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International Symposium
Food Fraud Prevention and Effective Food Allergen Management
Vienna-Vösendorf, Austria
7-8 June 2018
MoniQA Association 2018, www.moniqa.org

Symposium overview

Following the great success of and extremely positive response received at the First International MoniQA Symposium on 'Food Fraud Prevention and Effective Food Allergen Management' in Bari, Italy, 26-27 January 2017, the MoniQA Association would like to invite you to mark your calendars for the Second International MoniQA Symposium on 'Food Fraud Prevention and Effective Food Allergen Management' in Vienna-Vösendorf, Austria. 7-8 June 2018.

Food fraud undermines consumer confidence and threatens food safety. According to the 2016 US Pharmacopeial Convention, economically motivated adulteration (EMA) is a global economic problem and a public health issue. It is estimated to cost the industry \$ 10-15 billion annually and up to 10% of the global food supply appear to be affected. Food fraud typically involves a wide range of intentional fraudulent activities, usually for economic reasons (addition of non-authentic substances or removal or replacement of authentic substances) or counterfeiting (production of substandard goods sold as premium brands).

An exciting programme with practical litigation cases, with strategies and methods for detecting and combating food fraud, are presented by renowned speakers from around the world. Join us in Vienna and meet the 'Food Detectives' and get to know modern technologies that are looking for the 'unknown' or learn about legal regulations and get practical advice for protecting your branded products, for correct food labelling food, and ultimately to maintain consumer trust. The symposium includes speakers from the United Nation's FAO/IAEA, IFS, LGC, USP, MoniQA, as well as industry representatives from Nestlé, SQTS, Imprint Analytics, and others, as well as from various law firms and food research institutions.

This Second International MoniQA Symposium on Food Fraud Prevention and Effective Food Allergen Management will address food authenticity, food fraud and the need for simple labelling as major drivers for both the food industry and companies involved in rapidly developing new analytical technologies. The food industry must adapt to this changing landscape and be aware of the regulatory and legal issues that drive the change in the safety and quality of food. This workshop will bring together food industry experts, legal experts, regulators, academics and nongovernmental organizations (NGOs) to discuss the latest information on the implications of regulations and other burning issues in the food space.

Some of the areas we plan to cover include:

- What is on a label: 'free from...', 'may contain...', 'processed in a facility...'
- Interpreting legal limits vs laboratory accuracy
- Litigation: class action vs consumer deception

Science and technology involved in the analysis of food for authenticity will be shared by decision makers, legal advisors, marketing experts and lawyers. Case studies of successful brand protection, food fraud prevention and improved risk management will be provided in this timely and innovative Symposium.

We warmly invite you to attend this meeting in Vienna, which brings together international experts in the fields of food authenticity and food allergens, as well as various food industries, SMEs, research institutions, associations and regulatory bodies, all having a different stake in food safety. This special mix of scientific and practical input to the symposium will be a valuable opportunity to grow your knowledge base, learn from practical experiences, and exchange ideas with peers. Vienna has earned an outstanding reputation as meeting destination and is ranked top every year in the statistics kept by the International Congress & Convention Association. The central location of Vienna in the heart of Europe is beneficial to all delegates who are able to reach Vienna easily with direct flights from any European capital as well as from many other cities worldwide.

Dr Richard Cantrill
President, Canada

Dr Roland Poms
Secretary General, Austria

The global perspective of food fraud

ORAL 1

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Food fraud – intentionally causing a mismatch between food product claims and food product characteristics – though not a new problem, has become increasingly important in recent years with the trend towards global supply chains. There is greater incentive for food fraud when demand and/or prices are high, and greater opportunity when complex supply chains are involved. Food fraud is generally economically driven and rarely has malicious motivation, but there is often a food safety aspect associated with adulteration of food commodities, e.g. adulteration of milk powder with melamine. Because of its nature, the scale of the problem is difficult to gauge accurately but estimates of economic losses are generally of the order of US\$ 50 billion per year, and food crime can damage the reputation of entire commodity sectors leading to barriers to international trade. Food fraud is, therefore, of major concern to both the food industry and consumers. Control of food fraud is a difficult task which must involve multiple stakeholders in both the public and private sectors, including regulators, food producers, processors and distributors, research institutes and analytical laboratories. The Joint FAO/IAEA Division works with member countries to help improve their food control systems, including control of food fraud. The Division currently runs international coordinated research projects and capacity building projects focusing on food authenticity in more than 50 developing countries and also interacts in EU projects such as FoodIntegrity and Authent-Net. The focus of the projects is mainly on the development and application of analytical methods for food authenticity and to underpin traceability mechanisms, with the objectives of increasing confidence that food commodities reaching local consumers, and those destined for international trade, are safe and authentic. The experience of working with many institutes in different countries, with diverse capabilities and needs, both highlights the difficulties in controlling food fraud globally and presents opportunities to harmonise approaches and build networks to help identify and address the key issues.

Food authenticity from an industry perspective

ORAL 2

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Several serious food fraud incidents in recent years have led to the recognition that the risk of economically motivated food adulteration is higher than previously acknowledged. In some cases, the consequences were more than economic: melamine affected hundreds of thousands of children in China and, taken together, the many recent food fraud incidents damaged consumer trust in food. The detection, prevention and deterrence of food fraud is now a major priority for regulatory authorities and manufacturers alike. It has also stimulated intense research activity and the development of commercial solutions including software, tracking tools and analytical tools to protect consumers and the integrity of supply chains. Criminology approaches can help identify drivers of systemic risks in the food chain and can help to target action in the event of such risks being expressed. Factors influencing the emergence of food fraud risks include the growing complexity of the global food supply; longer supply chains and more rapid distribution; fluctuations in commodity prices; and consumer demands. Systemic vulnerability analysis, targeted audits, and early warning tools are now in use in both the private and public sectors to detect and to manage food fraud risks. Some facilities are employing sophisticated analytical tools to distinguish between 'normal' and 'abnormal' fingerprints which enable follow up action on a targeted basis. Such approaches have been shown to assist greatly in early management of issues affording greater consumer protection. However, there remain gaps in global coverage and in consistency of use of such tools.

Food Profiling: the value to assure food authenticity rather than to investigate for food fraud – experiences from the Swiss market

ORAL 3

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It is well known that food fraud is since many years present. In the past such issues were identified by 'luck', which means certain contaminants were added to food and identified by a target analysis. However, this identification procedure is not very effective as mainly one has to have someone who provides the information. Also, quite often discussed in this matter is a kind of food profiling to get a systematic approach for identification of manipulations related to targets or more over for non-targets. This sounds perfect, but here we actually face more wishes than facts as limitations in analytical chemistry show up. Nevertheless, with modern high-resolution mass spectrometer it is possible to come a step closer to a potential solution. Food profiling might be a good procedure, but food fingerprinting – a complex picture of food – is even more challenging, but might lead at the end to more success, but definitely not yet. As this topic is for the moment too complex to handle for one lab and collaborations are just going start, it might be useful to look at other areas, how it is handled here. One of these areas is the topic of food contact materials. We have here the same situation of targets (additives known) and non-targets (NIAS – non-intentionally added substances). Analytically here the situation is lesser complex as mainly food simulants like solvents used as matrix. Nevertheless, it is a very good 'training' area how to set-up non-target analysis on the basis of high resolution mass spectrometry. This is actually ongoing, but also the next step is already on its way driven by the mineral oil discussion in food. As mineral oils do have a high chemical similarity to natural saturated and aromatic hydrocarbons, the discussion and the analytical tools are more and more developed to handle it in a food fingerprint was which means for a clear identification of a mineral oil contamination it is necessary to know the hydrocarbon background of the analysed food. These kinds of activities are actually ongoing and explained by some examples from the Swiss market as well from other markets.

Assuring food authenticity – a standards developer's perspective

ORAL 4

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Assuring food authenticity is a complex problem that includes the need for information integrity and communication. Consumers expect foods to contain all of the constituents that should be present, to not contain anything inappropriate or harmful, and to have accurate and complete labels. These same expectations apply to all the participants in the supply chain, from harvest and to final sale. The only way to meet these expectations is for everyone in the supply chain to have access to reliable information on what constitutes a food grade ingredient and on how to determine whether a particular sample of an ingredient is food grade. Historically, standards development organizations (such as the US Pharmacopeia which publishes the Foods Chemicals Codex) met this need by publishing identity and purity standards for individual ingredients. The Food Chemicals Codex is an example of an independent third-party source that contains standards for individual ingredients along with analytic methods and acceptance criteria that can be used to confirm ingredient identity and purity. These acceptance criteria are expressed as threshold values for specific analytic tests (i.e. 'not more than' or 'not less than' values). While this approach continues to be appropriate for many ingredients, future standards will need to address complex ingredients (e.g. spices and high value oils) where composition can vary depending on environmental, agricultural, and other unpredictable factors. This will create a need to share information such as sets of spectra that are not amenable to publication as traditional documents. These changes mean that standards development organizations, and the food industry, will need to develop a new understanding of what constitutes an ingredient standard, of how to ensure data integrity, and of how to communicate and use these new standards along the supply chain.

Honey authenticity: when official controls are questioned

ORAL 5

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Honey is a product with definitions and compositional standards in Codex and in European law. However, there are many means of adulterating honey, including the addition of cheap sugars and syrups after collection from hives, overfeeding bees with saccharides or invert derivatives and the falsification of the floral or geographical origin. Honey mislabelling and fraud is a global issue. In the face of media reports that more mānuka honey is sold worldwide than is produced in New Zealand a set of high level characteristics for mānuka-type honey were developed. How appropriate are these criteria and should official control laboratories outside New Zealand supplement them? In 2015 the European Commission organised a control plan to assess market prevalence of adulterated honey. Over 2,000 samples of honey were collected, some 38% of which were non-compliant with authenticity criteria. Honey authentication requires a multifaceted approach which can be costly and time consuming. As well as classical analysis $\delta^{13}\text{C}$ EA/LC-IRMS is required. But is this sufficient? Nuclear Magnetic Resonance (NMR) can provide quantitative data and molecular structural information on key components in honey with little sample preparation and over the last five years a small but significant literature has emerged on this approach. However, in 2015 the UK Food Standards Agency wrote to UK enforcement authorities to state ‘... 1H NMR ...screening method gives indicative results and does not definitively prove that added sugar is present ... no enforcement action should be taken in relation to the NMR results alone with regards to added sugar at the present time ...’ Why did FSA do this, what were the consequences? In January 2018 the Joint Research Centre of the European Commission at Geel held a Technical Round Table on Honey Authentication. I will discuss these issues illustrated with real-world case examples and suggest possible next steps.

Lawyers perspective: how technologies and blockchain could be used to mitigate the risk of lawsuits and recalls

ORAL 6

C. Varallo

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The presentation will examine some of the most recent developments and application of AI (artificial intelligence), IoT (internet of things) and blockchain in the food supply chain. Any kind of technology, indeed, should be understood, before being applied: benefits and vulnerabilities should be carefully considered and aspects like systems’ interoperability cannot be ignored. After a brief focus on blockchain, the presentation will examine the contribution that such technologies can offer to strengthen the supply chain and to add value to the quality of the data and the records retained by the company, especially in terms of usability in case of litigation.

R. Van Laack

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Food fraud, the economically-motivated intentional misrepresentation of the true identity or contents of a food, is a threat to consumer confidence and public health. Food fraud comes in various forms, including dilution or substitution of ingredients, use of undeclared ingredients, misrepresentation of nutritional value, use of fraudulent processing claims, and false country of origin claims. Although food fraud is not a new phenomenon, modern food processing and the globalization of the food industry have created the circumstances for increasing opportunities to commit food fraud. Combatting food fraud requires collaboration between governments, global trade organizations, manufacturers, and the public. Unless related to food safety, food fraud has not generally been a priority for regulators. Although the U.S. Food and Drug Administration's regulations implementing the Food Safety and Modernization Act specifically mention food fraud, they address food fraud only to the extent that the fraud is a safety concern. Compositional standards help prevent and combat fraud to the extent that such standards can be verified. Frequently, standards cannot be verified by testing. For example, processing claims, such as organic claims, and country of origin claims, cannot be tested and largely rely on record-keeping throughout the supply chain. The Agricultural Marketing Service (AMS) of the U.S. Department of Agriculture verified a number of marketing claims. Verification generally relies on a combination of auditing and testing. This presentation will discuss the AMS National Organic Program's approach to combatting organic food fraud. In the United States, private litigation by competitors and consumers is used frequently to combat alleged fraud. Competitors know the market and more easily recognize circumstances of possible fraud. Many consumer protection organizations are on the lookout for potential fraud situations. In addition, many state laws provide for monetary recovery in consumer class actions for food fraud. Examples of food fraud litigation include lawsuits regarding pomegranate products and cases regarding extra virgin olive oil. In some cases, private litigation has resulted in the development of standards, either by independent third parties or by federal agencies.

The failure of analytical tests

ORAL 8

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For various reasons, analytical testing may 'fail' to detect food fraud. First and foremost, understanding the areas where potential fraud can occur is necessary. Without such knowledge, one cannot determine what to test for. Oftentimes, fraud is committed with the knowledge of what companies generally test. For example, in knowing that protein is generally tested based on its nitrogen content, adding a nitrogenous substance to replace protein is a likely scenario of committing fraud successfully. In the case of processing claims, analytical testing generally will be insufficient to detect fraud because there are no (known) compositional differences to determine if the product is actually processed as represented. For analytical testing to be useful in detecting fraud, at least three requirements must be met: (1) A clearly defined standard related to the chemical composition of the authentic product has been established; (2) There is a known compositional difference between the authentic and fraudulent food; and (3) The analytical test must be validated. The failure of testing is illustrated in litigation against retailers that were selling herbal dietary supplements which allegedly did not contain what they were represented to contain. An investigation by the New York Attorney General analysed six herbal supplements sold by retailers. DNA testing of these supplements allegedly revealed that all of the retailers were selling a large percentage of supplements for which modern DNA barcode technology could not detect the labelled botanical substance. The Attorney General brought actions against several retailers for the allegedly fraudulent marketing of the herbal supplements. However, investigators had failed to consider that the extraction processes applied to botanicals could have removed or destroyed genetic material. Moreover, they had failed to consider the sensitivity of the method. A positive result of a 'contaminant' was misinterpreted as evidence that the contaminant was an ingredient. In the context of nutrition labelling, for example, when the price of whey was high, companies started to add free amino acids to increase the protein content in sports supplements. Also, the new nutrition labelling regulations in the United States make it impossible to rely on analytical testing alone to determine whether the dietary fibre and added sugar content are properly represented. As a result, third party testing for these nutrients may no longer be sufficient to detect fraud. It is concluded that analytical testing can play a useful role in detecting and combatting food fraud. However, on its own (without more information) analytical testing may well lead to erroneous conclusions. Increasingly, knowledge of the supply chain is essential to assure that the product is indeed what it is represented to be.

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S7

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Unlike microbial hazards, chemical hazards in foods cannot usually be controlled by processing. This means that control of chemical hazards in a prevention-based food safety system is focused on the supply chain. Most supply chain control plans ultimately depend on laboratory testing to ensure that ingredients meet identity and purity standards. In the past, this testing has targeted a limited number of specific components and contaminants identified through prior experience. However, the number of targets or concern has increased and is constantly evolving as supply chains become longer and more international, as awareness of food fraud increases, and in response to consumer and regulatory concerns. This complexity makes it impractical to test each batch of a food or ingredient for all potential substances of concern. In response to this complexity, the food industry is turning to the use of non-targeted methods to characterize foods or ingredients. The advantage of well-designed non-targeted methods is that they can indicate whether a particular sample is 'out of range' without needing to know why. The disadvantage of these methods is that they require a great deal of data to determine the expected 'range' under a variety of growing, harvesting, and handling practices. In addition, each sample test can generate significantly more data than presence-absence or threshold-based tests. The volumes of data involved and the need for standards on how to generate and use these data present unique challenges and opportunities for standard development organizations and for food manufacturers.

Method validation and reference materials to assure the reliability of analytical results

ORAL 4

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In recent decades, safety and economical concerns have been the main driving forces for the development of rapid methods as well as the birth of a multitude of companies providing these technologies to industry and government. The need for rapid intervention for managing product tampering, bio-terrorism, and food contamination outbreaks have also led to the development of faster methods. Rapid tests and in recent years also non-targeted methods help industries in determining the effectiveness of food safety measures (e.g. in hazard analysis of critical control points – HACCP), assessing the integrity of foods, raw materials and ingredients, legal compliance as well as achieving logistical and operational goals while saving time and investments in complex instruments and staff qualifications. In some cases, they also reduce costs. Other drivers for the development of rapid methods in manufacturing have been the small sample size and quantities, portability of test systems, national and international regulations, and the potential universal use. There is always a need, however, for the laboratory to be able to demonstrate that a particular method works, is fit for purpose, gives indeed equivalent results to the reference method and can be used with confidence by the user. The minimum requirements for the quality of an analytical method are assessed in a validation study that usually involves some 8-16 laboratories to offer at least 8 valid results for statistical analysis. Parameters that are typically assessed in a validation study are the limit of detection, the limit of quantitation, repeatability and reproducibility (variability of results within and between laboratories), accuracy, specificity, false/positive negatives, and others. Additional information that can be drawn from a validation study concerns robustness, the acceptability and the handling of the method in the hands of different operators, and the possible to be identified influences on the results in a routine setting. Necessary steps towards assuring the reliability of analytical results in any laboratory are the preferred use of validated methods, the use of reference materials if available, method verification and participation in proficiency tests, training, considering the requirements for laboratory accreditation and following Good Laboratory Practice. Reliable analytical results are the basis for appropriate decision-making processes concerning product safety and adequate food safety management measures.

AOAC standard method performance requirements (SMPR) for allergen detection methods

ORAL 5

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The Association of Official Analytical Communities (AOAC INTERNATIONAL) has initiated a process to develop Standards Methods Performance Requirements (SMPRs) applicable for food allergen methods using ELISA-based techniques. AOAC SMPRs describe the minimum recommended performance characteristics to be used during the evaluation of a method. The evaluation may be on-site verification, a single laboratory validation or a multi-site collaborative study. SMPRs are developed and adopted by AOAC stakeholder panels composed of representatives of industry, regulatory organizations, contract laboratories, test kits manufacturers and academic institutions. AOAC SMPRs are used by AOAC expert review panels in their evaluation of validation study data for methods being considered for 'Performance Tested Methods' or 'AOAC Official Methods of Analysis', and can be used as acceptance criteria for verification at user laboratories. This presentation will review the process leading up to the prioritization of allergen methods for which SMPRs have been developed by AOAC's International Stakeholder Panel for Alternative Methods over the course of the last 18 months. Initial results leading up to the SMPRs for egg and milk will be briefly discussed. Future endeavours for the development of SMPRs for tree nuts will be relying upon the ability of the food allergen community to develop agreed-upon reference materials for the selected tree-nut(s) of priority.

Precautionary allergen labelling: do we address the needs of the allergic consumer?

ORAL 6

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Food allergy is a relevant health issue and may induce mild reactions up to life threatening conditions. Since there is no immunotherapy available to date, avoidance of the incriminating food is the method of choice. In parallel to patient tailored diagnosis and respective dietary recommendations, allergen labelling help to allergic consumer to make their choices. However, meaningful labelling needs generally accepted threshold levels, knowledge and expertise in food production aiming at reducing the carry over effects of allergenic components. Also hidden allergens may pose a risk for highly sensitized patients. Therefore, the current practice and examples how to help the food allergic patient will be presented and open needs that need to be addressed will be discussed.

Free-from foods – what does it mean for allergens?

ORAL 7

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Individuals with both immunoglobulin E (IgE)- and non-IgE mediated food allergies have to practise food avoidance, usually life-long. Having access to foods which are do not contain allergens – so called ‘free-from’ foods – makes an important contribution to the diet of allergic consumers. There is currently no consensus as to what constitutes a ‘free-from’ food with regards IgE-mediated food allergies. As a consequence regulators and food manufacturers alike have to rely on analytical testing to demonstrate the absence of an allergen in a food product. Since the protein components of allergenic foods causes food allergic reactions, this is the fraction that analytical methods need to target using either antibody or more recently mass spectrometry-based methods of analysis. The lack of agreed reference doses which are considered safe for the majority of allergic consumers means it is unclear how sensitive test methods need to be, although dose distribution modelling can provide guidance with regards the levels of allergens that are unlikely to cause a reaction. Inter-laboratory comparisons of immunoassay test methods for foods such as milk, egg and peanut have shown wide variations in test method performance regarding sensitivity and reproducibility of results. This leaves the possibility that manufacturers and enforcement bodies may obtain conflicting test results. The development of appropriate certified reference materials for allergen analysis can be used to help reconcile some of these differences, especially for test methods showing reproducible and consistent differences. Mass spectrometry (MS) methods have much to offer as a complementary, confirmatory method to the currently favoured immunoassay test methods. Through the iFAAM project a multi-centre study was undertaken using a method for determination of peanut in chocolate dessert using mass spectrometry and comparing it to commercially available ELISA test kits. Results of the ring-trial were converted to allergenic protein to assess their ability to quantify allergens in a way that allows their use in risk assessment processes. The trial showed significant divergence in the ability of ELISA tests to quantify peanut allergens and demonstrated that MS has the potential to detect and quantify peanut protein at similar levels to ELISA. Refinement is still required to improve the reproducibility of analytical workflows, especially sample preparation procedures which can take account of diverse food processing procedures and the interference of food components, together with harmonised approaches to calculate reporting units. These will be taken forward in the recently EFSA-funded project ThRAAll.

Improved reference materials for gluten analysis

ORAL 8

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Products bearing a gluten-free label must not exceed the regulatory threshold of 20 mg gluten per kg of the product laid down in Codex Alimentarius Standard 118-1979. The most commonly applied analytical methods for gluten detection used to assess regulatory compliance are enzyme-linked immunosorbent assays (ELISA), but alternatives such as polymerase chain reaction and liquid chromatography-mass spectrometry are being developed. Gluten analysis poses several challenges, because gluten is a complex mixture of 100+ proteins with additional variations caused by genetic and environmental factors as well as food processing. Other points to consider are the selection of relevant target sequences, protein polymorphism, sample preparation and removal of interfering substances from the food matrix. Well-characterized reference materials are essential to help address these challenges, which is why an international consortium is working on identifying wheat cultivars that are representative for the multitude of wheats grown worldwide. Several aspects were discussed: (1) use of a single wheat cultivar or a mixture of several; (2) importance of geographic origin and growing conditions; (3) economic importance and availability; (4) assurance of long-term stability, especially in case of flour; (5) use of flour, incurred materials or isolated protein fractions. In this study selection criteria for representative wheat cultivars as basis for the development of a new reference material for gluten(-free) analysis were defined. Grains of wheat cultivars from different geographical origins were collected, milled into white flours and characterized for chemical composition, wet and dry gluten content, ELISA response using two different antibodies and protein composition assessed by gel-permeation and reversed-phase high performance liquid chromatography and polyacrylamide gel electrophoresis. Based on the results, qualitative and quantitative selection criteria were defined and five wheat cultivars from four continents were selected. These cultivars were further investigated, and two reference materials are suitable: the single cultivar Carberry and the blend of the five cultivars.

ORAL 9

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We live in exciting times! Many new technologies have recently seen the light of day. And a number of manufacturers are focused on consumer devices. While many are in the consumer electronics field (fitness smart watches, Amazon Echo, Google Home), some devices were specifically designed to analyse health and nutritional values of foods, and also if pesticides or allergens / gluten are present. The presentation will look at some of these developments, the benefits and the challenges of the new generation of consumer analytical devices. It will report about stakeholder work, involving food industry, consumer advocates, government and device manufacturers to develop guidelines for such devices.

ORAL 10

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About 2-5% of the adult population suffers from one or more food allergies. Amongst children these numbers seem to be twice as high. Although not a food allergy, celiac disease is often mentioned in the same context. It is estimated around 1% of the population suffers from this gluten triggered auto-immune disease. Extrapolating this number to the number of households, this would mean that the number of people affected by a food allergy (directly or indirectly) is probably 3-4 times higher. On top of that if people are asked if they think they have a food allergy up to 25-30% of the people answers positively. Food allergies have a significant socio-economic impact. Individuals suffering from a food allergy and their family or household members are facing several additional costs. In the case of emergencies and hospitalization additional healthcare expenditures are made. But beyond those direct costs, individuals with a food allergy face various indirect costs like loss in productivity and quality of life. To protect allergic consumers and to assure consumers are informed in a correct way about the presence of allergens in their food, labelling legislation is in place in a growing number of countries all around the world. These legislations reflect to some extent the local differences in occurrence of allergens but are almost all based on the so called 'Big 8' of food allergens (milk, egg, fish, crustacean, tree nuts, peanuts, wheat and soybeans). For gluten (or 'gluten-free') separate legislation is in place in many countries. In order to meet this legislation and customer requirements, food producers should have a solid allergen management in place. This means of course the food industry has to invest in quality assurance with respect to food allergens, e.g. testing, but also invest in production capacities (dedicated production line or at least an effective cleaning regime). Also sourcing of raw materials and monitoring of suppliers may lead to higher production costs. On the other hand, the increasing number of consumers interested to buy 'allergen-free' (or gluten-free) products is increasing rapidly. In fact, this is one the growth drivers in the food industry and offers new possibilities for the food industry. This presentation will give an overview of the socio economical aspects of food allergens from the consumers and the industry's point of view. Current and potential future tools in allergen management and food allergen testing and their pros and cons are discussed.

Allergen analysis: customer solution case studies

ORAL 11

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Accurate detection of allergens in foods has never been more important, with an ever-increasing allergic population and an emerging market for allergen-free foods. ELISA-based methods are the most widely used for detection of allergens in food, but there are a number of challenges involved with the testing of such a wide variety of food types and how best to interpret the results given by the assays. Presented are three case studies which demonstrate how particular challenges faced by different users can be overcome to make sure that immunoassay-based testing for food allergens can best suits their needs.

iFAAM: new tools for allergen risk assessment and management

ORAL 12

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Allergens unintentionally present in a food product e.g. through cross-contact, can pose a risk for food allergic consumers. Good guidance (e.g. UK FSA, FoodDrinkEurope) exists to guide companies on allergen risk assessment and management, but few practical and readily accessible tools are available to help apply this guidance operationally, particularly for small and medium enterprises. The iFAAM project (Integrated Approaches to Food Allergy and Allergen risk Management) set itself a major objective of developing a suite of tools to fill that gap. The project team thus developed an allergen tracking tool, together with a tiered risk assessment approach, the first phase of which was purposely designed to operate with minimal inputs from users both in terms of expertise and data. The suite was completed by a tool which helps to select risk mitigation measures based on needs and resources. The tools and their application will be described.

Survey on the occurrence of allergens on food-contact surfaces from school canteen kitchensPOSTER 1

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According to Regulation (EU) 1169/2011, restaurants and catering services have to inform about the presence of certain ingredients that can produce an allergic reaction. One of the sources of hidden allergens in food could come from the cross-contact with surfaces or utensils. In order to gain knowledge about the current situation in such kind of establishments, the occurrence of 3 main allergen residues (milk, egg and gluten) has been evaluated in food-contact surfaces from 50 school canteens during a period of two academic years (2014-2016).. These food-contact surfaces were selected and analysed in situ by using a rapid LFIA test during the visits to kitchens. Leftover sample was sent to a laboratory where an ELISA test was performed to confirm results. Out of 621 analysed surfaces (213 samples for milk and egg and 195 samples for gluten) none of them were found to contain milk with the rapid tests. However, the presence of egg and gluten was detected in 15% and 45% of the food-contact surfaces, respectively. The results obtained with ELISA showed also a low occurrence for milk (6%) but higher for egg (24%) and gluten (57%). It has to be highlighted that for some specific utensils the occurrence reached up to 40%. Food-contact surfaces were classified in three groups (A, B and C) according to the rate of allergen occurrence. Ladles, pans, slotted spoons, tongs and trays presented the higher rates of allergen residues both for egg and gluten. These food-contact surfaces were classified within the group B (26-40% of positive surfaces) for egg and in group A (more than 40% of positive surfaces) for gluten. In order to control such sources of contamination an analytical strategy should be established to assure the correct cleaning as well as of the lack of post-cleaning contaminations. Our results indicate that the current cleaning procedures in kitchens of school canteens are not effective to remove allergens from food-contact surfaces. Therefore, processes should be improved to reduce the risk of allergen contamination. Validation of cleaning processes and verification of its effectiveness after each cleaning should be demonstrated by using the suitable tools of analysis. The use of exclusive food-contact surfaces to avoid allergen cross-contact during the preparation or serving meals is not a guarantee of the absence of allergens. Results as the presented in this study could help to apply a rational approach to manage the risk of food allergens.

Determination of 59 potential allergens in fragrances by comprehensive GCxGC (qMS)POSTER 2

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In fragrance products several compounds can cause allergic reactions and therefore referred to as potential allergens. According to the European Scientific Committee on Consumer Safety the list was suggested to be extended in 2012. In this work we analysed 59 compounds with potential to cause allergic reaction. Comprehensive GCxGC is the method with highest chromatographic separation. Here we used a RXI35 60 m × 0.25 mm × 0.25 µm column in the first and a WAX 1.5 m × 0.15 mm × 0.15 in the second dimension, respectively. Thermal modulation was achieved using a ZX-1 modulator. Typical peak width at the base were around 300 msec. Due to this fact the mass spectrometric detector need to supply 33 to 50 scans/sec at a mass range suitable for the compounds of interest. Here we set the quadrupole MS to 40-340 u at 50 scans/sec resulting in a scanning speed of 20,000 u/sec. Spectra quality and intensity were not reduced at this high acquisition speed due the patented advanced scanning speed protocol (ASSP US6610979) of the GCMSQP2020. The modulation frequency was set to 7 Hz. The first GC oven was set to 60 °C, 0.5 min and 3 °C/min to 260 °C. To control wrap around the second column was placed into an extra GC oven and the temperature was set +30 °C relative to the first dimension. Calibrations were done between 2 and 100 ppm. Regression coefficients were R²>0.9999 for most of the allergens. Several perfumes were analysed and the results were compared with the perfume supplier data. All expected compounds were separated from the matrix in all samples and quantified. The resulting concentrations were within 6% compared to the reference data. Other matrix materials are under test and will be reported elsewhere. Qualitative and quantitative determination of the extended list of allergens can be done using high speed quadrupole acquisition in scan mode over a mass range difference of 300 u. Comprehensive GCxGCMS supplies the necessary selectivity for quantification.

Development of an ELISA technique to detect soy protein in severe thermal processed food POSTER 3

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Soybean and soy by-products are widely used in food industry as ingredients due to their technological and nutritional properties and competitive price. However, soybean is one of the 'big eight' foods that are believed to be responsible for 90% of all food allergies. These ingredients are included in food labelling regulation of different countries. Moreover, soy proteins represent a particular insidious source of hidden allergens due to contamination during shipping, storage, and processing as well as to inadequate cleaning of processing equipment. Food industry needs analytical methods to assure the efficacy of the measures applied to control the risk of unintentionally soy presence in their products, thus protecting allergenic consumers. ELISA test is a suitable tool to detect allergens in food although thermal processing represents a challenge. Heat treatment can denaturalize target proteins causing the destruction of epitopes which are recognized by the antibodies used in immunoassays. In this work, a sandwich ELISA technique has been developed to detect a thermostable fraction of glycinin. This protein is one of the most abundant seed storage proteins (19-23% of total soy proteins). An in-house validation of the prototype ELISA test has been performed following AOAC guidelines. The test showed a limit of detection of 0.15 ppm of glycinin and a limit of quantification of 0.22 ppm of glycinin. No cross reactivity was found over a panel of 39 food commodities. Three incurred model foods incurred with different levels of soy protein isolate were manufactured at a pilot plant. Three different thermal processes were applied: baking (bread), pasteurization (sausage) and sterilization (pâté). A level of 0.001% of soy isolate was detected in sausage and levels of 0.05% in bread and pâté. Repeatability of the test was determined by analysing ten independent dilutions for each food extraction obtaining values of the coefficient of variation (CV) ranging between 7.4 and 8.7%. The intra-assay reproducibility provided values between 3.8 and 13.9%. The inter-assay reproducibility, provided values between 10.3 and 21.3%. These three precision parameters were assessed in UHT milk spiked with 0.05% of a mixture of soy drinks, a mix of 0.05% of cookies made with cookies with and without soy and in sausages incurred with 0.005 and 0.01% of soy protein isolate. This validation showed that the test is suitable for detecting an allergenic soy protein even after severe thermal processes where other tests fail.

Characterization of amylase-trypsin inhibitors (ATIs) of different wheat genotypes by MALDI-TOF mass spectroscopy POSTER 4

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Non-coeliac wheat sensitivity (NCWS) is getting more and more attention and about 5-10% of the population in Europe is affected by this kind of wheat related disorder. Amylase-Trypsin Inhibitors (ATIs) of wheat were recently identified as main triggers for NCWS, whereas FODMAPs (Fermentable Oligo-Di-Monosaccharides and Polyols) as wheat fructans only seems to influence the symptoms. ATIs are low molecular weight proteins, which have a mass of 12-18 kDa. They are soluble in water and a mixture of chloroform/methanol as well. In a first screening different genotypes of wheat, diploid einkorns, tetraploid emmers, and hexaploid wheat cultivars including spelt wheats were examined in respect to their ATIs composition. ATIs were dissolved in water and mixture of chloroform/methanol, applied on a metal MALDI plate and measured after addition of sinnapic acid as matrix on a MALDI Biotyper from Bruker. For verification, an alpha-amylase inhibitor purchased from Sigma-Aldrich was used. The molecular weights of intact proteins were compared with data from databases and literature, which resulted in identification of several ATIs. Results revealed stronger differences between einkorn and modern bread wheat samples, whereas ATIs composition of tetraploid emmers and hexaploid wheats were more similar. Since the A genome of modern hexaploid wheats originate from *Triticum urartu*, found results seemed to be consequential. Furthermore, other studies reported occurrence of less ATIs in emmer and einkorn as well. However, measurement of accurate masses by MALDI-TOF mass spectroscopy is a suitable method for ATIs characterization in untreated wheat samples.

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Effective food allergen risk assessment and food allergen management are important to protect allergic consumers and to comply with allergen labelling regulations. Such approaches require reliable analytical tools for the detection of allergens in food. Both, reference methods and reference materials are urgently needed to assure the quality, reliability and comparability of analytical results obtained with different methods. Ensuring the correctness of analytical results is crucial to laboratories, since incorrect results may trigger decisions that can cause economic damage or pose a risk to public health. The quality of reference materials is critical for accuracy and comparability of analysis results. Reference materials must be sufficiently homogenous, stable and traceable. Usually extensive material characterization and testing for homogeneity and stability of the material precede the availability of reference materials. Ideally a certified reference material shall be used, which has been validated by accredited institutions and is subject to strict quality testing. The first validated reference materials for food allergen analysis are now available and can be ordered from MoniQA Association www.moniqa.org or from authorized distributors. The first set of materials includes testing materials for milk allergen analysis comprising a Positive Control (SMP-MQA 092014, characterized dried skim milk powder, validated protein content), Negative Control (BLANK-MQA 082015, based on a gluten free cookie), and 2 Incurred Materials: LOW-MQA 102016 (SMP incurred in gluten free cookies, milled, 10 ppm skim milk powder, validated concentration 3.5 ppm milk protein) and HIGH-MQA 082016 (SMP incurred in gluten free cookies, milled, 50 ppm skim milk powder, validated concentration 17.5 ppm milk protein). These materials are the outcome of an international initiative (since 2013) led by MoniQA Association that has liaised with the EU funded project iFAAM, the Prolamin Working Group, Health Canada, FARRP, Australia's Allergen Bureau (Vital), and others. Additional food allergen reference materials are in preparation.

Combining reduced gluten content with good rheological properties: a feasibility study POSTER 6

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The identification of wheat genotypes with low toxicity could represent a valid alternative for the prevention of wheat intolerance onset. Over the last years, great efforts have been undertaken to develop effective gluten detoxification strategies mostly based on enzymatic strategies, which, however, involve a simultaneous detrimental alteration of the technological properties. In this frame, obtaining low-gluten wheat products without affecting their rheological properties is still a challenging issue. In this contribution, we present an integrated approach encompassing both proteomic characterization and grains yield/quality evaluation for the identification of durum wheat genotypes combining potential lower toxicity/immunogenicity with satisfactory rheological properties. A preliminary profiling of gluten proteins was accomplished by immunoassay-based quantification and liquid chromatography coupled to UV detection focusing on the gliadin fraction as main responsible for immunoreactivity in celiac disease patients. In addition, complementary information about productivity-related traits and quali-quantitative characteristics were collected. The pool of data was statistically evaluated confirming that durum wheat breeding programs improved the pasta-making quality (gluten strength) without causing an increment of toxic epitopes towards CD patients. The selected genotypes boasting medium and strong gluten strength, all presented a significantly lower number of toxic epitopes compared to commercial semolina. In perspective, such genotypes could represent an innovative alternative for preventive and therapeutic wheat-based foods in genetically predisposed individuals who may develop CD after prolonged wheat or gluten consumption. The research was funded by the Ministry of Education, Universities and Research (MIUR-Italy), program SIR 2014 within the project titled 'S. Wheat Pro. – Proteomic characterization of Selected durum Wheat cultivars for PROduction of low toxicity-food products towards celiac disease patients (RBSI14QQ1W)'.

Classification and discrimination of Indonesia wild honey using ATR-FITR combined with multivariate analysis technique

POSTER 1

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Wild honeys in Indonesia are still widely believed to be good for health with high economic value. The honey is naturally produced by *Apis dorsata* bee in Indonesia is a non-timber product of forest. In this study, authentication analysis by classification and discrimination of attenuated total reflectance-fourier infrared spectrometry (ATR-FTIR) spectra was conducted on several wild honeys from various places in Indonesia (n=186) which then compared to adulterated honey contained commercial sugars of aren, coconut, and cane sugar at 10-50% concentration (n=57). Combination of spectra measurement at 4,000-650 cm⁻¹ with Chemometric technique by several multivariate analyses resulted in visualization of honey grouping, classification, and regression model that differentiate these honeys, both partial and overall. principle component analysis multivariate analysis was able to visualize the differentiation of adulterated honey from the authentic ones. Discriminant analysis, a supervised classification technique, was used to differentiate the fake from the authentic honey among those from various origins at wave number range of 1,800-650 cm⁻¹ with 89.55% accuracy and performance index of 90.9, 90.32-100% sensitivity, and 95. 70-100% specificity. Partial least squares analysis was used to build a model provided quantitative results of commercial sugars content in honey allegedly added during adulteration. Commercial sugars content in authentic honeys was below 10% with measurable R² of aren, coconut, and cane sugar of 0.9995, 0.9980 and 0.9998, respectively, with their predictive R² values of 0.9977, 0.9983 and 0.9946, respectively.

Food and beverage fraud prevention using isotope fingerprints

POSTER 2

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In this presentation 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 labelling 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 suffer from fraudulent activities that include incorrect labelling 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 labelled 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 labelled 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.

EA-IRMS: tracing the geographical origin of roasted and green coffee using isotope fingerprints

POSTER 3

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Coffee is one of the most popular beverages worldwide, sourced from different geographical regions and exported through a commercial chain that usually involves several intermediates. To ensure that coffee beans come from labelled locations, laboratories need an analytical solution, enabling to discriminate geographical origin, with a special emphasis on the country of origin. Roasted and green coffee beans have a fingerprint, a unique chemical signature that allows them to be identified: isotope fingerprints of carbon, nitrogen, sulphur, hydrogen and oxygen have been reliably used for origin, authenticity and product label claim verification. In this poster, we report isotope measurements from green and roasted coffee beans measured using the Thermo Scientific™ EA IsoLink™ IRMS System. These data illustrate how isotope fingerprints can determine the origin of coffee beans. Consequently, it is evident that isotope fingerprint approach helps support legislation on food integrity and labelling (EC Reg. No. 1169/2011) and product geographical indication/origin (EC Reg. No. 510/2006) and therefore, protect consumers and brands.

