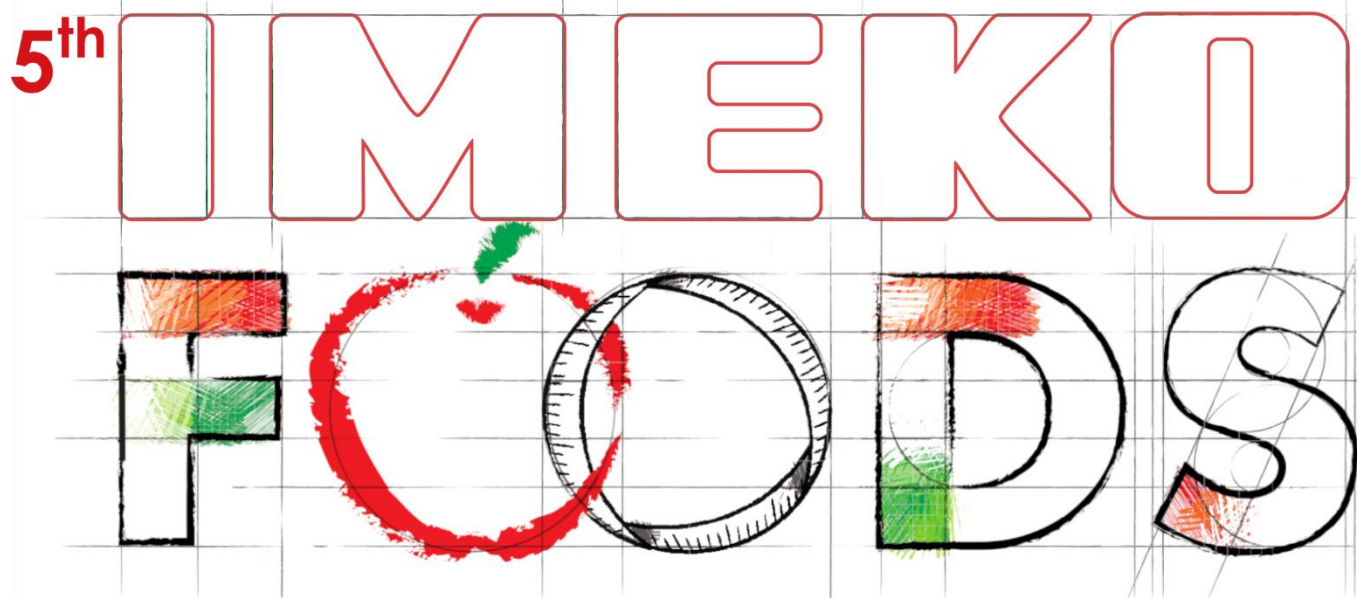


# BOOK OF ABSTRACTS

CORRECTED VERSION



Metrology for Sustainable Food Production

16-18 SEPTEMBER 2020 | PRAGUE | CZECH REPUBLIC



Czech University  
of Life Sciences Prague



5<sup>th</sup> international conference on metrology in food and nutrition

## **BOOK OF ABSTRACTS**

2<sup>nd</sup> version

5<sup>TH</sup> INTERNATIONAL CONFERENCE ON METROLOGY IN FOOD AND NUTRITION

16-18 SEPTEMBER 2020

CZECH UNIVERSITY OF LIFE SCIENCES PRAGUE | KAMÝČKÁ 129 | CZECH REPUBLIC

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# Welcome message

Dear conference participants,

When we took over the symbolic relay of the next IMEKOFOODS conference in Tervuren, Belgium, a year ago, we were looking forward to inviting participants to Prague as a beautiful metropolis of Europe, as well as to the campus of our university. But the coronavirus pandemic thwarted our plans. In the spring, when several organizers of symposia, conferences and seminars canceled their events, we were very much considering whether we would organize IMEKOFOODS at all. In the end, we decided not to cancel the conference, but to organize it in a hybrid way (with personal and online participation at the same time) so that we could respond flexibly to the current epidemiological situation.

At the beginning of the summer, we were a little worried about whether IMEKOFOODS would be interesting and attractive in such a way. In the end, reality exceeded our expectations. Thanks to everyone who disseminated information about the conference among their colleagues, about 120 participants from many countries of the world registered at the 5th IMEKOFOODS. The online form enabled the participation of those who could not arrive due to the long distance or time busyness. A reduced entry fee for online participants has also allowed many young researchers and doctoral students to participate. This great involvement of the young generation makes us very happy and enriches us with new perspectives and approaches to the scientific issues of metrology in food and nutrition.

Dear all participants, thank you for your interest and participation in the 5th IMEKOFOODS 2020. Thanks to all invited speakers for their very interesting contributions and willingness to raise the prestige of the conference. We thank the Czech University of Life Sciences Prague and the Faculty of Agrobiolgy, Food and Natural Resources for providing the rooms and equipment for organizing the event and all the sponsors who supported our event. I would also like to thank all of you who took part in organizing the conference, as well as IMEKO, namely TC 23.

It is a pity that we cannot show you round the picturesque places of Prague and the nice modern campus of our university, but I believe that IMEKOFOODS will bring you valuable and new information in the field of food and nutrition, as well as establishing contacts for your future scientific cooperation.

Welcome!



Lenka Kouřimská

Chair of the organizing committee



Czech University of Life Sciences Prague

**Faculty of Agrobiolgy,  
Food and Natural Resources**



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# Keynote speakers



## Paul Finglas (Quadram Institute)

### THE FOOD NUTRITION SECURITY CLOUD IN EUROPE

Head of the Food Database National Capability, and Research Leader in Food and Health. He is an experienced food and nutritional scientist on vitamins and bioactives in foods, their intake, absorption, metabolism and requirements. He is currently coordinating EuroFIR Nexus, and participating as a WPL and member of the management boards in PlantLibra and TDS, BaSeFoods, and MoniQA NoE.



## Selvarani Elahi (LGC Group)

### TECHNICAL DISPUTE RESOLUTION IN THE UK OFFICIAL FOOD CONTROL SYSTEM

Selvarani is the UK Deputy Government Chemist and Executive Director of the Food Authenticity Network at LGC. She has over 25 years' experience in the analysis of food and agriculture samples for authenticity, contaminants, additives, composition and nutrients, and has also managed teams delivering consumer product testing.



## Michael Sulyok (BOKU Vienna)

### PERFORMANCE OF LC-MS/MS BASED METHODS FOR THE SIMULTANEOUS DETERMINATION OF HUNDREDS OF COMPOUNDS DERIVING FROM DIFFERENT CONTAMINANT CLASSES

In his work there is a strong focus on method validation and participation in international interlaboratory comparison studies in order to characterize the performance of modern LC-MS based multi-analyte methods with minimal or even no sample clean-up which will be topic of his presentation.



## Ana Isabel Gomes Rito (INSA)

### THE WHO/ COSI EUROPE – THE ROLE OF SURVEILLANCE ON INVERSING TRENDS OF CHILDHOOD OBESITY IN PORTUGAL AND SOUTHERN EUROPE

Ana Rito has a PhD in “Nutrition and Public Health”. Since 2008 she undertook 2 Post-Doc at the NIH (INSA)-Portugal. She is the PI for Portugal, of the COSI/ WHO Europe study and a member of its Advisory Board. Collaborating at NIH -WHO Collaborating Center for Nutrition and Childhood Obesity.



## Eva Vlková (CZU Prague)

### PROBIOTICS AND PREBIOTICS IN HUMAN NUTRITION

Eva is a head of the Department of Microbiology, Nutrition and Dietetics at the CZU in Prague and a member of the Scientific committee of the Czech Association for Probiotics and Prebiotics. Her primary research interest is gastrointestinal microbiota and its modification by probiotics and/or prebiotics with focus on bifidobacteria, their application to food and feed and methods of their detection. She participated in the development of ISO/IDF standard for determination of bifidobacteria in food.



## Aleš Rajchl (UCT Prague)

### FOOD REFORMULATION – A CHALLENGE (NOT ONLY) FOR THE FOOD INDUSTRY

Aleš Rajchl is currently an Associate professor of Food Technology and a Head of the Department of Food Preservation at the University of Chemistry and Technology, Prague. His main research interest is focused on evaluation of food quality and authenticity and food analysis (ambient ionization mass spectrometry – DART).



## Reinhard Zeleny (European Commission, Joint Research Centre)

### RECENTLY DEVELOPED FOOD MATRIX CRMS FROM EC-JRC

PhD in food- and biotechnology (BOKU Vienna), fellow and temporary agent at EC-JRC-IRMM, reference material and research projects in the food area (allergens, food proximates, beef gender), senior post-doc at BOKU Vienna (glycobiology research work), now permanently employed at EC-JRC in Geel. Project responsible for food and biological reference materials and feasibility studies (veterinary drugs, food authenticity, food microbiology, biosecurity), past member of the Organic Analysis Working Group of the Consultative Committee for Amount of Substance.

5<sup>th</sup>

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# Program



## SESSIONS 17<sup>TH</sup> SEPTEMBER 2020

8:00-9:00 CEST

Registration and on-line participants connection

---

9:00-10:00

Welcome word of the symposium chair - **Kouřimská L**

Welcome word of the IMEKO nad TC23 chair - **Castanheira I**

Welcome word of the Czech National IMEKO - **Haasz V**

Ministry of Education, Youth and Sports CR and ESFRI representative **Vyšinka M**

Welcome word of the vice-dean of the Faculty of Agrobiolgy, Food and Natural Resources, Czech University of Life Sciences Prague **Tlustoš P**

### Session 1: Food safety, Food authenticity, Foodomics

Chair: **Lenka Kouřimská**

10:00-10:30

#### Keynote presentation:

Food safety – contaminants, trace elements, natural toxins

**Sulyok M**

---

10:30-11:00

#### Keynote presentation:

Technical dispute resolution in the UK Official Food Control System

**Elahi S**

---

11:00-11:20

Technical support for regional, PDO, PGI and TSG labels. The contribution of the distributed research infrastructure of the METROFOOD-RI – Greek node

**Tsimidou MZ, Tarantilis P, Trichopoulou A, Gerothanassis IP, Livieratos I, Kakoulides E**

---

11:20-11:40

Building up citizen awareness on food safety and sustainability through participation

**Di Giovanni B**

---

11:40-12:00

Combination of -omics strategies with traditional methods allows for comprehensive assessment of protective and adverse effect of  $\omega$ -3 polyunsaturated fatty acids in parenteral nutrition

**Kosek V, Bechyńska K, Heczko M, Dasková N, Cahová M, Hajslova J**

---

12:00-13:00 - Lunch break

## Session 2: Food security, Food authenticity

Chair: Pavel Klouček

13:00-13:30

### Keynote presentation:

The Food Nutrition Security Cloud in Europe

**Finglas P, Astley S, Korousic-Seljak B, Gibney E, Presser K, de la Cueva Gonzalez-Cotera J, Fillery-Travis A, Webb S, Frankte P**

---

13:30-13:50

Combined non traditional isotopes for quality, authenticity and high geographical resolution assessments of wines

**Donard FX**

---

13:50-14:10

Development of inorganic and isotopic analytical strategies for the geographical authentication of olive oil

**Nasr E, Epova E, Bérail S, Hammami M, Abderrazek H, Donard OFX**

---

14:10-14:30

Stable isotope ratios of herbs and spices commonly used as herbal infusions on the Italian market

**Khatri PK, Larcher R, Camin F, Ziller L, Tonon A, Nardin T, Bontempo L**

---

14:30-14:50

Laser photoacoustic spectroscopy for food fraud detection

**Fiorani L, Artuso F, Clai G, Giardina I, Lai A, Mannori S, Menicucci I, Nuvoli M, Pasquo A, Pistilli M, Pollastrone F, Puiu A**

---

14:50-15:15 - Coffee break



## Session 3: Public health, Food reformulation, Quality and safety of plant products

Chair: Eva Vlková

15:15-15:45

### Keynote presentation:

The WHO/ COSI Europe- the role of surveillance on inverting trends of childhood obesity in Portugal and Southern Europe

**Gomes Rito AI**

---

15:45-16:15

### Keynote presentation:

Food reformulation – a challenge (not only) for the food industry

**Rajchl A**

---

16:15-16:35

Quality assurance/quality control in cannabis products analysis

**Fenclova M, Benes F, Peukertova P, Binova Z, Hajslova J**

---

16:35-16:55

Essential oils in foods. Dead end or light at the end of the tunnel?

**Klouček P**

---

16:55-17:15

The use of tailored microbial consortia for enhancing crop yield and quality

**Bevivino A, Caldara M, Hett J, Passato S, Tabacchioni S, Frusciante S, Fiore A, Del Fiore A, Cantale C, Ambrosino P, Maestri E, Marmiroli N, Neuhoff N, Giovannetti G, Masoero G, Brunori A, Pihlanto A**

---

17:15-17:35

Improving crop plants growth and health with the help of microorganisms: SIMBA EU project

**Maestri E, Neuhoff D, Bevivino A, Passato S, Giovannetti G, Caldara M, Gullì M, Graziano S, Carlo S, Hett J, Tabacchioni S, Frusciante S, Fiore A, Del Fiore A, Ambrosino P, Giovannetti C, Masoero G, Brunori A, Pihlanto A, Marmiroli N**

---

18:30-22:00 - Social dinner

### Session 4: Food reference materials, Proficiency testing, Data handling in food samples

Chair: Paul Newman

8:30-9:00

**Keynote presentation:**

Recently developed food matrix CRMs from EC-JRC

**Zeleny R**

---

9:00-9:20

Open data in the context of FNS-Cloud and METROFOOD-RI

**Presser K**

---

9:20-9:40

Capacity development within METROFOOD RI Greek Node regarding food fraud. Chromatographic and spectroscopic expertise as a case study

**Ordoudi SA**

---

9:40-10:00

The first certified reference materials for somatic cell counting (SCC) in milk

**Zeleny R**

---

10:00-10:20 - Coffee break

## Session 5: Human nutrition

Chair: Aleš Rajchl

10:20-10:50

### Keynote presentation:

Probiotics and prebiotics in human nutrition

**Vlková E, Neužil Bunešová V, Killer J, Šubrtová Salmonová H, Musilová Š**

---

10:50-11:10

Research statistical study of various dietary supplements through measurements of their energy content

**Sapalidou G, Karampatzakis S, Kogia F, Angelidis G**

---

11:10-11:30

Nutritional features of Australian grown Feijoa (*Acca sellowiana*) fruit

**Phan ADT, Cozzolino D, Wright O, Rychlik M, Sultanbawa Y, Netzel M**

---

11:30-11:50

Personalised nutrition

**Žatečka L, Kouřimská L**

---

11:50-12:10

Natural food supplements from microalgae via supercritical CO2 extraction

**Marino T**

---

12:10-12:30

Nutritional value of edible insects and perspectives of their use as an alternative source of proteins

**Kouřimská L**

---

12:30-13:00

Best poster award

6th IMEKOFOODS announcement

Closing ceremony

---

13:00-14:00 - Lunch

---

13:00 - TC23 meeting – for TC23 members only

# Organizing committee

## **Lenka Kouřimská**

Chairman of the Organizing Committee, Department of Microbiology, Nutrition and Dietetics, FAFNR

## **Matěj Božik**

Department of Food Science, FAFNR

## **Laura Míčková**

Faculty Project Development Center

## **Lukáš Páček**

Faculty Project Development Center

## **Kateřina Jiralová**

Department of Food Science, FAFNR

## **Matyáš Krtička**

Department of Food Science, FAFNR

## Editor of the Event

## **Pavel Klouček**

Department of Food Science, FAFNR

# International program committee

**Nastasia Belc** (Bioresurse)

**Isabel Castanheira** (INSA)

**Nives Ogrinc** (IJS Environment)

**Karl Presser** (PREMOTEC)

**Michael Rychlik** (Technical University of Munich)

**Joris Van Loco** (Sciensano)

**Pavel Klouček** (CZU Praha)

**Claudia Zoani** (ENEA)

**Janaína Marques Rodrigues** (INMETRO)

**Maria Fernandes-Whaley** (NMISA)

**Kaoru Yoshida** (Sony Computer Science Laboratories)

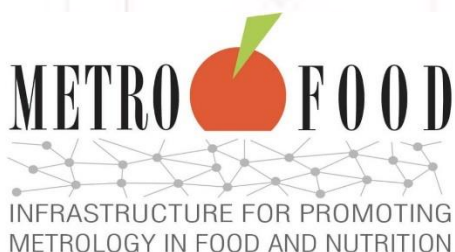
**Heather Greenfield** (TC23 Honorary Member)

**Maria Tsimidou** (Aristotle University of Thessaloniki)

# Support and sponsorship



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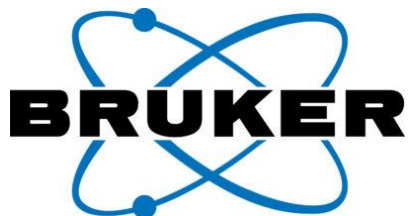


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# Oral presentations





# Technical support for regional, PDO, PGI and TSG labels. The contribution of the distributed research infrastructure of the METROFOOD-RI Greek node

Tsimidou, M.Z.<sup>1</sup>, Tarantilis, P.<sup>2</sup>, Trichopoulou, A.<sup>3</sup>, Gerothanassis, I.P.<sup>4</sup>, Livieratos, I.<sup>5</sup>, Kakoulides, E.<sup>6</sup>.

The Greek node partners: 1 ARISTOTLE UNIVERSITY OF THESSALONIKI (AUTH); 2 AGRICULTURAL UNIVERSITY OF ATHENS (AUA), 3HELLENIC HEALTH FOUNDATION (HHF), 4University OF IOANNINA (NMR CENTER-UoI), 5MEDITERRANEAN AGRONOMIC INSTITUTE OF CHANIA (MAICH), 6CHEMICAL METROLOGY LAB (EXHM/GCSL-EIM)

## AIM

METROFOOD-RI (<https://www.metrofood.eu/>) aims at providing high level metrology services in food and nutrition for the enhancement of food quality and safety at national, regional, European (ESFRI frame) and global level. During its preparatory phase, it will technically define how all the physical and electronic facilities will be made available in a coherent and effective way, as well as the organisation of the service provision. This will allow to effectively serve the very diversified and large user community, until the infrastructure full operation and exploitation. The overall working model of METROFOOD-RI as a pan-European distributed RI will be set up, determining the roles of the Central Hub and National Nodes and how the different components relate and interact. In this view, the Greek Node, organised in the Joint Research Unit METROFOOD-GR, makes efforts to organize itself at national level so that the involved research units and institutions to start co-operating and perform activities planned in a rather complementary way. The case study will exemplify the expertise and the services that can be provided by the Greek node regarding products bearing PDO, PGI, TSG and other regional labels. These labels are protected by intellectual property rights, specific EU regulations and are promoted worldwide as the tastes of Europe. The Greek node can support production, manufacturing, authentication, quality control and nutritional value of these products by means of its experimental fields and processing units (AUA, AUTH), high tech analytical laboratories and metrological expertise (NMR Center, EXHM, MAICH, AUA, AUTH) and available food composition databases (HHF). PDO & PGI virgin olive oils, table olives, honey, cheeses, yogurt, spices, wines, fruits, vinegar, traditional recipes are among those products for which the Greek node can provide technical services in its distributed installations that are spread all over Greece. Their expertise can be also extended to support new registrations for other products in Europe and third countries.

