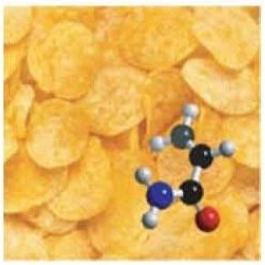
BOOK OF ABSTRACTS

International conference on new knowledge on chemical reactions during food processing and storage

CHEMICAL REACTIONS IN FOODS VII

November 14–16, 2012
Prague, Czech Republic

J. Pulkrabová, M. Tomaniová, V. Godulová, K. Cejpek and J. Hajšlová
Editors




INSTITUTE OF CHEMICAL TECHNOLOGY PRAGUE
VÚPP
DIVISION OF FOOD CHEMISTRY
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chemical reactions during food processing and storage

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Edited by
Jana Pulkrabová, Monika Tomaniová, Vanda Godulová, Karel Cejpek and Jana Hajšlová

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International conference on new knowledge on
chemical reactions during food processing and storage

CHEMICAL REACTIONS IN FOODS VII

November 14–16, 2012 • Prague • Czech Republic

Masaryk College Conference Centre

Organized by:

Institute of Chemical Technology, Prague, Czech Republic
Department of Food Analysis and Nutrition
&
Food Research Institute Prague, Czech Republic
&
Czech Chemical Society
Section of Food and Agricultural Chemistry
&
European Association for Chemical and Molecular Sciences
Food Chemistry Division

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CONFERENCE PROGRAM

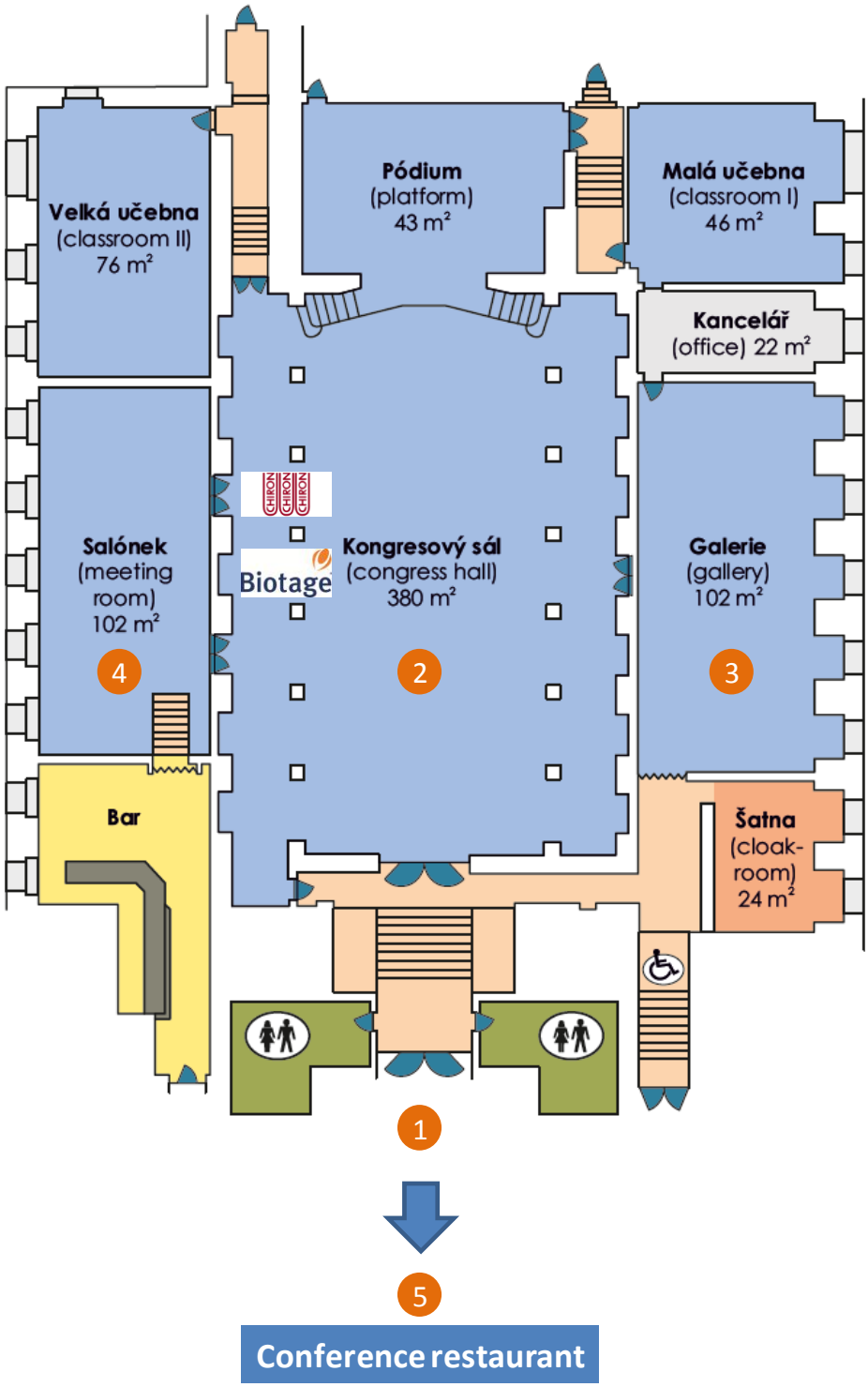
7th International Conference on Chemical Reactions in Foods

November 14–16, 2012

Masaryk College Conference Centre • PRAGUE • CZECH REPUBLIC

CRF 2012 Venue & Exhibition Floor plan

Masaryk College Conference Centre



1: Entrance to the conference centre & Registration desk
2: Conference hall
Exhibition area
Poster area
3: Poster area
4: Poster area
5: Conference restaurant
Catering area (coffee breaks, lunch, Welcome Cocktail)

CRF 2012 – PROGRAM AT A GLANCE

Time / Date	WEDNESDAY November 14, 2012	THURSDAY November 15, 2012	FRIDAY November 16, 2012
9:00–10:00		**Oral session 3** **STRATEGIES TO IMPROVE FOOD QUALITY AND SAFETY II**	**Oral session 6** **COMPOUNDS ASSOCIATED WITH NUTRITIONAL AND SENSORY QUALITY OF FOODS II**
10:00–10:30			
10:30–11:00	**Registration** for the conference Masaryk College Conference Centre	Coffee break / Exhibition	Brunch / Exhibition
11:00–12:30		**Oral session 4** **BIOLOGICALLY-ACTIVE CONSTITUENTS OF FOODS AND FOOD RAW MATERIALS II**	**Oral session 7** **COMPOUNDS ASSOCIATED WITH NUTRITIONAL AND SENSORY QUALITY OF FOODS III**
12:30–13:00		Lunch	**Final discussion panel** **& CRF 2012 poster award** **& Closing address**
13:00–13:30	**Opening of the conference** **& Welcome** **to the CRF 2012**		
13:30–15:30	**Oral session 1** **STRATEGIES TO IMPROVE FOOD QUALITY AND SAFETY I**	**Poster session / Exhibition**	
15:30–16:00	Coffee break / Exhibition	Coffee break / Exhibition	
16:00–18:30	**Oral session 2** **BIOLOGICALLY-ACTIVE CONSTITUENTS OF FOODS AND FOOD RAW MATERIALS I**	**Oral session 5** **COMPOUNDS ASSOCIATED WITH NUTRITIONAL AND SENSORY QUALITY OF FOODS I**	
18:30–19:30	**Welcome Cocktail** Masaryk College Conference Centre		
20:00–22:30		**Conference Dinner** Café & Restaurant Slavia	

Coffee breaks, lunch, Welcome drink will be served in the conference centre restaurant.

WEDNESDAY, November 14, 2012

10:30–13:00 **Registration for CRF 2012 conference**

13:00–13:20
Conference hall

OPENING of the conference and WELCOME

Karel Melzoch, Rector of ICT Prague
Tomáš Ruml, Dean of the Faculty of Food and Biochemical Technology, ICT Prague
Livia Simon-Sarkadi, Chair of the Division of Food Chemistry, EuCheMS
Jana Hajšlová, Chair of the CRF 2012 Scientific Committee

MUSIC WELCOME *(harp & flute)*

13:20–15:30
Conference hall

ORAL SESSION 1:
Strategies to Improve Food Quality and Safety I

Chairpersons: *Jana Hajšlová and Thomas Henle*

13:20–13:50 **L1**

RESEARCH AND INNOVATION IN FOOD INDUSTRY

Beate Kettlitz, *FoodDrinkEurope, Brussels, Belgium*

13:50–14:20 **L2**

INDUSTRIAL PRODUCTION OF FRUIT JUICES: CURRENT CHALLENGES FOR FOOD RESEARCH

Barbara Siegmund, *Graz University of Technology, Institute of Analytical Chemistry and Technology, Graz, Austria*

14:20–14:40 **L3**

DEEP ULTRAVIOLET LIGHT-EMITTING DIODES TO ENHANCE PLANT NUTRITIONAL VALUE AND MAINTAIN FRESHNESS AND PHYTOCHEMICAL COMPOSITION DURING POSTHARVEST STORAGE

Steven Britz, *USDA Food Components and Health Lab, Beltsville, USA*

14:40–15:00 **L4**

EFFECT OF A PREVIOUS HIGH HYDROSTATIC PRESSURE TREATMENT ON LIPID DAMAGE OF CHILLED JACK MACKEREL (*TRACHURUS MURPHYI*)

Santiago Aubourg, *Instituto de Investigaciones Marinas, CSIC, Vigo, Spain*

15:00–15:10 **L5***

CHANGES IN TANNIN SOLUBILITY AND MICROSTRUCTURE OF HIGH HYDROSTATIC PRESSURE TREATED PERSIMMON DURING STORAGE AT 4 °C

José Luis Vázquez-Gutiérrez, *Universitat Politecnica de Valencia, Spain*

15:10–15:30 **L6**

FORMATION OF SAFE CONTACT ACTIVE ANTIMICROBIAL SURFACES FOR FOOD PACKAGING

Elena Poverenov, *Agricultural Research Organization, The Volcani Center, Department of Food Quality and Safety, Bet Dagan, Israel*

15:30–16:00
Conference restaurant / Conference hall

Coffee Break / EXHIBITION

* Young scientist presentation

16:00–18:20 Conference hall

ORAL SESSION 2: Biologically-Active Constituents of Foods and Food Raw Materials I

Chairpersons: *Livia Simon-Sarkadi and Friedrich Bauer*

16:00–16:30 L7

WINE-FROM GRAPES TO A MATURED PRODUCT

***Erich Leitner**, Graz University of Technology, Institute of Analytical Chemistry and Technology, Graz, Austria*

16:30–16:50 L8

CONVERSION OF HISTAMINE IN HISTAMINOL: A NEW PERSPECTIVE FOR WINE QUALITY AND SAFETY?

***Marco Arlorio**, Dipartimento di Scienze del Farmaco and Drug and Food Biotechnology Center, Universite degli Studi del Piemonte Orientale "A. Avogadro", Novara, Italy*

16:50–17:10 L9

THE CASE STUDY OF ACRYLAMIDE FORMATION IN THERMALLY TREATED VEGETABLE IN RELATION TO TOTAL POLYPHENOLS CONTENT AND ANTIOXIDANT CAPACITY

***Kristina Kukurova**, Food Research Institute, Bratislava, Slovak Republic*

17:10–17:30 L10

ANTIOXIDATIVE CAPACITY OF *BETA VULGARIS* EXTRACTS AND THEIR EFFECT ON THE FORMATION OF HETEROCYCLIC AMINES IN A MODEL SYSTEM

***Audrius Pukalskas**, Kaunas University of Technology, Kaunas, Lithuania*

17:30–17:50 L11

INFLUENCE OF BAKING IN DIGESTIBILITY AND BIOACCESIBILITY OF CARBOHYDRATES, POLYPHENOLS AND FORMATION OF MAILLARD COMPOUNDS

***M. Elena Diaz-Rubio**, Institute of Food Science, Technology and Nutrition (ICTAN-CSIC), Madrid, Spain*

17:50–18:00 L12*

TRYPTOPHAN-CONTAINING DIPEPTIDES: BIOACTIVE COMPOUNDS IN FOODS WITH A BLOOD PRESSURE LOWERING EFFECT?

***Diana Lunow**, Institute for Food Chemistry, