounts of each freeze-dried iccurred material and a blank matrix were mixed in a Y-shaped powder homogenizer to obtain a final mass fraction of approximately 4 µg.kg-1 to 6 µg.kg-1 on a dry basis for each metabolite, corresponding to about 1 µg.kg-1 to 1.5 µg.kg-1 on wet basis. The obtained batch was bottled and freeze-dried again, in the bottles, for 24 h. In the homogeneity study, 11 samples were analyzed in duplicate and repeatability conditions. The short-term stability study was performed for 63 days at temperatures of 20 °C and 50 °C (critical transportation temperature), while the long-term study was performed for 360 days at a temperature of -20 °C (storage temperature). In both stability studies the reference temperature was -80 °C. The properties' values presented sufficient homogeneity and stability in the temperature storage conditions at -20 °C, however, the residual moisture content showed a tendency along the fill order. In the short-term studies the residual moisture content showed instability at both temperatures and the AOZ metabolite showed instability at 50 ° C. For this reason the samples will have to be transported frozen. The characterization of the CRM candidate was performed by HPLC-MS/MS, with exact matching calibration. The purity of the standards used in the preparation of the calibration solutions was determined by 1H-qNMR and 13C-qNMR. The values obtained by the characterization were compared with the consensus values in an intelaboratorial comparison involving laboratories with recognized competence.

Keywords: Certified Reference Material, Nitrofuran metabolites, Incurred matrix, Veterinary drug residues, Food safety, Liquid chromatography-tandem mass spectrometry.

OC20 -Proficiency testing scheme for benzoic acid in banana-based condiment to support the traceability of chemical measurements to SI units

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Banana based-sauce is widely used in every Filipino household thus quality measurement of preservative in this condiment is essential for food safety and health concerns. A reference material (RM) for benzoic acid in commercially available banana sauce was studied, spiked, produced and characterized according to ISO Guide 35 [1]. A validated HPLC-PDA procedure (2) with gravimetric sample preparation was used for the homogeneity and stability studies. Short-term stability test (n=20) for 3 weeks showed that transport condition is suitable up to 40 °C using isochronous approach. The absence of bottling trend and outliers were confirmed by regression analysis and Cochran's test, respectively. Likewise, stability tests for 12 months by regression analysis showed that the slope is not significantly different from zero at 95% confidence level. Isotope dilution mass spectroscopy (LCMSMS) was used to assign the reference value of this RM with the associated measurement uncertainty. The results of statistical evaluation carried out demonstrated that the developed RM for benzoic acid in banana sauce is appropriate for accuracy-based proficiency testing (PT) rounds and a good quality control material. The PT scheme using the RM developed was organized and provided to the local laboratories. A total of 6 participants joined the PT scheme. The performance of each participant using z-score was evaluated to which 50% gave satisfactory performance, 33.3% unsatisfactory performance and 16.7% gave a questionable performance. The performance of the participants can be attributed to the choice of the measurement method used.

Keywords: Reference material, banana sauce, benzoic acid, accuracy-based proficiency test.

OC21 - Detection of antibiotic resistance genes in microbial fermentation products

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Genetically modified microorganisms (GMM), harbouring commonly antimicrobial resistance (AMR) genes as selection markers, are frequently used by the food/feed industry to produce food/feed enzymes, additives and flavourings. According to the European legislation, these commercialized microbial fermentation products should not contain GMM, viable or associated recombinant DNA. However, such contaminations have been reported by enforcement laboratories in 2014, 2018 and 2019. Even if the use of AMR genes gives rise to public health and environmental concerns regarding their potential acquisitions by pathogens and gut microbiota through the ingestion of GMM, viable or associated recombinant DNA, harbouring AMR genes, no method targeting AMR genes harboured by such GMM is currently available for the enforcement laboratories. To overcome it, we propose a PCR-based strategy targeting AMR genes composed of two main successive steps. First, the potential presence of AMR genes is detected by real-time PCR. Second, the full-length of AMR genes is evaluated by a nested-PCR amplifying a large fragment of its sequence. The proposed strategy was successfully assessed in terms of specificity, sensitivity and applicability. The results generated by the proposed strategy allow to support the competent authorities for the decisions to be taken regarding the level of risks associated with the potential AMR gene acquisition in case of unexpected DNA contaminations from additives, enzymes and flavourings-producing GMM in commercialized food and feed products.

Keywords: Antimicrobial resistance gene, genetically modified microorganisms, PCR-based detection, food and feed microbial fermentation products.

OC22 - Reconstruction of plasmids carrying antimicrobial resistance genes in food, feed and human bacterial isolates using short and long read sequencing reads

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Detection and characterization of plasmids is crucial for public health. Plasmids containing antimicrobial resistance (AMR) genes transfer within and across bacterial species, facilitating the worldwide dissemination of AMR genes via multiple routes including food. However, classical molecular approaches are unable to identify carrier plasmids or search for new mutations. Even though next generation sequencing (NGS) seems promising for this purpose, many challenges remain in both the wet-lab and data analysis. We aimed to develop a generic NGS workflow addressing these challenges to improve the characterization of circulating plasmids carrying AMR genes.

Several DNA extraction methods for isolates were compared, targeting either the total genomic content or only the plasmid fraction. Additionally, plasmids were conjugated to E. coli with a known genomic content to theoretically facilitate downstream data analysis. Each of these approaches had its advantages and disadvantages, in terms of time investment, obtained coverage and complexity of data analysis. Both short (MiSeq) and long read (MinION) sequencing technologies and a combination (hybrid assembly) thereof were evaluated. The workflow was assessed using different case studies, i.e. plasmids involved in colistin and carbapenem resistance, and a genetically modified (GM) bacterium carrying plasmids with antibiotic resistance used in the production of feed and food additives. For each of the case studies, we demonstrated that hybrid assembly approaches result in the best characterization of the plasmids. The long MinION reads allowed to successfully span the repetitive plasmid DNA regions, while the high accuracy of the MiSeq reads allowed to correct the higher error rate of the scaffolds generated by the MinION reads. This resulted in fully circular plasmids and accurate detection of AMR genes, their variants and other expected genes. The AMR genes detected with NGS corresponded to the phenotypic results.

The elaborated NGS workflow will be crucial to give an adequate response to important public health issues related to AMR in a One Health context.

Keywords: plasmid, AMR, NGS, long read sequencing and GMO.

OC23 - Rationalizing the GMO analytical detection procedure: Optimization of subsampling, homogenization and milling steps.

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Bioanalytical procedures for the detection and quantification of genetically modified organisms (GMOs) consist of two main steps: (1) reduction and homogenization of the sample and (2) laboratory sample analysis. Step 1 aims at reducing the original aggregate sample towards a homogenous lab sample, which is further analyzed in step 2. The lab sample analysis includes sample preparation, DNA extraction and PCR analysis. Whereas guidelines and official methods exist for lab sample analysis, less is known about reliable and efficient subsampling (reducing), homogenization and milling. These steps are the most critical for reliable detection and quantification of the GMO(s). Moreover these are very time consuming steps, requiring highly sophisticated apparatuses and experienced personnel in the lab. The aim of this work is to offer some alternative, innovative and time saving solutions to the questions How to reduce and homogenize grain and flour samples in a practical and representative way? and How to mill the subsample in an efficient, practical way?

For the subsampling of large grain samples - “aggregate samples” - 3 practical ways of reduction and homogenization were compared. Mixtures of 3 different types of equal amounts of grains/seeds (rice, maize, wheat) were made in heterogeneous versus homogenous ways. The mixtures were poured out using a classical seed splitter, an in-house conical system and a Kenwood homogenizer. A subsample was divided back over the 3 types of grains/seeds and these were weighed, to verify if equal amounts of the 3 fractions and thus representative reducing was obtained. All tested alternative grain subsampling ways prove to be fast, easy and low-cost procedures, allowing total grain/seed sample amounts of max. 2 kg (Kenwood homogenizer) and even more than 2 kg (seed splitter, conical subsampling system) to be subsampled. The most representative subsampling was obtained with the conical system, while cleaning and disinfection were most practical with the Kenwood system.

For the reduction of large flour samples, Kenwood homogenization using different mixing times was tested on mixtures of flour (soybean, maize). Subsamples of 200 mg were taken at different mixing times and locations in the Kenwood recipient. DNA was extracted using NucleoSpin Food kit (Machery-Nagel) and tested in real-time PCR measuring maize and soybean Ct values. No significant differences in DNA yield and Ct measurement were observed after homogenization and subsampling with a kitchen Kenwood mixer, irrespective of the mixing time.

Milling a subsample seems to be very easy, fast and practical using a Kenwood AT320A chopper (kitchen robot). This alternative milling was compared with the standard Retsch ZM200 analytical milling. Kenwood or kitchen robot milling delivers DNA yield and amplifiable DNA (comparable Ct values for maize and soybean) at least as good as with the more time-consuming and expensive Retsch ZM200 milling.

Keywords: GMO analysis, analytical milling, sample preparation, subsampling, homogenization.

OC24 - Development of new stable isotope reference materials for food authentication and traceability

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Metrology represents an essential part of stable isotope analysis (SIA). However, some methods and approaches are not yet standardised and Certified Reference Materials (CRMs) are still missing for some disciplines where SIA is applied routinely. To place isotope-delta values on the internationally agreed reporting scales it is necessary to normalise the raw stable isotope ratios using RMs that are, ideally, very similar in their chemical composition and analytically treated the same as samples. Two-point normalisation along isotopic scales is mandatory to account for 'scale compression' of individual mass spectrometers, which calls for at least pairs of isotopically different RMs. Most laboratories have prepared their own food matrix laboratory standards for day-to-day quality control, however their proprietary standards are typically stored in air or even light, which can promote oxidation and slow changes or drift in chemical and isotope characteristics. This presentation outlines the development and characterisation of new isotopic RMs of plant and animal origin for food analysis including:
(i) honey RMs from tropical Vietnam and subarctic Canada, (ii) flours from C3 and C4 plants, (iii) vegetable oils from C3 and C4 plants, and (iv) collagen powders from marine and terrestrial origins. After thorough homogenization of the 'mother' supply of each of these materials, multiple aliquots are being sealed in glass under vacuum or noble gas to exclude oxygen and to extend their shelf life to decades when stored frozen in the dark. The end-user will be able to obtain 0.5 to 1 g or mL aliquots of RMs with consensus isotopic compositions based on a current ring-test with seven participating laboratories. The ring-test benefits from the availability of liquid RMs (also international reference waters) from the U.S. Geological Survey (USGS) that are sealed in segments of silver tubing for direct EA-measurements. This enables users to reproducibly measure hydrogen stable isotope ratios of bulk honeys and vegetable oils without interference from atmospheric moisture. Our well-characterised food isotope RMs will support forensic efforts to test for food authenticity and food provenance verification. In addition, the food RMs will strengthen the metrology system by providing traceable and comparable measurement data, e.g. in the framework of the European Metrology System (EURAMET).

Keywords: SIRA, reference materials, plant, animal, authenticity, food.

OC25 - ICAR proficiency testing scheme and a novel calculation model to compare proficiency testing schemes

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The ICAR proficiency testing (PT) scheme for milk analyses is involving 85 laboratories representing 38 countries from 4 continents. In the processing of the resulting data ICAR is applying a statistical approach, developed by Luginbühl & Berger (2016), for comparing PTs by assessing them using a quality index PQ and assessing participating laboratories using a quality index PL. These are deriving from probabilities, the probability of a laboratory achieving the assigned value of a specific PT and the probability of achieving the precision figures as specified in ISO/IDF standards. The resulting indices do indicate the capability of laboratories and PT schemes to meet a common level.

The statistical approach is applied to the parameter somatic cell count as a first example . Somatic cell count is a key indicator for udder health and relevant for farm management, animal health and milk quality. It is one of the most frequently performed analytical tests worldwide. Somatic cell count is determined either with the microscopic reference method or routinely with fluoro-opto-electronic instruments and by charge-coupled devices that produce digital images.

This statistical tool should be helpful to promote analytical equivalence in somatic cell counting at a global scale.

Keywords: calculation model, proficiency test, somatic cell counting.

OC26 - International co governance of food safety based on quality infrastructure

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Due to the globalization of food trade, food safety has become the focus of global concern. At present, in order to promote the international co-governance of food safety, there are still some problems such as the lack of international co-governance mechanism and the urgent need to strengthen the quality infrastructure. Therefore, this paper proposes measures such as establishing international food safety co-governance organization and actively promoting the construction of international quality infrastructure to promote the smooth realization of international food safety co-governance.

Keywords: food safety, International governance, Globalization, Quality infrastructure

OC27 - Mineral oil in food: How is the situation in Belgium and what are the risks?

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Researchers reported presence of mineral oil in various food. Mineral oil can enter the food via different routes: (i) certain mineral oils are allowed as additive (e.g. E905a), (ii) as a pollutant from atmospheric precipitation or aquatic pollution, (iii) due to processing of food (e.g. use of machine oils and anti-dusting products) and (iv) as a residue coming from ingredients from pesticides or components from printing inks on paper and board packaging.

In 2013, the European Food Safety Authority published a scientific opinion, thereby indicating that occurrence data on mineral oil are only available for a limited number of food groups and from few countries. In Belgium, data on the contamination of food by mineral oil were lacking. Another point of concern is that the potential health effects related to the contamination of the food chain by mineral oil are largely unknown.

The analysis of mineral oil in food is further complicated since it consists of MOSH (saturated hydrocarbons) comprising a complex mixture of linear, branched and cyclic compounds and variable amounts of MOAH (aromatic hydrocarbons), mainly alkylated. Both MOSH and MOAH form “humps” of unresolved peaks in the chromatograms with the same range of volatility. Since these two fractions have a different toxicological relevance, it is important to quantify them separately. Commonly, an on-line technique existing of a combination of Liquid Chromatography and Gas Ghromatography (LC-GC) with Flame Ionization Detection (FID) is used for quantification of MOSH and MOAH. The tested matrix has also an important impact, not only on data integration and interpretation but also on the sample preparation. Due to the presence of olefins and natural alkanes, some matrices require auxiliary methods such as epoxidation and aluminum oxide. The on-line technique was used to perform analyses of a wide variety of different food samples such as dry food, vegetables, fish and meat products, …

Within this research, the presence of mineral oil in food sold on the Belgian market and subsequently the exposure of the Belgian population was evaluated. Afterwards, the hazards associated with mineral oil in terms of genotoxicity and endocrine activity were investigated. The data obtained within the project will be used to support the Belgian authorities in the implementation of new rules to minimize the exposure to mineral oil and prevent health issues caused by mineral oil.

OC28 - Effect of food composition on the migration of surrogate contaminants from paperboard packaging

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Introduction

When foods are packaged in recycled paperboard, migration of many non-intentionally added substances such as mineral oils can occur. This can be prevented by using a functional barrier. However, it is not always clear how much protection such a barrier should provide. Certain components have a higher tendency to migrate than others. Depending on their fat content, particle size or shelf life, certain types of foods may be less sensitive to migration than others, so a weaker barrier may be appropriate. Gaining more knowledge on migration to specific foods may provide valuable insights on these matters.

Purpose

The current research uses a simple test to monitor migration from spiked paperboard to foods by extracting the paperboard, rather than the food itself. This allows evaluating the migration of surrogates too hard to extract from matrices.

Methods

Virgin board was spiked with 9 surrogate components: 4 saturated hydrocarbons, 3 aromatic hydrocarbons and one plasticizer. Samples were stored at 22°C, in tightly sealed bottles containing Tenax® (legal simulant) or one of 9 different foods as receptor. After 2, 4, 10 and 16 weeks of storage, paperboard samples were extracted by immersion in ethanol/hexane (1/1) to evaluate recovery of the surrogates. The non-recovered fraction of surrogates is assumed to have migrated towards the receptors. This was validated by comparing the recovery from paperboard in contact with Tenax® with the recovery from the Tenax® itself.

Results and discussion

The extent of migration of the surrogates was found to be related to their vapor pressure. Paperboard in contact with wheat pasta showed high recovery, indicating low migration. Paperboard in contact with starchy or particulate foods such as egg pasta, wheat flour and rice flower showed intermediate recovery. Paperboard in contact with fatty foods such as biscuits and chocolate showed low recovery, indicating high migration. These results show that the fat content affects the extent of migration more than the particle size of the food. Migration to Tenax® was in some cases a strong overestimation of the migration to foods. For these cases, a correction factor for using Tenax® as a simulant may be appropriate.

Keywords: Migration, MOSH/MOAH, packaging.

OC29 - Investigation of migrating substances from food fabrics

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With the polarized debate about paper and board packaging materials, food contact materials (FCM) made from fabrics/textiles has gain importance on the market. This trend is expected to rise with the prohibition of the Walloon and Brussels-Capital Region for single use plastic bags. In this context, the aim of the research was to evaluate the safety of these “new” food fabrics.

A simple ultra-sound assisted extraction of substances from the fabrics was done with acetonitrile to avoid any compound losses. Both a GC-MS full scan screening method and targeted quantitative assays were used to determine the levels of substances migrating from food fabrics. These protocols were carried out for 43 food fabrics carefully selected on the Belgian market. Well known food contaminants such as butylated hydroxy toluene, phthalates and photo-initiators were detected, but at very low levels. On the contrary, one bread bag sample contained higher amounts of bisphenol A.

It is well characterized that the migration of contaminants from food contact materials in foodstuffs is challenging due to the complexity and large variety of foodstuffs. To overcome this issue, migration experiments were carried out using food simulants. Consequently, the bread bag was brought into contact with the simulants for bread using accelerated time and temperature conditions to simulate migration. Analysis of the simulants afterwards showed no significant migration for Bisphenol A. Although this particular bread bag contains Bisphenol A, the migration thereof is limited and the article is considered to be food safe.

In conclusion, 43 food fabrics surveyed on the Belgian market were investigated for the migration of possible contaminants towards foodstuffs. Different compounds were detected but measured levels are very low and do not trigger safety concerns.

Keywords: Food contact materials, Textile/Fabric, screening, Migration, Market survey.

OC30 - MYCOSUGAR: Investigation on mycotoxins and their producing fungi in sugarcane and its by-products

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Sugarcane (Saccharum officinarum L.) is placed among the most susceptible commodities for fungal attack, especially by the mycotoxigenic fungi. The plant has great agro-industrial and economic values and sugarcane juice is a traditional beverage consumed on a daily basis, by millions of people, in different countries such as Brazil, Egypt, India and Pakistan. Despite this knowledge, very little is known about mycotoxin contamination in the sugarcane and other sugarcane-based products (1). The current study investigated the co-contamination of aflatoxins and fumonisins in sugarcane grass, juice and sugarcane jaggery. Further, a polyphasic approach was applied in order to identify and classify the isolated fungal species (2). Mycotoxin contamination results have been used to estimate the dietary exposure of the Egyptian population to mycotoxins through juice consumption.

Sugarcane juice samples (n=89) were randomly juice shops in Egypt, while the grass were collected from Malawi (n=55) and Kenya (n=80) from different agroecological areas. Jaggery samples (n=50) were collected from different cities in Kenya. Quantification of mycotoxins were performed by a validated LC-MS/MS analytical method. Morphological and molecular identification of mycotoxigenic fungi and other microorganisms was performed according to Samson et al., 2010 (2). Dereplication strategy was also applied to unravel the diversity of the produced secondary metabolites using an in-house screening library with a fast DDA UHPLC–TOF-MS profiling method that can screen for hundreds of mycotoxins and other metabolites. Risk assessment of mycotoxins using probabilistic and deterministic approaches at various scenarios for adult male and female Egyptian juice consumers was performed. So far the obtained results for the targeted analysis, exhibit the contamination of sugarcane juice samples with AFB1 and FB1 mycotoxins in 63% (n=56) of the samples. Juice collected during winter had a contamination range of 0.1-3 µg.L−1 for AFB1 and 4-58 µg.L−1 for FB1, while samples from the summer season had a contamination range of 0.3-1.3 µg.L−1 for AFB1 only. Furthermore, the risk assessment using probabilistic and deterministic approaches for adult male and female Egyptian juice consumers pinpointed a remarkable difference in levels of exposure to mycotoxins between the two seasons and between males & females. Other results will be presented during the conference.

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OC31 - Occurrence, toxicokinetics and toxicity of citrinin and risk assessment

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Citrinin (CIT) is a nephrotoxic mycotoxin produced by *Penicillium*, *Aspergillus* and *Monascus* species. The toxin is considered as an important food contaminant, mostly found in cereals and cereal products. Until now, the risk assessment for CIT is incomplete, due to a lack of data on occurrence and toxicity. The main aim of this study is therefore to gather more data on CIT occurrence in Belgium, and to investigate the toxicity and toxicokinetics using animal models.

To study the CIT occurrence, a survey in Belgian food products was performed during two years (2018-2019). Following foodstuffs were based on occurrence data in literature: cereal products, herbs, spices, nuts, seeds, vegetarian products, meat products, baby food, alcoholic beverages, food supplements, fruit and vegetable juices. CIT was detected in a high number of food products (60%), the highest concentration was found in a red yeast rice food supplement (<LOQ - 1787 µg/kg). Other contaminated food sources were cereal products (<LOQ - 1 µg/kg), vegetarian products (<LOQ - 1.5 µg/kg), spices (<LOQ - 4.5 µg/kg) and meat (<LOQ - 1 µg/kg).

Toxicokinetic studies were performed on two relevant animal species, namely pigs and broiler chickens. Both species are important in the livestock industry, and pigs serve as a reliable model for man. A two-way cross-over experiment was performed on 8 animals. Compartmental open toxicokinetic model was applied for the analysis of the toxicokinetic parameters. The absolute oral bioavailability of CIT was determined to be 52% in pigs and 100% in chickens. A longer mean elimination half-life was observed in pigs compared to chickens (12h versus 4h respectively). These results showed a remarkable inter-species variability.

Concerning food safety, it is important to screen for residues of CIT in edible tissues of animal origins. Therefore, steady-state studies were performed on 16 pigs, 16 broiler chickens and 16 laying hens. CIT contaminated feed was administered to the animals for 3 weeks. After this period, the animals were euthanized and all edible tissues were collected, including eggs from laying hens. Carry-over to tissues for human consumption was demonstrated for all species. In addition, toxicity was evaluated by histopathology.

This study contributes to achieve a refinement of the risk assessment of CIT, since new crucial data concerning occurrence and toxicity was gathered.

Keywords: Citrinin, Occurrence, Toxicity, Carry-over, Risk Assessment.

Acknowledgements: This research was funded by the Belgian Federal Public Service of Health, Food Chain Safety and Environment through the contract RT 16/6308 (CITRIRISK).

OC32 - Analytical methods for mycotoxin determination

B. Huybrechts

Mycotoxins are secondary metabolites (MW 300-700) produced by microfungi that are capable of causing disease and death in humans and other animals. Ochratoxin A and Aflatoxin B1 are probably the two toxins of major significance. Given their potential impact on human health and on agricultural production including food, feed and livestock there has been significant research on a wide range of analytical techniques. Most methods used rely on the correct extraction from the biological matrix and often require some kind of clean-up before analysis. These steps are time consuming: sample preparation remains the main time factor in an analysis. A suitable detection method should be robust, sensitive over a wide range of compounds, but needs to be very specific in the same time. Due to the variety in structure of these toxins, it was not straightforward in the past to use one standard analysis technique. Immuno-assay based methods offer a reliable, fast and easy solution allowing them to be used by non-scenically trained personnel for in-the-field testing although they are somewhat limited to a yes/no answer and these tests are usually limited to the determination of one (group) of toxins. To our knowledge analytical liquid chromatography hyphenated to mass spectrometry, and thus enabling the analysis of multiple toxins in one run, has become the golden standard for reliable quantitative determination. The complexity of the technique limits its use to a controlled laboratory environment.

OC33 - Calcium and vitamin D intake from foods and supplements in the Dutch population

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A sufficient intake of calcium and vitamin D is important for musculoskeletal health. To ensure an adequate intake, the Dutch Health Council recommends young children and older adults to use vitamin D supplements combined with a high dietary calcium intake. This study aimed to evaluate intake of calcium and vitamin D from foods and dietary supplements in the general Dutch population.

The Dutch national food consumption survey 2012-2016 included 4313 participants aged 1-79 years. Dietary data were collected using two nonconsecutive 24-h dietary recalls and questions on frequency of dietary supplement use. Food consumption data were linked to the food composition database NEVO 2016 and supplement consumption data to the dietary supplement database NES 2018. Habitual intake distribution was modeled using SPADE-software. The prevalence of inadequate intake was assessed using the EAR-cut point approach. This could be done for calcium intake of men aged 19-70 and women aged 19-50, and for vitamin D intake of adults 70+ years. In all other cases, only an adequate intake level (AI) was set by the Dutch Health Council, and adequacy was evaluated qualitatively with possible outcomes adequate (median intake above AI) or inconclusive (median intake below AI).

The median intake of calcium increased with age until middle age. On average, dietary supplements contributed 2% to calcium intake. Among boys aged 1-8 years and girls aged 1-3 years, calcium intake was adequate, but 17% of men aged 19-70 years and 30% of women aged 19-50 years had a low intake. For other age groups, calcium adequacy was inconclusive.

Due to higher intakes of vitamin D supplements in young children and older adults, mean intake of vitamin D decreased with age in children and increased with age in adults. On average, dietary supplements contributed 15% to vitamin D intake. In case of sufficient sunlight exposure and light skin, intake of vitamin D seemed sufficient for boys and for adults until the age of 69 but inconclusive for girls. About a quarter of adults aged 70+ years had a low vitamin D intake.

In conclusion, low calcium intake was observed for part of the Dutch adult population. Nutritional status research is recommended to verify if calcium intake is inadequate. In Dutch older adults, suboptimal intake of vitamin D is already verified and reinforcement or reconsideration of policies for adequate intake is recommended in order to prevent bone fractures.

Keywords: calcium, vitamin D, intake, dietary supplements, nutrient adequacy, DNFCS.

OC34 - School meals in light of the regulation - Assessment of the public catering decree in primary schools in Hungary

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In Hungary the Public Catering Decree entered into force in 2015, addressing dietary risk factors and setting standards for food provided by public caterers.

The present assessment was carried out in 2017 and supported by the World Health Organization. The goal was to evaluate the implementation of the Regulation in terms of achieving compliance and acceptance, and to detect changes compared to the National Nutritional Environment Survey in Schools, 2013.

In a national representative sample of 139 primary schools self-administered questionnaires were filled out by school administrators. In the sub-sample of 33 schools (the sub-sample as in the 2013 assessment) food allotment sheets for 10 school meal days and menus were collected and analyzed for raw materials. The compliance of meals were assessed against the specifications of the Regulation.

Public caterers could comply with the following provisions of the Regulation in 90% or more of the primary schools: sugar should not be added to ready for consumption and flavoured dairy products; salted food powder, salted soup powder, salted flavouring creams or pastes should not be used for other than flavouring or consistency improvement purposes; drinks containing caffeine – except for hot chocolate or tea – should not be given to children under the age of 18. The proportion of primary schools increased significantly where fruits and vegetables were provided once or more times a day from 2013 to 2017, although the amount provided still not reaches the number of portions specified by the Regulation.

Overall the survey revealed high levels of implementation at almost all evaluation areas of the Regulation. In addition to the legislation our Institute helps to promote healthy public catering with various campaigns and programs.

Keywords: public catering, regulation, children.

OC35 - Probiotic properties of lactic acid bacteria isolated from household fermented sorghum slurries

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Fermented cereals/foods have been produced and consumed for thousands of years in South Africa. These products include a fermented sorghum porridge called ting. Ting is loved for its unique acidic taste but there has been claims that it is used to treat gastrointestinal related problems like diarrhoea. This suggest that ting might contain probiotics, which are living microorganisms that exhibit beneficial effect on the health of human by the intestinal microbial balance. Most widely used probiotics are Lactic Acid Bacterial (LAB) group found in milk and milk products.

In this study the probiotic properties of LAB isolated from household fermented sorghum slurries were characterized. Nine LAB strains were isolated and purified from household fermented sorghum slurries using MRS agar medium. The isolates were further identified using PCR and 16S RNA sequencing. To determine the probiotic properties of these nine isolates, different tests such as tolerance to acid and bile, antibacterial activity and antibiotic resistance were done. Seven (81,8%) of the bacterial strains were able to thrive in stimulated gastric conditions in vitro, meaning that they possess a high tolerance to stomach pH. All the isolates survived the 0.3% bile conditions which simulates the gastrointestinal tract. When the antibiotic susceptibility of the isolated LABs were evaluated on 9 different antibiotics, it was observed that all the strains were susceptible to five antibiotics, Ampicillin, Erythromycin, Mupirocin, Tetracycline and Chloramphenicol, whereas susceptibility to Polymycin B and Streptomycin was observed from only two strains. Seven strains showed resistance to Polymycin B, Streptomycin, Kanamycin and Oxacillin. All the nine strains showed different levels of antibacterial activity against Escherichia coli ATTC8739, Salmonella typhimurium ATTC 14028 and Staphylococcus aureus ATTC 6538. Four of the nine strains were therefore identified as the good probiotics that would be use to prepare different traditional probiotic products that would be also be fortified to meet the nutritional and healt benefits of poor communities.

Keywords: Lactic Acid Bacteria, probiotics, fermented sorghum, antimicrobial activity, antibiotic susceptibility.

OC36 - Method validation for determination of microplastics in food

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Many studies focused on the presence of microplastics in the marine environment, including seafood such as mussels, crab and fish. However, awareness has grown that other food items, such as sea salt, beer and honey, can also be contaminated with microplastics. To our knowledge, no extensive screening has yet been conducted on the wide variety of food items humans consume, thus lacking a view on the total exposure to microplastics by ingestion. In the 'Plastic in Food' project (financed by the Belgian FPS Health and Environment), ILVO and Sciensano will closely work together to generate data on microplastics in multiple food items, representing the potential overall microplastics intake of a Belgian consumer. Special attention will be paid to specific sources of microplastic contamination, such as uptake of microplastics present in water and air, and uptake through packaging and food processing.

We will optimize and validate an analytical method for the determination of microplastics in food, comprising the destruction of the matrix tissue, followed by active filtration and detection of microplastics by means of a stereomicroscope. The destruction protocol presented by Dehaut and co-authors showed that a destruction at 60 °C during 24h with 10% KOH is highly performant for the analysis of microplastics in mussel tissue. This destruction method allows for an efficient matrix degradation with limited impact on the polymer structure of most types of plastics. With no mussel tissue remaining visible, filtration and microplastics detection is fast and efficient.

Method optimization and validation is exclusively done with spiked samples and procedure blancs, as no certified samples exist. For spiking, polystyrene and polyethylene beads are used, differing in size and color. Validation is done with and without mussel tissue as matrix, using red and colorless beads in two size classes (106 – 125 µm and 500-600 µm). Accuracy and precision are determined, next to quantification limits, specificity, cut-off size and robustness. More tests need to be performed on a broad variety of matrices, ensuring the feasibility of the analytical protocol for multiple food items.

Keywords: microplastics contamination, method validation, food.

OC37 - Characterization of the nano-sized fraction of silver particles in food additive E174 by EM and sp-ICP-MS

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Metallic silver is an EU approved food additive referred to as E174. The recent EFSA opinion dealing with the re-evaluation of E174 indicated lack of data on toxicity studies on E174, unknown particle size distribution of E174 and evidence of the release of silver ions from elemental silver, which are specific concerns for risk assessment [1]. In this perspective, the fraction of silver nanoparticles in food additive E174 was characterized by EM and sp-ICP-MS, based on ten pristine E174 food additives and ten E174-containing products in line with EFSA's guidance document for nanotechnology and nanoscience in the food and feed chain [2].

Samples were purchased from different European web shops and local supermarkets. Descriptive TEM analysis revealed that all samples consisted of a combination of single (nano)particles and larger flakes. A higher amount of nanoparticles was measured in the silver products than in the pristine materials. In addition, the general appearance of larger flakes was altered, possibly due to the processing of E174. The chemical composition and the crystallographic structure of silver particles were confirmed by EDX and electron diffraction, respectively. Number-based particle size distributions were obtained by quantitative TEM for all products and for five pristine materials. In the five other pristine materials the amount of particles was insufficient to obtain reliable distributions. The expanded measurement uncertainty for the size measurement of the primary particles was 8%. The size distributions obtained for sp-ICP-MS did not significantly differ from the distributions obtained for TEM taken into account the limit of detection, and the measurement of agglomerated particles by sp-ICP-MS.

Effects of sample preparation on primary particle size, shape and morphology were determined. Chemical reaction between silver particles and certain dispersion media containing e.g. acetone and PVP, and the addition of too high levels of sonication energy possibly lead to alterations in particle size, and artificial introduction of nanoparticles. These results urge to include the modification of particle properties induced by processing of food additive E174 as an important factor for risk analyses.

Keywords: E174, characterization, electron microscopy, particle size distributions, silver.

OC38 - Inhibition of pathogenic bacteria in duck meat using nanoclay encapsulated oregano essential oil

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The shelf life problems and microbial safety of duck meat is a common issue. In RASFF portal there are more than 50 cases of serious risk notifications connected to the pathogenic bacteria contamination of duck meat products. Besides strict hygienic conditions through the whole processing chain, natural compounds such as essential oils (EO) with antibacterial properties could be used to increase safety and shelf-life of such products. However, their effectivity depends on several factors, including chemical composition and formulation. In our research, we investigated the effectiveness of oregano essential oil encapsulated into clay nanoparticles (nanoEO) for antibacterial properties on chilled duck breasts, as well as the residual concentration of the main compound carvacrol and sensorial properties of the nanoEO treated grilled meat. The application of nanoEO caused significant decrease of Escherichia coli and Salmonella enteritidis numbers and in some cases led to their complete inhibition after 15 days of storage. The effect of nanoEO was also superior to the application of oregano EO water emulsion. The main component of the oregano EO was carvacrol (71 %), and its concentration in the meat slowly decreased to app. 25 % of the applied dose during the experiment. The sensorial analysis revealed that the application of nanoEO did not change the overall acceptability, the pleasantness of taste and the pleasantness of smell of grilled samples. While the application of nanoparticles and the essential oils into food remains controversial, their positive effect on chilled meat microbial safety should be considered for practical use.

Keywords: food safety, nanomaterials, natural preservatives, pathogenic bacteria, antimicrobial.

OC39 - Physicochemical characterisation of several types of the E171 food additive

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The application of E171 (titanium dioxide) as a food additive has been an issue of debate in the European Union. A detailed physicochemical characterization of the E171 particles can objectify the discussions and is essential in the context of risk analysis.

This work focuses on the physicochemical characterization of 15 pristine E171 materials by transmission electron microscopy (TEM) and single particle inductively coupled plasma mass spectroscopy (sp-ICP-MS) following CEN/TC 352 guidelines. The E171 samples were purchased on the Belgian market or were obtained from European producers.

In optimized conditions, representative TEM micrographs could be recorded and the ParticleSizer image analysis software succeeded in applying noise reduction and background subtraction, allowing robust automatic thresholding and constituent particle detection. The large majority of constituent particles, confirmed to be TiO2 by energy dispersive X-ray spectroscopy (EDX), were reliably detected and measured by the software. The measurement uncertainty budgets of particle sizing by TEM and sp-ICP-MS are in the order of 10% and 16 % (Ucx, k=2), respectively, based on validation studies of a series of representative test materials. The phase of the E171 particles was determined by powder electron diffraction.

Several types of TiO2 particles were found in pristine E171. These types were shown to be applied as well in food products containing E171. All examined E171 food additives contained a significant amount of nanoparticles. In the most dispersed state, the particle size measurements by TEM and sp-ICP-MS agreed well. Eleven E171 materials consisted of anatase. Three materials consisted of smaller rutile TiO2 particles (20-40 nm) coated on mica. One material contained a mixture of anatase and rutile particles.

In future research, the methodology will be implemented in a systematic and larger scale study of E171 food additives and food items containing E171, available on the market.

Keywords: E171, characterization, titanium oxide, electron microscopy, single particle inductively coupled plasma mass spectroscopy, particle size distributions.

OC40 - Towards a routine method for the characterisation of TiO2 nanoparticles in food by single particle-ICP-MS

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Titanium dioxide (TiO2), which is largely used nowadays as a food additive (E171) in order to make products whiter and/or shiny can be partially present under nano particle (NPs) form. Despite the existing European regulation in terms of nanoparticles imposing to report the presence of NPs in foodstuffs, accurate characterisation of the nanoparticle fraction in food products is still an analytical challenge, particularly for routine food analysis laboratories.

This study addresses the development of a novel approach for the characterization of TiO2 in NPs in the E171 food additive by Inductively coupled plasma–mass spectrometry using the single particle approach (Sp-ICP-MS) in self-aspiration sampling mode. The mains analytical parameters, such as the dwell time, the suspension solvent, the most appropriate isotope, etc. were optimised. Moreover, the data obtained by using the ICP-MS software were compared with the data treatment using an improved internal calculation spreadsheet. The size limit detection obtained with the isotope 48Ti is around 25-30 nm. Finally, the method was applied for the analysis of a selection of food samples containing E171; the results in terms of NPs distribution and their fraction in the real life samples are addressed.

Keywords: Titanium dioxide, E171 food additive, nano particles, single particle-ICP-MS.

OC41 - Validation of single particle ICP-MS for routine sizing and quantification of the fraction of silver nanoparticles in E174 food additives and confectionery products

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Silver (Ag) is a food additive (E174) approved by the European Commission to be used for the external coating of confectionery, for decoration of chocolates, and in liqueurs [1]. It is commercially distributed in its pristine powder and sheet form, and in confectionery products. Due to its nature, E174 may contain silver nanoparticles, which implies a need for validated methods to size and quantify these particles. Single particle inductively coupled plasma-mass spectrometry (spICP-MS) is thereby a promising technique as it is capable of sizing and counting particles at the same time.

A spICP-MS method was developed and validated for sizing and quantifying the fraction of silver nanoparticles in E174 food additives and in products containing E174. The samples were prepared for analysis according to a slightly modified version of the method of Jensen et al. [2]. The E174 food additives showed a large silver background concentration combined with a relatively low number of nanoparticles, making the quantification of the nanoparticles more challenging than in the products containing E174. Validation of the method showed good performance with respect for the size distribution compared to the size distribution obtained from transmission electron microscopy. Depending on the sample and the background silver concentration, particles with an equivalent spherical diameter (ESD) down to 11 nm could be detected. The performance in terms of repeatability (size 4-11%, concentration 16-29%), and intermediate precision (size 2-8%, concentration 18-31%) depended on the type of sample. The large repeatability compared to the intermediate precision demonstrates the need to analyze multiple independent replicates under routine conditions. When analyzing three replicates, the extended measurement uncertainty (k = 2) on the mean ESD is 20% for E174 food additives and 11% for products containing E174. The quantification of the mass and number concentration is more challenging with extended measurement uncertainties up to 45%.

Keywords: Nanomaterials, Food additives, E174, Silver.

Acknowledgements: This research was funded by the Belgian Federal Public Service of Health, Food Chain Safety and Environment.

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ABSTRACTS: POSTER COMMUNICATIONS

A1 - Isolation and identification of microorganisms from processed milkfish products for the development of matrix-based PT material for salmonella SP. in milkfish

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Processed milkfish are common export products from the Philippines but are oftentimes rejected by importing countries due to high frequency of Salmonella sp. being detected. Aside from improving the quality of the production process, it is of great importance to improve the accuracy in detecting Salmonella sp. in food products in the country. One approach is to increase the level of competence of local testing laboratories through regular participation in proficiency test (PT) schemes, which is also an ISO/ IEC 17025:2017 requirement. Though some laboratories are able to comply, suitable PT materials for food products such as processed milkfish are unavailable. Moreover, the Philippines lack local PT providers which compels laboratories to participate in PT schemes provided by other countries which tends to be more expensive.

This study aimed to isolate and identify Salmonella sp. from processed milkfish which would be used as the analyte for the development of matrix-based PT material for Salmonella sp. in milkfish. For initial study, samples of processed milkfish products – marinated deboned milkfish and salted milkfish belly – were analyzed using FDA-BAM methods for total viable count, coliform count and Salmonella sp. detection. As a screening process, bacterial isolates were identified via VITEK® 2 Compact, an automated biochemical identification system. Identities of representative enteric isolates were confirmed using 16S rDNA sequence analyses. Salmonella enterica was found in marinated deboned milkfish while Proteus mirabilis and Escherichia coli were found in salted milkfish belly. Other enteric isolates such as Citrobacter freundii, Enterobacter spp., Klebsiella oxytoca and Klebsiella pneumoniae ssp. pneumoniae were isolated in both samples. The Salmonella enterica isolate would further be characterized for succeeding studies on PT material development. Furthermore, as Proteus mirabilis showed almost similar biochemical reactions to Salmonella enterica in conventional Salmonella sp. detection, it could be a good background microflora for the desired PT material.

Keywords: Salmonella, PT material, biological metrology, milkfish, food microbiology, food safety.

A2 - Antioxidant and antimicrobial activities of leaves extracts of urtica dioica L. from Algeria: application in apple fruits

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Increasing interest in replacing synthetic food antioxidants by natural antioxidants has intensified the demand for plant materials for the identification of new antioxidants and also antimicrobials molecules.

The aim of this work is to study the chemical composition of essential oil and determination of total phenolic content of different leaves extracts of Urtica dioica collected in the province of Tizi-ouzou, North of Algeria. Also, their antioxidant and antimicrobial activities. The total content of phenolic compounds was determined by Folin-Ciocalteau reagent. The antioxidant activity was evaluated using free radical DPPH reduction method. DPPH. The essential oil was analyzed using GC-MS technique. The study of the antibacterial activity was carried out by the method of agar against bacterial and fungal strains. Major compounds found in essential oil of nettle were Carvacrol (27.42 %) and Naphtalene (6.86 %) as principal chemical components. Essential oil revealed an antibacterial activity with respect to the bacterial and fungal strains tested. The total content of phenolic compounds was 8.44, 1.41, and 1.02 mg GAE/g of dry weight of the aqueous extract, ethyl acetate and ethanol extracts respectively. All the studied extracts showed a good antioxidant activity than ascorbic acid, used as standard. The values of antioxidant activity determined by DPPH method increased in the following order: ethyl acetate, aqueous and ethanol extracts. It was revealed that U. dioica extracts, exhibited the antimicrobial activity at least one of the tested microbial strains. The essential oil was tested in apple fruits; experimentally inoculated with Botrytis cinerea at a concentration of 107 spores/ml. Results demonstrated that U. dioica essential oil exerted an antifungal effect against pathogen tested. This effect was evident from 24 hours of incubation; showing significant differences with untreated samples. The data suggest a possibility that essential oils could be used as natural preservatives for improving life storage of apple fruits.

Keywords: Essential oil, Polyphenols, Antioxidant activity, Antimicrobial activity, Apple fruits.

A3 - Occurrence of polyether ionophores residues in Brazilian Minas cheese

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Polyether ionophores are hydrophobic antibiotics widely used in animal feeding to prevent and treat coccidiosis, as growth promoters in swines and ruminants, and to increase milk production in lactating cows. Consequently, residues of these drugs may be found in animal foods and analytical methods for their determination are available for different matrices, including milk, egg, tissues and feed. However, no occurrence data has been reported so far for dairy products, such as cheese, regarding these analytes. This work aimed to evaluate the occurrence of three polyether ionophores (lasalocid, monensin and salinomycin) in Brazilian Minas cheese using an ultra-high performance liquid chromatography coupled to tandem mass spectrometry (UHPLC–MS/MS) validated analytical method. Limits of detection and quantitation were, respectively, for monensin and salinomycin (0.25 ng g−1 and 1 ng g−1) and for lasalocid (0.25 ng g−1 and 0.5 ng g−1). The method was applied to 60 samples of Minas Frescal cheese available in the retail market in the region of Campinas, SP, Brazil. Only monensin residues were found among the analyzed samples. This antibiotic was detected in 55% of the samples and it was quantified in 5 samples at concentrations ranging from 1.1 to 2.05 ng g-1. Even though no maximum residue limit (MRL) has been established for these substances in this matrix, the results are very close to the MRL for milk (2 ng g-1) recommended by the Codex Alimentarius and the European Commission. We hope this work will contribute to the establishment of an effective food safety system in Brazil in relation to veterinary drugs residues in foods.

Keywords: polyether ionophores, Minas cheese, mass spectrometry, monensin.

A4 - Use of additives in Brazilian dairy products and compliance with current legislation

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The use of food additives is justified by technological reasons. They are essential for the development of industrialized foods, ensuring process quality, large-scale production, transportation over long distances and, mainly, capacity to increase shelf life. Without additives, food industrialization would be very restricted and limited. However, due to the current high demand for natural products, the use of food additives has been questioned in relation to inadequate applications and adverse effects to human health. The present study aimed to evaluate the commonly used additives in dairy products from the Brazilian market and verify compliance with the current legislation according to information provided on the label. For that, labels of 292 dairy products commercially available in the city of Campinas-SP, Brazil, were photographed and the additives declared by manufacturers were registered according to their technological function. In order to check compliance with current legislation, the products were divided into 14 categories: fluid milk, milk powder, infant formula, toddler formula, whey products, fermented milk, cream, condensed milk, dulce de leche, dairy-based dessert, butter, requeijão and cheese. A total of 83 food additives were reported in the label of the products. Among them, the most used were: potassium sorbate (INS 202), citric acid (INS 330), xanthan gum (INS 415), carrageenan (INS 147), sodium citrate (INS 331iii), guar gum (INS 412), annatto (INS 160b), cochineal carmine (INS 120), gelatin (no INS) and sodium carboxymethylcellulose (INS 466). Regarding the toxicological aspects of these additives, most of them have a not limited or not specified Acceptable Daily Intake (ADI), which means that they may not represent a health risk if used according to the good manufacture practices. It was observed that most of the mentioned additives are being employed according to the legislation, but some irregularities of use and declaration on the labels have been found. Ensuring that additives are properly used is important to improve food quality and promote food safety.

Keywords: additives, dairy products, legislation, food safety, labeling.

A5 - Ultra-low level quantification of pesticides in baby foods using an advanced triple quadrupole GC-MS/MS

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The EU maximum residue level (MRL) for the majority of pesticide-commodity combinations is set at the default level of 10 µg/kg. For a small number of the more toxic pesticides, MRLs for baby foods are set as low as 3 µg/kg calculated as the sum of the parent compound and relevant metabolites. In some of these multi-component MRLs the quantification limits of individual compounds can be 1 µg/kg or lower.

In this study, the quantitative performance of the Thermo ScientificTM TSQTM 9000 Triple Quadrupole GC-MS/MS system was assessed for the analysis of ~200 pesticides in baby food at very low concentrations (as low as 0.025 µg/kg). A complete evaluation of method performance included, sample preparation, overall method suitability measured from pesticides recoveries, selectivity, sensitivity, linearity and long term robustness.

The method performance was tested in accordance to the SANTE/10518/2017 guidance document. All detected compounds, at the three spiking levels in both matrices satisfied all SANTE requirements. More than 97% of the target pesticide residues had recoveries between 70 – 120% at the 1 µg/kg spiking level. Over 90% or the target compounds had a Limit of Identification below 0.5 µg/kg and over 60% below 0.1µg/kg – 100 times lower than the default MRL. All results in compliance with all SANTE criteria for method validation. Compound linearity was assessed by injecting matrix matched standards in the range of 0.025 to 250 µg/kg in duplicate for two composite Baby foods; carrot/potato and apple/pear/banana. Both sets of linearity data showed R2 > 0.990, and % RSDs of <20% for over 96% of component peaks. Robustness of the AEI source was demonstrated by maintenance of SANTE compliance at the default MRL throughout a sequence of ~400 consecutive injections of sample matrix (1 g/mL).

Keywords: Pesticide Residues, Baby Food, GC-MS/MS, QuEChERS , TSQ 9000, Advanced Electron Ionization.

A6 - Fluorimetric method and trends in aflatoxins determination

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Mycotoxins are toxic compounds produced by different types of fungus, belonging mainly to the Aspergillus, Penicillium and Fusarium genera. Aflatoxins are metabolites of the fungi Aspergillus flavus and Aspergillus parasiticus. Under specific environmental conditions, temperature and moisture these fungi may produce mycotoxins. They commonly enter the food chain through contaminated food and feed crops, cereals. There are four types of aflatoxins: B1 B2 G1 and G2. Aflatoxin B1 is the most toxic aflatoxin and the most frequently encountered. According to EU Regulation the concentration of B1 and the concentration for total aflatoxins are limited. There has been an increase in demand for development of sensitive, accurate, simple and fast method which is reliable to detect total aflatoxins at low concentrations. The most used methods in practice are enzyme linked immunosorbent assay (ELISA), high performance liquid chromatography with fluorescence detector (HPLC-FLD; HPLC-UV light-FLD) and high-performance liquid chromatography-tandem mass spectrometry (LCMS/MS). From the literature investigation and according statistical evaluation of the report from participation in Proficiency testing (PT) fluorometric determination is not very used technique for determination of total aflatoxins. From the PT report for aflatoxins in corn, 339 laboratories from 40 countries have participated. Two laboratories only have reported results obtained by flourimetric determination. According statistical evaluation of the PT participation 50% of laboratories used ELISA method, 23% HPLC-FLD, 21% HPLC-MS and 2% other method. With ELISA and LC/MS technique 21% unsatisfactory results were obtained while two results obtained with fluorimetric determination were satisfactory with z-score below 2.

Furthermore, results from fluorimetric determination possess very high reproducibility. The Laboratory 1 has obtained concentration 5.7 ppb for total aflatoxines and the Laboratory 2, 5.8 ppb. There are not significance difference between these two laboratories. The method validation for fluorimetric determination was performed using spiked corn samples and sample from PT participation. The obtained validation parameters indicate that the fluorimetric method is suitable for determination of AFs in different kind of products according EU Regulation.

Keywords: aflatoxins, corn, fluorimetric, proficiency testing, EU Regulation.

A7 - Optimization and validation of HS-SPME GC/MS to measure furan and alkylfurans in babyfood

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Furan and its derivatives are volatile compounds formed in foodstuffs during heating process. They were first reported in the late 70s 1. These compounds contribute to the food flavour but have been reported as toxic within the last years 2. To date, several reports have highlighted furan occurrence in many foodstuffs like coffee, baby food, snacks and beverages containing caramel 3. Those data were used by risk assessors to show that there is a risk related to the furan ingestion by babies 4. In 2017, the EFSA published a scientific opinion related to the risk of the presence of furan, 2-methylfuran, 3-methylfuran, and 2,5-dimethylfuran in food 5. In that framework, food safety authorities within EU Member States were asked to provide additional data regarding the occurrence of alkylfurans in foodstuffs.

The HeadSpace Solid Phase MicroExtraction coupled to Gas Chromatography/mass spectrometry (HS-SPME GC/MS) using the isotope dilution for the quantitation has been reported to be suitable for furan analysis. Nevertheless, as it is an equilibrium limited extraction technique, the extraction conditions have been optimised for the new set of analytes in cereal based baby food through a Central Composite Design approach. This experimental design highlights that each compound has different but closed optimal conditions. The results indicated that the compromised optimal conditions were found at an extraction temperature of 30 ℃ during 35 minutes.

The HS-SPME GC/MS has been validated in cereal based baby food at three levels (10, 30, and 60 µg/kg) for three days in triplicate through spiking experiments. The method shows a very high sensitivity with LOQs lower than 2 µg/kg for all methylfurans, and within-lab reproducibility RSD between 2 and 13%.

Keywords: furan, 2-methylfuran, 3-methylfuran, SPME, DoE, Validation.

Acknowledgments: The research that yielded these results was funded by the Belgian Federal Public Service of Health, Food Chain Safety and Environment through the contract RT19/05 MEFURAN.

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A8 - Investigation of the polycyclic aromatic hydrocarbons contamination in spices and dried herbs available on the Belgian market

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Polycyclic Aromatic Hydrocarbons (PAHs) are a very broad class of processing contaminants showing genotoxic and carcinogenic effects. For many years, high amounts of PAHs have been found in spices and dried herbs due to bad drying and smoking practices. Therefore, in 2015, European Commission (Reg. (EU) 1933/2015) fixed Maximal Residue Level (MRL) for PAHs in these matrices at 10 µg/kg for the benzo[a]pyrene and 50 µg/kg for the sum of four PAHs focused by the EU legislation (benzo[a]anthracene, chrysene, benzo[b]fluoranthene and benzo[a]pyrene). Although this legislation has been adopted 3 years ago, very few methods dedicated to PAHs analysis in spices and herbs have been found in the literature nor paper focused on the monitoring of this food category for the Belgian market. Hence, there was a need to validate a method to quantify these compounds and investigate to check the compliance of spices and dried herbs available on the Belgian market.

This work contains an exhaustive analysis of spices and dried herbs available on the Belgian market. 86 samples (44 spices and 42 dried herbs) have been purchased in April/May 2019 in local stores the most frequented by the Belgian population.

Samples have been analyzed by an in-house method specifically designed for these food categories and validated according to the EU regulation (Reg. (EU) 836/2011). Before extraction, 4 isotopic labelled internal standards are spiked. Then samples are extracted with a mixture of acetone/dichloromethane, followed by a purification based on solid phase extraction. The sorbent type depend on the nature of the matrix considered. Then samples are injected on a gas chromatographic system coupled to a triple quadrupole mass spectrometer.

Firsts results shows that PAHs contamination is very variable throughout samples analyzed. No trends has been observed yet between the two groups spices and dried herbs nor between the different subclasses of spices/herbs. Some samples contain no PAHs higher the individual detection limit (0.5 ng/g) and only one sample exceed slightly the EU legislation. Regarding preliminary results, PAHs contamination of spices and dried herbs seems not to be a concern for consumers in Belgium.

Keywords: Polycyclic aromatic hydrocarbons, spices, herbs, gas chromatography, mass spectrometer.

A9 - Exposure to pesticides and metals following tea consumption in Belgium

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Tea is one of the most popular non-alcoholic beverages in the world. Tea consumption contributes to the daily dietary requirements of essential elements but also to toxic overdose conditions of certain elements and contaminants [1, 2]. Pesticide residues and (heavy) metals are some of the major chemical contaminants found in tea matrices.

A Belgian market study is performed by sampling 53 teas and herbal infusions in local supermarkets. To calculate exposure to elements and contaminants from tea consumption, it is important to analyse not only the total concentrations in tea, but also the extracted concentration in the teas once brewed. Moreover, when teas are brewed, not all elements are extracted with the same efficacy [1-3].

Total element concentrations (Manganese (Mn), Iron (Fe), Cobalt (Co), Nickel (Ni), Copper (Cu), Zinc (Zn), Arsenic (As), Selenium (Se), Cadmium (Cd), Tin (Sn) and Lead (Pb)) in dry tea leaves are analysed by ICP-MS after acid (50% HNO3) closed-microwave digestion. About 400 different pesticides residues and metabolites were monitored by LC-MSMS and GC-MSMS after methanolic or acetone extraction depending the chromatographic technique used. Teas are brewed using ISO guidelines [4]: 2g of dry tea is poured in blank tea bags and 100 mL of boiling water is added. The teas are left to stand for 6 minutes after which the tea bags are removed (no stirring). Elements in these extracts are analysed by ICP-MS in these brewed tea extracts after acidification with HNO3.

The potential risk is estimated by comparing the measured element concentrations with the drinking water guidelines of the WHO [5]. If total element concentrations in the tea leaves are considered, there is a concern for Ni, As, Cd and Pb in respectively 6, 2, 3 and 24 teas. However, in the brewed teas, there is only a concern in one tea for Ni and two teas for As. Elements like Cd, Pb and Ni might be included in primary silicates in teas leaves and therefore not easily extracted. On the other hand, As is easily extracted in water. Teas with elevated As concentrations are teas containing algae. In algae, As is mainly present as arsenosugars which are As species for which the toxicity is still not well documented. Concerning pesticides residues in tea leaves content and concentration were very variable throughout all the samples. In average, 5 different pesticide residues were found in tea leaves with a maximum of 15 different residues in the same sample. Most pesticides found were neonicotinoid (acetamiprid, imidacloprid), pyrethroid (bifenthrin, cyhalothrin-L), one organochlorine pesticide (OPP) and an insect repellant: DEET.

This study will continue by analyzing the As species in the tea extracts. In addition, more algae based teas will be purchased to estimate the risk when consuming this kind of infusions. For pesticides, the next step will be the characterization of the transfer of these pesticides from the tea leaves to the infusion in order to better estimate the risk for the consumer.

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A10 - Development and application of a novel analytical methodology for simultaneous speciation analysis of Cr(III) and Cr(VI) in foodstuffs by HPLC-ICP-MS using species-specific isotope dilution

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Chromium speciation analysis at trace and ultra-trace levels in foodstuff has received great attention in the last years. Chromium is a peculiar element whose different species show opposite behaviour. Whereas Cr(VI) has been recognized for several decades as being carcinogenic, Cr(III) was for a long time considered as having beneficial health effects. Nevertheless, the European Food Safety Authority (EFSA) stated relatively recently (2014) that there is no convincing evidence of beneficial effects of Cr(III) for healthy people. Therefore the interest in chromium speciation shifted relatively recently form focusing solely on Cr(VI) to the determination of both Cr(III) and Cr(VI). Nevertheless, performing simultaneous chromium speciation analysis in food is very challenging, mainly because of the high instability of Cr(III) and Cr(VI) species depending on the temperature and pH. In addition, in food samples chromium is present at ultra-trace levels hence requiring the use of both sensitive and highly selective analytical tools. Online coupling of HPLC with ICP-MS by using species specific isotope dilution (SSID) has become in the last years the state-of-the art method for accurate speciation analysis at low concentration levels.

This work aims at the development of an accurate method for simultaneous speciation analysis of chromium (Cr(III) and Cr(VI)) in foodstuff by SSID in combination with HPLC-ICP-MS. For this purpose, species-specific sequential complexation of Cr(III) with EDTA (Ethylenediaminetetracetic acid) and of Cr(VI) with DPC (Diphenylcarbazide) was performed. Species separation was carried out by using a Dionex ION PAC AG7 (2x50mm) HPLC column and and a mobile phase composed by 10 mM HNO3 + 2.5% Methanol + 32 mM EDTA.

The results in terms of method development and validation by means of the accuracy profile approach using several food matrices such as half-fat milk, baby milk, bread and beef are presented. Additionally, its application for the analysis of a selection of meat and milk samples are also addressed.

Keywords*:* chromium speciation; food; HPLC-ICP-MS; species specific isotope dilution; meat and milk analysis.

A11 - SIMBA: Design, formulation and optimization of plant growth-promoting microbes for their use as microbial consortia inoculants

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The interactions between plant-roots and the surrounding soil, including the resident microbial populations, play an essential role on crop productivity. A growing body of evidence demonstrates the potential of various microbes to enhance plant productivity in cropping systems although their successful field application may be impaired by several biotic and abiotic factors. In this context, the activities of Work Package 2 of SIMBA (Sustainable Innovation of MicroBiome Applications in the food system) project were dedicated to exploit the full potential of Plant Growth-Promoting Microorganisms (PGPMs) for sustainable crop production by optimising the efficacy and reproducibility of field applications. In order to identify the PGPMs to be applied as bioinoculants on different crop plants (wheat, maize, potato and tomato) in Italy and Germany, a comprensive literature survey was performed by examining peer-reviewed articles and results from European related projects. The following functional groups of microorganisms were considered; i.e., phosphate solubilizing microbial strains, nitrogen-fixing bacteria, biocontrol strains, endophytic bacteria. To guarantee the development of compatible microbial consortia, selected PGPMs were preliminary screened in vitro for their ability to coexist and exhert a PGP activity.

Keywords: microbiome application, PGPM, crop, sustainability, food system.

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A12 - Toxic element mobility in soil-wheat system: comparison between 6 wheat varieties

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Food quality is defined by a set of properties such as: healthiness, nutritional value, shelf-life, organoleptic properties, etc. Many of these properties depend on the quality of the production environment and in particular of the growing soil. High level of toxic or potentially toxic elements in soils can compromise food safety due to the risk of translocation to the edible parts and/or potential bioaccumulation along the food chain. It can affect also food security, in relation to phytotoxic effects with a consequent reduction of crop yield. Several studies demonstrated that biological effects linked with the risk of contamination from toxic elements in the soil-plant system are related not to their total content, but to the bioavailable fraction. This fraction is regulated by complex mechanisms and several factors, such as: the element, its physical-chemical form and its concentration, the presence of other elements with synergic or antagonist effects, the soil and its physical-chemical characteristics, the plant (species, variety, etc.), the soil microbial population, and the effects in co-cultivation
.
The present work is focused on durum wheat and concerns the set up of analytical methods aimed at studying the relations between the plant and its rhizosphere. Samples of soil-plant specimens (i.e. the plant with its own rhizosphere) of durum wheat collected at the CREA experimental fields (Azienda Inviolatella, Roma - Italy) were characterised by evaluating: elemental total content and mobile fraction in soils, the soil parameters that can influence the absorption in the different plant parts (roots, aerial part and kernels). Furthermore, differences between cultivars grown in the same area (therefore with the same pedo-climatic conditions) were investigated. In particular, the following varieties were considered: Iride, Simeto, Achille, Antalis, Santograal, Egeo. Elemental analysis in soils was performed by ICP-AES, after complete microwave digestion for the total content, and after EDTA extraction for the mobile fraction. Toxic and potentially toxic element distribution in the different plant parts was determined by ICP-MS and the Bioaccumulation Factor (BAF) and Translocation Factor (TF) were compared in order to highlight relations between elemental bioavailability and soil characteristics or specific behaviours of some variety. Measurement uncertainty was calculated taking into account all the sources, from sampling to final analytical results.

Keywords: bioavaiability, toxic elements, durum wheat, soil-plant sysem.

A13 - Accomplishment of the EU regulations 2017/644-771 for PCDD/Fs and PCBs in food by using a novel triplequadrupole MS generation

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In 2014, European Regulations laying down methods of sampling and analysis for the EU official control of levels of polychlorodibenzo-p-dioxins (PCDDs), polychloro-dibenzofurans (PCDFs), dioxin-like (DL) and non-dioxin-like (NDL) PCBs in food and feed have been amended by EU Regulations No 589/2014 [1] and 709/2014 [2]. As a direct consequence, based on validations studies [3], gas chromatography (GC) coupled to triple quadrupole mass spectrometry (GC-QqQ MS/MS) was recognized as a confirmatory tool for checking compliance with maximum levels (ML) following specific analytical criteria [4]. Later EU Commission Regulations 2017/644-771 [5-6] further confirmed the use of GC-QqQ MS/MS and a significant number of laboratories have nowadays implemented QqQ approaches to replace, or in parallel to, their classical high resolution (HR)MS approaches based on the use of sector instruments.

In this study, the performance of a novel triple quadrupole GC-QqQ MS/MS system equipped with a programmable temperature vaporization (PTV) injector was evaluated for the analysis of PCDD/Fs and PCBs in food and feed. The MS analyzer was equipped with a titanium ionization chamber and a new short collision cell capable to accumulate and eject ions by means of very narrow pulses that allow to minimize the noise and to adapt accumulation times for sensitive selected reaction monitoring (SRM). The analytical capability of the system was confronted by the strict requirements set by the EU Regulation for a range of standards, quality control (QC) and food/feed samples and compared with a routine GC-HRMS method.

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A14 - Aluminium in food

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Humans, in general, exposed to aluminium (Al) through consumption of drinking water and food, and using pharmaceuticals and consumer products. The European Food Safety Authority (EFSA) assessed the risk of Al exposure in their Scientific Opinion on the Safety of aluminium from dietary intake (2008). In the general European population, the mean weekly dietary exposure ranged from 0.2 to 1.5 mg Al/kg body weight (bw), while for the high consumer the exposure was up to 2.3 mg Al/kg bw/week. EFSA established a TWI (tolerable weekly intake) of 1 mg Al/kg bw per week, which is likely to be exceeded in a large part of the European population. The main sources to the dietary intake of Al appeared to be cereals and cereal products, vegetables, beverages and some infant formulae.

The uses of Al as a food additive and in food contact materials are regulated in the European Union (EU); however, there are currently no regulations for Al content in food. Official food controls requires validated methods for the analysis of Al in food. Studies suggest that there are challenges with the extraction of aluminium using only HNO3. Higher recoveries were obtained when using HNO3 and HF, than when using only HNO3 in the digestion . Two CEN (European Committee for Standardization) candidate methods for the determination of Al in foodstuffs using ICP-MS or ICP-OES (prEN 17264 and prEN 17265) addresses the challenges with extraction of Al compounds.

We have establish the CEN candidate method for the determination of Al in foodstuffs using ICP-MS and are currently validating it. Once the method is validated, it will be applied to a set of food samples. The selection of samples will be based on a search in the RASFF (Rapid Alert System for Food and Feed) database, i.e. samples with an expected elevated level of Al will be selected. Results from the method validation and the investigation of Al in food will be presented and discussed with regards to method performance and food safety.

Keywords: Analysis, CEN, EURL-MN, validation.

A16 - Pilot study for developing an interactive platform to facilitate communication between main actors in the food packaging chain

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Packaging has an important role in identifying, containing and protecting the food product throughout the supply chain from the production to its consumption. Without packaging the product would be hard to handle in the different supply chain operations and it would be exposed to a great risk of damage and contamination. Packaging production is a global industry, which is characterized by its internal diversity and each of its sectors individually affects the situation on the market. The requirements towards packaging and materials intended to come into contact with food are systematically growing. The packaging industry will continue to face relentless and growing pressures from manufacturers, wholesalers, logistics and supply chain companies, retailers, authorities and above all from consumers, to look great, prevent damage, reduce cost, cut waste and reduce energy use.

In order to face changes and trends, main actors in the food packaging chain will be invited to interact using an interactive online tool - platform - which will be developed by involving as much as possible the food packaging producers from Romania. The main aim of the platform will be, beside facilitating the communication of the producers with other actors involved in the food packaging chain, to promote safe packaging materials, to have a sustainable consumption of it and to reduce food waste while protecting environment. Based on a detailed questionnaire which will be developed after a comprehensive study of the food packaging market in Romania, food packaging manufacturers will be invited to join the platform and interact with the entire food packaging chain stakeholders. The project, through its results, will ensure the consumers health and trust in food packaging chain.

Keywords: packaging materials, food contact materials, food packaging chain, packaging industry, online platform.

A17 - Fast Analysis of a Multi-class Pesticides Panel in Wine and Olive Oil Extracts by LC-MS/MS

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Introduction:
Increasing food safety concerns and growing agricultural trade has resulted in more stringent pesticide regulations globally. To comply with such regulatory standards, screening methods for large numbers of pesticide residues are becoming more common in the routine food safety laboratory. Tandem quadrupole mass spectrometry offers a highly sensitive and selective detection in complex matrices. This poster describes a method for analysis of multi-class pesticides in food samples (wine and olive oil) using liquid chromatography coupled with a triple quadrupole mass spectrometer.

Method:
Wine and olive oil samples were extracted with acetonitrile and buffer salts using a simplified QuEChERS method. In the case of olive oil, and additional clean-up step was added utilizing a lipid removal cartridge to remove unwanted matrix co-extractives. An Accucore aQ column (2.1 x 100 mm, 2.6□m) was used for the separation of all analytes within 15 minutes. The mobile phases were A: 0.1% formic acid/5 mM ammonium formate in water and B: 0.1% formic acid/5 mM ammonium formate in methanol. Injection volume was only 1uL to increase robustness of the method for both column and mass spectrometer. Detection was performed using a TSQ Quantis triple quadrupole mass spectrometer, coupled to a Vanquish Flex UHPLC system.

Preliminary Data:
A multi-residue method was developed for screening (over 550 residues) and quantitation of approximately 300 pesticides in one 15-minute run with polarity switching. One or two ion ratios were used to confirm each analyte (±30%), plus accuracy of retention time to ±0.1 min to show robustness of the method which are required under EU SANTE Guidance 11813_2017. This single method was applied to the analysis of pesticide residues in wine and olive oil extracts. All pesticides analyzed showed excellent limits of quantitation and detection between 0.5 to 10ppb. Precision was excellent with calibration R2>0.9900. Utilization of a lipid removal cartridge showed good percent recovery on spiked levels between 10 and 50 ppb of 70-120%, which is within the SANTE Guidance. Samples of wine and olive oil were also screened to check for pesticides beyond the required target compounds to confirm that a single multi-method can be used to greatly increase the scope of residues monitored using a triple quadrupole. Furthermore, the method was developed using software with built-in workflows for streamlining method development and routine analysis. The experimental results will be discussed in detail.