**Keywords:** METROFOOD-RI, Research Infrastructure, GREEK NODE, PDO, PGI, TSG, regional foods

**Contact person:** Tsimidou, M.Z. (tsimidou@chem.auth.gr)

## ACKNOWLEDGEMENT

METROFOOD-PP project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 871083.

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Kazou, M., Tzamourani, A., Panagou, E.Z., Tsakalidou, E. (2020), Unravelling the microbiota of natural black cv. Kalamata fermented olives through 16S and ITS metataxonomic analysis, *Microorganisms* 8(5),672

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Trichopoulou, A., Soukara, S., Vasilopoulou, E. (2007). Traditional foods: a science and society perspective, *Trends in Food Science & Technology* 18, 420e427

Tsimidou, M.Z. (ed). (2019) The PDO and PGI products of the olive tree (special issue). *EJLST*, 121 (3).



# Combination of -omics strategies with traditional methods allows for comprehensive assessment of protective and adverse effect of $\omega$ -3 polyunsaturated fatty acids in parenteral nutrition

Kosek V.<sup>1</sup>, Bechyńska K.<sup>1</sup>, Heczko M.<sup>2</sup>, Dasková N.<sup>2</sup>, Čáhová M.<sup>2</sup>, Hajslová J.<sup>1</sup>

<sup>1</sup> University of Chemistry and Technology, Prague, Czech Republic, <sup>2</sup> Institute of Clinical and Experimental Medicine, Czech Republic

## AIM

Parenteral nutrition (PN) is a type of nutrition where the nutrients get into the body intravenously contrary to normal route. It is the only way to bring nutrients to people who suffer from nutrient absorption failure such as patients with short bowel. This type of treatment bears a number of risks of which deterioration of liver function is one of the most problematic ones. In practice  $\omega$ -3 fatty acid containing lipids originating from fish oil are added to the PN to improve liver function. However, due to their multiple double bonds, they are suspected to contribute oxidative stress which is undesirable. In two performed studies, variety of analytical methods was employed to assess the effect of PN composition on rat plasma and liver. To assess the effect of long term PN, human plasma and erythrocytes were studied.

## MATERIAL AND METHODS

The methods used involved "classic" markers of inflammation and oxidative stress such as MDA analysis and ORAC assay, in addition lipidomics and proteomics were employed for better insight into PN related processes with better molecular resolution.

## CONCLUSIONS

In rat model, the composition of PN had great effect on the lipidome and "classic" markers. Because of short timescale of the experiment, increased oxidative stress was not evident, however, inflammation markers were reduced in  $\omega$ -3 PUFA administered rats. On the other hand, in humans, increased oxidative stress in  $\omega$ -3 administered patients was very evident and was demonstrated by a wide array of markers including plasmalogens, endogenous antioxidants which were reduced in this group of patients. However, the decrease of inflammation was demonstrated by lower levels of both traditional markers as well as of fatty acyl esters of hydroxylated fatty acids. It is evident that the effect of  $\omega$ -3 FAs is complicated and these new data could lead to better design of a new generation of PN lipid emulsions.

**Keywords:** parenteral nutrition, omics, lipidomics, rat model

**Contact person:** Vit Kosek, kosekv@vscht.cz

## ACKNOWLEDGMENT

This study was supported by the Ministry of Health of the Czech Republic, grants no. 15-28745A AZV CR. This work was also supported by the "Operational Programme Prague – Competitiveness" (CZ.2.16/3.1.00/21537 and CZ.2.16/3.1.00/24503) and the "National Programme of Sustainability I" - NPU I (LO1601- No.: MSMT-43760/2015)

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# The food nutrition security cloud in Europe

Finglas P.<sup>1,2</sup>, Astley, S.<sup>2</sup>, Korousic-Seljak, B.<sup>3</sup>, Gibney, E.<sup>4</sup>, Presser, K.<sup>5</sup>, de la Cueva Gonzalez-Cotera, J.<sup>6</sup>, Fillery-Travis, A.<sup>7</sup>, Webb, S.<sup>8</sup> and Frankte, P.<sup>9</sup> on behalf of the FNS Consortium

<sup>1</sup> Quadram Bioscience Institute, UK, <sup>2</sup> EuroFIR AISBL, BE, <sup>3</sup> Jozef Stefan Institute, SI, <sup>4</sup>University College Dublin, IE, <sup>5</sup>Premotec, CH, <sup>6</sup>Javier de la Cueva Gonzalez-Cotera, ES, <sup>7</sup>University of Wales Trinity Saint David Royal Charter, UK

## AIM

To overcome today's food and nutrition safety, security, and sustainability challenges, multi- and trans-disciplinary research questions need to be addressed and, increasingly, this requires combining and analysing data from different domains (e.g. food composition, food intake, food authenticity, nutritional epidemiology and metabolism). Food Nutrition Security Cloud (FNS-Cloud, GA No. 863059, [www.fns-cloud.eu](http://www.fns-cloud.eu)) aims to launch a first-generation 'food cloud', federating existing and emerging datasets and develop new services to support re-use by researchers.

Facilitating access (i.e. finding and having the abilities, permissions) and integration (tools and services) of data and its re-use (i.e. combining data) requires that resources from different domains are harmonised and standardised. These goals face several obstacles: each dataset uses different food classification and coding system; datasets are scattered globally, and compiled using different language, both human and machine, making integration problematic.

FNS-Cloud has developed an 'food nutrition security (FNS) data map', creating a structure to cluster FNS data and support formulation of research questions and identify their data needs, underpinned by FAIR (findable, accessible, interoperable and re-usable). Interoperability, however, demands that datasets from heterogeneous sources can be assembled, offering more than their individual worth, with minimal effort. Currently, systematic and semi-automatic curation of datasets, harmonising and structuring data against defined requirements and criteria, is lacking. Through a series of use cases and field studies, designed to collect and analyse different data types, FNS-Cloud will identify gaps, inconsistencies, and needs in these process and published criteria that can support interoperability and, ultimately, reuse of FNS datasets. Recommendations for implementing these criteria will be tested in through the development of Demonstrators, drawing together use cases and field studies with external datasets to address research questions covering agri-food (food traceability, food labelling and reformulation, and risk assessment), nutrition and lifestyle (food intake and consumer behaviour) and non-communicable diseases and the microbiome (healthy diet for a health microbiome, type 2 diabetes cohort data risk tool, and diet-food-drug interactions).

Currently, fragmented resources not only result in knowledge gaps that inhibit public health and agricultural policy, and the food industry from developing effective solutions, making production sustainable and consumption healthier, but also do not enable exploitation of FNS knowledge for the benefit of European citizens. FNS-Cloud will begin to overcome this fragmentation by integrating existing and emerging resources essential for high-quality, pan-European FNS research addressing food, nutrition, and health questions and lead to more sustainable agriculture, robust bioeconomies, and access to safe, affordable and nutritious foods.

**Keywords:** Food and nutrition security, FAIR Data, e-Infrastructure, web and information & database systems, information retrieval and digital libraries, and Cloud computing & services.

**Contact person:** Paul M Finglas, paul.finglas@quadram.ac.uk

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# Combined non-traditional isotopes for quality, authenticity and high geographical resolution assessments of wines

Donard O.F.X.<sup>a</sup>, Cellier R.<sup>b</sup>, Epova E.<sup>c</sup>, Barre J.<sup>c</sup>, Bérail S.<sup>a</sup>, Coelho I.<sup>d</sup>, Ors P.<sup>b</sup>

<sup>a</sup>Institut des Sciences Analytiques et de Physicochimie pour l'Environnement et les Matériaux CNRS / University of Pau & Pays Adour, UMR5254, Hélioparc, 2, Avenue du Président Angot, 64000 Pau Cedex 9, France, <sup>b</sup>MHCS Epernay 51200 France., <sup>c</sup>Advanced Isotopic Analysis, Hélioparc 2, Avenue du Président Angot, 64000 Pau, France, <sup>d</sup>Dep. of Food and Nutrition, National Institute of Health Doutor Ricardo Jorge, Portugal

## AIM

The authenticity of wines is of growing considerable importance on a worldwide basis. The geographical origin is now also of paramount importance and is now often related to the authenticity and quality of the products. These questions are addressed by a large variety of analytical parameters. We have currently developed the combined use of different isotopic systems of non-traditional isotopes such as of Sr, Pb or later B. Each of them translates into different origins of the elements and its isotopes under consideration. Both Sr and Pb isotopes are strongly related to the origin of the products. Boron is more related to the typology of the processes involved during the winemaking. We have applied this approach that reveals an unrepresented high geographical resolution to a large array of either red wines or sparkling wines including Champagne to samples coming from Europe, but also from other countries.

## MATERIAL AND METHODS

Wines from France, Italy, Spain, Slovenia, Portugal and China were investigated with this approach. The isotopic ratios of elements of interest were measured after purification on specific columns with the appropriated and selective resins by MC ICP/MS or High-Resolution MC ICP/MS.

## RESULTS

The use of chemometrics allowed to enhance discrimination for authenticity assessment. Then 3D diagrams allowed to clearly discriminate most of the different origins of the wines.

## CONCLUSIONS

Combined non-traditional isotopic systems provide an exceptional potential for high geographical resolution for wines addressing to the critical issues associated with authenticity.

**Keywords:** Sr, Pb and B isotopic systems, MC ICP-MS, geographical authentication

**Contact person:** Olivier F.X. Donard, olivier.donard@univ-pau.fr

# Development of inorganic and isotopic analytical strategies for the geographical authentication of olive oil

Emna Nasr <sup>1,2,3</sup>, Ekaterina Epova <sup>4</sup>, Sylvain Bérail <sup>3</sup>, Mohamed Hammami <sup>1</sup>, Houyem Abderrazek <sup>1</sup>, Olivier F.X Donard <sup>3</sup>

<sup>1</sup> Institut National de Recherche et d'Analyse Physico-chimique, Pôle Technologique de Sidi Thabet 2020, Tunisia, <sup>2</sup> Université de Tunis El Manar - Campus Universitaire Farhat Hached Tunis- ROMMANA 1068, Tunisia, <sup>3</sup> Institut des Sciences Analytiques et de Physicochimie pour l'Environnement et les Matériaux CNRS / University of Pau & Pays Adour, UMR5254, Hélioparc, 2, Avenue du Président Angot, 64000 Pau Cedex 9, France, <sup>4</sup> Advanced Isotopic Analysis, Hélioparc 2, Avenue du Président Angot, 64000 Pau, France

## AIM

Olive oil occupies a considerable place in worldwide consumption and more particularly in Mediterranean diet due to its many virtues. Its quality and taste are closely linked to production practices and geographical origin. Besides, it is among the foods most affected by fraud and counterfeits leading to economic repercussions and even affecting consumer's health. Therefore, this complex lipid matrix represents a focus for scientific research at the geographical traceability axis. The challenging aim of the present work is to develop a reliable analytical approach for authentication of Tunisian extra virgin olive oil (EVOO) known for its unrivaled quality.

## MATERIAL AND METHODS

Multi-elemental fingerprinting and Strontium isotopic composition ( $87\text{Sr}/86\text{Sr}$ ) have been adapted for the analysis of olive oil as proven tools for agrifood authentication 1,2. In this study, six Tunisian EVOOs were collected from oil mills in different regions of the country and two others were obtained from a supermarket. A sample preparation technique was enhanced to destroy organic matter in all the samples prior to quantifying low concentrations of trace elements by means of quadrupole ICP-MS. Then marketed foreign EVOOs from different geographical areas were analyzed in the same way in order to compare cross-country results. A multivariate analysis (PCA) was applied to the obtained results. Later the study focused on developing a method to extract Strontium from olive oil in order to measure the isotopic ratio  $87\text{Sr}/86\text{Sr}$  by Multicollector ICP-MS (MC ICP-MS) knowing that strontium is present at trace levels in a high organic matter matrix thus making its separation a critical step.

## RESULTS

PCA has demonstrated that Tunisian EVOO could be distinguished with a high probability from the samples of other origins. Besides, the Sr isotopic ratio, when detected, allowed authenticating each sample group by geographical area.

## CONCLUSIONS

Olive oil is a complex matrix that requires advanced pretreatments prior to analysis. Multielemental profile combined with Strontium isotopic signature has proven to be a reliable tool for distinction between olive oil samples from different geographic areas. This preliminary study has provided promising results for the geographical authentication of olive oil.

**Keywords:** olive oil; trace element; ICP-MS;  $87\text{Sr}/86\text{Sr}$ ; MC ICP-MS; geographical authentication



**Contact person** : Emna Nasr, emna.nasr@univ-pau.fr

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<http://dx.doi.org/10.1016/j.foodchem.2014.08.121>



# Stable isotope ratios of herbs and spices commonly used as herbal infusions on the Italian market

Khatri P.K<sup>1,2</sup>, Larcher R<sup>2,3</sup>, Camin F.<sup>1,2</sup>, Ziller L.<sup>1</sup>, Tonon A.<sup>1</sup>, Nardin T.<sup>3</sup>, Bontempo L.<sup>1</sup>

<sup>1</sup> Department of Food Quality and Nutrition, Research and Innovation Centre, Fondazione Edmund Mach (FEM), Via E. Mach 1, 38010 San Michele all'Adige, Italy, <sup>2</sup> Center Agriculture Food Environment (C3A), University of Trento, via Mach 1, 38010 San Michele all'Adige (TN), Italy, <sup>3</sup> Experiment and Technological Services Department, Technology Transfer Centre, Fondazione Edmund Mach (FEM), Via E. Mach 1, 38010 San Michele all'Adige, Italy

## AIM

Spices and herbs are one of the top adulterated food commodities due to their complex supply chain and high price. Traceability and authenticity of spices and herbs have been investigated only in a limited number of plant species. Stable isotope ratio analysis has widely been used to verify the authenticity of various food commodities. However, stable isotope profiles of spices and herbs have been not extensively explored and have been limited to few plant species. The aim of this study is to start exploring and determining the characteristic ranges of values for stable isotope ratios of spices and herbs, focusing in particular on those commonly available on the Italian market.

## MATERIAL AND METHODS

One-hundred-sixteen plant species belonging to sixty plant families were purchased from different physical shops and online stores in Italy (Nardin et al., 2018). Plant samples were dried, ground and homogenized for stable isotope ratio analysis. Stable isotope ratios of carbon, nitrogen, sulphur, oxygen and hydrogen ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ,  $\delta^{34}\text{S}$ ,  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$ ) of the plant samples were analysed by Isotope Ratio Mass Spectrometry (IRMS) coupled with an Elemental Analyser (EA) or with a High Temperature Pyrolyser (TC/EA).

## RESULTS AND CONCLUSIONS

For the first time the characteristic ranges of values of the stable isotope ratios of the bio-elements as a whole ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ,  $\delta^{18}\text{O}$  and  $\delta^{34}\text{S}$ ) were determined in spices and herbs collected on the Italian market and here presented. These first results are encouraging, and further investigations are recommended involving a more extensive sampling and focusing on specific plant species.

**Keywords:** stable isotopes, herbs and spices, origin and authenticity

**Contact person:** Purna Kumar Khatri, purnakumar.khatri@unitn.it

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# Laser photoacoustic spectroscopy for food fraud detection

Luca Fiorani, Florinda Artuso, Giulia Clai, Isabella Giardina, Antonia Lai, Simone Mannori, Ivano Menicucci, Marcello Nuvoli, Alessandra Pasquo, Marco Pistilli, Fabio Pollastrone, Adriana Puiu

Nuclear Fusion and Safety Technologies Department (FSN) of the Italian National Agency for New Technologies, Energy and Sustainable Economic Development (ENEA)

## AIM

The global food market is huge (hundreds of billions of dollars). It has been estimated that “seasonings and spices”, only one of its sectors, reached about \$ 17 billion at the end of 2019. Consequently, also the world market of analytical tests for food safety is great: by 2022 it is expected to exceed \$ 18 billion.

Laser photoacoustic spectroscopy (LPAS) has been applied for years to food safety by the Diagnostics and Metrology Laboratory (FSN-TECFIS-DIM) of the Italian National Agency for New Technologies, Energy and Sustainable Economic Development (ENEA). CO<sub>2</sub> lasers were used in the first prototypes because these sources are discretely tunable from 9 to 11 μm, just inside the fingerprint region, thus allowing the detection of organic compounds.

The advent of quantum cascade lasers (QCLs) provided us with sources in the fingerprint region that can be deployed in the field, because significantly smaller. Although their power is still lower than CO<sub>2</sub> lasers, their tunability is continuous and broader, thus allowing to record spectra that provide more information on the sample.

For this reason, we developed a QCL based photoacoustic spectrometer deployable in the field, having in mind that control authorities that routinely verify food quality will greatly benefit from a portable and user-friendly system.

The food sample is inserted in a photoacoustic cell where it is irradiated by the continuous wave emitted by the QCL chopped at an audio frequency. The sample absorbs the radiation, with the consequent temperature increase, adiabatic expansion and simultaneous generation of a pressure wave that is amplified by acoustic resonance and detected by a sensitive microphone coupled with a lock-in amplifier. A power meter monitors a small part of the laser beam coming from partial reflection by a beam splitter.

Fast detection of adulterated saffron will be discussed in this paper as a case study. Saffron is the most expensive spice, costing about \$ 20 per g. The authentic spice is part of *Crocus sativus*, commonly known as saffron crocus, a flower bearing three stigmas that are collected and dried. Frequent falsifications consist in adding flower fragments from other species to stigmas of *Crocus sativus*. Ground saffron is often cut with turmeric powder, a yellow spice obtained from the rhizome of *Curcuma longa*, a flowering plant known as turmeric.

**Keywords:** QCL application, Laser spectroscopy, Photoacoustic technique, Agrifood chain, Food fraud

# Food reformulation – a challenge (not only) for the food industry

Rajchl A.<sup>1</sup>

<sup>1</sup> University of Chemistry and Technology, Prague, Department of Food Preservation

## AIM

The main goal of the food industry is to provide enough safe and quality food for a wide range of consumers. The industry is responding to the increased interest in a healthy lifestyle through a number of measures, in which changes to recipes – *reformulation* – are a key part. Reformulation is meant to be positive, meaning that it should offer the consumer some benefits, especially with regard to nutrition, while considering the current state of knowledge about nutrition. Given the current rising trend of overweight and obesity and related diseases, it is expected that interest in food reformulation will increase. Food producers are responding to these problems by changing the formulations of their products, but any recipe change is a demanding process involving not only the production of the reformulated product itself, but also re-evaluation of the product's shelf life or marketing studies to determine whether the product will be accepted by the consumer. When changing food product formulations, technological procedures must often be optimized so that the product can be produced, which can often result in the need to make investments in production equipment.

**Keywords:** salt, sugar, obesity, non-communicable diseases

**Contact person:** Assoc. prof. Aleš Rajchl, PhD, ales.rajchl@vscht.cz, UCT Prague, Department of Food Preservation, Technická 5, 166 28 Prague 6 - Dejvice

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# Quality assurance / quality control in cannabis products analysis

Fenclova M.<sup>1</sup>, Benes F.<sup>1</sup>, Peukertova P.<sup>1</sup>, Binova Z.<sup>1</sup>, Hajslova J.<sup>1</sup>

<sup>1</sup> Department of Food Analysis and Nutrition, University of Chemistry and Technology Prague, Technická 3, 16628 Prague 6, Czech Republic; marie.fenclova@vscht.cz

## AIM

The cannabis and products thereof have long time stated apart from the official control and regulation. As cannabidiol (CBD), the main cannabis component believed to be associated with health benefits, has recently been approved in a number of countries worldwide, the remarkable amount of various cannabis-based products has been placed on the market. This situation raised an urgent demand for quality and correct potency control of cannabis products and resulted in a number of laboratories providing such services. However, the inter-laboratory variability in results is often fairly large. This is mainly due to the high complexity and variability of the cannabis matrix, use of analytical methodologies not always suitable for the purpose and not properly validated, together with the lack of tools for analytical quality assurance and control (QA/QC), and standardization. Currently, neither the certified matrix reference materials (CRM) as important QA/QC tools, nor the proficiency tests (PT) for external quality assessment of generated data are available in Europe, and those provided by US seem to be rather disputable. We present here the experience with matrix CRM and PT organized this year for cannabis potency by US provider, and our approach employed for cannabis-based matrix reference material preparation.