Isotope fingerprints: origin of tequila with GC coupled with isotope ratio MS

POSTER 4

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The blue agave (Agave tequilana Weber var. Azul) is a native plant of the Jalisco region in Mexico and is an important economic product that, by law, is the only one allowed to be used in the production of tequila. Globally, tequila is a popular alcoholic beverage, which has led to increasing demand and thus production, with a subsequent increase in export value to the Mexican economy. This provides for an opportunity of economically motivated fraud either by adulteration and mislabelling of original tequila or production of fake tequila. Gas chromatography/isotope ratio mass spectrometry provides a powerful tool for determining carbon, oxygen and hydrogen isotope fingerprints in beverages and food. Thermo Scientific™ TRACE™ 1310 GC coupled with Thermo Scientific™ GC IsoLink II™, Thermo Scientific™ ConFlo IV™ Universal Interface and a Thermo Scientific™ DELTA V™ isotope ratio mass spectrometer offers a solution for identifying the purity and adulteration of products. Biosynthesis of organic molecules in A. tequilana requires water that comes principally from rainfall. Therefore, oxygen isotope fingerprint of the A. tequilana plant, and local sugars used in mixed tequilas, is primarily given by the rainfall water in those regions and can provide a geographical tool for origin. Here we report carbon and oxygen isotope fingerprints from commercial tequila, sugar cane and the A. tequilana plant. Coupled $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$ values of ethanol allow differing the original branded mixed tequila from A. tequilana and sources of sugar (corn and cane). This indicates that mixed tequila can be clearly differentiated from pure tequila, which derives 100% from A. tequilana. In addition, it also shows the difference between A. tequilana, original mixed tequila and sugar sources, meaning that adulterated and mislabelled tequila can be differentiated from original tequila and original source ingredients.

Determination of origin and authenticity of fish by elemental and Sr-isotopic fingerprints POSTER 5

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Misrepresentation of the origin of food exploits known regional quality by substitution with products from cheaper production areas. EC No 1379/2013 regulates the labelling of the country of origin of fish and defines labelling requirements of fish from aquaculture. Conventional methods are unable to determine the time a fish has spent in a certain environment. However, the use of fish otoliths as a record of the ⁸⁷Sr/⁸⁶Sr and Sr/Ca ratios gives time resolved information. Otoliths record the uptake of Sr and Ca via the gills, whereas the distribution of these elements from food is less pronounced. The potential shift of the chemical information in otoliths for fish bred in aquaculture needs consideration. The otolith can only be used if the whole fish is available. Since the head is often removed for retail, a method was developed for filets and fish bones to assess if data can indicate the site of origin. However, in filets and bones the detailed spatial resolution of the otolith data is not accessible. Water, fish feed, fish otoliths, bones and filets as well as fish eggs were analysed by ICP-MS. Elemental composition was analysed by ICP quadrupole mass spectrometry, while the ⁸⁷Sr/⁸⁶Sr isotope ratio was determined by a multicollector sectorfield ICP-MS (MC ICP-SFMS). Spatially resolved data in fish otoliths were assessed by split stream laser ablation ICP-QMS/MC ICP-SFMS analysis. Sr/Ca and ⁸⁷Sr/⁸⁶Sr ratios in all investigated fish reflect the composition of the ambient water. The influence of feed on the resulting Sr isotope pattern in otoliths was determined to be around 20% in fish from aquacultures. The contribution of feed was corrected mathematically using isotope pattern deconvolution. The temporal resolution within an otolith assessed by LA-ICP-MS analysis is defined by the laser spot size and the monthly otolith accretion rate and results in about 2-4 months in salmonids. The temporal resolution of the data is different in other fish species with different otolith sizes and growth rates. However, it should be possible to determine if a fish has spent half of its life in the habitats provided as habitat of origin given that site specific information on the ⁸⁷Sr/⁸⁶Sr and Sr/Ca ratios are available. The information found in fish bones and filets are suited to establish the relation to the indicated habitat of origin, if water samples or reference samples are available.

Identification of animal species and their detection in food and feed by multiplex real-time PCR

POSTER 6

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Food adulteration arises when food distributors want to attract customers by low price and, put pressure on food producers to lower the price of their products. This situation leads to a replacement of expensive food ingredients with cheaper variants or to an addition of non-declared additives. Processed meat products may often be composed of meat from several species. The aim of this study was to develop simultaneous and sensitive detection of the majority of significant animal species used for the production of meat and dairy products by multiplex quantitative PCR (qPCR). Methodology for the identification of selected animal species (pig, cattle, chicken, sheep, goat, horse, buffalo, rabbit, donkey, turkey, duck, goose, ostrich, quail) was based on the qPCR amplification of polymorphous sequences of genomic DNA. The species selected were sorted out to the five logical multiplex qPCR assays. Unlike mitochondrial DNA, the genomic DNA approach allowed quantification of DNA in the sample according to a standard. The first-choice multiplex qPCR assays (i.e. each sample must be examined by this multiplex) were equipped with the plasmid internal amplification control (IAC), which was introduced to distinguish between truly negative and false negative (inhibited) samples. The efficiency of each multiplex qPCR assay was about 95% and the analytical detection limit was determined to be 0.25 ng of DNA per qPCR reaction. A combination of up to five fluorescent dyes (pentaplex) was selected in order to be applicable for the measurement on LightCycler[®] 480 Instrument. In conclusion, we have developed first-choice multiplex qPCR assays for the determination of the presence of pork, beef and chicken in animal-by products (food and feed) and multiplex qPCR assays for detection of the most frequently processed meat such as: pork, beef, chicken, sheep, horse, rabbit, buffalo, turkey, duck, goose, ostrich or quail. Both first-choice multiplex qPCR assays contained IAC. Remaining multiplex qPCR assays did not require to have IAC included as their conduction will always follow after the first choice multiplex qPCR assay for meat species identification. Quantification of each species according to the plasmid quantification gradient is planned in the future. Our method is fast, showing a high specificity and sensitivity and it is suitable for routine identification not only of raw tissue (raw materials), but also for technologically processed food and feed.

IsoPROTECT – towards a terrestrial and aquatic physicochemical landscape of Austria POSTER 7 for testing the provenance of food by citizen involvement

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The Citizen Science project 'IsoPROTECT: Protecting regional food production in Austria by isotopic and multi-elemental fingerprinting' aims at the establishment of a tool to verify the provenance of Austrian primary agricultural products. The tool is based on physicochemical distribution maps of chemical parameters of soils and waters across the major food producing areas in Austria. Currently, the chemical information includes the ⁸⁷Sr/⁸⁶Sr isotopes as well as the multielement pattern of the bioavailable fraction in soils and water of a part of the anticipated areas. The scientific concept is based on the fact that the chemical fingerprint of the soil and water is reflected by the Sr isotopic composition as well as the multielemental pattern. This fingerprint leaves a fingerprint in agricultural products grown on the respective soils or in fish bred in an according water. The assignment of agricultural products to the area or origin can be accomplished via the analysis of the fingerprint by methods based on inductively coupled plasma mass spectrometry. The major advantage of the provided tool is the fact, that these distribution maps can be applied independent of the type of plant or fish and can be considered as time-stable (which is in contrast to many database-based approaches). In addition to the major growing areas in Austria, water samples from the three most important fish farms in each federal state will complement an aquatic isoscape including also the major lakes for commercial fish production. Based on these terrestrial and aquatic physicochemical distribution maps, a statistical online tool for the assignment of food to its origin by measured chemical data will be developed. The prerequisite of a working tool is a comprehensive sample collection of soils and water covering the main food production regions in Austria. Therefore, regional producers, schools and interested citizens have been invited to participate as Citizen Scientists, register at the web-platform and collect samples and information ('crowdsourcing'). All necessary materials, instructions and tools for collaboration, are provided via an online platform. First results show distinct regions based on ⁸⁷Sr/⁸⁶Sr isotope ratios and elemental patterns in soils and water. As a major outcome of the project on a social level, producers are made aware of the existing opportunities to detect fraudulent products. The interest in participation is steadily increasing.

Authenticity and origin of sturgeon caviar – towards an European database for caviar POSTER 8 traceability

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Sturgeon caviar is one of the most expensive food commodities in the world. While aquaculture production of caviar is emerging, there is also a decrease in the population of wild sturgeons mainly due to over-exploitation, poaching, illegal trade and destruction of natural habitats. As a consequence, sturgeon caviar trade was put under regulation by the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES). These measures include legal caviar identification using a uniform labelling system. However, currently implemented measures to control label compliance are primarily based on administrative controls which have been vulnerable to fraud. Sound analytical methods based on isotopic and elemental fingerprints have high potential for the unambiguous verification of sturgeon caviar provenance fostering legal caviar trade and sustainable farming. In this pilot study, raw and salted caviar samples along with water, fish feed and salt, representing the main sources to the chemical composition of caviar, from seven sturgeon farms were analysed for their ⁸⁷Sr/⁸⁶Sr isotopic and elemental composition using multi-collector inductively coupled plasma mass spectrometry ICP-MS. We identified twelve environmental factors taken up by the sturgeon from fish farm water (⁸⁷Sr/⁸⁶Sr, Na, Mn, Cu, Mo, Fe/Ca) and feed (Mg, As, Rb, Mg/Ca, K/Ca, Co/Ca) allowing to discriminate raw and salted caviar according to its source. Salting altered the ⁸⁷Sr/⁸⁶Sr isotopic and Fe, Mn, and As elemental composition of samples depending on the elemental content of salt. Therefore, a mathematical procedure based on mixing model calculations was applied to correct the ⁸⁷Sr/⁸⁶Sr isotope ratio for the influence of salt as well as to retrieve the biogenic Sr isotopic composition of water. Raw and salted caviar samples were attributed to their corresponding fish farm of origin with 88 and 94% accuracy using multi-variate statistics. This study forms the basis for establishing a European caviar database for the verification of CITES label compliance. Further research will focus on the combination of the site specific environmental factors with light isotopes such as $\delta^{13}\text{C}$, $\delta^{15}\text{N}$ or $\delta^{18}\text{O}$ providing additional discrimination power.

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Food allergies are currently one of the major issues in the field of food safety. The development of methods based on liquid chromatography coupled to mass spectrometry (LC-MS) for the analysis of allergens is receiving increasing attention. Several multi-methods that follow a bottom-up proteomics approach have been published, most of which use multiple reaction monitoring or single-stage high-resolution mass spectrometry to target proteotypic marker peptides. A 'First Action Method' for allergens was reviewed and accepted by the AOAC Stakeholder Panel on Strategic Food Analysis Methods (SPSFAM). The method was reviewed against AOAC Standard Method Performance requirements (SMPR 2016.002) and published by New *et al.* The method demonstrated good recovery and repeatability, with an analytical range of 10-1000 mg/kg for all tested allergen commodities and was able to meet the minimum performance requirements of the SMPR. Nevertheless, the reproducibility between different laboratories was not tested. Corresponding standardization concepts are not yet available for the routine application of these methods in more than one laboratory. In order to meet the requirements of the official authorities responsible for food surveillance in Germany and Europe, it is requested to validate and to transfer these methods to the 'Official Collection of Methods of Analysis and Sampling (ASU)' and to CEN. Following an invitation from the Federal Office for Consumer Protection and Food Safety (BVL) on March 13, 2018, experts from relevant fields gathered to discuss application of new LC-MS based methods for the detection and quantification of peptides in the field of nut allergens and fish or plant species differentiation and to exchange their experiences. Within the framework of the first meeting, possible new topics with regard to the application and standardization of these methods were discussed. The aim of the newly founded working group 'Mass spectrometric analysis of proteins' is to create a general guideline for interlaboratory validation studies and to validate existing methods. The poster presents the design of experiments for pilot studies. The aim of these pilot studies, each with at least four laboratories participating, are the standardization of the sample preparation for mass spectrometric analyses and the evaluation of the results with different mass spectrometry equipment under a uniform procedure. The data collected for these pilot studies will also be used for the optimization of the search for useful marker peptides with high specificity and sensitivity.

***In silico* approach for identification of bioactive peptides in jack bean**

POSTER 2

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Jack bean is one of underutilized legume in Indonesia, although it contains high protein. Jack bean tempeh and fried jack bean are two common products using jack bean as a raw material in Indonesia. The protein in jack bean especially globular proteins such as concanavalin A, concanavalin B and canavalin can be hydrolysed to several bioactive peptides that can be beneficial for human health. Several functional properties of bioactive peptides are correlated to reduce the potency of hypertension and also as antioxidant. So far, there is a limited investigation for using *in silico* approach for evaluating several potential proteins in jack bean as precursors of bioactive peptides. The purpose of this research is evaluating several proteins in jack bean as precursors of ACE inhibitory and antioxidant bioactive peptides using *in silico* approach, and thus to establish the rationale for choosing the appropriate substrates proteins in preparing ACE inhibitory and antioxidant peptides. Based on our pre-preliminary results, we can conclude that specific protein from jack bean e.g. canavalin has potency as precursors of ACE Inhibitory and antioxidant bioactive peptides using *in silico* analysis.

Jack bean (*Canavalia ensiformis*) tempeh, a traditional fermented product from Indonesia

POSTER 3

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Tempeh is traditional food from Indonesia as an example of soybean solid-state fermentation. Fermentation will represent a valuable and cost-effective approach for food stabilization and also for nutritional improvement. Jack bean tempeh is one of local product in Indonesia. Its contain high protein that can contribute to human nutrition. During the fermentation, peptides in fermented legume products e.g. jack bean is either released by the hydrolysis of precursor of proteins during fermentation or produced by the microorganisms associated with fermentation. Individual microbial strains will also in the formation of specific bioactive peptides with respective health benefits depending on the sequence and composition of amino acids. Such bioactive peptides may act as regulatory compounds and exhibit bioactive properties such as antihypertensive, antimicrobial, antioxidant, antidiabetic and also anticancer activities. The objective of our research is to investigate the properties of jack bean tempeh during fermentation We fermented several formula of tempeh for 24, 36, 48, 60 h with two different packaging e.g. banana leaves and LDPE Plastic. After 24 h miselia of *Rhizopus* hasn't been formed., but it fully formed after 36 h fermentation for all formulated tempeh with two different packaging. The miselia started the change to form brownish after 36 hours fermentation.

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