Conclusion:
A multi-class, multi-pesticide residue method with simplified sample preparation and broad applicability to a wide variety of matrices is required to address growing regulatory requirements. The LC/MS/MS method presented here is compliant with EU SANTE regulations and can also be used for screening additional residues.

Keywords : Multi-residue analysis, pesticides, LC-MS/MS.

A18 - Food and Beverage Fraud Prevention Using Isotope Fingerprints

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In this poster the application of stable isotope fingerprints in food and beverage fraud detection is explored. Data are shown that show how stable isotopes offer conclusive answers on questions associated with origin, adulteration and correct labeling of food and beverage products. An overview of the interpretation of isotope fingerprints and the technology used is also provided.

The food and beverage industry suffers from fraudulent activities that include incorrect labeling of products and adulteration, which has a significant impact on food and beverage safety, brand names and reputation and the market economy. Preventing food and beverage fraud is a key challenge that requires a reliable, cost-effective analytical process that can detect whether the labeled product is authentic or if it has been changed after the final manufacturing process, or alternatively if it has been independently produced, using alternative ingredients, but labeled as an original product.

Detecting food and beverage fraud can be achieved using stable isotope measurements because stable isotopes can differentiate between food and beverage samples which otherwise share identical chemical composition: this is called the isotope fingerprint. Using the isotope fingerprint of food and beverage products is a reliable technique in food and beverage fraud prevention and food safety.

IRMS, stable isotope fingerprinting, food fraud, food safety

B1 - Smart Store-Keep your sample and data safe forever

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Food quality includes all the attributes that influence a product value to the consumers. This refers positive attributes such as appearance (size, shape, color, gloss, and consistency), origin, texture, nutritional aspects and food processing methods, and negative attributes, like contamination with filth, discoloration, off-odors and spoilage. Food safety, according to the FAO and WHO (2003) [1], refers to all those hazards, whether chronic or acute, that may make food harmful to the health of the consumers.

Therefore, definition of food quality is a global issue. The elemental composition of food is fundamental to assess the presence of nutrients elements and the absence of contaminants. Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-AES) and Inductively Coupled Plasma Mass Spectrometry (ICP-MS) are the common techniques employed for foodstuff elemental analysis [2]. Sample preparation is a main critical step of chemical analysis, which affect directly the precision and the accuracy of the results. Main problems faced during the destructive sample preparation procedures are: contamination and losses, the time required for total mineralization, and the use of hazardous mixtures of acids, which may also give interference.

In this contribution, we propose SMART STORE™ device as a solution for sampling and classification, assemblage. Foodstuff sample is deposited on acetate cellulose filter and inserted in the SMART STORE™. The device sandwiches the sample between two polymeric foils, protecting it from further contamination and preserving it from environmental degradation. SMART STORE™ is connected to the web application that classifies the samples by QR code, simultaneously creating a database. Labelling is allowed directly in the field. Back to the lab the system automatically transfers the data to the cloud and updates your database. At EXPO 2015 it was presented a successful application of SMART STORE™ for food quality and safety: chemical analysis of chocolate by Total reflection X-Ray Fluorescence (TXRF) for screening purposes.

SMART STORE™ is a useful tool for sample preparation with many advantages with respect to conventional approaches, with the unique feature of infinite sample storage capability. Moreover, thanks to its friendly interface and software, it is easy to use by non-experienced staff.

Keywords: Sample preparation, SMART STORE, foodstuff, screening, direct analysis.

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B2 - The use of Biotechnology to improve the use of indigenous complementary food

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Complementary feeding (weaning) is the provision of foods or fluids to infants in addition to breastmilk between the ages of 6-24months. Poor weaning practice and malnutrition are still a public health problem in developing countries, South Africa inclusive, since the commercial weaning foods may not be quite affordable to everyone. The aim of this study was to develop protein and nutrient-enhanced, cereal-based complementary food products and young children's foods that have incorporated locally available protein sources, green leafy vegetables. The nutritional, functional, microbial and sensory acceptability of traditional South African complementary foods were investigated.

A survey of weaning foods in all the nine Provinces of South Africa revealed that unfermented maize or sorghum are used as grain sources for the preparation of weaning foods. In some cases, the cereal flour (sorghum or maize) is mixed with the readily available protein source being soya or bambara nuts or peanut or cowpea. Most communities use green leafy vegetables such as amaranthas and okra for use by adults, which are high in minerals and vitamins. However, this are not served to children. The low dry matter content of the traditional complementary foods limits the daily protein and energy intake to around 70 and 40%, respectively the RDA and may thus contribute to the high stunting rate of children in South Africa.

The optimization of the ideal formulations of the "flour+ protein+ vegetable" were investigated using the sorghum or maize + bambara nuts/ cowpea/peanuts + amaranthas and according to the RDA guidelines for infants and children...

So far the project has been able to achieve a composite flour to make weaning food products with formulations for cereal: legume: vegetable 60:30:10). The addition of 5% sorghum malt for 10 minutes at room temperature significantly ($p < 0.05$) increased protein digestibility (20%–50%), and total sugars (2.0%–4.5%), while phytate content decreased (3.5–2.9 mg/g) significantly ($p < 0.05$). Sorghum malt was also able to reduce the viscosity of the porridges to 2600cp. The effect of the use of probiotic cultures on the microbiological and nutritional quality of the food is still being investigated. In addition, the assessment of mineral contents particularly, Ca, Mg, Na, and Zn is also still underway and the results will form part of this presentation. It is expected that the formulated diets will have the desired characteristics of weaning food, hence could be used for alleviating protein energy malnutrition (PEM) in infants and children under 5 years of age.

Keywords: Protein Energy Malnutrition, Complementary foods, traditional foods, nutrition, Recommended Daily allowances.

C1 - Comparison between ELISA and qPCR kits for detection of hazelnut incurred in rice, cookies and chocolate samples

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Patients having a hazelnut (Corylus avellana) allergy can depict several different symptoms from mild, e.g. oral allergy syndrome (OAS), up to potential life-threatening, i.e. anaphylactic shock. According to EU Regulation 1169/2011, hazelnut, just like other tree nuts and 13 other food ingredients, need to be mandatory labelled as ingredients on the food product. To monitor and minimize the risk for unsuspected contamination in food products, different ELISA and qPCR kits exist to detect hazelnut trace levels.

Protein-based kits often target known hazelnut allergens such as Cor a 1.04, Cor a 11, Cor a 8 and or a 9, which are the most important characterized allergens linked to hazelnut. Processing steps like heating, cooking, roasting may affect the conformation of thermolabile allergenic proteins. This lowers but does not exclude allergenicity. Processing may also render the targeted proteins less detectable due to modification of the allergens during food processing, matrix interferences, inhibiting agents, or weak extractability of proteins. DNA-based methods may offer an alternative, however are linked to detection of the species of the ingredient, and not of the allergenic protein(s) as such.

Annually, the Belgian National Reference Laboratory (NRL) for Allergens organizes validation studies on the detection of primary food allergens. The aim of this study was to evaluate the performance and fitness for purpose of five ELISA and two qPCR hazelnut detection kits on home-made reference materials. To produce these reference materials, rice powder, cookies with and without gluten, and chocolate paste were incurred with well known levels of whole defatted roasted or unroasted hazelnut powder. In total, 4 blank matrices and 11 matrices incurred with levels from 0.6 to 4.2 mg total hazelnut protein per kg product were tested with 7 commercial kits in the 2 NRL labs (CER and ILVO).

Additionally, some reference materials were tested by the kit providers themselves. Results of this validation study are shown in detail in this poster.

Tested qPCR kits seem to be less useful for hazelnut detection and quantification than tested ELISA kits. Indeed, false negative, suspicious and false positive results are obtained with qPCR kits. All the evaluated ELISA kits gave a correct indication of hazelnut presence or absence (except for two suspicious samples). There is no significant influence of the presence of gluten on hazelnut detection and quantification. Finally, ELISA kits are useful for HN quantification in chocolate but less useful for cookies (influence of processing).

Keywords: hazelnut, food allergens, ELISA, qPCR, comparative study, incurred reference material.

D1 - Assessment of physicochemical, antioxidant and antimicrobial characteristics of Algerian honeys

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Honey, as a natural product well known for its nutritional and therapeutic characteristics, has become the subject of several studies. In Algeria, a wide range of honeys of various botanical and geographical origins have not yet studied, and some are also less well known.

Ten samples of honey (n=10) of various botanical origins (assumed according to beekeepers) collected from different regions of the Algerian territory, were analyzed for certain physicochemical properties as well as their content of phenolic compounds and tested for their antiradical activity using DPPH method.

Also, their inhibitory activity against five microbial species, including Klepsiella pneumonia, Pseudomonas aeroginosa, Staphylococcus aureus, Escherichia coli and Candida albicans were investigated. The analysis of the physicochemical parameters of the samples shows that the majority of them comply with the international standards with the exception of a few samples which have HMF contents higher than the tolerated limits. Quantitative estimation of the phenolic compound reveals that honeys are rich in total polyphenols, however, they are poor in flavonoids. The quantitative evaluation of the antiradical effect shows that the darkest and the citrus honeys have the most important activity, that the PCA statistical analysis confirmed its closed relationship with electrical conductivity and total flavonoid content. The in vitro evaluation of the antimicrobial activity reveals that all the bacterial species have been sensitive by different significative levels, for the activity of the honeys. The most sensitive species were (E.coli and Staphylococcus aureus), while the yeast (Candida albicans) considered as the most resistant.

From the obtained results, it can be concluded that the varieties of Algerian honey tested with their floral and geographical diversification, can be used as much as natural antioxidant and antibiotic.

Keywords: Honey, Physicochemical, Antioxidant properties, Antimicrobial activity, HMF.

D2 - Estimation of flavonoid and chlorophyll changes in several micro-green species due to different LED light regimes with multiparametric fluorescence indexes

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The aim of our research was to investigate the effects of different LED light regimes on the growth and the epidermal content of flavonols, anthocyanins and chlorophylls in microgreens monitored by a non-destructive fluorescence measurement.
Microgreens that are leafy vegetables harvested as seedlings, 8-15 days after germination, highly acceptable by consumers as Ready-To-Eat (RTE) food because tender, tasty, visually attractive and promoted by scientific reports as a highly nutritious, being an excellent source of vitamins and antioxidants in concentrations from 4 to 40 times higher than in adult plants. LED light affects growth and nutritional quality of microgreens particularly LED irradiance and spectra. In our study we tested two photosynthetic photon flux density (PPFD) set at 150 and 300 µmol·m-2·s-1 and two different light treatments with Red and Blue (RB) and Red, Blue and White (RBW).
Cultivation was carried out in four separated grow rooms each equipped with DYNA RX30 (Heliospectra, Sweden) LED multispectral light source and on a jute-kenaf-fiber mat (Greenfelt, Manifattura Maiano, Italy) as a substrate in an ebb and flow hydroponic system.
The microgreen species selected were: Lactuca sativa var. augustana - Celtuce, Brassica napus - Kale Red Russian, Brassica oleracea var. acephala - Cavolo Nero di Toscana and Brassica oleracea var. capitata - Cabbage Red (CN Seeds Ltd., Pymoor, England). Seeds were evenly distributed at a density of one plant/cm2 onto the hydrated pad (25 x 25 cm for every species) inside the grow room tray. During the experiment air temperature was set at 25±1°C during day while at 16±1°C during night. Germination of the seeds was realized in the dark in the first three days. Growth and morphology data were collected 10 and 15 days after sowing. Ten randomly selected seedlings of each species were used to determine mean fresh weight and dry weight, cotyledon area with the AM350 Portable Leaf Area Meter (ADC Bioscientific Ltd., Herts, UK) and hypocotyl length with a digital caliper (Scienceware Digi-Max Sigma-Aldrich).
The adaxial (AD) and abaxial (AB) side of the microgreen cotyledons were separately analyzed, one plant at time, with the Multiplex Research (Force-A, Orsay, France) a handheld multi-parametric fluorescence sensor based on light-emitting diode excitation and filtered-photodiode detection.
In this study we have used: SFR_RT, FLAVT, ANTHT indexes as the sum of the adaxial (SFR_RAD, FLAVAD, ANTHAD) and abaxial (SFR_RAB, FLAVAB, ANTHAB) values registered and NBIC_RT.
The total simple fluorescence ratio SFR_RT (SFR_RAD+ SFR_RAB) index is linked to the chlorophyll concentration of the leaves calculated as the ratio of far red chlorophyll (FRF, 735 nm) emission, divided by red chlorophyll emission (RFR) under red excitation (AD+AB). This index increases with increasing sample chlorophyll concentration. The Flav index (FLAVT= FLAVAD + FLAVAB) compares the chlorophyll fluorescence intensity emitted as far red fluorescence (FRF_UV) under ultraviolet and red excitation (FRF_R) and is proportional to the flavonols concentration of the epidermis (AD+AB). The NBI (NBIC_RT = SFRRAD + SFRRAB/FLAVAD + FLAVAB) index is based on the ratio between the SFR_R and FLAV indices and takes into account the total chlorophyll concentration of the leaf and the total of the epidermal FLAV of the leaf (AD+AB). The ANTHT = ANTH_RGAD+ANTH_RGAB is the ratio of far-red fluorescence (FRF) excited at two different wavelengths with red FRF_R (635 nm) and Green FRF_G (516 nm) and is proportional to epidermal anthocyanin content [ANTH_RG=log(FRF_R/FRF_G)].
To evaluate changes of the physiological status in microgreens species due to different light treatments we have used this promising technique for non-destructive real-time monitoring and analysis and as a tool to implement during the production process for nutritional quality improvement.

Keywords: flavonols, anthocyanins, chlorophylls, microgreens, fluorescence, non-destructive real-time monitoring.

D3 - Validation of HPLC method with pre-column derivitization for the determination of histamine in dried fish

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Histamine, a biogenic amine produced from decarboxylation of histidine, is a parameter for detection of decomposition of food products, especially in seafoods. High levels of histamine can cause severe allergic reactions to some people. To address this food safety concern, a reversed phase high performance liquid chromatography developed and validated for the detection and quantification of histamine in dried fish. Pre-column derivatization using dansyl chloride after extraction with 0.1 N HCl and liquid-liquid extraction using hexane were optimized for dried fish matrix. Acceptable separation of histamine from selected biogenic amines was achieved using Agilent Zorbax Eclipse XDB-C18 with gradient program of acetonitrile and water as mobile phase. Histamine derivative is detected by photodiode array detector at 255 nm. Processed sample stability was determined before the method was validated for its linearity, sensitivity, repeatability and recovery. Dansylated histamine was found to be stable in autosampler for 48 hours at 10 °C. Good linearity was achieved from 0.49 – 13 mg/kg prepared in matrix solutions and the repeatability ranged from 3.7 – 4.9 % at three concentration levels. Average recovery of 98 % was obtained. The validated method was suitable for the determination of histamine in locally consumed dried fish.

Keywords: method development, histamine, dried fish, dansyl chloride.

D4 - Comparison of ascorbic acid and nitrates content in fruit and vegetables from farmers´ market and supermarket

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The growing interest of people in healthy lifestyle and food quality also increases the popularity of farmers´ markets. Fruit and vegetables are the best food sources of ascorbic acid, which is essential nutrient involved in many important body functions. They are also sources of substances potentially harmful to the human body such as nitrates.

The aim of this work was to compare ascorbic acid and nitrates contents in selected fruit and vegetables from farmers´ markets and supermarkets in the Czech Republic. The contents of ascorbic acid and nitrates were determined in apples, plums, strawberries, red pepper, spinach and tomatoes by the HPLC-DAD.

The highest content of ascorbic acid was in red pepper samples (922 mg.kg-1). The ascorbic acid content in different fruit and vegetables from farmers´ market and supermarkets was not significantly different ($p = 0.9140$). However, when individual types of fruit and vegetables were compared, strawberries and tomatoes had significantly higher, and red peppers significantly lower, ascorbic acid content ($p < 0.05$).

The highest content of nitrates was found in the spinach samples (2969 mg.kg-1). The nitrates content in fruit samples was lower in comparison to vegetables samples. Products from farmers´ market had a 29% higher content of nitrates than similar products from supermarkets ($p = 0.0349$). Statistical analyses of each kind of fruit and vegetable samples showed, significantly higher nitrates content in tomatoes from farmers´ markets ($p = 0.0002$).

To sum it up fruit and vegetables from farmers´ markets may not always be more suitable choice than fruits and vegetables from supermarkets from ascorbic acid and nitrate content point of view.

Keywords: ascorbic acid, nitrates, fruit, vegetables, farmers´ market, supermarket.

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D5 - The use of a biofixators for the removal of AFM1 from milk

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Introduction

The EU RASFF (Rapid Alert System for Food and Feed) system observed the significant presence of mycotoxins in food and feed. It is considered that 25% from the total quantity of produced food in the world, mainly of plant origin, are contaminated with mycotoxins. Due to their toxicity, it is important to control their presence and quantity in foods. Aflatoxin M1 is formed as the product of biological conversion of aflatoxin B1 in the mammary glands of mammals fed with fodder containing aflatoxin B group. Therefore, mycotoxins are entering in human food chain, not only through cereals and cereal products, but also through the meat, eggs, milk and dairy products. It is important to find accurate, reliable and efficient strategy for removal of inherent mycotoxins in food and/or fodder supplements. In this study, removal of mycotoxins with microbial cultures and their cellular components are imposed as an alternative to existing physical and chemical methods of detoxification. The project aims to explore the application of lactic acid bacteria (LAB) isolated from traditional dairy products and yeast cell components (β-glucan), and possible removal of resulting complex with mycotoxin from the milk. The research included quantification of the amount of AFM1 in milk samples using HPLC-FLD and LC-MS/MS followed by application of different concentrations of live and dead cells of LAB, lyophilized live and dead cells of LAB, as well as application of β-glucan (commercial and isolated from yeast cells). Formed biofixator - AFM1 complex was removed using the membrane filtration. Quality of milk regarding the quantity of macronutrients (fat, fatty acid composition, protein and carbohydrates and lactose) were determined.

Results

It was isolated and identified 10 native LAB species/strains, incubated their viable or heat-treated cells (108 CFU mL-1) in milk spiked with 0.5 µg L-1 of AFM1 at 4 °C for 0, 2, 4, and 24 h, and quantified the amount of unbound AFM1 with HPLC. AFM1 binding efficiency ranged from 21 to 92% for viable cells and from 26 to 94 % for the treated ones. Since both viable and heat-treated Lactobacillus plantarum showed the best results, were used for the next step in AFM1 removal from milk. Heat treatment in combination with filtration and centrifugation yielded removal as high as 96%. The treatment of AFM1-contaminated milk with LAB and beta-glucan as mycotoxin binders does not affect the content of macronutrients in milk to the point that it becomes unfit for human or animal consumption, or a variety of milk products. The only treatment method that stands out in the reduction of macronutrients is the one with live LAB cells combined with centrifugation and filtration. However, even in this case, the quality parameters of milk were acceptable for consumption and further processing. We believe that our findings are encouraging for the dairy industry to continue abating milk contamination with AFM1 with the investigated mycotoxin binders and prevent economic damage caused by AFM1.

Conclusion

Results of this study will be proposed for application of biological methods in reducing and removing AFM1 in / from milk in special situations of milk contamination, without significant impact on the quality of the product itself. This solution can be of great interest for the dairy industry and all its products regarding quality and safety demands and satisfy allegations on specification (nutritional table) which is an integral part of labeling of the products, all in accordance with the EU Regulation on the provision of food information to consumers (1169/2011).

Keywords: AFM1, lactobacillus, biofixator, milk.

D6 - Could the antioxidant profile be related to the presence of toxic elements in Olea Europea L. leaves and in drupes? A study on 11 Italian cultivars

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Olive growing in Italy generates 3,151 million euros and accounts for 2.4% of agrifood production value (Ismea) through the olive oil sector, without overlooking the market of table olives. This crop of considerable agricultural and economic relevance is appreciated for its nutritional properties. In fact, Olea Europea L. contains high amount of polyphenols, such as oleuropein and hydroxityrosol, which are strong antioxidants and radical scavengers and confer bitterness and pungency to the oil taste. The concentration of these compounds either in leaves or drupes is known to be influenced by biotic and abiotic factors (pH, redox potential, water regime, clay content, organic matter content, cation exchange capacity, nutrient balance). Toxic or potentially toxic elements, absorbed from the environment, are among the most effective abiotic factors that may modulate antioxidants concentration.

The present work aims to study potential correlations between the antioxidant profile and the content of toxic or potentially toxic elements in leaves and drupes of different olive cultivars. In particular, eleven Italian cultivars with a different commercial use were compared:

- oil cultivars: cipresso, canino, frantoio, itrana, moraiolo, pendolino;
- table cultivars: ascolana, uovo di piccione;
- dual-purpose cultivars: leccino, maurino, ortice.

The trial was carried out taking advantage of the olive tree collection located at the ENEA Casaccia Research Centre (Rome – Italy). The availability of different cultivars in the same field limited the variability associated to the pedoclimatic and agronomic conditions, thus allowing to focus on the cultivar-related effects. The following parameters were determined, in both olive drupes and leaves:

1. Total polyphenols (UV-Vis spectroscopy)
2. Antioxidant activity (UV-Vis spectroscopy)
3. Oleuropein and hydroxityrosol (HPLC-DAD)
4. Toxic and potentially toxic elements (ICP-AES and ICP-MS).

Results, processed by Principal Component Analysis (PCA), provide a tool that may enable valorisation of olive oil and table olives in terms of quality and safety - and promote the use of leaves for the extraction of bioactive compounds. Further experiments are foreseen to extend the data set to the same cultivars as well as to other cultivars, in order to evaluate the correlations with the collection period/phenological phase, and to verify potential correlations with different pedoclimatic conditions.

Keywords: Olea Europea L., antioxidant profile, toxic elements.

D7 - Sustainability in the food industry: managerial and technological actions to reduce the environmental footprint of Tinaia wine and Taleggio

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According to IPCC, agricultural and food systems are estimated to account for one-third of global greenhouse gas emissions, more than twice that of the transport sector). In the last years, agri-food companies and consumers have increased their awareness on sustainability criteria. Many innovations and technological advancements have been emerging to carry out new organizational frameworks and production systems as well as to guarantee food and energy security from and environmental and social point of view. The European project “PEFMED – Uptake of the Product Environmental Footprint across the MED Agri-food Regional Productive Systems to Enhance Innovation and Market Value”, running from November 2016 till July 2019, co-financed by the Interreg Mediterranean Programme and coordinated by ENEA, promoted eco-innovation in the food and drink industry in the Euro-Mediterranean area, by applying the "Product Environmental Footprint" (PEF) method, which evaluates the environmental impact of a product throughout its life cycle, from the primary production to domestic consumption and waste treatment. This method, coupled with environmental and socio-economic aspects, was tested in nine agri-food product chains and clusters located in different Mediterranean regions. Two Italian companies and their supply chains, producing Taleggio cheese and Tinaia wine were involved in the project. The PEF studies of Taleggio cheese and Tinaia wine highlighted the most relevant life cycle phases in relation to their environmental impacts. On the basis of the outcomes of the PEF studies, some eco-innovative interventions able to potentially reduce the environmental impact of the two product and their value chains as well as to improve its socio-economic performance were selected. The proposed actions that are presented in this work, taking into account their effective applicability and cost-benefit ratio, are cross-cutting eco-innovative interventions as well as some specific technologies or methods applicable to one stage of the production chain. The goal of these intervention is to reduce the products environmental footprint without modifying the quality of food and its safety index. In conclusion, PEF application process seems to be an interesting opportunity to help farmers and food producers to better understand their environmental and social impact and to identify improvement solutions which could decrease the impact per unit of food.

Keywords: Food systems Sustainability, PEF, environmental and social impact eco-innovative interventions.

Acknowledgments: This work was partially supported by “PEFMED – Uptake of the Product Environmental Footprint across the MED Agri-food Regional Productive Systems to Enhance Innovation and Market Value”, Project co-financed by the European Regional Development Fund.

D8 - Evaluation of antifungal activity of essential oils against Penicillium spp. in in vitro and in vivo conditions by integrated approaches

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In recent years, consumers prefer safe and high quality food, that is easy to prepare, natural or low processed, but with a longer shelf life. The Safe & Smart project refers to the safety of the food system through the development of innovative technologies involved at different stages of the supply chain, integrating systems for risk prevention and for the rapid diagnosis of chemical and biological contaminants or other unwanted substances. To realize smart, active packaging systems that, following an external stimulus such as light irradiation release natural antimicrobial agents, two commercial essential oils were tested; thyme and basil oils, given their known biocidal activity against food microorganisms were chosen. The aims of the present study were i) to assess the inhibition activity of thyme and basil oils on the outgrowth of some common food-spoiling fungi in vitro and in vivo, ii) to detect fungal growth in tomato sauce by using a combined approach of conventional culture-based method and smart system. Some Penicillium spp. strains isolated from tomato, being tolerant to low temperatures, are able to contaminate and grow also in fridge stored tomato sauce. Here, a Penicillium sp. strain, identified as Penicillium expansum by both culture-based and molecular methods, was used. The in vitro-inhibition assays performed on this strain revealed that the thyme oil showed a significantly higher antifungal activity than that of basil oil. Preliminary "fitness" tests were also carried out in the same commercial tomato sauce to monitor the inhibitory effect of the essential oil of thyme on fungal growth in vivo. The Microbial Challenge Test was performed by inoculating P. expansum strain into jarred tomato sauce, and the microbial development was evaluated both in the presence of thyme oil and in its absence (control), by enumerating fungal and total bacterial load over time. Overall, our results showed a low level of fungal mycelium in samples artificially inoculated with P. expansum in presence of thyme oil, thus confirming its antifungal effect. In parallel, detection of P. expansum was performed by measuring the CO2 produced by the fungal metabolism, in order to develop a method useful for early detecting and monitoring the qualitative decay of tomato sauce due to possible contamination after opening the jar, as aiming to allow a proper and waste free domestic management of the product.

Keywords: Mould early detection, antifungal activity, essential oils, Penicillium sp.

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D9 - METROFOOD-CZ: state-of-the-art national research infrastructure in the fields of food quality & nutrition

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METROFOOD-CZ represents the Czech national node of the European Research Infrastructure METROFOOD-RI “Infrastructure for Promoting Metrology in Food and Nutrition”, listed in the ESFRI Roadmap 2018 – Domain “Health & Food”. The main aim of the METROFOOD-CZ is to operate a state-of-the-art research infrastructure in the fields of primary agricultural food production, food processing and technology, food quality & safety and traceability of raw materials and products, as well as to support a new interdisciplinary research approach by bridging all agricultural and food sciences disciplines to cover the whole agri-food sector the entire supply chain and all its actors, from producers up to consumers. The food industry sector is one of the largest and most important manufacturing sectors in Europe. The situation in the Czech Republic is however different. There is still a burden from the times of transition from planned to open economy. In the highly competitive environment Czech agricultural and food companies lost their innovation potential and until now they are strongly dependent on the import of know-how and technologies.

Measurements play a key role in every aspect of control and evaluation of food quality and safety: from the determination of nutritional value, assimilability and biological value of nutrients, to the evaluation of freshness, nutraceutical properties and sensory characteristics, up to chemical and microbiological safety checks, detection of food adulteration and control of raw materials and products traceability.

METROFOOD-CZ represents a very important opportunity for the development of the agri-food sector and for strengthening the presence of the Czech Republic at European and International level. METROFOOD-CZ brings together the main research institutions in the field of agri-food production and agricultural products quality (the Czech University of Life Sciences Prague), food processing and technology (Food Research Institute Prague) and food chemistry, food analysis, quality, safety and nutrition (University of Chemistry and Technology, Prague). This infrastructure works in close cooperation with the Czech Agriculture and Food Inspection Authority, State Veterinary Administration, Central Institute for Supervising and Testing in Agriculture, and national reference laboratories. RI is strictly linked with other Czech Institutions (such as: Czech Chemical Society, Milk and Dairy Research Institute, Czech Society for Nutrition, Federation of the Food and Drink Industries of the Czech Republic, etc.). Each partner brings its wide and consolidated network of international collaboration, which ensures a very broad range of action, open to EU Member States and Associated Countries, but to Developing Countries and new markets and able to meet the needs of the scientific community and all stakeholders at a national, EU and global level.

Keywords: metrology, food quality, food safety, nutrition, food processing, agriculture.

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D10 - Researches on the impact of minimal processing on the antioxidant potential of cabbage varieties

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Antioxidants are compounds that have an important role in reducing the concentration of free radicals (especially peroxyl radicals) responsible for lipid peroxidation and autooxidation of organic substances. In present, antioxidants are a very important class because of their potential as prophylactic and therapeutic agents against many diseases as cancer, diabets and cardiovascular diseases. Also, they can successfully reduce adverse damage caused by oxidants by eliminating them before they react with biological targets, preventing chain reactions or preventing oxygen activation in highly reactive products.

The Cruciferous family acts as a source of natural antioxidants due to high levels of vitamins, polyphenols and pigments, protecting the human body from damage caused by reactive oxygen species. Vegetables from the Brassicaceae family are highly nutritious, providing nutrients and phytochemicals such as vitamins, carotenes, fiber, soluble sugars, minerals, glucosinolates and phenolic compounds. Cabbage (Brassica oleracea L. var. capitata) is one of the most important vegetables grown worldwide. The level of vitamin C, polyphenol constituents, Trolox equivalent antioxidant activity (DPPH) and content of chlorophyll and carotenoids were determine for four processed cabbage varieties (Buzau, Buzoiana, Magura, Isalnita). The effects of minnimum processing on stability of bioactive components and antioxidant activity are discussed below.

Keywords: antioxidant activity, Brassicaceae, cabbage, minimal processing.

E1 - Development of a bioinformatics pipeline for routine analysis of whole genome sequencing data of Escherichia coli isolates

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The adaptation of whole genome sequencing (WGS) and bioinformatics for routine molecular typing and pathogen characterization in a public health setting remains problematic, which is partly due to the lack of user-friendly and validated data analysis tools that can be used for routine typing in the National Reference Laboratories (NRLs) and peripheral laboratories. In collaboration with the Belgian NRL for Escherichia coli, we developed a pipeline for the routine analysis of E. coli isolates that was specifically designed to tackle the aforementioned challenges. The push-button pipeline is executed through a user-friendly interface in Galaxy and can use Illumina or IonTorrent WGS data to characterize E. coli isolates. The pipeline performs automated data processing and quality control of sequencing data before several bioinformatics assays are executed: kmer-based taxonomic classification, variant calling and filtering, resistance characterization based on the presence of genes from several antimicrobial resistance (AMR) gene databases and based on specific point mutations, virulence characterization, serotype determination, plasmid replicon detection and sequence typing using several typing schemes (including core genome Multilocus Sequence Typing). The pipeline performance is currently being characterized by means of a set of performance metrics and definitions that were specifically adapted towards bioinformatics assays, and which evaluate repeatability, reproducibility, accuracy, sensitivity, precision, and specificity. Preliminary results on a representative set of samples demonstrate high performance, indicating the feasibility of using WGS in routine public health settings to replace classically employed pathogen typing and characterization techniques. Similar pipelines can be developed for other pathogens and case studies, making bioinformatics analyses less complex and more time-efficient for both expert and non-expert users.

Keywords: NGS, STEC, whole genome sequencing, validation, public health, national reference laboratory.

E2 - Detection of food enzyme-producing microorganisms in food enzyme preparations

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The use of food enzymes (FE) in commercial food applications is growing continuously. Nowadays, the producing industry is responsible for the safety of the FE introduced on the food market. Currently, no strategy exists for an efficient and accurate control and monitoring of contaminants in FE (preparations). The presence of micro-organisms (living and/or related DNA) producing the FE is one of the possible contaminants. Here, we propose a general strategy that can be used by enforcement laboratories to detect this type of contaminant(s). First the list of 304 FE submitted to EFSA for safety evaluations was analyzed, of which 87% are produced by micro-organisms (fungi (53%), yeast (2%) and bacteria (32%)), resulting in 71 different species. Thereafter, in order to identify these micro-organisms in FE, we have developed a simple and fast workflow based on PCR and Sanger sequencing. Samples of the 71 species were collected, after which their 16S (bacteria) and ITS (fungi/yeast) regions were amplified by PCR. The obtained amplicons were then sequenced through the Sanger platform to subsequently allow characterization for most of these strains using 16S and ITS sequences collected from public databases (UNITE and NCBI). Lastly, the relevance of the sequence analysis using the collected 16S and ITS sequences was verified through a consensus tree analysis. The proposed strategy allowed the identification of the FE-producing microorganisms at the genus level and, for most of them, at the species level.

Keywords: Identification, PCR, sequencing, food enzymes, producing organisms.

Acknowledgements: "The research that yielded these results, was funded by the Belgian Federal Public Service of Health, Food Chain Safety and Environment through the contract [RT 17/5 SPECENZYM]".

E3 - An integrated strategy combining DNA walking and NGS to detect GMOs

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In order to strengthen the current genetically modified organism (GMO) detection system regarding unauthorized GMO, we have developed a new strategy based on DNA walking to amplify by PCR unknown sequences surrounding a known DNA region. This DNA walking is performed on key transgenic elements, commonly found in GMO, that were earlier detected by real-time PCR screening. In coupling this DNA walking to the Next-generation sequencing technology, we demonstrated the ability of this approach to detect unauthorized GMO via the identification of unique transgene flanking regions and the unnatural associations of elements from the transgenic cassette. This approach was successfully applied on different food and feed samples.

Keywords: GMO, detection, real-time-PCR, DNA walking, Next-generation-sequencing, food and feed chain.

E4 - The added value of WGS for foodborne outbreak investigation and surveillance of STEC and Staphylococcal enterotoxins

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Foodborne illnesses can be caused by pathogens or their toxins. Nearly all foodborne pathogens can cause outbreaks. Controls are routinely performed on food as part of surveillance systems. The classical characterization methods, however, show a limited fingerprinting profile of outbreak strains, thereby insufficiently discriminating the outbreak isolates from the circulating background. Moreover, several sequential analyses are required for a full characterization, which is costly and time-consuming. Detection of e.g. Staphylococcal enterotoxins (SEs) with the currently available commercial ELISA kits is expensive and limited to a set of 5 out of 22 toxins known to cause food intoxication. Therefore, failed toxin detection in a food sample with a suspected isolate can be due to the absence of the genomic potential to produce the toxin, absence of toxin expression or detection limitations. Whole Genome Sequencing (WGS) has the potential to address these issues. With its single nucleotide resolution, WGS is the ultimate tool for pathogen typing, including rapid identification of antimicrobial resistance, virulence and toxin genes, and source tracking. However, the benefits of this technology are accompanied by challenges which must be addressed before it can be used in routine public health practice. This study aims to assess the added value and bottlenecks of WGS for its application in foodborne outbreak investigation and surveillance, using Shiga-toxin producing Escherichia coli (STEC) and Staphylococcus aureus as case studies.

For the development of the WGS workflow, selected isolates were cultured from food and human fecal samples originating from sporadic and outbreak cases, or which had the genomic potential, as previously assessed with PCR, to produce one or more of the SEs most prevalently causing food poisoning in Belgium. All the different steps in the WGS workflow are currently being assessed. To start, different commercially available DNA extraction kits were evaluated and compared.

The choice of the most appropriate DNA extraction kit for each case study was based on different criteria i.e. quality, quantity and integrity of DNA; and the potential influence of confounding factors in the DNA extract. Using the WGS data of the investigated strains with a priori known characteristics, it was evaluated whether the choice of the extraction kit influenced the potential to accurately track the relationship between the STEC isolates and whether sufficient information could be obtained on the presence of SNPs, virulence genes, antimicrobial resistance genes, and se- genes. Next, library preparation, sequencing and rapid real-time data analysis will be evaluated. An optimized WGS workflow addressing the different bottlenecks will be crucial to fully take advantage of WGS and to assist the competent authorities in evidence-based decision taking in case of foodborne outbreaks.

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F1 - Proficiency test round for the determination of deoxynivalenol, ochratoxin A and zearalenone in cereals

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The reliable determination of mycotoxins in commodities are essential in order to protect consumers, to comply with regulations and international trades. In 2018, the first proficiency test aiming at providing interested laboratories in Asian and Pacific regions with an opportunity to evaluate their performances regarding to the determination of deoxynivalenol (DON), ochratoxin A (OTA) and zearalenone (ZEN) in maize and barley malt flours.

All procedures were done in accordance with ISO/IEC 17043. Naturally DON and ZEN contaminated maize was used while naturally OTA contaminated barley malt was mixed with DON and ZEN contaminated maize materials, produced by inoculating cereal grains with Fusarium graminearum strain. No detected OTA concentration was found in maize flour. Homogeneity assessment was performed according to IUPAC 2006 and showed that DON, OTA and ZEN were sufficiently distributed in these materials. The stability demonstrated that no significant loss of DON, OTA and ZEN occurred during the timescale of the PT...

Twenty-six international laboratories were registered in China, Romania, Republic of Moldova and India. Only 9 laboratories were ISO/IEC 17025 accredited.

Statistical assessment of the results was done according ISO 13528 using the robust statistical method to evaluate the assigned values and their uncertainties. Standard deviations for proficiency assessment were calculated using the modified Horwitz equation. No false positive results were reported for OTA in maize flour. Satisfactory z-scores were ranged from 81.0 to 100% of the participants. By combining z-scores for both maize and barley malt flours, rescaled sum of z-scores analyses showed that acceptable results were mostly obtained by participants. Nevertheless, up to 71.4% of the results were supplied without estimate of the MU. Hence, no ζ-score was calculated for these participants. Efforts should be devoted in estimating and reporting the MU for their analytical method, to allow the assignment of compliance towards the maximal accepted limits and the calculation of ζ-scores in inter-comparison laboratory trials.

Keywords: Proficiency test, Cereals, Mycotoxins, Deoxynivalenol, Ochratoxin A, Zearalenone.

F2 - Certified reference materials and quality control materials …… Valuable tools for laboratory management

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Quality management for ISO 17025 accredited facilities is an inherent part of daily operations. Utilizing both Certified Reference Materials and Quality Control Materials as a cornerstone of the quality system is an excellent method for implementing and then maintaining solid quality laboratories. These materials are crucial for calibrations, validations, training, run acceptance criteria, blind testing, troubleshooting, and proficiency testing. New uses for materials have the potential to further enhance quality systems. This poster details some recommendations for incorporating CRM's and Quality Control materials into the overall system.

Keywords: Quality control, certified reference materials, mycotoxins.

F3 - The role of scientific research in increasing the performance of agro-food testing/analysis

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Agro-food products globalization generated difficulties in quality monitoring of food products which have a negative impact in food safety. In this respect, at European Union level, there is an increase in the activity of regulating and harmonizing of the instruments for assessing the quality of food products. An important role is kept by European Food Safety Authority (EFSA) and Joint Research Center (JRC).
These main actors support innovation involved in testing techniques area by developing and standardizing new methods and, on the other hand, increase of measurements performance. Legislative requirements for testing the quality of agro-food products increase the demand for specific products and services.. Organizations involved in these activities must comply with the requirements of EN ISO/ IEC 17025 “General requirements for the competence of testing and calibration laboratories”. One of the mandatory requirements of EN ISO 17025 regarding the quality of the results evaluation, refers to the organization involvment in inter-laboratory schemas and to constantly determination of the uncertainty of measurements. Even if generating high costs, measurement performance depends on the organization's access to domain-specific tools. To balance the costs, the involvement of the organization in inter-laboratory comparisons and the use of certified materials in testing activity is a traceable evidence of testing/ analysis performance. This study shows the complexity in manufacturing multi-parameter reference materials which is correlated to the need of developing analytical test techniques. In the same time, the study identifies risk factors associated with the measurements performance and the production of multi-parameter reference materials.

A comparative analysis of the required financial effort in producing of reference materials highlights a risk factor in development in the food testing field. National R&D Institute for Food Bioresources - IBA Bucharest, as a scientific research unit with experience in cereals area, implements the standards EN ISO 17034 “General requirements for the competence of reference material producers” and ISO 17043 “Conformity assessment. General requirements for proficiency testing”, which complies with the EN standard ISO / IEC 17025. Thus, the scientific research supports the increase of the performance in the testing of agro-food at national level and implements the principles of the METROFOOD project.

Keywords: food testing performance, certified reference materials, risk factors in producing certified reference materials.

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F4 - Primary reference material for somatic cell counting in milk

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In the dairy analytical sector for some parameters it is difficult to build traceability because stable reference materials are lacking and the reference method shows a poor precision.
To try to solve this issue a joint Project Group of the International Dairy Federation (IDF) and the International Committee on Animal Recording (ICAR) has been given the task to design a workable global reference system for somatic cell counting (RSSCC) in milk as a model to promote analytical equivalence.
Somatic cell counting in milk is a clear example. It is one of the most frequently performed measurements, estimated at over 500.000.000 tests/year worldwide. It serves as an indicator for the udder health status of lactating animals, is relevant in food legislation, in payment of raw milk and also has a considerable impact for farm management and animal breeding programs. The analytical performance of nowadays fluoro-opto-electronic routine methods in terms of precision is superior to the reference method based on microscopy. Laboratories have therefore adopted various solutions for anchoring their counting level.
In the frame of the RSSCC with the EC Joint Research Centre a stable primary reference material for somatic cell has been produced and is going to be characterized by qualified laboratories representing research centres, dairy industries and governmental laboratories from European, American and Asiatic continents.
The reference materials can be used for calibration or calibration checks of routinely operated somatic cell counters and as quality control samples for reference method performance verification. It is also possible to use the reference materials to establish assigned values for locally applied secondary materials and as a blinded control sample in proficiency testing schemes.
The bottom-up approach of the RSSCC and the involvement of the different dairy stakeholders during the development and characterization of the reference material, offers a fertile soil for a better acceptance of this important quality assurance tool to optimally safeguard comparability of routine testing results in laboratories worldwide and would serve as an example to build metrological traceability in the food sector.

Key words: Reference System, Primary reference material, Metrological traceability

F5 - Establishment of the international joint research center of reference material for mycotoxins: contexts and fields of activities

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Reference materials enable the traceability of results to appropriate measurement standards and are important tools for laboratories during the development of analytical methods, chart control building, uncertainty measurements and interlaboratory comparisons such as proficiency test and method validation study. Commercially available reference materials are often dedicated to the regulated mycotoxins whilst less known or emerging mycotoxins are gaining interest. In this context and in recognition of mutual interests, the Shanghai Academy of Agricultural Sciences and Sciensano join their forces in creating an “International Joint Reserach Center of reference materials for mycotoxins” based on the following main fields of activities :

- Preparation of matrix reference materials for mycotoxin analyses
- Preparation of pure standards of mycotoxins
- Organisation of proficiency tests
- Establishment of the analytical methods for mycotoxins

The establishment of this international joint research center will impact the research levels on reference materials and subsquently improve the availability of exotic standards and matrix reference materials for analytical purposes, animal experiments or food processing behaviour research on mycotoxins. Providing ring tests will also benefit from these material produtions. The development of these metrological tools is an important step towards food and feed quality and risk assessment.

Keywords: Reference Materials, Standards, Mycotoxins, Quality, Safety, Proficiency test.

F6 - Preparation of dimethylarsinic acid reference material

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In this study, dimethylarsinic acid reference material was prepared using high purity sodium dimethylarsinate as raw materials by weighting method. On basis of quality analysis, the certified value was defined by the comparing with primary reference material by means of HPLC-ICP-MS. In addition, the checking of uniformity and stability test were systematically studied .The result revealed the certified value of the reference material was 22.1 μg/g with expanded uncertainty of U = 0.8 ug/g (k = 2) while its valid was 12 months.

Keywords: dimethylarsinic, elemental speciation, reference material, uncertainty.

G1 - Mineral oil migration from cardboard food contact materials: assessing the endocrine activity of mineral oils (PAHs) using the DRE-, ERE- and PPARγ CALUX bioassays

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Introduction

Mineral oils are a complex mixture of organic compounds, consisting of two main types: the saturated hydrocarbons (MOSH) and the aromatic hydrocarbons (MOAH). The occurrence of mineral oil in food has been reported in the past, with concentrations that often reach 0.1% and can even approach 1% in some exceptional cases. These compounds can enter the food chain either intentionally (as an authorized food contact material or additive), or unintentionally (printing inks, recycling, lubricating oils, …). The biggest concern for mineral oil regarding human health is the MOAH fraction, since these compounds are potentially genotoxic, carcinogenic and may have endocrine disrupting properties. An important group of compounds in the MOAH fraction are the polycyclic aromatic hydrocarbons (PAHs). PAHs consist of fused benzene rings, which are formed during incomplete combustion of organic matter (e.g. in engines, incinerators, forest fires…). An important concern linked to PAHs is their potential endocrine disrupting activity. Endocrine disrupting chemicals (EDCs) are a structurally diverse class of both synthetic and natural compounds which can interfere with our hormonal system by binding to endocrine receptors, resulting in adverse health effects such as reproductive damage, developmental impairment, obesity, diabetes and cancers. The aim of this study was to determine the endocrine activity of 9 PAHs using three different CALUX bioassays: DRE-CALUX, ERE-CALUX and PPARγ CALUX.

Materials and methods

Three CALUX (chemically activated luciferase gene expression) bioassays were used to determine the endocrine activity: 1)The PPARγ CALUX® system of BioDetection systems (BDS, Amsterdam) uses U-2OS cells (human osteoblast) that are stably transfected with human PPARγ2. 2)DRE-CALUX (dioxin responsive element) uses a third H1L7.5c1 generation cell line, derived from the mouse hepatoma hepa1c1c7 wild-type cells, which contain the AhR receptor and 3) The ERE-CALUX (estrogen responsive element) uses a human breast cancer cell line (VM7Luc4E2), which contains the estrogen receptor. All three cell lines are also stably transfected with the firefly luciferase reporter gene. Potential activity is determined by measuring light emission after exposure to the compounds and adding luciferin.

Results and discussion

1)PPARγ CALUX®: Anthracene, fluoranthene, pyrene and fluorene showed a weak agonistic activity for the PPARγ receptor. Only BaP showed a weak antagonistic activity. In both the agonistic and antagonistic tests, no effects were observed in presence of S9 metabolic fraction. Larger compounds like chrysene and benzo(ghi)perylene did not show any activity on the PPARγ receptor which may be the result of sterical hindrance preventing them to interact with this receptor (>4 rings). 2)DRE-CALUX: BaP is known to have an AhR agonistic effect and was used as reference compound. Out of the other eight non-alkylated PAHs, only chrysene exhibited significant agonistic activity. 3) The ERE-CALUX: So far, only preliminary range finding results were obtained, showing that phenanthrene, naphthalene, chrysene, fluoranthene and pyrene all show agonistic estrogenic activity.

G2 - Analysis of individual (MOAH) by APGC- QTOF-MS and comparison to the conventional method LC-GC-MS

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Mineral oil hydrocarbons (MOH) consist of a mixture of chemical isomers of saturated hydrocarbons (MOSH) and aromatic hydrocarbons (MOAH). Their analysis is very complex, as there are hundreds of isomers both linear, branched and with multiple aromatic rings, with or without sulphur. The contamination of food from food contact materials by MOH, mainly by MOAH, represents a risk to human health as MOAH are mutagenic agents and genotoxic carcinogens [1].

The contamination of food can come from different stages of food processing or packaging. Recycled materials, printing inks or some adhesives are common sources of MOAH.

Conventional methods to determine mineral oils involves the separation of MOSH and MOAH fractions either by silica column with AgNO3 (0.33%) and subsequent quantification by GC-FID or by LC-GC-FID or LC-GC-MS. A hump of unresolved peaks, which is quantified by GC-FID using an internal standard, gives the final result [2].

However, MOSH and MOAH contain a large number of compounds, which have not been identified and require a deeper evaluation before being quantified, to avoid overestimating or underestimating them. The analysis of mineral oil samples treated with solid phase extraction cartridges (SPE) followed by GC-MS and APGC–QTOF revealed the presence of different aromatic and polyaromatic compounds, such as alkylated benzenes, alkylated naphthalenes, benzothiophenes, compounds up to four aromatic rings, and dinaphthothiophenes.

GC-MS(EI) is commonly used for identification of unknown substances, since the large number of fragments generated by this technique and its high reproducibility allow identification of unknown substances through the comparison of mass spectra to those from the scientific libraries. APGC is a soft ionization that generates less fragmentation and allows obtaining the exact mass of molecular ion. APGC coupled to QTOF-MS allows the structural elucidation and identification of a large number of unknown substances.

This research explores these two techniques as complementary tools for identification and quantification of mineral oil components. Qualitative and quantitative analysis of individual MOAH present in several mineral oils and in a series of recycled materials were carried out by APGC-QTOF-MS and the results obtained are shown.

Topics: Food safety, minerals oils.

Keywords: Food safety, mineral oils, migration, analysis, APGC-QTOF-MS.

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G3 - Release of trace elements from porcelain enamelware

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Introduction

Currently, the European Commission is revising the Directive 84/500/EEC on ceramic food contact materials (FCM) and will extend the scope of this directive with vitreous enameled articles and glassware. New release limits for Pb and Cd as well as limits for other metals are being discussed. Ceramics themselves are rather well studied, on the contrary, little data exist on the metal release from vitreous enameled cookware. Vitreous enamelware consists of a metal substrate and an enamel coating (frit), this creates a very specific material. The substrate can be composed of several metals and alloys (aluminium, steel or cast iron) with possible contaminants and impurities, more specifically oxides of Ni, Co and Li are important components of the frit.

Purpose

The goal of this research is to collect some data as an answer to the call of the Europe. Which trends on metal release for enameled tableware can be seen compared to ceramics. These differences could have important consequences when setting (new) limits for Pb, Cd and other metals in the Directive 84/500/EEC.

Methods

Samples (in quadruplicate) for enameled table-, cook- and bakeware were collected from the Belgian market. Migration was performed using the testing conditions of the Ceramics Directive 84/500/EEC (4% Acetic acid, 24h, 22°C). Three consecutive migrations (with cleaning in between) were performed and compared. The simulants after migration (and the blancs) were measured by ICP-MS/MS with a fully validated method for 20 elements: Li, Be, Al, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Se, Mo, Cd, Sn, Sb, Ba, Tl, Ti and Pb.

Results

Preliminary results showed that for some elements differences can be seen between ceramic or enameled tableware. Enameled tableware seemed to mostly release Li (3 - 800ppb), Al (0.1 - 6 ppm), Co (0.2 – 200 ppb) and Ti (0 - 1ppm), with Co and Li exceeding in some cases the existing limits of the Resolution for Metals and Alloys (CM/Res(2013)9).

Conclusion

Release of trace elements from both materials showed different trends for some elements. This can have important implications when setting limits in the revision of Directive 84/500/EEC, where enamelware will be included.

Keywords: Trace elements, metal release, enamelware.

I1 - Level of minerals and inorganic contaminants in meat, fish and mixed dishes: an exploratory analysis developed in Portugal

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Due to their nutritional attributes, meat and fish products are an integral part of dietary guidelines. However, their daily intake should be moderate when compared to other food groups. Globally, meat and fish consumption have been increasing for the past few decades. Portugal is no exception, meat and fish are consumed daily in quantities that exceed the national recommendations. The main objective of this exploratory study was to develop a methodology, based on chemometric studies, to characterize, compare and detect patterns in food groups and subgroups of meat, fish and meat or fish mixed dishes, from the Portuguese TDS (Total Diet Study), according to their concentrations of minerals and contaminants. The methodology consists of an exploratory analysis rooted in univariate, multivariate, parametric and non-parametric statistical techniques. The concentrations of 9 minerals (Cr, Mn, Co, Ni, Cu, Zn, Se, Sr and Mo) and four contaminants (As, Cd, Pb and Sn) within ten meat samples, thirty-one fish samples and nineteen mixed dishes samples, were used as an input for the exploratory analysis. Multielement analysis was performed using an ICP-MS. The application of the exploratory analysis was successful and allowed the development of a framework suitable for situations with similar purposes.

ANOVA and Kruskal-Wallis' test managed to prove and dismiss the existence of several similarities and differences, firstly observed with descriptive statistics techniques. These analyses also provided evidence of a homogenization effect on the concentrations caused by the inclusion of additional ingredients, besides meat and fish, in the mixed dishes group. Red meat and shellfish were the subgroups responsible for the majority of the significant differences between their counterparts.

The analysis of Pearson and Spearman correlations, between minerals and contaminants, was conducted to enhance TDS' security aspect. Overall, fewer correlations were identified in the resulting dishes food group than in the meat and fish groups.

Lastly, cluster analysis and factor analysis allowed to conduct a pattern recognition analysis in the data. These analyses provided further proof about the homogenization effect and the clear separation of red meats and shellfish when compared to their respective subgroups.

Keywords: Total Diet Studies, Pattern Recognition, ANOVA, Correlations, Cluster Analysis, Factor Analysis.

I2 - Influence of fat types on the fatty acids and trans fatty acids composition of biscuits

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This aim of this study was to evaluate the influence of the type of fat used in the manufacture of biscuits on the fatty acids (FA) and trans fatty acids (TFA) composition of end products. Five types of fat (margarine, butter, sunflower oil, palm oil, and pork fat) have been used in the biscuit manufacturing. All five assortments of biscuits were analyzed for: the total lipid content, individual and total content of saturated (SFA), monounsaturated (MUFA), polyunsaturated (PUFA) fatty acids and TFA.

The FA profile was determined by gas chromatography coupled with mass spectrometry (GC-MS).
The following fatty acids were identified and quantified:

- 24 FA in biscuits with margarine (the main ones were oleic, palmitic, and linoleic acids);
- 27 FA in biscuits with butter (the main were palmitic, oleic, and myristic acids);
- 18 FA in biscuits with sunflower oil (the major ones were linoleic, and oleic acids);
- 20 FA in biscuits with palm oil (the main ones were oleic, and palmitic acids);
- 26 FA in biscuits with pork fat (the main ones were oleic, palmitic, and stearic acids).

The fatty acid content (% of total fat) varied as follows:

- biscuits with margarine: SFA – 45.87, MUFA – 38.18, PUFA – 15.95;
- biscuits with butter: SFA – 70.68, MUFA – 23.13, PUFA – 6.19;
- biscuits with sunflower oil: SFA – 10.74, MUFA – 20.23, PUFA – 69.03;
- biscuits with palm oil: SFA – 45.10, MUFA – 41.51, PUFA – 13.39;
- biscuits with pork fat: SFA – 37.82, MUFA – 51.42, PUFA – 10.76.

In terms of total TFA content, the values ranged from 0.21 (biscuits with pork fat) to 1 g/100 g fat (biscuits with butter). The TFA found were elaidic and trans vaccenic acids. The results showed that the fatty acid composition of biscuits is influenced by the type of fat used and TFA was below 1% when pork fat and butter were used.

Keywords: fatty acids, trans fatty acids, biscuits, composition, GC-MS.

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I3 - Soy protein hydrolysates in bakery products

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Soy protein is an important food protein source owing to its nutritional benefits and functional properties. The nutritional composition of soy proteins includes essential amino acids, calcium, iron, magnesium, fiber, polyunsaturated fats etc.

The aim of the study is to develop soy protein based products (pasta and cookie) using as ingredient soy protein hydrolysates (SPHs). Different SPHs were obtained from soy protein concentrate by enzymatic hydrolysis using the following proteolytic enzymes: Neutrase+Flavorzyme (NeuFla), Papain (Pap) and Umamizyme (Uma). The enzymatic hydrolysis determined an increase in the antioxidant capacity of the hydrolysates and improved content of amino acids (especially, lysine which is deficient in most cereal grains) and also improved the oil binding capacities of the SPHs.

Pasta and cookies were manufactured using wheat flour as base ingredient. Wheat flour was substituted with 5% SPH. The sample made with 100% wheat flour was the control.

Cookie and pasta samples with SPH addition had almost 1.2-and 1.3-fold higher protein content than control pasta, respectively. By SPH addition, the color of pasta was changed. The color variation of the pasta from the darkness to whiteness was: Uma > NeuFla > Pap > Control. There were positive correlations between L* (lightness parameter) of pasta and the corresponding SPHs (r = 0.91).

In order to investigate the volatile composition of the samples, an electronic nose system was used. A discrimination index of 93 was achieved between control and cookies with SPHs, which explains a very distinct odor of samples. All the samples had different aroma compounds, but NeuFla and Uma cookies were more alike in volatile composition being situated in the opposite side of the PCA plot (Principal Component Analysis) compared to Pap cookie. The same conclusion was also observed for pasta samples. Regarding the taste, the products containing NeuFla and Pap had lower bitterness intensity than Uma.

Soy protein hydrolysates obtained from SPC using the enzymes Papain or the combination between Neutrase and Flavorzyme can be used as supplements in wheat flour to enhance the nutritional value of pasta and cookie products.

Keywords: soy protein hydrolysates, proteases, pasta, cookie, electronic nose.

Acknowledgements: This work was supported by a grant of the Romanian National Authority for Scientific Research and Innovation, CCCDI – UEFISCDI and the Ministry of Education, Science and Technological Development, Republic of Serbia, project number E!9936.

I4 - Analysis of the amylose content of starch from different gluten-free flours

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The starch molecule consists of 2 polymers of D-glucose: amylose (linear chain and completely amorphous) and amylopectin (branched chain and it is associated with the crystallinity of the starch granule). The amylose / amylopectin ratio is a characteristic of cereal starches that influence the final product by varying gelatinization, solubility, resistant starch formation and textural characteristics. The amylose / amylopectin ratio has an important role in bread making process. Amylose content influences the nutritional and technological properties of starch (i.e. gelling and pasting behaviour). Commonly, the quantification of amylose content is based on the iodine binding capacity of amylose (amylose/iodine complex) determined by potentiometric, amperometric or colourimetric measurements.

This study aims to investigate the amylose content of starch from different gluten-free flours. The amylose content was measured using an amylose/amylopectin assay kit (Megazyme, Wicklow, Ireland) and expressed as % of total starch. The method principle is based on the precipitation of amylopectin with lectin concanavalin A and its removal by centrifugation; while the amylose from the supernatant is enzymatically hydrolyzed (by amyloglucosidase/α-amylase enzyme mixture) to D-glucose and analyzed using glucose oxidase/peroxidase reagent through colourimetrical measurement.

The following gluten-free flour were analyzed: white rice, oat, millet, chickpea, maize, brown rice, buckwheat, plantain and tiger nut (all with origin from Hungary), teff (Germany) and quinoa (Ecuador). Tiger nut flour had the highest amylose content of 28% of total starch, followed by chickpea and teff flours (25%). The amylose percentage of total starch is in the same range for rice, millet and buckwheat flours (22%) and for oat, maize and plantain flours (20%). Brown rice had a lower percentage of amylose (13%), while quinoa flour showed the lowest amylose content of 6.7% of total starch compared to the other gluten-free flours. From the sample analyzed, tiger nut, chickpea and teff flours (high amylose content) are preferred for manufacturing of gluten-free dried pasta due to their tendency to retrogradate and ability to form a network that resists on cooking.

The measurement of the amylose content of starches is an important quality parameter for food processing.

Keywords: amylose, amylopectin, starch, gluten-free, flour.

Acknowledgements: This study was achieved through Core Program, with the support of the Ministry of Research and Innovation (MCI), contract 22N/2019 (project PN 19 02 02 02).

I5 - Characterization of Jerusalem artichoke as ingredient in bakery products

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Jerusalem artichoke (Helianthus tuberosus L.) has a high interest due to its complex biochemical composition that gives various beneficial effects on health. Jerusalem artichoke has an important functional potential given by its high content of inulin, minerals and amino acids. These represent an important group of biologically active compounds, which give to these tubers their therapeutical properties and functional potential. Numerous studies have shown that inulin provides a unique combination of interesting nutritional properties, it conferring functional role in human nutrition.

In this paper, Jerusalem artichoke – cultivated in Romania – was nutritional characterized in order to be used in bakery products (croquettes) development.

Jerusalem artichoke was dry and finely ground at low temperature. Total protein was analysed following Kjeldahl method. Inulin was determined by spectrophotometric method. The minerals was determined by atomic absorption spectrophotometer.

Jerusalem artichoke flour was chemical analyzed for its content in (d.m.): proteins (11.8-19.50%), ash (3.59-4.56%), lipids (0.30 -0.75%) and inulin. Inulin is the major component 52.55% - 63.10% (d.m). These data confirm that Jerusalem artichoke should be considered a source of interesting added value carbohydrate compounds, particularly inulin with potential known prebiotic properties, useful to formulate functional foods as well as nutraceuticals.

Jerusalem artichoke has high mineral content, among which the most important (expressed as mg/100g d.m.) are: Potassium, (2700 - 3300), Iron (17.00-19.50) and Magnesium (220 - 240). From the performed analyses regarding minerals content, it can be observed that Jerusalem artichoke represent a material having important minerals contents. 100 g of Jerusalem artichoke assures the daily intake for some of these elements according to the Reference Daily Intake (RDI) of macronutrients and micronutrients recommended by the FDA/2016. Specifically, 100 g of this ingredient contain more than the necessary daily intake of iron, a half of the daily demands of potassium and magnesium.

The chemical characterization performed in this study proved that the Jerusalem artichoke is a valuable source of nutritional components, mainly inulin and minerals and it is a potential functional ingredient in bakery products.

Keywords: Jerusalem artichoke, functional ingredient, inulin, mineral.

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I6 - HPLC/DAD Method for Determination of Flavonoids Rutin and Quercetin in Herbal Supplements

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Flavonoids are a large group of polyphenolic components possessing benzo-γ-pyronic structure and are widely distributed in plants. The chemical nature and the biological activity of flavonoids depends on the structural class to which they belong, the degree of hydroxylation, the degree of polymerization, and the presence of other substituents and bonds. *Ginkgo biloba* is one of the top-selling botanicals in the world. It has supported efficacy for the treatment of cerebrovascular disease and dementia. *Ginkgo biloba* leaf extract is said to contain more than 20 kinds of flavonoids. We have analyzed quercetin and rutin as representative flavonoids in the commercially available herbal supplements containing *Ginkgo biloba* leaf extract. HPLC analyses were performed using a Schimadzu LC-2010 chromatographic system (Schimadzu, Kyoto, Japan) consisting of a LC-20AT Prominence liquid chromatograph pump with a SPD-M20A Prominence Diode Array Detector. Chromatographic separation was performed on a Purospher® STAR RP-18e reversed-phase column (250 X 4.0 mm I.D.; particle size 5 µm) in a gradient mode with a mobile phase constituted of: acetonitrile: 3% phosphoric acid (85% phosphoric acid was used). The elution was carried out at a flow rate of 1.50 ml /min. All analyses were performed at room temperature (24 +/- 2°C). Rutin was monitored at 255 nm, while quercetin at 375 nm. Data analyses were done using Class VP 7.3 Software. The proposed method was validated according to the guidelines set by the International Conference on Harmonization for validation of analytical procedures. The identification of flavonoids was done by comparison of retention times of the analyzed components, their UV spectra and by standard addition method. Calibration curves were obtained using standard solutions of rutin and quercetin with concentrations ranged from 0.01 – 0.08 mg/ ml. Correlation coefficients were 1.0 and 0.9998 for rutin and quercetin, respectively. The precision of the method was confirmed by assessment of repeatability and reproducibility. Relative standard deviations obtained in the investigation of repeatability were: 0.52 % and 0.05 % for rutin and quercetin, respectively. Relative standard deviations obtained in the investigation of reproducibility were: 0.82 % and 0.95 % for rutin and quercetin, respectively. The average recovery for samples containing rutin and quercetin were 99.2 % and 101.2 % for rutin and quercetin, respectively. The limits of detection for rutin and quercetin were 0.95 ng/ml and 1.25 ng/ml, respectively, which indicates an excellent sensitivity of the proposed method. From the results presented, it can be concluded that this method is simple, easy to perform and specific for routine determination of flavonoids rutin and quercetin in herbal preparations containing *Ginkgo biloba* extract.

Key words: flavonoids, rutin, quercetin, *Ginkgo biloba*, HPLC, validation

J1 - Optimization and validation of quantitative TEM analysis of pristine titanium dioxide powders in a regulatory context

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Transmission electron microscopy is essential to implement the regulation of Belgian NanoRegistry, EFSA guidance2018 and Reach annexes on (nano)material characterization to provide precise and accurate data for the risk assessment of the nanotechnology.

To determine the physicochemical characteristics of pristine, powdered titanium dioxide materials, standard operating procedures for the analysis of the size and shape properties of the constituent particles and of aggregates/agglomerates were developed.

In an intra-laboratory validation study, the measurement of the size, shape and surface topology of primary particles and of the AA of titanium oxide powders by a combination of dispersion, specimen preparation, TEM imaging and image analysis were validated based on the analysis of JRCNM10200a and JRCNM10202a, provided by the Joint Research Center (JRC) repository.

The TEM imaging conditions, including the selection of images and the magnification, and the number of particles to be analyzed, were objectified to assure a high precision, accuracy and robustness of results. The lower and upper limits of detection and quantification (working range) were determined. A systematic random sampling strategy was applied to control the selectivity.

The ruggedness of the method against variation in the number of measured particles is evaluated by determining the measurement uncertainty from sub-datasets of measurements in function of the number of analyzed particles.

An uncertainty balance was set up to evaluate several sample preparation and imaging conditions. The selection of an optimal pH during sample preparation assured a high state of dispersion and was critical to precisely and accurately measure the AA and constituent particle properties. For the evaluated relatively pure materials, measurement of the zeta-potential was shown instrumental to determine the pH conditions were the dispersion was (un)stable. Probe sonication at 700 kJ/l was further applied to reach the most dispersed state, where image analysis was most precise.

The results of TEM analysis were compared to the results obtained by dynamic light scattering and particle tracking analysis.

Keywords: E171, characterization, titanium dioxide, electron microscopy, particle size distributions.

K1 - Shortening the traceability chain for Africa: purity assignment of organic calibrators

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High purity calibrants are essential for quantification of organic contaminants and residues in food and the environment; drugs and their metabolites in clinical and forensic chemistry. To support global measurement equivalence, it is necessary that these high purity substances be fully characterized, having measurement values assigned with traceability to SI units (typically the amount of substance (mol); mass fraction concentration in mg/g).

Purity can be determined either by direct assay of the main component using a primary method, or by the indirect approach “mass balance” approach. This involves the identification and quantification of the mass fraction of individual impurities/ minor components present in the material. The sum of impurities is subtracted from the total mass to obtain the mass fraction of the main component in the material.

Quantitative nuclear magnetic resonance spectroscopy (Q-NMR) is a rapid, quantitative primary-ratio analytical technique for quantifying the main component. The amount of time and material required to value assign, conduct homogeneity and stability studies over the duration of the material lifetime, made the establishment of this new capability at NMISA worthwhile.Since 2006, NMISA has participated in several international comparisons to benchmark purity assignment capabilities, that would allow NMISA to certify high purity calibrants for regional analytical laboratories in Africa. A mass balance approach has typically been applied, which involves various analytical techniques to identify and quantify all minor components. These currently include: GC-FID; GC-TOFMS; LC-(ESI/APCi/APPI)-MS/MS; LC-PDA; LC-CAD; DSC; TGA-FTIR; headspace-GC-TOFMS (residual solvent), proton NMR and Karl Fischer Coulometric moisture determinations. NMISA has also recently implemented Q-NMR for the purity assignment of mycotoxins: zearalenone and ochratoxin-A.

An overview of the value assignments performed by the NMISA on high purity substances such as theophylline (anti-asthmatic drug), 17□-estradiol (hormone), Aldrin (pesticide), L-valine (Amino acid) and folic acid (vitamin B9) and mycotoxins will be presented. The approach in obtaining the final measurement result and uncertainty contributions from each analytical technique is described. Final purity uncertainties range from 0.8-1.6% relative for organic calibrator purities of 980 mg/g to 1000 mg/g.

Keywords: Purity assignment, QNMR, metrological traceability, mycotoxins.

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