## MATERIAL AND METHODS

The matrix CRM (oil) and PT (hemp bud) for phytocannabinoids analysis were ordered from US provider. The material was analyzed using ultra-performance liquid chromatography and high-resolution tandem mass spectrometry method for quantification of 17 phytocannabinoids. As an internal reference material, cannabis flowers and full spectrum CBD oil were prepared at various levels of target phytocannabinoids. Both the homogeneity and stability of the reference materials have been controlled by the above-mentioned method.

## CONCLUSIONS

We highlight the importance of the appropriate tools for QA/QC in the analysis of cannabis products and propose an approach for preparation of reliable materials with potential to be used as matrix CRM and for PT organization. Without strict adhering on QA/QC principles, not only in phytocannabinoids analysis, but also in the control of possible contaminants, the poor reliability of generated laboratory data may increase the risk of placing hazardous cannabis products on a global market.

**Keywords:** cannabis, phytocannabinoids, QA/QC, matrix reference material, proficiency test

**Contact person:** Marie Fenclova, marie.fenclova@vscht.cz

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# Essential oils in foods. Dead end or light at the end of the tunnel?

Pavel Klouček<sup>1</sup>

<sup>1</sup>Department of Food Science, Czech University of Life Sciences Prague, CZ – 165 21 Prague 6, Czech Republic

## AIM

Historically, essential oils were of tremendous importance in form of natural food preservatives and/or spices. What is the situation today? Could they be once again used as natural preservatives in food production chain?

The antimicrobial activity of EOs has been extensively studied and demonstrated against a number of microorganisms, mostly in vitro and recently also in model samples. Yet, their commercial applications are rather scarce. This could be due to their high hydrophobicity, volatility, and intensive impact on sensory properties, and this limits their application in a realistic context. Deeper knowledge of their mechanism of action leads to the improved ways of EO application. There are attempts to utilize them in a vapour phase, as a part of modified atmosphere packaging, incorporated into the packaging materials, or formulated by different encapsulation methods. The number of products based on EO is slowly increasing mainly in primary agricultural production, while the food sector is still waiting for some major breakthrough.

**Keywords:** essential oils, food preservation, food safety, foodborne pathogens

**Contact person:** Pavel Klouček, [kloucek@af.czu.cz](mailto:kloucek@af.czu.cz)

# Improving crop plants growth and health with the help of microorganisms: SIMBA EU project

Maestri E<sup>1</sup>, Neuhoff D<sup>2</sup>, Bevivino A<sup>3</sup>, Passato S<sup>4</sup>, Giovannetti G<sup>5</sup>, Caldara M<sup>1</sup>, Gullì M<sup>1</sup>, Graziano S<sup>1</sup>, Carlo S<sup>1</sup>, Hett J<sup>2</sup>, Tabacchioni S<sup>3</sup>, Frusciante S<sup>3</sup>, Fiore A<sup>3</sup>, Del Fiore A<sup>3</sup>, Ambrosino P<sup>4</sup>, Giovannetti C<sup>5</sup>, Masoero G<sup>5</sup>, Brunori A<sup>3</sup>, Pihlanto A<sup>6</sup>, Marmioli N<sup>1</sup>

<sup>1</sup>SITEIA.PARMA, Interdepartmental Centre for Food Safety, Technologies and Innovation for Agri-food - Department of Chemistry, Life Sciences and Environmental Sustainability, Parma, Italy, <sup>2</sup>Institute of Crop Science and Resource Conservation, Dept. Agroecology & Organic Farming Rheinische Friedrich-Wilhelms-Universität Bonn, Germany, <sup>3</sup> ENEA, Italian National Agency for New Technologies, Energy and Sustainable Economic Development, Department of Sustainability, Via Anguillarese 301, Rome, Italy, <sup>4</sup>AGRIGES srl, San Salvatore Telesino (BN), Italy, <sup>5</sup>Centro Colture Sperimentali, CCS-Aosta S.r.l., Quart, Italy, <sup>6</sup>SIMBA COORDINATOR - Natural Resources Institute Finland (Luke), Myllytie 1, FI31600 Jokioinen, Helsinki, Finland

## AIM

An intensive agriculture is still required to satisfy the needs of a large human population. The current crisis due to the COVID-19 pandemics has evidenced even more the strategic role of food production for a safe and secure sustainment of citizens. However, reliable food production can no longer be achieved maintaining the same agricultural practices which damage the environment, and this had been reported in the EU document “From farm to fork” (EC 2020). A more sustainable agriculture is needed (Janni et al. 2020). Greenhouse and field experiments are ongoing in the Horizon2020 SIMBA project to verify the beneficial effects of microbial consortia on crop plants of relevance for EU agriculture: maize, tomato, wheat. Beside the role of microbes, amendments and carriers were tested, and in particular biochar has been considered as a way of returning to the soils the carbon formerly trapped in plant residues, and at the same time providing a “niche” for the growth of microorganisms.

## MATERIAL AND METHODS

In Italy and Germany greenhouse trials were carried out growing plants in pots and in the presence of microbial consortia developed within the SIMBA project as biostimulants (see Bevivino et al., this conference) and as alternative to chemical fertilizers. Consortia were delivered as powder, as liquid suspension or entrapped in wood biochar (Marmioli et al. 2018). Consortia composed of bacteria and fungi were compared with commercial products (MICOSAT F) and mycorrhizae. Cultivars of wheat and maize were selected to allow growth in both countries. The same cultivars were then tested in field trials starting from spring 2020: wheat Svevo and Bramante in Italy, maize DKC6587 in both countries, tomato H1648, H1766, Sailor in Italy.

In all experiments, soil analysis addressed to detect the main physico-chemical parameters, the degree of mycorrhization, the persistence of microorganisms, were performed. Plant growth has been estimated with non-destructive measurements of transpiration and photosynthesis efficiency (SPAD) and measurement of biomass, height and vegetative growth. Yield for grain and fruits, and quality parameters, are currently under evaluation and results will be presented during the conference. For grain cereals, the protein amount and gluten content are analysed.

## CONCLUSIONS

In the first year of the SIMBA project, different crop plants have been challenged with microbial consortia designed to include bacteria and fungi with different beneficial properties. Results obtained showed significant effects of microbes and biochar combinations on plant growth and



development. Changes in quality of the food products are under consideration with molecular methods.

**Keywords:** biochar; *Solanum lycopersicum*; sustainability; *Triticum aestivum*; *Zea mays*;

**Contact person:** Nelson Marmiroli, nelson.marmiroli@unipr.it

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FOODS

# Open data in the context of FNS-Cloud and METROFOOD-RI

Presser K.<sup>1</sup>, Matuszczak A.<sup>2</sup>, Kapela W.<sup>2</sup>, Zoani C.<sup>3</sup>, Finglas P.<sup>4,5</sup>, Astley, S.<sup>5</sup>, Koroušič Seljak B.<sup>6</sup>

<sup>1</sup> Premotec GmbH, Winterthur, Switzerland, <sup>2</sup> Premotec Poland sp. z o.o., Krakow, Poland, <sup>3</sup> Italian National Agency for New Technologies, Energy and Sustainable Economic, Development, Dept. for Sustainability - Biotechnologies and AgroIndustry Division (SSPTBIOAG), Rome, Italy, <sup>4</sup> Food Databanks National Capability, Quadram Institute Bioscience, Norwich, UK, <sup>5</sup> EuroFIR AISBL, Brussels, Belgium, <sup>6</sup> Computer Systems Department, Jožef Stefan Institute, Ljubljana, Slovenia

## AIM

A publication in 2011 defined open science based on six principles including open data, open source, and open access (Kramer 2011). The idea behind all these is to make research outcomes and data accessible to others, facilitating and/or accelerating other investigations (Woelfle 2011). The European Commission is supporting these principles through European Strategy Forum on Research Infrastructures (ESFRI) and European Open Science Cloud (EOSC). METROFOOD-RI is funded through ESFRI, promoting metrology in food and nutrition and providing high level services for enhancing food quality and safety. Within the context of EOSC, FNS-Cloud is building a first-generation 'food cloud', federating existing and emerging datasets, and developing new services to support finding, accessing, comparing and re-use more widely.

## METHOD

The first challenges in developing frameworks for open data are technical and financial. The technical challenge is creation of an electronic infrastructure with multiple data repositories. Larger data-producing organisations have repositories, via which data might be made openly accessible, but need to invest continually in hardware and software to ensure sustainability of these resources. This investment should return increased visibility, namely citations and reputation. In parallel, other initiatives will generate centralised data repositories, where data can be uploaded and made accessible. These are large scale projects and appropriate funding must be provided by Member States, governments, partners, or projects. However, where personal data is included, ethical, legal and societal aspects (e.g. consent, privacy, GDPR) must also be considered. Personal data must be fully anonymised and, according to GDPR, individual have control over what, how, and where data is used. Thus, one challenge for these initiatives is how citizens are able to identify what data are included and revoke access at any time. Another challenge is convincing research communities to contribute data, a valuable personal and professional asset. Data are used to write publications, but not published in full. Sufficient incentive is needed so that datasets are published and contribute to scientific excellence. Both approaches are developing roadmaps and strategic agendas to create and maintain open data infrastructures and convince the community to actively participate to open data, and working with user communities to address the various challenges.

**Keywords:** open data, open science, METROFOOD-RI, FNS-Cloud

**Contact person:** Karl Presser, karl.presser@premotec.ch

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# The first certified reference materials for somatic cell counting (SCC) in milk

Zeleny R.<sup>1</sup>

<sup>1</sup> European Commission, Joint Research Centre, Geel, Belgium

## AIM

SCC is an important parameter for hygienic quality of raw milk and for animal health. Elevated SCC levels in milk can serve as an indicator for mastitis in lactating cows, pointing at insufficient hygiene practices on farms. In addition, SCC is used for milk payment as well as milk recording for genetic evaluation and farm management. Worldwide, approximately 500 million analyses are performed per year. - In early 2020, EC-JRC released the first certified reference materials (CRMs) for the concentration of somatic cells (somatic cell count, SCC) in milk.

## MATERIAL AND METHODS

Raw cow's milk was converted into milk powder by means of defatting, microfiltration, concentration, spray-drying and mixing, using a tailor-made procedure to minimise alteration of the cells during processing. Two cell count concentration levels were produced (low count, high count). The resulting milk powders were filled into amber glass bottles under inert gas atmosphere and packaged in sets (one set consists of one bottle with low count and one bottle with high count material). Between-unit homogeneity was quantified and stability during dispatch and storage were examined in accordance with ISO Guide 35. Characterisation of the materials was accomplished by means of an inter-laboratory comparison involving 32 laboratories with demonstrated competence and adhering to ISO/IEC 17025.

## RESULTS

Upon technical and statistical data evaluation, certified values were assigned and their respective uncertainties calculated in accordance with the Guide to the Expression of Uncertainty in Measurement. The materials are intended for calibration of fluoro-opto-electronic routine methods according to ISO 13366-2 and for the assessment of the method performance of SCC reference methods based on microscopy according to ISO 13366-1 and routine methods operating fluoro-opto-electronic cell counters according to ISO 13366-2. In addition, the materials can be used for establishing quality control charts or validation studies.

## CONCLUSIONS

The CRMs will constitute an integral part of the currently established reference system for somatic cell counting in milk, aiming to improve the comparability of SCC measurement results over space and time

**Keywords:** somatic cell count (SCC), milk, certified reference material (CRM), method calibration, ISO 13366-1, ISO 13366-2, microscopy, flow cytometry

**Contact person:** Reinhard Zeleny (reinhard.zeleny@ec.europa.eu)

## ACKNOWLEDGMENT

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# Research statistical study of various dietary supplements through measurements of their energy content

Sapalidou G.<sup>1</sup>, Karampatzakis S.<sup>2</sup>, Kogia F.<sup>3</sup>, Angelidis G.<sup>4, 1</sup>

Physics Department, International Hellenic University, St. Lucas, 654 04, Kavala, Greece, <sup>2</sup>Physics Department, International Hellenic University, St. Lucas, 654 04, Kavala, Greece, <sup>3</sup>Physics Department, International Hellenic University, St. Lucas, 654 04, Kavala, Greece, <sup>4</sup> Faculty of Medicine, Medical University of Pleven, Sv. Kliment Ohridski Str., Pleven, Bulgaria

## AIM

In the present work dietary supplements such as Raspberry Ketone (weight-loss aid), Neuropan (insomnia, anxiety, and psychological stress) and Ubiquinol (cellular energy for the organs to function) are used. Their energy content is estimated using a Parr 6400 bomb calorimeter and a statistical analysis of the data follows, aiming to drawing conclusions about their structural stability, calorific value, and the possibility of exploiting their waste to generate electricity.

## MATERIAL AND METHODS

As was mentioned above Parr 6400 bomb calorimeter is used for the measurement of the gross calorific value of Raspberry Ketone, Neuropan and Ubiquinol. Different masses of each sample are burned in the bucket of the bomb calorimeter and the data after each experiment is the temperature change and the energy content of the sample. For each mass, the temperature change is measured three times for the same mass, and the average value of these three values is calculated. Ten measurements for different masses are taken for each sample in study. Least Squares Method is applied in equation 1, for the estimation of the energy content of each dietary supplement with the maximum possible precision.

$$\Delta T = \frac{(GH)_s}{k_c} \cdot m_s \quad (1)$$

From the measured values of the energy content of the sample, the graph of the normal or Gaussian distribution is plotted, by using equation 2, where  $\sigma_x$  is the standard deviation or standard error of the distribution, given from equation 3.

$$\rho_x(x) = \frac{1}{\sigma_x \cdot \sqrt{2\pi}} \cdot e^{-\frac{(x_i - \bar{x})^2}{2\sigma_x^2}} \quad (2)$$

$$\sigma_x = \sqrt{\frac{\sum_{i=1}^N (x_i - \bar{x})^2}{N-1}} \quad (3)$$

## RESULTS

The obtained data show that the energy content of Raspberry Ketone is 4078 cal/g, of Neuropan is 4076 cal/g and of Ubiquinol is 7351 cal/g, while the plotted graphs show the high accuracy of the measurements.

## CONCLUSIONS

From the graph of Least Squares Method and from the graph of the Gaussian distribution, conclusions are drawn as to the accuracy of the measurement method. From the stability of the measurements, thus from their deviation from the average value, conclusions can be resolved about the homogeneity of the sample in terms of its composition. From the value of the energy content of the sample, conclusions are drawn about its caloric value, which is of direct interest to every

consumer. Finally, from the value of the energy content of the sample, in combination with the combustion residue, conclusions are drawn about the possibility of utilizing its waste for electricity production.

**Keywords:** Dietary supplements; Gross Heat; Least Squares Method; Normal Distribution

**Contact person:** Georgia Sapalidou, georgiasapalidou@gmail.com, +30 6984766493

## **ACKNOWLEDGMENT**

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# Nutritional features of Australian grown Feijoa (*Acca sellowiana*) fruit

Phan ADT.<sup>1</sup>, Cozzolino D.<sup>1</sup>, Wright O.<sup>2</sup>, Rychlik M.<sup>1,3</sup>, Sultanbawa Y.<sup>1</sup>, Netzel M.<sup>1</sup>

<sup>1</sup> ARC Training Centre for Uniquely Australian Foods, Centre for Nutrition and Food Sciences, Queensland Alliance for Agriculture and Food Innovation, The University of Queensland, Coopers Plains, QLD 4108, Australia, <sup>2</sup> School of Human Movement and Nutrition Sciences, The University of Queensland, Brisbane, Qld 4072, Australia, <sup>3</sup> Chair of Analytical Food Chemistry, Technical University of Munich, 85354 Freising, Germany

## AIM

To determine the nutritional composition of Australian grown feijoa (*Acca sellowiana*) in order to assess the dietary value of this emerging subtropical fruit.

## MATERIAL AND METHODS

State of the art analytical methods such as UHPLC-PDA-MS/MS were used to determine the nutritional composition (including vitamins, polyphenols, minerals, sugars, dietary fibre, fatty acids) of feijoa whole fruit, fruit peel and pulp.

## RESULTS

Feijoa fruit not only contains high amounts of antioxidant flavonoids, but is also a valuable source of vitamin C (63 mg/100 g FW (fresh weight)) and pantothenic acid (0.2 mg/100 g FW). The fruit is also a good source of dietary fibre (6.8 g/100 g FW) and potassium (255 mg/100 g FW). The edible fruit peel possesses significantly ( $p < 0.05$ ) higher amounts of antioxidant flavonoids and vitamin C than the fruit pulp.

## CONCLUSIONS

The consumption of feijoa fruit can deliver a considerable amount of bioactive compounds such as vitamin C, flavonoids and fibre, and therefore, may contribute to a healthy diet. However, short-term clinical trials and intervention studies are warranted to determine the actual bioavailability of the main nutrients and bioactive compounds as well as the potential health benefits of feijoa fruit consumption for humans.

**Keywords:** *Acca sellowiana*; feijoa; nutritional composition; bioactive compounds; diet

**Contact person:** Michael Netzel, m.netzel@uq.edu.au

## ACKNOWLEDGMENT

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# Personalised nutrition

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Žatečka L.<sup>1</sup> Kouřimská L.<sup>1</sup>

<sup>1</sup> Czech University of Life Sciences Prague (CZU)

## AIM

Personalized nutrition is a concept that has been created lately. Nowadays there is no acknowledged definition. This concept is understood as an effort for creating personalized nutrition recommendations based on personal characteristics. For this purpose is used nutrigenetics, nutrigenomics, microbiome analysis, and metabolomics. Even though this concept looks promising, there are not enough studies to make these personalized nutrition recommendations precision enough.

**Keywords:** personalized nutrition, microbiota, nutrigenetics, nutrigenomics

**Contact person:** Ladislav Žatečka, ladislav.zatecka@zateckal.cz

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# Nutritional value of edible insects and perspectives of their use as an alternative source of proteins

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Kouřimská L.

Department of Microbiology, Nutrition and Dietetics, Czech University of Life Sciences Prague, Kamýcká 129, 165 00, Praha 6, Suchbát, Czech Republic

## AIM

The aim of the presentation was to describe and compare the nutritional quality and bioactive compounds present in different kinds of insects. The issue related to entomophagy and the eventual risks that should be taken into account when consuming insects were also discussed.

## MATERIAL AND METHODS

Sustainability is becoming increasingly important in the world we live in. Edible insects belong to the possible alternative food source replacing traditional ingredients. With the growing population and increasing demands on world food production, research on this topic has intensified over the last 20 years. While 59 articles containing the keyword "edible insects" were published on the WOS in 2000, there were already 1182 of such articles in June 2020. However, there is still insufficient information on the composition of some edible insect species and on the benefits or disadvantages of their breeding, processing and use as food or feed. Therefore, the recent published data on the nutritional value and composition of insects were compared with our experimental values and critically evaluated. Attention was focused on basic nutrients: content and composition of proteins and lipids, as well as on micronutrients: minerals and vitamins. In particular, we compared the fatty acid profiles of selected insect species, the proportion of saturated and unsaturated fatty acids and the indices of atherogenicity and thrombogenicity. Some biologically active substances are also mentioned, as well as microbiological or chemical risks and allergenicity of insects.

## RESULTS

From a nutritional point of view, insects contain an easily digestible quality protein with an optimal representation of most essential amino acids, as well as lipids containing a number of essential fatty acids. The nutritional value of insects is subject to a number of factors including breeding technology. Although insects are a nutritionally valuable and potentially ecological resource, there are still some concerns about placing insects on the European market. The biggest concerns relate to the safety and acceptance of this food source.

## CONCLUSION

In the view of the growing market demand for edible insects and considering the inclusion of this new source of food, it is very important to analyse the available literature data and to increase the number of studies providing new data on the quality and safety of edible insects. There will also be challenges in legislation and the regulation of the edible insect sector.

**Keywords:** Entomophagy; insects' composition; food safety; novel food

**Contact person:** Lenka Kouřimská, kourimska@af.czu.cz

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# Natural food supplements from microalgae *via* supercritical CO<sub>2</sub> extraction

Leone G. P.<sup>1</sup>, Marino T.<sup>2</sup>, Casella P.<sup>2</sup>, Larocca V.<sup>3</sup>, Zoani C.<sup>1</sup>, Molino A.<sup>2</sup>, Balducchi R.<sup>3</sup>

<sup>1</sup>ENEA, Italian National Agency for New Technologies, Energy and Sustainable Economic Development, Department for Sustainability - Biotechnology and Agroindustry Division (ENEA – SSPT-BIOAG) C.R. Casaccia, Via Anguillarese 301, Rome (RM), 00123, Italy, <sup>2</sup>ENEA, Italian National Agency for New Technologies, Energy and Sustainable Economic Development, Department for Sustainability- Biotechnology and Agroindustry Division (ENEA – SSPT-BIOAG) C.R. Portici Piazzale Enrico Fermi, 1 - 80055 Portici (NA), Italy, \*tiziana.marino@yahoo.it, <sup>3</sup>ENEA, Italian National Agency for New Technologies, Energy and Sustainable Economic Development, Department for Sustainability- Biotechnology and Agroindustry Division (ENEA – SSPT-BIOAG) CR Trisaia, SS Jonica 106, km 419+500, 75026 Rotondella (MT), Italy

## AIM

Microalgae can be used as natural and sustainable components that, thanks to their nutritional and functional properties, offer opportunities for producing bioactive compounds to be used for several food and non-food purposes, including food supplements and additives, nutraceuticals and animal feed (Leone et al, 2019; Molino et al, 2020; Marino et al., 2020). In this work, *Nannochloropsis gaditana* (*N. gaditana*) microalga was selected for performing pilot scale extraction tests via supercritical CO<sub>2</sub> (ScCO<sub>2</sub>) with the aim to enrich the final extract in bioactive compounds.

## MATERIAL AND METHODS

Lyophilized biomass of *N. gaditana* was collected from Archimede Ricerche srl, A&A F.lli Parodi Group (Italy). CO<sub>2</sub> (industrial grade) and fatty acids (FAs, analytical grade) were purchased from Rivoira (Italy) and Sigma Aldrich (USA), respectively. Microalgae biomass underwent pretreatment step -mechanical cell disruption through ball mill- prior ScCO<sub>2</sub> extraction. Bioactive compounds extraction was performed at pilot scale by using CO<sub>2</sub> as solvent in supercritical conditions. After the extraction stage of bioactive compounds, both the extract and the exhausted biomass were analyzed in terms of their composition. In particular, for FAs profile the identification was done by using gas chromatography (Agilent 7820A GC-FID, USA) of the corresponding methyl esters (C8-C24 FA methyl esters). Other biomass characterizations were carried out *via* conventional standard methods.

## RESULTS

*N. gaditana* allowed to obtain after the extraction tests a low content of ash (2%) and high protein level, which increased from 47% in the biomass to 79% in the extract. Moreover, lipids contained in the biomass almost completely passed in the extract, with a content of about 12%. Lipids were almost all FA methyl esters, and –interestingly– among them in the extract the 33% was represented by eicosapentaenoic acid (EPA). The remaining exhausted biomass, enriched of total dietary fiber and carbohydrates, might be used as animal feed. Consequently, *N. gaditana* might represent an encouraging source of precious food supplement and feed in the near future.

**Keywords:** bioactive compounds; microalgae; *Nannochloropsis gaditana*; supercritical CO<sub>2</sub> extraction; fatty acids; EPA.

**Contact person:** Marino Tiziana, tiziana.marino@yahoo.it

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# Posters

FOODS

# Omega-3 and omega-6 fatty acids from sustainable crops

Del Fiore A.<sup>1\*</sup>, Colonna N.<sup>1</sup>, Crinò P.<sup>1</sup>, Cuna D.<sup>2</sup>, Stamigna C.<sup>1</sup>, Tolaini V.<sup>1</sup>, De Rossi P.<sup>3</sup>

<sup>1</sup> ENEA, Italian National Agency for New Technologies, Energy and Sustainable Economic Development, Department of Sustainability, Casaccia Research Centre, Via Anguillarese 301, 00123 Rome (Italy), <sup>2</sup> ENEA, Italian National Agency for New Technologies, Energy and Sustainable Economic Development, Department of Sustainability, Brindisi Research Centre, SS7 KM 706+000, 72100 Brindisi (Italy), <sup>3</sup> ENEA, Italian National Agency for New Technologies, Energy and Sustainable Economic Development, Energy Efficiency Unit Department, Casaccia Research Centre, Via Anguillarese 301, 00123 Rome (Italy)

## AIM

Fats are essential for living organisms, having not only energetic but also structural and metabolic functions. They are constituents of cell membranes and tissues, and precursors of some hormones. The main structural components of fats are fatty acids. Among these, polyunsaturated fatty acids (PUFAs) are fat components of great interest, as they have many beneficial effects on health, allowing to preserve cellular and metabolic balance. PUFAs include omega-3 and omega-6 essential fatty acids such as docosahexanoic (DHA), eicosapentanoic (EPA), and arachidonic (AA) acids, along with their precursors linoleic and  $\alpha$ -linolenic acids. They cannot be synthesized by the human organism and, therefore, must be taken with the diet. A balanced intake of omega-3 and omega-6 fatty acids can protect against cardiovascular and inflammatory diseases. Linoleic acid, precursor of omega-6 fatty acids, is the most abundant polyunsaturated fatty acid in nature. It is present in many seed oils and cereal grains (e.g. sunflower, sesame, peanut, corn oil, walnuts, almonds, wheat germ, rice, barley). The primary sources of  $\alpha$ -linolenic acid, precursor of omega-3 fatty acids, are instead mainly fish, fish liver oil, molluscs and crustaceans; it is present also in flax and soybean seed oil and in algae. Due to poor sustainability of both fishing and conventional agriculture, it would be essential to have alternative sustainable sources of these essential acids. The present work aimed at studying the suitability of different genotypes of some sustainable crops (artichoke, hemp, safflower) to be exploited as source of omega-3 and omega-6 fatty acids.

## MATERIAL AND METHODS

The fatty acid profile analysis has been applied to seed samples in order to characterize and valorize some sustainable and alternative crops. The fatty acids were extracted and derivatized into fatty acid methyl esters following a hydrolysis and methylation procedure.

## CONCLUSIONS

Exploiting plant biodiversity as renewable biological source of nutraceutical compounds meets the need for sustainable development from a bioeconomy perspective.

**Keywords:** polyunsaturated fatty acids (PUFAs); essential fatty acids; omega-3 and omega-6 acids; sustainability; alternative crops.

**Contact person:** Antonella Del fiore; email: antonella.delfiore@enea.it

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# Characterization and assays of wild growing plants used in herbal medicine

Kubeš J., Sýkorová T., Skalický M., Lhotská M., Česká J., Hnilička F.

Department of Botany and Plant Physiology, Faculty of Agrobiolgy, Food and Natural Resources, Czech University of Life Sciences Prague, Czech Republic

## AIM

Harvesting of wild medicinal plants, beside agriculture production, represents another important resource of various herbs for domestic manufacturers of herbal medicines and mixtures. Purchased plant parts have to meet several requirements and the herbs that are given over to curative application should comply with corresponding tests as stated in Czech Pharmacopoeia. Chosen plants are well-known in folk medicine, like as leaves of *Plantago lanceolata* that contains polysaccharides and other metabolites and they are used against cough (Stojilkovski K. et al. 2018). Other plant, *Hypericum perforatum*, is used for its flowering tops possessing antidepressant activity (Tian J. et al. 2014). Flowers of *Sambucus nigra* and *Matricaria recutita* have also positive effect on health (Mahboubi M 2020; McKay DL and Blumberg JB 2006). Aim of this work is determine quality of these harvested wild plants for potential application according the requested standards (ČR, M. Z. 2017).

## MATERIAL AND METHODS

Plant parts were harvested in Vestec near Prague (49.9804967N, 14.5048744E) during the spring and summer 2019 and dried and processed according to the instructions of companies buying the herbs. Individual herbal substances were identified by macroscopical and microscopical characterisation. The tests like water content and total ash were conducted in drying oven and muffle furnace. Specific assays for secondary metabolites were performed by UV/Vis spectrophotometry according to the pharmacopoeia or other required methods.

## RESULTS

Majority of these evaluated plant drugs met required tests, however *Hyperici herba* had higher water content and *Matricariae flos* did not contained enough of typical blue essential oil.

## CONCLUSIONS

The chosen locality provided mostly appropriate herbal substances usable for application.

**Keywords:** herbal substances, macroscopical and microscopical identification, secondary metabolites content

**Contact person:** Kubeš Jan, [kubes@af.czu.cz](mailto:kubes@af.czu.cz)

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# Protein quality of foods of plant origin available in Portugal

Motta C.<sup>1</sup>, Coelho M.<sup>1</sup>, Tavares N.<sup>2</sup>, Diniz D.<sup>2</sup>

<sup>1</sup> National Health Institute Doctor Ricardo Jorge, Food and Nutrition Department, Lisbon, Portugal, <sup>2</sup> CBIOS Centro de Investigação em Biociências e Tecnologias da Saúde, Universidade Lusófona de Humanidades e Tecnologias, Lisboa, Portugal

Based on the growth of concerns with environmental and food sustainability associated with health, a recent and gradual change in the Portuguese dietary paradigm occur. According to the National Food, Nutrition and Physical Activity Survey, an increase to vegetarian food options as an alternative to consumption of animal products.

## AIM

This study aims to define the protein quality of 59 different foods of plant origin belonging to five different nutritional groups, which are consumed by the Portuguese population. For that, the analysis of protein and the profile of amino acids, including the essential amino acids, was performed.

## MATERIAL AND METHODS

Sampling plan of the study was organised by nutritional groups, namely cereals, vegetables, fruits, legumes and vegetable meat substitutes. The amino acid profile was determined by ultra-performance liquid chromatography with photodiode array detector (UPLC / PDA). Quantification of the essential amino acids was performed to access the protein quality of each sample. The amino acid score of each nutritional group was calculated based on recommended intake portions, according to the Mediterranean Food Wheel, a base reference for the Portuguese dietary recommendation.

## RESULTS

The amino acid scores of the analysed food groups present a high variability of the scores between the analysed groups for the different amino acids. Concerning all the essential amino acids histidine presents the highest levels, especially in fruits, like bananas and mango (scores between 6 and 2) and cereals as rice (score >1.6). Comparing scores in the tested food groups, we observed a similar trend. Aromatic amino acid and histidine presented scores higher than 1 in all food groups while lysine, valine and isoleucine present scores higher than 1 representing in this way the limiting amino acids in the vegetables.

## CONCLUSIONS

The results obtained demonstrate the importance of a varied, yet correctly weighted intake of foods from different nutritional groups. To ensure complementarity of daily protein levels, such as, for example, a combination of cereals or fruits and legumes needs to be taken in to account. The correct combination of portions of different nutritional groups can be an effective way to obtain the essential amino acids required for a balanced diet.

**Keywords:** amino acid scores; recommended intake; Portuguese population; protein intake

**Contact person:** Carla Motta – carla.motta@insa.min-saude.pt

# Determination of fat content and profile of fatty acids in new varieties of oats

Pokhrel K.<sup>1</sup>, Kouřimská L.<sup>1</sup>, Božik M.<sup>2</sup>

<sup>1</sup>Department of Microbiology, Nutrition and Dietetics, <sup>2</sup>Department of Food Science, Czech University of Life Sciences Prague, Kamýcká 129, 165 00, Praha 6, Suchbát, Czech Republic

## AIM

The aim of the study was to determine the total lipid content and the fatty acid profile of husked and naked oat varieties and to make a comparison of these characteristics.

## MATERIAL AND METHODS

Four hulled spring oat cultivars (Cavaliere, Kertag, Selodon, Gregor) and two naked oats (Kamil, and Otakar) were provided by the Selgen a.s. breeding station. Hulled oat samples were dehulled manually. Dry matter was determined using infrared moisture balances. The determination of the fat content was carried out by Soxhlet extraction according to the ISO 659: 2009. Fatty acid methyl esters were prepared according to the ISO 12966-2:2011 and determined using GC-MS. The proportions of fatty acids were calculated by the area normalisation method.

## RESULTS

Dry matter of oat samples was from 87.44 ±0.78 g/100 g to 90.46 ±0.15 g/100 g. The result of fat content showed that naked oats cultivars Kamil and Otakar had significantly higher fat content (5.12 g/100 g) than hulled oats (3.02 g/100 g) ( $p < 0.0001$ ). Regarding fatty acid composition, linoleic acid was the predominant fatty acid on all analysed cultivars (38.40%), followed by oleic (32.88%) and palmitic (19.80%) acids. The highest oleic acid content was found in the Kamil variety which significantly differs from all hulled oat cultivars ( $p < 0.05$ ), but not from the naked oat Otakar. The lowest percentage of oleic acid (28.69%) was observed in the dehulled group compared to the original hulled oats with husks (32.88%) and naked varieties (35.81%). Dehulled samples Cavalier B ( $p = 0.0321$ ) and Seldon B ( $p = 0.0138$ ) had higher linoleic acid contents than original samples with husks. Overall, the highest proportion of MUFA were found mostly in naked variety Kamil (38.61 ±0.36), and PUFA were found significantly higher in dehulled variety Cavaliere (47.67 ±0.62). There was no significant difference in SFA in all analysed varieties. Comparatively, n-3 and n-6 were found in higher amount in hulled variety Gregor (2.99 ±0.56) and dehulled variety Seldon (44.89 ±0.71) respectively.

## CONCLUSION

This study provided a good information regarding the lipid content and composition of new oat varieties harvested in the Czech Republic to have better estimation on breeding and yield improvement for the purpose of animal feed as well as for both food producers and consumers.

**Keywords:** *Avena sativa*; *Avena nuda*, fat content, fatty acid profile

**Contact person:** Kshitiz Pokhrel, pokhrel@af.czu.cz

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# The effect of microwave treatment on quality and oxidation stability of nuts and almonds

Škvorová P.<sup>1</sup>, Kouřimská L.<sup>1</sup>

<sup>1</sup>Department of Microbiology, Nutrition and Dietetics

## AIM

Dry shell fruits are common parts of our diet. They contain large amounts of health-positives compounds. On the other hand, during their storage attacks of pests and microorganisms are very common. Many conservation techniques are used for extension of the shelf life as well as for eradication of pests. Microwave radiation represents a modern preservation technique enabling drying, pasteurization, sterilization, defrosting, tempering or sealing of food materials. The main advantage of microwave radiation is energy saving, shortening of the time and cost reduction. The main disadvantage is uneven temperature distribution in food, which is solved by the addition of hot air heating zone after microwave treatment. The aim of this research was to evaluate the effect of microwave radiation of on the quality and oxidation stability of dry shell fruit during their storage. The effect of the radiation was monitored by colour changes, dry matter content and peroxide value (PV) determination.

## MATERIAL AND METHODS

Three types of dry shell fruit (peanuts, almonds and hazelnuts) were treated by the newly constructed large-scale industrial microwave line. The content of dry matter and the colour of fruits were analysed immediately after the treatment. Oxidation stability was observed for three months during the storage at temperatures 20, 40 and 60 °C.

## RESULTS

There were statistically significant differences in dry matter content and colour between treated and untreated samples. Significant effect of the microwave radiation treatment on PV was observed in peanuts. There were also significant influences of temperature and the time of storage on PV. Our results showed that microwave radiation treatment and higher storage temperature resulted in the increase of PV of tested samples.

## CONCLUSION

The microwave radiation caused the changes of the dry shell fruit stability against rancidity. The changes primarily depend on treated kind of sample than on the storage conditions. However, it was found that the quality of nuts and almonds treated with microwave radiation was still acceptable throughout storage even at elevated temperatures. We can therefore assume that it will be preserved until the expiration date on the shelves in stores.

**Keywords:** Microwave treatment; rancidity; nuts; almonds; peroxide value

**Contact person:** Petra Škvorová, xskvp002@studenti.czu.cz

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# Behaviour of polyphenols and antioxidant activity after boiling of heads from a globe artichoke improved genotype

Tolaini V.<sup>1</sup>, Salazar Sanclemente K.C.<sup>2</sup>, Crinò P.<sup>1</sup>

<sup>1</sup> ENEA (Italian National Agency for New Technologies, Energy and Sustainable Economic Development) – Division Biotechnologies and Agroindustry - Laboratory Bio-products and Bio-processes, Casaccia Research Centre, Via Anguillarese 301 - 00123 Rome, Italy,

<sup>2</sup> Università Campus Bio-Medico di Roma, Via Alvaro del Portillo, 21 - 00128 Rome, Italy

## AIM

The Mediterranean diet is characterized by a high consumption of different fruits and vegetables. In many South-Europe and North-Africa countries, the cultivation and consumption of globe artichoke (*Cynara cardunculus* var. *scolymus* L.) is widespread. In particular, Italy is the main worldwide producer of this vegetable (FAOSTAT, 2017). Immature flower heads (capitula) are rich in minerals, vitamins, dietary fiber and phenolic compounds with many healthy properties (Lattanzio *et al.*, 2009). In particular, polyphenols are secondary bioactive metabolites responsible for the antioxidant activity and *Cynara* spp. are a good source of hydroxycinnamic acids (chlorogenic acid, cynarin and 1,5-dicaffeoylquinic acid) and flavonoids (luteolin and apigenin) (Maietta *et al.*, 2017). Different factors can affect polyphenol content such as genotype, soil and climatic conditions, crop management, harvest time (Lombardo *et al.*, 2010). Furthermore, heat treatments tend to increase the concentration of phenolic compounds and the antioxidant capacity also in some vegetables (Juániz *et al.*, 2016). The inner portion of the artichoke head (receptacle) is traditionally used for many typical dishes, both raw and cooked. Up to our knowledge, there are few data (Ferracane *et al.*, 2008) on the influence of heat treatment or cooking on polyphenols and antioxidant capacity in commercial globe artichoke heads. The aim of our work was to chemically characterize a globe artichoke improved genotype compared to a commercial one, both as raw and boiled.

## MATERIAL AND METHODS

The total polyphenol content and antioxidant activity were measured by spectrophotometric assay. Polyphenol profile was carried out by HPLC.

## RESULTS

Results showed an increase of total polyphenols in cooked heads, mainly for the improved genotype, whereas the antioxidant activity did not change. About the polyphenol profile, boiling determined especially a 1,5-dicaffeoylquinic acid increase.

**Keywords:** globe artichoke head; polyphenols; antioxidant activity; heat treatment

**Contact person:** Valentina Tolaini, valentina.tolaini@enea.it

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# A method for determination of acrylamide in bread by UPLC-MS/MS

Copeto Sandra, Jesus Susana, Motta Carla, Coelho Inês, Castanheira Isabel

Department of Food Safety and Nutrition, National Institute of Health Doctor Ricardo Jorge, Lisbon, Portugal

## AIM

Acrylamide is a contaminant be present, especially in baked and fried rich in starchy food products, such as french fries, bread, cookies and coffee. It is a product of the Maillard Reaction formed during food processing, at high temperatures by the reaction between free amine groups (amino acids/proteins) and the carbonyl group (sugars/carbohydrates). Acrylamide is classified by the International Agency for Research on Cancer as a carcinogenic substance to humans, and its quantification is essential to control the exposure risk. Since the discovery of its presence in food, several studies have been conducted to developed extraction procedures for acrylamide quantification. The present study aims to develop a cleaning method suitable for several food matrices, like bread and chips followed by liquid chromatography coupled to a mass detector method (LC-MS/MS) for acrylamide quantification. To achieve this aim, the use of solid-phase extraction and several salts mixtures, such as QuEChERS type, were used.

## MATERIAL AND METHODS

For the extraction of acrylamide the application of two different salts mixtures was studied: magnesium sulfate, aluminum oxide and sodium chloride (mixture 1) and magnesium sulfate, primary and secondary amine (PSA) and sodium acetate (mixture 2), which remove the fat and purify the sample. A combined method with SPE Oasis HLB was also tested. The D3-acrylamide internal standard was individually added to each sample before extraction and clean-up. The method accuracy was demonstrated through the evaluation of the recovery of the analyte in samples spiked with acrylamide standard of known concentration and, also by the evaluation of the bias, using ERM-BD274 ( $74 \pm 7 \mu\text{g}/\text{kg}$ ). All samples were analyzed in triplicate. For the detection and quantification of acrylamide, an Ultra Performance Liquid Chromatographic coupled to a mass detector (UPLC-MS/MS) was used.

## RESULTS

For the extraction method using mixture 1 the results were satisfactory, where it was obtained a spiked recovery between 85 - 112% and the results of the certified reference material vary between 71.2 – 80  $\mu\text{g}/\text{kg}$ , which is within the range of uncertainty. The correlation coefficient of the calibration curve ranged between 0.995 - 0.999, and the limit of quantification (LOQ) was 5 mg/kg.

## CONCLUSIONS

The presented method, LC-MS/MS, with mixture 1 is adequate for the determination of acrylamide in bread , accomplishing the performance criteria of the regulation published by the European Commission, COMMISSION REGULATION (EU) 2017/2158 of 20 November 2017.

## ACKNOWLEDGEMENT

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# Characterization of food from burnt areas in the Central Region of Portugal

Gueifão S.<sup>1,2</sup>, Moreira T.<sup>1</sup>, Ventura M.<sup>1</sup>, Delgado I.<sup>1</sup>, Celho I.<sup>1</sup>, Castanheira I.<sup>1</sup>

<sup>1</sup> Food and Nutrition Department, National Institute of Health Doutor Ricardo Jorge, I.P. (INSA), Avenida Padre Cruz 1649-016 Lisboa – Portugal, <sup>2</sup> Department of Chemical Engineering, Instituto Superior Técnico (IST), Av. Rovisco Pais, 1049-001 Lisboa - Portugal

## AIM

Forest fires constitute an environmental problem with adverse and diversified impacts on atmospheric, terrestrial, and aquatic ecosystems, with economic and social repercussions, and a significant impact on flora. Fire mineralizes soil nutrients and returns plant-locked nutrients to the soil. Nutrients are lost from the ecosystem through the release of particulates and volatile compounds during the fire, and later through surface transport and leaching by rains<sup>1</sup>. Wildfires can play an important role in the environmental distribution of major and trace elements, including through their mobilization by fire-induced runoff and associated transport of soil and ash particles. Also, wildfires have been found to release and deposit contaminants on the soil surface, either directly by combustion of vegetation and mineralization of soil organic matter or indirectly through interactions of ashes. This fact can lead to the addition of potentially significant amounts of these elements in the soil and, thereby, can alter its chemical properties<sup>2</sup>.

This study aims to evaluate the content of arsenic (As), cadmium (Cd) and chromium (Cr) in cabbage, potatoes, eggs and fruit samples cultivated in recently burnt areas.

## MATERIAL AND METHODS

A total of 24 pools composed of 3 samples each was analysed at three different harvest periods. Cabbage samples from burnt zones were compared with a designated control sample from a non-burnt zone. Analytical determinations were performed by Inductive Plasma Coupled Mass Spectrometry (ICP-MS). Previously, samples underwent a heating block acid digestion with a validated time and temperature program. Speciation analysis was carried out by High Performance Liquid Chromatography (HPLC) coupled to ICP-MS. Before analysis, samples were extracted using nitric acid. Analyses were performed in accordance with ISO 17025:2005.

## RESULTS

The concentrations of As in fruits and Cd in eggs were below the limit of quantification (2.2 µg/kg and 3.1 µg/kg, respectively). The variation of Cr content between matrices was as follows: fruits > eggs > cabbages > potatoes. On the other hand, As contents was verified as: cabbage > eggs > potatoes > fruit. Cabbage presented the highest Cd content, while eggs had the lowest values for this element. When compared to the control sample, the most pronounced rise was observed for As. Speciation studies show that the predominant arsenic species in cabbages are also the most toxic ones, that is, arsenite (As (III)) and arsenate (As (V)).

## CONCLUSIONS

The values of the inorganic elements present in this study are not yet properly legislated so it is important to performed studies able to support future legal references. Monitoring studies based on accredited methods are a powerful aid in assessing the risk exposure of affected populations.

**Keywords:** Forest fires, ICP-MS, contaminants, foods

**Contact person:** Sandra Gueifão (sandra.gueifao@insa.min-saude.pt)

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# Mackerel seasonality effect on Se content

Rego A.<sup>1</sup>, Ventura M.<sup>1</sup>, Gueifão S.<sup>1</sup>, Coelho I.<sup>1</sup>, Cardoso C.<sup>2</sup>, Afonso C.<sup>2</sup>, Bandarra N.<sup>2</sup>, Isabel Castanheira<sup>1</sup>

<sup>1</sup> Department of food and nutrition, National Institute of health Doutor Ricardo Jorge, Lisbon

<sup>2</sup> Portuguese Institute for Sea and Atmosphere, Lisbon

## AIM

Selenium (Se) is an essential micronutrient that can become toxic when ingested in high concentrations. The human being can obtain Se through the consumption of food, dietary supplements, water and exposure to ambient air. The food source is the most abundant and the one that most contributes to the daily intake of Se.

Fish is one of the best sources of Se, and the Portuguese Mackerel is one of the most important and abundant species in Portugal, being rich in several vitamins and Se, due to being a predator. The work presented integrates part of the first task of the project Newfood4thought that aims at the prevention of cognitive decline through combinations of nutrients. One of the subtasks is the study of the influence of seasonality in the levels of Se in Mackerel samples collected throughout the year.

## MATERIAL AND METHODS

Mackerel samples were collected monthly, from March 2019 to February 2020, and prepared in twelve pools. Se was determined by an inductively coupled plasma mass spectrometer (ICP-MS) preceded by acid digestion in a graphite hotplate using a previously validated time and temperature program. All standards and reagents were of high purity (over 99.5%).

As quality control, a minimum of three replicate analyses was performed for each sample. Se calibration curve was made with no less than five standards in different concentrations with a correlation coefficient superior to 0.9995. Spiked samples and a certified reference material from fish muscle (ERM-BB422) were also used. All analyses performed were in accordance with ISO 17025:2017.

For statistical analysis, One-Way ANOVA and the Least Significant Test were used.

## RESULTS

Results showed insignificant variation of Se content between months, ranging from 462 µg/Kg to 605 µg/Kg, with an average of approximately 540 µg/Kg except for a particular month, May. The Se content from May was significantly (715 µg/Kg). Cabral et al. (2002) showed that the mackerel diet consisted mainly in zooplankton (rich in Se and other vitamins) in spring and summer, fish and decapods in autumn and decapod larvae in winter. Martins et. al. (1996) referred that in the Portuguese continental coast the spawning season of Spanish mackerel, runs between February to March and April to May.

## CONCLUSIONS

With the exception of May, Se content in mackerel remained constant throughout the year. The high Se content found in May samples may be related to mackerel's spawning season and feeding habits. However, further studies are needed to confirm these preliminary conclusions, namely studying the Se content monthly for several years.

**Keywords:** selenium; Portuguese mackerel; ICP-MS; seasonality.

**Contact person:** Andreia Rego, [andrea.rego@insa.min-saude.pt](mailto:andrea.rego@insa.min-saude.pt)

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# Assessment of the mechanisms and pathways of fish contamination and mercury-selenium antagonism

Ribeiro M.<sup>1,2,3</sup>, Zephyr N.<sup>1</sup>, Leufroy A.<sup>1</sup>, Coelho I.<sup>2</sup>, Danion, M.<sup>4</sup>, Castanheira I.<sup>2</sup>, Guerin T.<sup>1</sup> and Jitaru P.<sup>1</sup>

<sup>1</sup> Université Paris-Est, Anses, Laboratory for Food Safety, F-94700 Maisons-Alfort, France, <sup>2</sup> Department of food and nutrition, National Institute of health Doutor Ricardo Jorge, Lisbon, <sup>3</sup> Structural Chemistry Center, Instituto Superior Técnico, University of Lisbon, Lisbon, <sup>4</sup> Agency for Food, Environmental and Occupational Health & Safety (ANSES), Ploufragan-Plouzané Laboratory, Fish Viral Pathology Unit, Technopôle Brest-Iroise, 29280 Plouzané, France

## AIM

Mercury (Hg) is one of the most dangerous chemical contaminants for humans and the environment. This is due to its extreme toxicity, particularly in the methylated form, denominated methylmercury (CH<sub>3</sub>Hg), which is easily absorbed by the body (bioaccumulation). CH<sub>3</sub>Hg is present in low concentrations in water and in water sediments, and can greatly increase its concentration in aquatic organisms, often reaching more than 85% of total Hg. Therefore, the main source of exposure to CH<sub>3</sub>Hg is fish consumption, in particular fish from higher trophic levels, since its content tends to increase along the food chain (biomagnification). Nevertheless, it is interesting to note that mercury's bioavailability seems to be linked to selenium (Se) levels. The Se protective effects against Hg bioaccumulation and toxicity has been identified for the last decades, although the mechanism is still poorly understood. The aim of this study is to accurately assess the Hg-Se interaction in fish and the human dietary exposure.

## MATERIAL AND METHODS

A bioassay was carried out in a representative freshwater fish (rainbow trout) in collaboration with the fish viral pathology unit of ANSES (Ploufragan-Plouzané laboratory). Fish were dietary exposed to CH<sub>3</sub>Hg under controlled conditions, in the presence or absence of SeMet. The final concentration of CH<sub>3</sub>Hg and SeMet in the diets was respectively 0,2 µg g<sup>-1</sup> and 5 µg g<sup>-1</sup>.

Briefly, six feeding protocols were compared: a non-supplemented diet (control group), a diet supplemented with SeMet, a diet supplemented with MeHg, a diet supplemented with both MeHg and SeMet, a diet supplemented first with MeHg for 1 month and then with SeMet for 2 months and a diet supplemented with SeMet for 1 month and then with MeHg for 2 months.

The edible part of fish (muscle) was analysed for SeT and Hgt concentrations by ICP-MS after different time of exposure (1 month, 2 months and 3 months).

**Keywords:** mercury; selenium; methylmercury; selenomethionine; dietary exposure

**Contact person:** Mariana Mendes Ribeiro, mariana.ribeiro@insa.min-saude.

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## Open access to METROFOOD-RI\* services

Subhalakshmi Sharma, Karine Vandermeiren, Nastasia Belc, Jan Mast, Karl Presser, Eveline Verleysen, Claudia Zoani, Joris Van Loco

### AIM

METROFOOD-RI ([www.metrofood.eu](http://www.metrofood.eu)) is an EU-funded ESFRI project, aiming to establish a new distributed European Research Infrastructure (RI) to increase efficiency in food quality and food safety. It strives to provide and coordinate high-level metrological services on a European scale for researchers, laboratories, food inspection instances and policymakers. As part of the preparatory phase towards the legal statute of ERIC (European Research Infrastructure Consortium), two open calls have been recently launched to provide the opportunity for free access to some RI's facilities.

Projects that align well with the aims of METROFOOD-RI and meet the technical, scientific and ethical requirements, will be eligible. The two current open calls are for (a) remote access to the Transmission Electron Microscope (TEM) facility for physicochemical characterization of nanoparticles in food (Sciensano, Belgium), and (b) physical access to the food pilot demonstrating technical solutions and adaptations of food processing technology to minimize acrylamide in bakery products (National Research & Development Institute for Food Bio-resources - IBA, Romania). In the near future, a third open access will be made available for two electronic services for open data use. We encourage all interested parties to request further information and apply via the website.

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# Screening for Microcystins and Nodularin in food supplements on the Belgium market

W.H.R. Van Hassel<sup>1,2</sup>, B. Huybrechts<sup>1</sup>, A. Wilmotte<sup>2</sup>, M. Andjelkovic<sup>3</sup>

<sup>1</sup>Organic contaminants and additives, Sciensano, <sup>2</sup>InBios-Centre for Protein Engineering, University of Liège, <sup>3</sup>Risk and health impact assessment, Sciensano

## AIM

During the last years, the financial turnover of the non-registered consumer health market and the share of food supplements in this market has grown at a steady pace<sup>1</sup>. This growth implies an increased exposure of the public to these products and their ingredients. Some ingredients, mainly from natural sources, have beneficial effects but might exert adverse effects due to the presence of non-intentional compounds, like natural toxins.

Food supplements produced by microalgae or cyanobacteria could be one of these examples. Within this group of products, three organisms are responsible for the production. First of all, the green algae called *Chlorella vulgaris*, for which commercialization started post-WW2. Second, the cyanobacterium *Aphanizomenon flos-aquae*, which is known to form massive blooms in Klamath Lake in Oregon US. Third, we have *Arthrospira sp.*, better known under its trade name “spirulina”. This organism has been harvested from lakes in Africa and South America for a long time. Retailers advertise these food supplements as protein-rich food sources with additional health benefits.

However, the scientific community devoted to cyanobacteria has expressed doubts surrounding the safety of some of the supplements available on the market. Research has shown that some cyanobacterial species can produce toxins harmful to humans. These toxins are commonly detected in blooms of cyanobacteria in lakes all over the world. In Western Europe, microcystin is frequently detected. Microcystin congeners (MC's) are hepatotoxic and cause nausea, vomiting and, in severe cases, liver damage when ingested.

Although the most species used to produce the food supplements (except *Aphanizomenon*) should not produce these toxins themselves, published data suggest that there could be contamination of the products by toxin-producing cyanobacteria. Production steps vulnerable for contamination are cultivation and processing of the organisms (cyanobacteria).

## MATERIAL AND METHODS

To acquire initial data of the presence of microcystins and the related nodularin in food supplements, we optimized and validated an analytical method capable of quantification of eight microcystin congeners and nodularin based on the protocol by Turner et al., 2018<sup>2</sup>. With this method, we analyzed 40 samples from the Belgium market. The samples were bought from the major retailers in Belgium.

## RESULTS AND CONCLUSION

The results show MC's in several products. Some of them were above the guideline values suggested by the WHO<sup>3</sup>, which poses a possible risk for public health. To better assess this risk, exposure studies should be executed to evaluate the consumption of these products in Belgium. The consumption data of food supplements are not readily available and include several assumptions, which is another hurdle for this research.

**Keywords:** LC-MS/MS, Food Supplements, Microcystin, Nodularin

**Contact person:** Wannes H.R. Van Hassel, wannes.vanhassel@sciensano.be

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# Magnetic aptamer-based oligoprecipitation as innovative sample treatment strategy for food allergen determination: egg white lysozyme as case study

Toma L.<sup>1</sup>, Franzoni A.<sup>1</sup>, Mattarozzi M.<sup>1</sup>, Cavazza A.<sup>1</sup>, Giannetto M.<sup>1</sup>, Ronda L.<sup>2</sup>, Careri M.<sup>1</sup>

<sup>1</sup> Department of Chemistry, Life Sciences and Environmental Sustainability, University of Parma, Italy, <sup>2</sup> Department of Medicine and Surgery, University of Parma, Italy

## AIM

Development of an innovative sample treatment strategy involving magnetic aptamer-based oligoprecipitation of target proteins from complex food extracts for efficient protein extraction to ensure detection of hidden allergen in processed foods. For this purpose, the potential of aptamer oligonucleotides as recently discovered recognition elements alternative to antibodies is being studied for egg white lysozyme as case study, since lysozyme is known as a dominant egg-white allergen.

## MATERIAL AND METHODS

Four different sequences of aptamers binding to white egg lysozyme were selected from literature; circular dichroism (CD) spectra were acquired as preliminary assessment of their interaction. Tryptic peptide selection was carried out using liquid chromatography-mass spectrometry (LC-MS) in data dependent acquisition mode followed by bioinformatics data processing. Electrochemical assays were performed to study the aptamer immobilization on the surface of carboxyl-functionalized micro magnetic beads using a (5')-amino-C6 modified and (3')-biotin-labelled sequences. Protein extraction was carried out using a thermostatic water bath, beat beating and immersion probe sonication. Total protein concentration was assessed by spectrophotometric Bradford assay.

## RESULTS

The significant deviation between algebraic sum and experimental CD spectra of lysozyme and aptamer mixture (1:1) demonstrated the establishment of protein-aptamer interaction for all the aptamers under investigation. Unique peptides were selected and a target LC-tandem mass spectrometry method was developed for lysozyme analysis. Preliminarily experiments carried out on magnetic beads functionalization at aptamer levels equal to 500 nM, 1  $\mu$ M and 2  $\mu$ M allowed to observe a signal increase corresponding to a concentration attributable to a greater occupation of the available sites. In order to assure maintenance of lysozyme structure without compromising the interaction with the aptamer receptor, different innovative protein extraction strategies such as beat beating and sonication immersion probe are under investigation, avoiding the denaturing conditions commonly used in incurred materials and processed food analysis. The approach also paves the way for protein extraction involved in emerging aptasensing platforms. **Keywords:** aptamers; hidden food allergens; magnetic beads; protein extraction; complex food matrix

**Contact person:** Lorenzo Toma, lorenzo.toma@studenti.unipr.it

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# Characterization and discrimination of salt samples of different geographical areas and kind by Neutron Activation Analysis and chemometric methods.

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Bergamaschi L., Mandrile L., Rossi A.M.

INRIM - Istituto Nazionale di Ricerca Metrologica, Strada delle Cacce 91, Torino (Italy)

## AIM

Salt is essential for human health, when eaten in the appropriate amount. It is considered one of the most common seasoning as salt is used in many cuisines around the world, but the numerous kinds with special colours and tastes available on the market make it a food speciality for fine cuisine. From a chemical point of view, refined sea salt and peculiar mine salts present characteristic elemental composition which influences the taste and the commercial value.

In this study, Neutron Activation Analysis (NAA) was applied on rock and sea salt samples of different origin and kind for the determination of major and trace elements. This technique allows to determine more than 30 elemental concentrations, guaranteeing results traceable to the SI. The elemental concentration were obtained to characterize the salts and point out possible nutrient or hazardous elements, as well as to figure out the elemental fingerprint of different mine salts for their authentication. Data were analysed by multivariate statistics, including in the chemometric modelling, with the aim of discriminate the samples by geographical origin and salt kind.

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# Building a database for provenance studies of origin wines from the Douro Valley

Coelho I.<sup>1</sup>, Matos A.<sup>2</sup>, Nascimento A.<sup>1</sup>, Bordado J.<sup>3</sup>, Donard O.<sup>4</sup>, Epova E.<sup>5</sup>, Barre J.<sup>5</sup>, Berail S.<sup>4</sup>, Castanheira I.<sup>1</sup>

<sup>1</sup> Dep. of Food and Nutrition, National Institute of Health Doutor Ricardo Jorge, Portugal, <sup>2</sup> UNIDEMI, Dep. de Engenharia Mecânica e Industrial, Faculdade de Ciências e Tecnologia, Universidade Nova de Lisboa, <sup>3</sup> CERENA, Dep. de Engenharia Química e Biológica, Instituto Superior Técnico, Universidade de Lisboa, <sup>4</sup> MARSS-IPREM, Center of Mass Spectrometry for Reactivity and Speciation Sciences, Université de Pau et des Pays de l'Adour, France, <sup>5</sup> Advanced Isotopic Analysis, Hélioparc, Pau (France)

## AIM

The aim of the present work is to produce high quality analytical data suitable for evaluating the provenance of Port wine and Douro wine and integrating an authentic wine reference database. Both wines are produced within the same geographical area, the Douro Valley region located in the northeast of Portugal but follow different vinification procedures. The fermentation of Port wine is interrupted by the addition of a grape based spirit producing a sweet fortified wine typically consumed as digestif and dessert wine. Douro wine, on the other hand, is a table wine.

The mineral composition of plants reflects the soil and the environmental conditions under which plants were grown. Minerals and trace elements, as well as some of their isotopes, are potential biomarkers in authenticity studies.

We followed two analytical strategies, multielemental analysis and stable isotopes, to understand the parameters common to the region and fundamentally dependent on the soils and the ones influenced by processing methods or environmental contamination.

## MATERIAL AND METHODS

Fifteen samples of each type of wine, Port and Douro, were characterized for 26 elements. <sup>7</sup>Li, <sup>9</sup>Be, <sup>11</sup>B, <sup>27</sup>Al, <sup>52</sup>Cr, <sup>55</sup>Mn, <sup>59</sup>Co, <sup>60</sup>Ni, <sup>65</sup>Cu, <sup>66</sup>Zn, <sup>75</sup>As, <sup>77</sup>Se, <sup>85</sup>Rb, <sup>88</sup>Sr, <sup>95</sup>Mo, <sup>107</sup>Ag, <sup>111</sup>Cd, <sup>118</sup>Sn, <sup>205</sup>Tl, <sup>208</sup>Pb were determined by inductively coupled plasma mass spectrometer (ICP-MS) while Fe, Mg, P, Ca, Na, K were analysed by inductively coupled plasma optical emission spectrometer (ICP-OES). In addition, two isotopic systems were also studied: <sup>87</sup>Sr/<sup>86</sup>Sr and <sup>18</sup>O determined by Multicollector-ICP-MS (MC-ICP-MS) and Isotope Ratios Mass Spectrometry (IRMS), respectively.

For all analysis reagents used were of high purity to prevent for contaminations. Ultra pure water of grade I, as defined in EN ISO 3696, was obtained using a Milli-Q plus Millipore System. Concentrated nitric acid, analytical grade (65%; Merck), was purified in the laboratory, using an acid distillation system (Milestone SubPUR), and compared to a commercial ultrapure nitric acid solution (Merck), on a routine basis.

Analytical results were combined with chemometric tools, Principal Components Analysis (PCA) and Hierarchical Cluster Analysis (HCA), capable of performing multivariate data analysis.

## RESULTS

Ag, Cd and Tl were mostly below the level of quantification and were therefore removed from statistical analysis. <sup>87</sup>Sr/<sup>86</sup>Sr varied between 0,71652 and 0,72290. PCA revealed 6 principal components that explained 84,6% of the total variance.

## CONCLUSIONS

The vinification method did not affect significantly stable isotopes, either  $^{87}\text{Sr}/^{86}\text{Sr}$  or  $^{18}\text{O}$ . On the other hand, some elements; namely Cr, Co, Cu, Rb, Se, Ca and Na; showed significant differences between the two types of wine. The produced data can be integrated into an authenticity system for Port and Douro wines.

**Keywords:** Port wine; Douro wine, provenance, multielement analysis, stable isotopes

**Contact person:** Inês Coelho, ines.coelho@insa.min-saude.pt

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# Determination of genetically modified DNA in foods by PNA-functionalized magnetic microbeads as substrates for enzyme-labelled amperometric genoassay

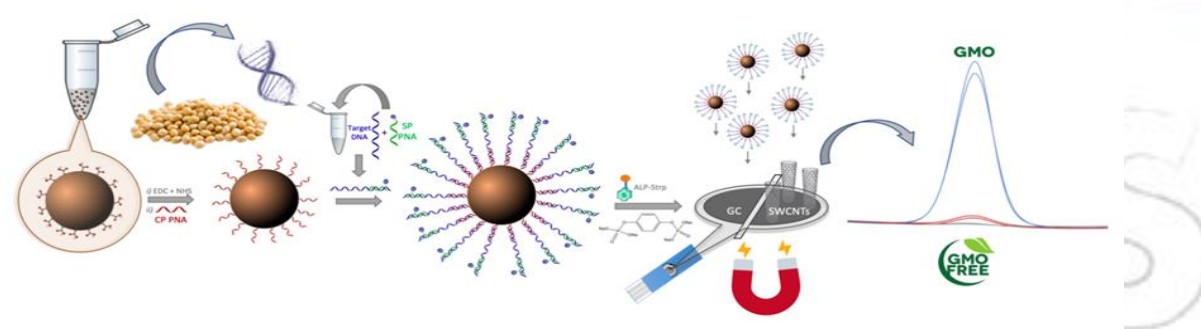
Fortunati S.,<sup>1</sup> Giannetto M.,<sup>1,2</sup> Rozzi A.,<sup>1</sup> Corradini C.,<sup>1,2</sup> Careri M.<sup>1,2</sup> <sup>1</sup> Department of Chemistry, Life Sciences and Environmental Sustainability, University of Parma, Italy, <sup>2</sup> Center on Safety, Technologies and Agri-food Innovation (SITEIA.PARMA)

## AIM

Determination of genomic DNA from genetically modified *Roundup Ready* soy with a rapid and cost-effective amperometric magnetogenoassay.

## MATERIAL AND METHODS

The amperometric magnetogenoassay was implemented on carboxyl-functionalized magnetic microbeads (mMBs) through covalent immobilization of a PNA capture probe (CP) complementary to a target DNA sequence different with respect to the sequence recognized by a biotin-labelled PNA signaling probe (SP) in order to generate a sandwich CP/DNA/SP complex. The assay was applied to genomic DNA extract from European Reference Material soy flours containing different percentages of Roundup Ready soy in wild-type soy flours.



## RESULTS

Enhanced performance with respect to other genoassays requiring preliminary amplification of target DNA *via* PCR was demonstrated. The analytical performance was also improved compared to the immobilization of the same PNA probes on screen-printed electrode (SPE) substrates. Results compared with those obtained using carbon and single walled carbon nanotubes SPE as readout platform highlight the best performance of the nanostructured substrate, reaching LOD and LOQ values of 415 and 995 fM, respectively.

## CONCLUSIONS

The developed electrochemical magnetogenoassay showed outstanding performance if compared to similar approaches recently reported in literature for the determination of DNA from GMO at trace levels, not requiring PCR preamplification of the target sequence. The method overall performance allows to detect and quantify DNA from genetically modified RR soy, below the threshold level established by European Union regulation for labelling of GMO-containing foods.

**Keywords:** Magnetic microbeads; electrochemical genoassay; GMO; soy; food authenticity

**Contact person:** Simone Fortunati, [simone.fortunati@studenti.unipr.it](mailto:simone.fortunati@studenti.unipr.it)

## ACKNOWLEDGMENT

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# Antioxidant properties of white wines from different geographical origins

Jakabová S.<sup>1</sup>, Fikselová M.<sup>2</sup>, Ševčík M.<sup>3</sup>, Aláčová Z.<sup>2</sup>

<sup>1</sup>BioFood Centre, Faculty of Biotechnology and Food Sciences, Slovak University of Agriculture in Nitra, <sup>2</sup>Department of Food Hygiene and Safety, Faculty of Biotechnology and Food Sciences, Slovak University of Agriculture in Nitra, <sup>3</sup>Department of Ecology and Environmental Science, Faculty of Natural Sciences, Constantine the Philosopher University in Nitra

## AIM

The antioxidant properties of white wines from four wine-growing regions in Slovakia were evaluated using the total content of polyphenols and antioxidant activity.

## MATERIAL AND METHODS

Samples of 12 quality quality white wines with PDO (Pinot blanc, Pinot gris and Chardonnay) from 4 wine-growing regions and 3 producers in Slovakia were directly analysed for the total phenolic content and total antioxidant activity. Total phenolic content (TPC) was performed by the Folin–Ciocalteu assay (Lachman et al., 2003). Absorbance was measured with use of UV/VIS spectrometer at 765 nm and results were expressed as gallic acid equivalent (GAE). Total antioxidant activity was measured at 515.6 by modified methodology of Brand-Williams et al. (1995). The results were expressed as a percentage of inhibition DPPH radical. Due to repeated measures design, differences among wine-growing regions were tested using the linear mixed model.

## RESULTS

Phenolic compounds belong to the important characteristics of wines and they are responsible for many differences among red and white wines, even among the wine varieties. The content of phenolic compounds differs considerably in different kinds of wines, depending on the grape variety, environmental and manufacturing factors. The total polyphenolic content ranged from 256.1 mg GAE .dm<sup>-3</sup> in Pinot gris (Malokarpatska wine-growing region) to 403.6 mg GAE .dm<sup>-3</sup> in Chardonnay (Južnoslovenska wine-growing region) samples. The total antioxidant activity ranged from 51.1% (Pinot gris, Malokarpatska wine-growing region) to 72.5% (Pinot gris, Stredoslovenska wine-growing region). By selected statistical evaluation we observed the non-significant difference among wine-growing regions for both monitored parameters: GAE ( $F_{3,8} = 0.56$ ,  $p = 0.66$ ) and DPPH ( $F_{3,8} = 0.33$ ,  $p = 0.81$ ).

**Keywords:** white wines, antioxidants, total polyphenolic content, total antioxidant activity

**Contact person:** Martina Fikselová, [martina.fikselova@uniag.sk](mailto:martina.fikselova@uniag.sk)

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# Authenticity of dairy products by capillary electrophoresis

Masci M.<sup>1</sup>, Zoani C.<sup>2</sup>, Navigato T.<sup>1</sup>, Turrini A.<sup>1</sup>, Jasionowska R.<sup>3</sup>, Caproni R.<sup>1</sup>, Ratini P.<sup>3</sup>

<sup>1</sup> Council for agricultural research and economics (CREA), Research Centre for Food and Nutrition, Rome (Italy), <sup>2</sup> Italian National Agency for New Technologies, Energy and Sustainable Economic Development, Department for Sustainability - Biotechnology and Agroindustry Division (ENEA- SSPT-BIOAG), Rome (Italy) <sup>3</sup> Università degli Studi di Roma La Sapienza, Rome (Italy)

## AIM

After the first studies of Arne Tiselius Electrophoresis had a remarkable development and currently Capillary Electrophoresis (CE) is one of the most powerful and versatile analytical techniques. Thanks to the small use of solvents, fast analysis time and ease of operation, CE showed competitive with conventional separation techniques such as LC or GC, or to better say, it may be complementary to them. Its main value is flexibility. Due to the multiple adjustable parameters it is possible that a wide range of small and large molecules, especially if charged such as proteins, are successfully analyzed.

Milk and derivatives are a very important part in the diet of the world population. Products from goat, buffalo and sheep species have a greater economic value than the cow ones therefore authenticity frauds occur frequently: dairy products are among the seven more attractive foods for adulteration. Each milk from the above-cited animal species has its own definite pattern of whey proteins, essentially variants of b-lactoglobulin ( $\beta$ -LG) and a-lactalbumin ( $\alpha$ -LA) that can be usefully exploited as markers of authenticity by means of CE.

The Research Infrastructure METROFOOD-RI (ESFRI Roadmap 2018, domain Health and Food) includes among its main focuses research activities on authenticity of raw materials and products. In this frame, the application of Capillary Electrophoresis is also planned in support to protection and valorisation of typical products of great economic and cultural value. Such a facility is planned to be included specifically in the Italian Node of the infrastructure.

## MATERIAL AND METHODS

The unique CE feature of electroosmotic flow (EOF) minimizes band broadening. Because the EOF essentially originates near the capillary walls it has a flat flow profile which is different from the parabolic flow profile usually observed in the pressure-induced flows, such as in the case of HPLC for example. The flat profile of EOF is the main reason for the high resolution and efficiency in CE. From the few works on dairy products CE showed itself effective in separating the genetic variants of  $\beta$ -LG and  $\alpha$ -LA.

This work presents case studies on the authenticity of cheeses found by local producers and on the profile of some milk mixtures. Sample preparation simply required the adding of an acidic solution for milk samples and of distilled water for cheese samples. After centrifugation and filtration, samples were ready for CE analysis in CZE mode (Capillary Zone Electrophoresis). Electrophoretic runs were carried out by means of an alkaline buffer and the UV detection which allowed the complete CE resolution of proteins of interest in few minutes.

**Keywords:** Food authenticity; Capillary Electrophoresis; METROFOOD-RI; Dairy products

**Contact person:** Maurizio Masci, maurizio.maschi@crea.gov.it

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METROFOOD-RI, Infrastructure for promoting metrology in food and nutrition.  
<https://www.metrofood.eu/>



# Optimization of HPLC method for determination of cholesterol

Kolarič L.<sup>1</sup>, Šimko P.<sup>1</sup>

<sup>1</sup> Slovak University of Technology, Faculty of chemical and food technology, Institute of Food Science and Nutrition, Radlinského 9, 812 37 Bratislava, Slovakia

## AIM

The aim of this work was to evaluate the optimal conditions for determination of cholesterol by HPLC chromatography when optimal composition of mobile phase, its influence on the retention time, absorption wavelength, and limit of quantification have been studied.

## MATERIAL AND METHODS

HPLC analysis was performed using an Agilent Technologies 1260 infinity system equipped with a UV-DAD detector. Isocratic elution was performed at a flow rate of 0.5 mL.min<sup>-1</sup> using the various mobile phase composed of water, methanol, and acetonitrile. Cholesterol standard was from Sigma-Aldrich with a purity ≥99%. The injection volume was 10 µL and the temperature was set at 30 °C. As a stationary phase, a Zorbax Eclipse Plus SB-C<sub>18</sub> column (2.1x100 mm, 3.5 µm particle size) was used with the guard column Zorbax SB-C<sub>18</sub> (4.6x12.5 mm, 5 µm particle size). The results were recorded using the OpenLab CDS software, ChemStation Edition for LC and LC/MS systems (product version A.01.08.108).

## RESULTS

The main parameters, which influence the effectivity of liquid chromatography are the composition of mobile phase, type of stationary phase, or the size particle of column. The cholesterol content can be determined by several analytical methods but the most used are gas or liquid chromatography. In the HPLC method, most of the mobile phases are composed of acetonitrile, isopropanol, or methanol<sup>1</sup>. In our study, various mixtures of mobile phases were tested including water, acetonitrile, and methanol. The best results were achieved using acetonitrile/methanol (60:40, v/v). Ahn et al.<sup>2</sup> used ethanol with methanol (50:50), and Borkovcová et al.<sup>3</sup> methanol with water (95:5). The retention time of cholesterol was 5,701 min, what is similar to previous study<sup>1</sup>. The absorption maximum of cholesterol peak was set at 205 nm.

## CONCLUSIONS

It can be concluded that the optimal mobile phase for cholesterol determination by HPLC is mixture of methanol and acetonitrile (40:60, v/v), with the retention time of cholesterol in 5,701 min at the flow rate of 0.5 mL.min<sup>-1</sup> and the absorption wavelength at 205 nm. At these conditions, the LOQ was 24.21 µg.ml<sup>-1</sup>.

**Keywords:** HPLC; cholesterol; optimization

**Contact person:** Lukáš Kolarič, lukas.kolaric@stuba.sk

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# Variations in the fatty acids profile of the meat by adding hempseed cake in the diet of multiparous cull cows

Pianezze S.<sup>1,2</sup>, Perini M.<sup>2</sup>, Sepulcri A.<sup>1</sup>, Baldini M.<sup>1</sup>, Fabro C.<sup>1</sup>, Piasentier E.<sup>1</sup>, Corazzin M.<sup>1</sup>

<sup>1</sup> Department of Agricultural, Food, Environmental and Animal Sciences, University of Udine, Udine (UD), <sup>2</sup> Fondazione Edmund Mach, Technology Transfer Centre, San Michele All'Adige, Trento (TN)

## AIM

In the present work, eighteen multiparous cull cows belonging to Italian Simmental breed were considered. The cows were divided into two groups, group A whose diet was hay-based and group B whose diet was corn silage-based. Within the same group, whether A or B, the diet of half cows was supplemented with a concentrate containing hempseed cake (5% of the diet dry matter DM), whereas the others were provided with soybean meal instead. The hempseed cake, a by-product of hempseed oil production process, is rich in fat (around 10% of the DM) and has a favourable fatty acids (FA) profile, having unsaturated FA for over the 80% of the total FA (Baldini et al., 2018).

The aim of this work was to check whether the hempseed cake could improve the fatty acids profile of multiparous cull cows with respect to soybean meal.

## MATERIAL AND METHODS

The total lipid fraction was extracted from samples of *m. longissimus thoracis* by following the procedure reported elsewhere (Folch et al., 1957). Fatty acids were esterified, methylated and quantified through GC-MS (GCMS 5977E, Agilent Technologies). The separation was performed through a HP 88 column (100 m x 0.25 mm x 0.25 µm). The FA composition was expressed as percentage of total FA and reported as mean ± standard error (SE). The statistics was carried out by using the software R vers. 4.0.0 with a model that considered the experimental factor (hempseed cake vs soybean meal) as fixed, and the type of forage (whether hay or corn silage) as block factor.

## RESULTS

The results obtained for the hempseed cake group, considering the FA mainly present in meat, C16:0 (27.08 ± 0.59%), C18:0 (19.44 ± 0.78%), C18:1n-9c (35.31 ± 0.49%) as well as the FA favourable for the human health such as C18:3n-3 (0.36 ± 0.02%) and total polyunsaturated fatty acids (PUFA) (3.98 ± 0.17%), were comparable to those found for the soybean meal group (P>0.05). The n-6/n-3 ratio was not affected by the use of hempseed cake either (7.79 ± 0.74%; P>0.05). On the other hand, C14:0 turned out to be statistically higher (3.47 vs 3.03; P<0.05) for the group provided with hempseed cake. In conclusion, from an overall point of view, the FA profile of meat was not improved by adding hempseed cake in the diet of Italian Simmental cull cows.

**Keywords:** fatty acids; cows; gas chromatography-mass spectrometry

**Contact person:** Pianezze Silvia, mail: [silvia.pianezze@fmach.it](mailto:silvia.pianezze@fmach.it), tel.: +39 0461615160

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# The effect of frying oil on the fat content of fish fingers and potato croquettes and its stability

Soukup J.<sup>1</sup>, Kouřimská L.<sup>1</sup>

<sup>1</sup> Czech university of Life Sciences (CULS), Faculty of Agrobiolgy, Food and Natural Resources, Department of Microbiology, Nutrition and Dietetics, Kamýcká 129, 165 00, Suchdol, Prague, Czech Republic

## AIM

Examine whether the type of frying oil and repeated frying have an effect on the fat content in the fried food.

## MATERIAL AND METHODS

Three different types of oils (rice, rapeseed, sunflower) were used as a frying medium, and pre-fried fish fingers and potato croquettes were prepared in them. After frying, the samples were frozen at -80 °C and then lyophilized. Lyophilized samples were homogenized for 1-2 minutes using the IKA laboratory grinder. In total, 66 samples were analysed.

### Determination of dry matter and ash content

Determination of dry matter and ash content was done gravimetrically. About 2.5 g of the homogenised sample was dried in the laboratory oven at 103±2 °C to the constant weight. Additionally, the sample was mineralised in a muffle furnace to the temperature 550 °C. Control samples were calculated from three parallel samples.

### Determination of fat content

Determination of fat was based on the Soxhlet method using the Soxhlet extractor. About 2.5 g of the homogenised samples were extracted by 70 ml of petroleum ether. The residuum of extraction solvent was then evaporated at 103 °C for 2 h and weighted after cooling down. Fat content analysis was carried out in duplicate.

### Determination of oil stability during storage

The oils were stored in open 100mL beakers at 60 °C and the weight changes indicating the amount of oxygen absorbed in the oil were recorded. The increase in mass reflects the degree of oxidation of the monitored oil.

## RESULTS

The results of individual samples after frying showed neither increasing nor decreasing tendencies in any of observed factors. During the storage, rice oil and rapeseed oil showed the highest stability, the sunflower oil was on the other hand really unstable and easily oxidised. Because the rice oil also soaked the least amount of fat in case of fish fingers, it seemed to be the best option. It was not shown in case of potato croquettes – sunflower oil soaked less.

## CONCLUSIONS

The difference between fat content before and after frying was not significant. The least amount of fat was soaked in rice oil when frying fish fingers and sunflower oil when frying potato croquettes. There were no differences in samples during the repeated frying. Rice oil showed the highest oxidative stability during the storage at increased temperature, sunflower oil was the least stable.

**Keywords:** frying, oil, fat, fish fingers, potato croquettes

**Contact person:** Josef Soukup, soukupjosef@af.czu.cz, +420 777 572 119

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# Food Research Institute Prague – part of the European Research Infrastructure METROFOOD

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Šmídová Z., Šavrdová M., Laknerová I., Urban M.

Food Research Institute Prague, p.r.i., Radiová 7, Prague 10200, Czech Republic

## AIM

METROFOOD-RI as a global multidisciplinary research infrastructure joins together 18 European countries with 48 partner institutions. It is aimed at metrology in food and nutrition and operates in various areas – agriculture, sustainable development, food quality and safety, food traceability and authenticity, environmental safety and human health. The Czech national node of METROFOOD-RI is represented by METROFOOD-CZ, Infrastructure for Promoting Metrology in Food and Nutrition in the Czech Republic. METROFOOD-CZ makes part of large research infrastructures in the Czech Republic. The partners in the Czech Republic include Czech University of Life Sciences, University of Chemical Technology and Food Research Institute Prague (FRIP).

The equipment of the FRIP Institute allows research in the field of new products development and new technological challenges in the area of food processing, storage and preservation. Within FRIP various facilities are included in the Metrofood infrastructure: Accredited Testing Laboratory, devices for food analysis and processing - high-pressure processing machine, spray drying, rheological instruments, bioreactors. The Accredited Testing Laboratory (ISO 17025) promotes scientific excellence in the field of food allergens analysis, e.g. gluten, casein, soya proteins, buckwheat allergen, mustard allergens and peanut allergens. This Laboratory operates with approx. 100 analyses a month of products from the leading Czech food producers. The innovative method high-pressure processing enables delicate conservation of foods (e.g. juices) without heating. Thus, the products keep and preserve their natural colour, appearance, taste, smell as well as nutritional value. The products have prolonged lifetime and are stored in a cold place.

**Keywords:** Metrofood, Food Research Institute Prague, Accredited Testing Laboratory, food, allergens, high-pressure processing

**Contact person:** Zuzana Šmídová, zuzana.smidova@vupp.cz

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# Policies and strategies in food and healthy lifestyles

Jesus s.<sup>1</sup>, Coelho M.<sup>1</sup>, Dias G.<sup>1</sup>, Castanheira I.<sup>1</sup>

<sup>1</sup> National Institute of Health Doctor Ricardo Jorge

## AIM

The rapid growth in industrialisation, urbanisation and market globalisation had a significant impact on population diets and lifestyles. At the same time, the living standards improved, the food availability expanded and became more diversified. However, it occurred an increase in dietary patterns and physical inactivity, which led to a rise in the chronic non-communicable diseases (NCDs), including obesity, diabetes, cardiovascular disease, hypertension, stroke and cancer. It was determined that in 2001 chronic burden diseases contributed 46% in the world and that in 2020 will increase to 57%. So, the present study aimed to analyse the strategies and policies in health and food lifestyles in Europe. The study was developed in the project "A Quadruple Helix Atlantic Area Healthy Food Ecosystem for Growth of SME's" (AHFES). This project aims to improve the competitiveness and growth of SMEs in the value chain of healthy eating and lifestyles, contributing to the development of a transnational innovation ecosystem that helps entities to access information, partners and markets and align their products and services to the needs and expectations of the European consumer/community.

## MATERIAL AND METHODS

In the present study was used different approaches and tools. It was made a literature review with the terms of "healthy food policies", "healthy lifestyles policies" and "food policy". Another approach was the information collected from the partners about the existing regional policies and strategies in healthy food, where it was made some questions about healthy food and lifestyles and innovation policies, innovation tools and existing support structures and reforms in each country (Portugal, Spain, Ireland, France and the United Kingdom).

## RESULTS

The countries increasing the policies and measures to be implemented in the area of health. It was found that although most countries have very similar national plans, their policies and levels of implementation vary between them.

In general, the implemented measures and policies are organized into two groups: health and lifestyles programmes and policies and support structures and reforms. These groups have high relevance in changing dietary patterns and lifestyles. Besides, in most countries, there was a consensus on the most important and priority age groups for a more effective outcome of measures and policies in the future. These two age groups were children, who are still open to new knowledge and knowledge in the area of food, and the elderly who corresponds to a group that requires the effects of NCD risk factors to be controlled. The measures and policies applied to increase and encourage society to modify its diet and lifestyle were diverse and can be grouped into five groups: nutrition and diet counselling, portion size control, regulation of marketing to children, reformulation and dietary guidelines.

## CONCLUSIONS

To improve healthy food and lifestyles of the population, it is necessary to regularly re-evaluate strategies and consequent effects on the health status of the community. Besides, to the

implementation of strategies and policies to be successful, there must be cooperation between the government-university-industry and community. AFHES project is an important tool to assist SME in the production of foods framed by new health guidelines.

**Keywords:** Healthy food, Food policies, Healthy lifestyles

**Contact person:** Susana Jesus, [susana.jesus@insa.min-saude.pt](mailto:susana.jesus@insa.min-saude.pt)

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# EPA and DHA content in fish oil supplements on the Italian market: A preliminary study

Navigato T.<sup>1</sup>, Masci M.<sup>1</sup>, Turrini A.<sup>1</sup>, Zoani C.<sup>2</sup>, Caproni R.<sup>1</sup>

<sup>1</sup> Council for agricultural research and economics (CREA), Research Centre for Food and Nutrition, Rome (Italy), <sup>2</sup> Italian National Agency for New Technologies, Energy and Sustainable Economic Development, Department for Sustainability - Biotechnology and Agroindustry Division (ENEA- SSPT-BIOAG), Rome (Italy)

## AIM

Despite a consistent fish consumption is widely accepted as preventing coronary heart diseases, positively acting towards other important pathologies (such as cancer) and helping brain and retina development, food supplements fish oil-based are not universally recognized to have the same beneficial effects. Some authors suggest that the easy oxidation that these supplements go through invalidates the goodness of the experiments giving conflicting results. The food supplement market in Italy has grown a lot in recent years with a turnover of 3.3 billion Euros in 2018. Italy ranks first in Europe in the consumption of food supplements with a 23% share of the European market being adults the main consumers, with a prevalence of women. Omega3-based supplements are a good part of such a market with the two Omega3 fatty acids Docosahexaenoic (DHA) and Eicosapentaenoic (EPA) as the molecules of the most value in these products since mainly to them are attributed the positive effects on health. There are very few papers in the literature about the quality control of fish oil capsules and none of them refer to the Italian market. The existing works cite that in some cases significant high amounts of linoleic acid are detected, which indicates a probable product adulteration. In any case, the EPA and DHA content as declared on the label needs to be specifically checked given the related health implications and the money involved. In the present work, an analytical control on fish oil capsules purchased on the market was conducted: to our knowledge this is the first time such a control is performed on products from Italian commercial distribution.

## MATERIAL AND METHODS

Three Omega3 supplements of different brands were purchased from retailers. The capsules were opened and approximately 10 mg of oil were methyl derivatized. For each sample the instrumental analysis was conducted by using Gas Chromatography-Mass Spectrometry.

## RESULTS SAMPLES

1 and 2 both showed an EPA content of about 80% and a DHA content of about 100% of what was declared on labels. Sample 3 showed EPA and DHA content in compliance with the label. It was already observed by other authors an EPA content of about 80% of what was declared for commercial Omega3 supplements.

## CONCLUSIONS

This preliminary study shows that a more thorough check on Omega3 supplements would be appropriate because it is possible that there are products on the market with a composition differing from what was declared by the producer.

**Keywords:** Omega3 supplements; Fatty acids; EPA; DHA; Quality control;

**Contact person:** Teresina Navigato [teresina.navigato@crea.gov.it](mailto:teresina.navigato@crea.gov.it)



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# Is the nutrition of the Czech badminton representatives adequate?

Sabolová M., Bašová A.

Department of Microbiology, Nutrition and Dietetics, Czech University of Life Sciences Prague

## AIM

The aim of this work was to compare real and recommended energy and nutrients intake of the Czech professional badminton players.

## MATERIAL AND METHODS

The one-week food intake records were obtained from 6 professional badminton players (3 men, 3 women). Their intake of energy and selected nutrients were calculated using Nutriservis application. The obtained data were compared with the recommended intake values for athletes and for the general population.

## RESULTS

The intake of energy, carbohydrates, fiber, vitamin D, iron, calcium, and sodium was significantly lower than the recommended values for athletes ( $p < 0.05$ ). Insufficient intake of the energy and vitamin D was also reported when compared to the recommendation for the general population ( $p < 0.05$ ). On the other hand, intake of proteins, fats, and vitamins C was optimal.

## CONCLUSIONS

The results showed that the energy and nutrients intake of the Czech badminton representatives corresponded to the recommended intake of the general population. However, their real dietary intake did not meet the dietary needs of athletes. **Keywords:** nutrient intake; sport; diet plan; performance

**Contact person:** Monika Sabolová, sabolova@af.czu.cz

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# Non-coeliac gluten sensitivity

Žatečka L.<sup>1</sup> Sabolová M.<sup>1</sup>

<sup>1</sup> Czech University of Life Sciences Prague (CZU)

## AIM

The aim of the study was to investigate the effect of consumption of gluten and easily fermentable carbohydrates on the intensity of negative symptoms, anthropometric parameters and blood pressure.

## MATERIAL AND METHODS

This was investigated using an observational clinical study involving 9 volunteers. The study lasted 14 weeks and consisting of two phases. The first phase included a gluten-containing diet and a gluten-free diet, as well as double-blind exposure weeks and the second phase included a diet with a reduced amount of easily fermentable carbohydrates followed by an exposure test. The intensity of negative symptoms using a visual analogue scale, anthropometric parameters and blood pressure of all volunteers were regularly measured.

## RESULTS

The results of the observational clinical study showed that neither gluten nor easily fermentable carbohydrates had a significant effect on the intensity of negative symptoms in our volunteers. Although a positive effect on symptom intensity has been shown in the case of gluten-free diet, this effect is not confirmed in a double-blind placebo challenge test. The intensity of the negative symptoms was assessed subjectively using a visual analogue scale. The results of the study also showed that there was no significant change in anthropometric parameters and blood pressure due to the consumption of a gluten-free diet and a diet with a low content of easily fermentable carbohydrates.

## CONCLUSIONS

According to our study, it appears that role of gluten and easily fermentable carbohydrates in non-coeliac gluten sensitivity is overemphasized, however further studies are needed.

**Keywords:** Non-coeliac gluten sensitivity, gluten, gluten-free diet, easily fermentable carbohydrates, visual analogue scale, anthropometric parameters

**Contact person:** Ladislav Žatečka, ladislav.zatecka@zateckal.cz

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# 5<sup>th</sup> IIMEKO FOODS

## Omega-3 recovery from food processing waste

Massimo Lucarini, Barbara Benedetti, Alessandra Durazzo, Stefano Nicoli, Aida Turrini, Gabriella Di Lena and Ginevra Lombardi-Boccia

CREA-Research Centre for Food and Nutrition, Via Ardeatina 546, 00178 Rome, Italy

The current food production system generates a large amount of waste and nutrients loss. This productive approach has a strong environmental impact and now different strategies are addressed toward a more rational exploitation of food resources. This objective involves the recovery and recycle of nutrients and molecules of interest from food waste.

Regarding the sustainable management of food value chain, in this study an approach to recycle wastes coming from marine food-chain is described. By now it is of increasing importance to look at food waste as a sustainable resource of nutrients and molecules of nutritional interest to be recycled and re-utilised as active principles. In particular, the growing awareness to increase the levels of n-3 PUFA intake, has focused the attention on the importance of fish as a natural supplier of these molecules to the diet. The case-study here reported deal with the recovery of omega-3 fatty acids (EPA, DHA) from the fish food-chain, valorising fisheries wastes unsuitable for human consumption but rich of valuable and healthy nutrients. Chemical analyses aimed at achieving a valorisation of fish waste through the recovery of fish oil and bioactive compounds (omega-3: EPA, 20:5 n-3 and DHA, 22:6, n-3) were carried out. The fish by-products utilised (heads, fins, bowels, bones, skin) were analysed for their fatty acids profile by GC/MS and, subsequently and by Fourier Transformed Infrared Spectroscopy (FTIR) on Attenuated Total Reflectance (ATR). The application of these methodologies allows development of standardised methodology, the harmonisation of data and, as well, data management. The green-type of extraction and analyses of omega-3 fatty acids is part of the innovation of the sustainable process adopted, addressed to reduce the introduction of hazardous substances in food products in order to give safer products to the end-users. Sustainable development of agri-food sector can highly gain from metrology on characterization of agri-food matrices particularly concerning the valorisation of by-products and food wastes to promote circular economy and industrial symbiosis. METROFOOD-RI, as research infrastructure aimed at improving the quality and reliability of measurement results in food science and make available and share data, information and metrological tools, comprises an important cross-section of highly interdisciplinary and interconnected fields throughout the food value chain. In this frame, it addresses the concept of circular economy with reference to the potential in valorising by-products of food processing, re-including them in the food production system and transforming them into high added value, food grade and fully functional food ingredients, as well as the concept of industrial symbiosis with their re-use in non-food production chains (e.g. supplements, pharmaceutical, cosmetics). Results of the present activity provide some interesting information as scientific background for the implementation of the research infrastructure currently in its Preparatory Phase.

**Keywords:** metrology, research infrastructure, food quality, food safety, food waste.

# METROFOOD: an Infrastructure for Promoting Metrology in Food and Nutrition

Massimo Lucarini, Barbara Benedetti, Alessandra Durazzo, Stefano Nicoli, Aida Turrini and Ginevra Lombardi-Boccia

<sup>1</sup>CREA-Research Centre for Food and Nutrition, Via Ardeatina 546, 00178 Rome, Italy

## AIM

The Project METROFOOD-PP (Preparatory-Phase) aims to promote Metrology in Food and Nutrition. METROFOOD includes a consortium of about 50 Research Institutes from 18 different countries that sees Italy, in the figure of ENEA, as coordinator. METROFOOD is intended as an infrastructure aimed at improving the quality and the reliability of measurements in the field of food quality and safety; it also aimed to share data, strengthening scientific knowledge and promoting cooperation and integration of the whole scientific community, providing services at territorial level working on the reliability and of the harmonization of the procedures, adopting the FAIR approach (Findable, Accessible, Interoperable, Re-usable) in data management. The project has a multidisciplinary approach that integrates the skill of multiple actors in the entire supply chain, comprising an important cross-section of highly interdisciplinary and interconnected fields throughout the food value chain, including agrifood, sustainable development, food safety, quality, traceability and authenticity, environmental safety, and human health. The METROFOOD network will be a physical and electronic infrastructure. The Physical Infrastructure consists of a network of facilities for the production of reference materials and analytical laboratories for the characterization of foods and matrices of interest for the agro-food sector (METRO) and a network of experimental fields and technological systems for the production and transformation of food products (FOOD).

Regarding the sustainable management of food value chain, in this study an approach to recycle wastes coming from marine food-chain is described. By now it is of increasing importance to look at food waste as a sustainable resource of nutrients and molecules of nutritional interest to be recycled and reutilised as active principles. In particular, the growing awareness to increase the levels of n-3 PUFA intake, has focused the attention on the importance of fish as a natural supplier of these molecules to the diet. The case-study here reported deal with the recovery of omega-3 fatty acids (EPA, DHA) from the fish food-chain, valorizing fisheries wastes unsuitable for human consumption but rich of valuable and healthy nutrients. Chemical analyses aimed at achieving a valorization of fish waste through the recovery of fish oil and bioactive compounds (omega-3: EPA, 20:5 n-3 and DHA, 22:6, n-3) were carried out. The fish by-products utilised (heads, fins, bowels, bones, skin) were analysed for their fatty acids profile by GC/MS and, subsequently and by Fourier Transformed Infrared Spectroscopy (FTIR) on Attenuated Total Reflectance (ATR). The application of these methodologies allows development of standardised methodology, the harmonisation of data and, as well, data management. The green-type of extraction and analyses of omega-3 fatty acids is part of the innovation of the sustainable process adopted, addressed to reduce the introduction of hazardous substances in food products in order to give safer products to the end-users.

**Keywords:** Metrology, research infrastructure, food quality, food safety, food waste.

# Joint Research Unit – METROFOOD-MK and its Contribution to Food Safety and Quality

Najdenkoska A.<sup>1</sup>, Arsova Sarafinovska Z.<sup>1</sup>, Velkoska-Markovska L.<sup>2</sup>, Jankulovska S. M.<sup>2</sup>.

<sup>1</sup>Institute of Public Health of the Republic of North Macedonia, 50 Divizija 6, Skopje, North Macedonia, <sup>2</sup>“Ss. Cyril and Methodius” University in Skopje, Faculty of Agricultural Sciences and Food – Skopje, 16-ta Makedonska Brigada, 3, 1000, Skopje, North Macedonia

## AIM

The North Macedonian Node within the European Research Infrastructure METROFOOD-RI consist of two institutions: Institute of Public Health (IJZRSM) and Faculty of Agricultural Sciences and Food (FASF), named as Joint Research Unit – METROFOOD-MK. The node's laboratories are equipped with: gas chromatography systems with different type of detectors: MS, NPD, ECD, FID; liquid chromatography systems with different type of detectors: DAD, UV, RI than, Graphite Furnace Atomic Absorption Spectroscopy, Flame Atomic Absorption Spectroscopy system, Cold vapor (flow injection mercury system). Within the METROFOOD-PP activities, it is defined what services METROFOOD-MK Node will provided in characterization of RMs and food safety&quality analysis. For the food safety the following parameters are selected: inorganic contaminants (trace elements), organic contaminants (pesticides and mycotoxins), allergens, additives, and microbiological parameters.

As the most important parameter in estimating food quality is determination of food composition in terms of content of proteins, fat, fatty acids, carbohydrates, fibers (total, crude), vitamins, micro- and microelements and providing information about nutritional values, physico-chemical analysis, bioactive compounds and adulteration. Determination of nutritional value is important as consumers could be informed which nutrients are present in the certain food and how much energy they provide. General food labelling is governed by Directive 2000/13/EC, while Nutrition labelling of food is regulated by Directive 90/496/EEC. Standardised methods for determination of food composition, are used. Kjeldahl method has been applied to determine proteins / nitrogen content. For determination of total fats has been used: Soxhlet, Weibull-Stoldt and Rose-Gottlieb Method. Fibers will be determined through the enzymatic method. Total carbohydrate content is calculated by difference, rather than analyzed directly. Under this approach, the other constituents in the food (protein, fat, water, alcohol, ash) are determined individually, summed and subtracted from the total weight of the food.

According to ISO/IEC 17025 laboratories shall have quality control procedures for monitoring the validity of tests. This monitoring may include the participation in interlaboratory comparisons or proficiency testing programmes. By these mechanisms the laboratory can provide evidence of its competence to its clients, interested parties and the accreditation body. In order to maintain accreditation and prove competence the MK - Node laboratories participated in many PT schemes, organized by the international PT providers. The evaluation of the PT results showed that the results, about the food quality and safety, are with z – score below 2. Furthermore, during the "Early Phase" of the METROFOOD-RI, the MK - Node have participated in a pilot service dedicated to characterization of a new RMs of rice grains, rice flour and oyster tissue, particularly for

determination of food composition. Given that accredited methods were used for that purpose, the new food matrix-Reference Materials (RMs) will be characterized with acceptable reference value.

**Keywords:** METROFOOD-RI, metrology, food quality and safety.

**Contact person:** Anita Najdenkoska; email: a.najdenkoska@iph.mk

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FOODS



# Use of a certified reference material (CRM) of metals in hydrobiological products in a laboratory intercomparison program

Sandoval S.<sup>1</sup>, González K.<sup>1</sup>, Muñoz L.<sup>2</sup>

<sup>1</sup> Designated Metrology Laboratory- RNM, Sección Metrología Ambiental y de Alimentos, Subdepartamento de Metrología y Desarrollo Tecnológico, Departamento de Salud Ambiental, Instituto de Salud Pública de Chile (ISP), Santiago, Chile., <sup>2</sup> Chemical Metrology Laboratory, Comisión Chilena de Energía Nuclear (CCHEN), Nueva Bilbao 12501, Santiago, Chile.

## AIM

Prepare and evaluate a certified reference material for arsenic and cadmium in a hydrobiological food matrix for purposes of its use in an interlaboratory-type proficiency test for national laboratories of control of fish and fishery products.

## MATERIAL AND METHODS

A batch 4 kg of fishmeal was dried, sieved and homogenized by Laboratory of ISP. Homogeneity (n=10) and stability (n=5) was controlled by the ISP. For characterization 10 samples of the material were analyzed by ISP and CCHEN. The ISP was in the charge by Inductively Coupled Plasma-Mass Spectrometry (ICP-MS) for microwave digestion, the CCHEN was responsible for the execution of the tests following the Neutron Activation Analysis (NAA), ICP-MS, and Isotope Dilution Mass Spectrometry (ID-MS). They were used calibration standards traceable to the SI: Standard Solution of Cadmium SRM® 3108 and Arsenic SRM® 3103a. For quality control of analysis to samples used the *Fish protein certified reference material for trace metals* DORM -4 of NCR-CNRC. Humidity was determined by gravimetric method and the coulometric Karl Fischer method using a certified standard oven 1%, Apure®, Merck.

## RESULTS

Certified values were established for the processed reference material of arsenic 2.64 +/- 0.35 mg/kg (U k = 2) and cadmium 0.86 +/- 0.11 mg/kg (U k = 2). Its homogeneity and stability is suitable for use in a round of proficiency tests for national food control laboratories. The results for the mean of the 11 participating testing laboratories were 2,160 mg/kg for arsenic and 0.863 mg/kg for cadmium. The results for the median of the participants were 2,114 mg/kg for arsenic and 0.863 mg/kg for cadmium. The performance values of the participants were evaluated with the z-score statistic, obtaining a satisfactory result of 60 % for the arsenic element and 73 % for cadmium.

## CONCLUSIONS

The material demonstrated its adequacy by meeting the requirements established in the ISO/IEC 17034 and ISO/IEC 17043:2010 standard, to be used in intercomparison tests. The results of the performance assessment revealed that only 60% of the laboratories obtained unsatisfactory results for arsenic and 73 % for cadmium, which reveals that control processes in laboratories that conduct this assays must be improved, according to requirements of the ISO/IEC 17025 standard, they should set corrective actions to improve their process. Intercomparison tests are tools that allow to State and accreditation organizations to evaluate the performance of the authorized or accredited laboratories and this way request the necessary improvements so that their results are reliable and comparable for decision making

**Keywords:** reference material; CRM; fish; proficiency testing.

**Contact person:** Soraya Sandoval, soraya@ispch.cl, +56 2 25755498.

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# Active food packaging joins quality by design for the development of antimicrobial materials containing essential oil–based cocrystals

Bianchi F.<sup>1,2</sup>, Fornari F.<sup>1</sup>, Riboni N.<sup>1,3</sup>, Spadini C.<sup>4</sup>, Cabassi CS.<sup>4</sup>, Carraro C.<sup>1</sup>, Mazzeo PP.<sup>1</sup>, Bacchi A.<sup>1,5</sup>, Orlandini S.<sup>6</sup>, Furlanetto S.<sup>6</sup>, Careri M.<sup>1,7</sup>

<sup>1</sup> Department of Chemical Sciences, Life Sciences and Environmental Sustainability, University of Parma, Italy, <sup>2</sup> Interdepartmental Center for Packaging (CIPACK), Parma, Italy, <sup>3</sup> Center for Energy and Environment (CIDEA), Parma, Italy, <sup>4</sup> Department of Veterinary Sciences, University of Parma, Italy

## AIM

Use of a Quality by Design (QbD) strategy to develop plastic packaging containing essential oil (EOs) cocrystals exerting antimicrobial properties towards bacterial strains involved in fruit and vegetable deterioration.

## MATERIAL AND METHODS

The EO-based cocrystals, namely carvacrol, eugenol, thymol and cinnamaldehyde, were prepared in bulk by grinding or direct mixing methods [Mazzeo, 2019]. Antimicrobial activity of the EOs was tested over a pool of both Gram-positive and Gram-negative bacteria, namely *Escherichia coli*, *Salmonella Typhimurium*, *Staphylococcus aureus*, and Methicillin-resistant *Staphylococcus aureus* [European Commission, 2002]. Both minimal inhibitory concentration and time-kill assays were performed. The effect of critical parameters represented by the concentration of the four EOs in a mixture on critical quality attributes related to the percentage of microbial inhibition was investigated by QbD. For this purpose, a Box-Wilson central composite face-centered design was performed. Monte-Carlo simulations were carried out to draw probability maps, then a Plackett-Burman design was used to validate the design space. Pack prototypes were prepared using LDPE substrates coated with chitosan and EO-based cocrystals. GC-MS analyses were carried out to evaluate the release of EOs along the time. The antimicrobial activity of the prototypes was assessed both by contact and in gas phase. Finally, the shelf-life of fruit samples was investigated for a 5-days period.

## RESULTS

The QbD strategy allowed the optimization of the concentration of the EO-based cocrystals to achieve the simultaneous inhibition of the investigated microorganisms. The use of cocrystallization resulted in the prolonged release of the antimicrobial agents compared to the use of pure EOs, obtaining a steady concentration for a minimum of 14 days. The antimicrobial activity of plastic packaging was proved with and without the need of a direct contact with the culture medium. Finally, an improvement of the organoleptic properties and a reduction of the total microbial charge were obtained for fresh fruit stored using the cocrystal-based pack.

## CONCLUSIONS

For the first time, QbD was applied to food packaging materials development, making it possible to manage the optimization by a systematic risk-based approach. The developed active packaging can be considered a promising candidate for shelf-life prolongation of perishable foodstuffs.

**Keywords:** Quality by design; Cocrystals; Essential oils; Packaging; Shelf-life

**Contact person:** Federica Bianchi, federica.bianchi@unipr.it

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# ***Clitoria ternatea* extract as a source of antioxidant compounds in chitosan based edible films and their intelligent properties**

Jancikova S.<sup>1</sup>, Dordevic D.<sup>1</sup>, Tesikova K.<sup>1</sup>, Antonic B.<sup>1</sup>, Tremlova B.<sup>1</sup>

<sup>1</sup> Department of Plant Origin Food Sciences, University of Veterinary and Pharmaceutical Sciences Brno, Palackeho tr. 1946/1, 612 42, Brno, Czech Republic

## **AIM**

The aim of the study was to add a water based *Clitoria ternatea* (also called blue tea) extract to the chitosan films and measured antioxidant potential of experimentally produced edible packaging. Another part of the research is focused on the determination of intelligent properties due to the presence of anthocyanins responsible for changing colour in different pH (Kungsuwan et al., 2014; Netramai et al., 2020).

## **MATERIAL AND METHODS**

Edible films consisted of chitosan, glycerol and *Clitoria ternatea* extract in the following concentrations 5, 10, 20, 30 and 40 % were prepared. DPPH assay was used as a determination of antioxidant properties. DPPH solution in ethanol was mixed with 3 mL of ethanolic extract of films and incubated for 30 minutes in dark. The absorbance was measured at 517 nm and DPPH scavenging activity was expressed as a percentage. The polyphenol content was measured with Folin-Ciocalteu solution. The absorbance was measured at 765 nm and the polyphenol content was expressed as an amount of gallic acid (mg/g). The intelligent properties were analysed by dipping films in 1M HCl and 1M NaOH.

## **RESULTS**

It was found out that films with *Clitoria ternatea* extract do not have a high antioxidant activity, the highest results for DPPH was around 3 %, which is very low, in comparison with the article by Jancikova et al. (2019):  $87.84 \pm 0.07$  % for carrageenan films with 20 % of dried rosemary extract. Total polyphenol content as an indicator of antioxidant activity is also not too high (max.  $14.85 \pm 0.65$  mg gallic acid/g of sample). The best intelligent properties were confirmed in the films with 40 % of extract. Film's colour in neutral pH was blue, in acidic pink and in basic green.

## **CONCLUSIONS**

Prepared films with addition of blue tea extract are not a good source of polyphenolic and antioxidant compounds. In the future researches the concentration of extract should be increased or the amount of *Clitoria ternatea* used for extract preparation should be higher. Concentration 40 % of blue tea extract is sufficient for the intelligent properties, so the film could potentially work as an indicator of foodstuff's shelf life. **Keywords:** plant extract; DPPH, total polyphenol content, packaging

**Contact person:** Simona Jancikova, jancikovas@vfu.cz

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# Capacity development within the Greek Node of METROFOOD RI regarding food processing. Challenges and opportunities for fermented food product reformulation

Tsimidou, M.Z.<sup>1</sup>, Mantzouridou, F.Th.<sup>1</sup>, Trichopoulou, A.<sup>2</sup>, Tsakalidou, E.<sup>3</sup>, Kazou, M.<sup>3</sup>, Tarantilis, P.<sup>3</sup>

<sup>1</sup> ARISTOTLE UNIVERSITY OF THESSALONIKI (AUTH); <sup>2</sup>, <sup>2</sup>HELLENIC HEALTH FOUNDATION (HHF), AGRICULTURAL UNIVERSITY OF ATHENS (AUA)

## AIM

Food reformulation may be defined as the action of “reformulating existing foods to remove (e.g. trans fatty acids) or reduce (e.g. sugars, saturated fat, salt) certain food components while maintaining characteristics such as flavour, texture and shelf life” [1]. Reformulation might also focus on helping the maintenance of nutrients that are normally removed during conventional processing. Critical aspects of choosing foods suitable for reformulation and setting feasible goals of reformulation are related to nutrient profiling, technological and safety aspects, consumer acceptance and food legislation.

Fermented foods account for approximately one-third of all foods and beverages consumed. Food microbiota is the driving force in food fermentations. In any food microbial ecosystem, the surveillance and control of the balance, in favor of the technological and functional microbial members over the spoilage and pathogenic ones, guarantee the quality, nutritional value and safety of the final product. In recent years, high-throughput sequencing technologies, among them metagenomics, are widely applied to monitor microbial dynamics in food fermentative processes and uncover the functionality of complex microbial consortia. The new knowledge acquired will help to understand how to make a profitable use of microbial genetic resources and modulate key activities of beneficial microbes in order to ensure process efficiency, product quality and safety. On the other hand, as salt and sugar contribute greatly in the manufacture of fermented products, their reduction in foods is not a straightforward process despite the dietary recommendations and regulatory restrictions [1]. The impact of process modifications on safety issues as well as physical, chemical and nutritive changes of the final products needs study and experimentation. For this, properly controlled process in the reformulated environment as well as appropriate metrological tools for assessing the quality and safety of the fermented products are required. Finally, updating of the food composition tables to include the data for newly launched reformulated foods is mandatory. In this presentation the expertise and the services that METROFOOD – RI Greek node members [2-5] can provide for reformulating fermented foods and beverages (e.g. bread, table olives, dairy products, sausages, bread, vinegar) toward healthier alternatives will be illustrated and the prospect of further capacity development in collaboration with other METROFOOD - RI partners will be discussed. **Keywords:** METROFOOD –RI, Research Infrastructure, GREEK NODE, Fermented Foods, Food Reformulation text

**Contact person:** Mantzouridou, F.Th. (fmantz@chem.auth.gr)

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# Data sharing to improve food supply chain management

Palocci C.1, Zoani C.2

1 Department of Enterprise Engineering, University of Rome Tor Vergata, Rome, Italy, 2 ENEA, Italian National Agency for New Technologies, Energy and Sustainable Economic Development, Department for Sustainability, Biotechnologies and Agro-Industry Division (ENEA SSPT-BIOAG), Casaccia Research Center, Rome, Italy

## AIM

The agrifood sector is a very complex system and owns many responsibilities. A food product should have nutritional and sensorial quality, should be safe, authentic, sustainably and ethically produced, as well as traceable all along the supply chain. Nowadays, there are still many issues worldwide in all those aspects, indicating that the Food Supply Chain Management (FSCM) approaches used can be improved. It is fundamental to deeply understand how food supply chain is structured, operates, performs and is managed to increase its competitiveness and sustainability, identifying suitable management tools and indicators, along with the main influencing factors. Many companies, and their quality laboratories, are taking many efforts in improving FSCM, and inspection and control agencies have a key role, but further improvements are still necessary in order to properly tackle the problem. In addition, there is a lack of technology transfer from research institutions to industries should be improved. , while it would be relevant to practically introduce the latest scientific know-how in new products and solutions. The result is that knowledge is fragmented, with the risk of duplication of efforts, time and money waste and product losses, decrease of the productions' value. It is very relevant to strengthen tThe synergiesy can be useful both between public and private sectors; in fact, as an example,; on one hand, real-time data availability from industries can support scientific researches and,; on the other hand, scientific know-how can improve food production (in terms of security, safety, quality, etc.)s, also resulting in a greater consumers' confidence and consequently in an increase of supply.

Digital technologies can enhance information sharing, enabling all parties of the supply chain and relevant stakeholder to collaborate on advancing in sustainable productions. Sharing data, following FAIR principles (1), between companies, control agencies, academic and research communities could represent a relevant solution for improving Food Supply Chain ManagementFSCM to predict, plan, monitor, and rapidly and efficiently solve many issues related to agrifood production and preserve human health. This network can be supported by blockchain technology and big data analytics: the first one ensuring integrity and reliability of data, the second one allowing their efficient use. Furthermore, the Nutrient hazard Analysis and Critical Control Point (NACCP) process (21), which was developed as an evolution of HACCP with the purpose to ensure nutritional quality - in addition to food safety - along the food chain, can be used as a model with the final purpose to create an integrated system of interoperable data combining analytical results related to nutrition and food quality, safety and authenticity parameters quantified in the different production steps, with data about movements of goods, as well as to better integrate academic knowledge with food industries business management.

**Keywords:** food chain; nutrition; big data, food supply chain management, blockchain, NACCP

**Contact person:** Caterina Palocci, [caterina.palocci@uniroma2.it](mailto:caterina.palocci@uniroma2.it)

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# The effect of the addition of cricket flour on the quality of bakery products

Petříčková D.<sup>1</sup>, Kouřimská L.<sup>1</sup>, Ferusová Ž.<sup>1</sup>, Faměra O.<sup>2</sup>

<sup>1</sup>Department of Microbiology, Nutrition and Dietetics, <sup>2</sup>Department of Food Science, Czech University of Life Sciences Prague, Kamýcká 129, 165 00, Praha 6, Suchbát, Czech Republic

## AIM

The research was focused on bakery products quality after enrichment a dough by cricket flour. This work deals with the final products quality and their sensory perception.

## MATERIAL AND METHODS

The experimental part was focused on a rheological measurement and comparison of quality of wheat and enriched dough. The dough was enriched by two types of cricket flour produced by a different type of drying process (conventional or spray drying). The amount of added cricket flour was 5, 8 and 10 %. The dough properties were measured by farinograph, sensory quality between standard and enriched bakery products was compared by sensory profiling method.

## RESULTS

The obtained results suggested the significant rheological difference between the only wheat dough and the dough with cricket flour addition. The stability of enriched dough remained the same as the wheat flour (5.5 minutes) or decreased up to 3.5 minutes. Decrease in dough consistency of the enriched buns was about 70-120 BU, which was larger than 60 BU (wheat dough). Enriched dough decreased the volume of the baked buns to 45-80 % of wheat dough and height also decreased to 58-76 % of wheat dough.

The assessors noticed significant difference in sensory quality between the only wheat products and buns enriched by cricket flour. The difference was observed in intensity of colour, acceptability of smell, toughness, taste, and overall rating. No difference was observed in acceptability of texture and crumbliness, acceptability of taste and intensity of taste. PCA analysis showed, that the assessors considered the wheat buns sweeter. On the other hand, the enriched buns were more often evaluated as the ones with unusual tastes, like nutty and earthy taste. Buns with 5% addition of cricket flour had the most pleasant texture. Buns with 8 and 10 % addition of cricket flour were more tough.

## CONCLUSION

These results suggest that enrichment of wheat flour by cricket flour effected the dough properties, the size as well as sensory quality of the final bakery products.

**Keywords:** Edible insects, rheological dough properties, volume of bakery products, sensory quality

**Contact person:** Dora Petříčková, petrickova@af.czu.cz

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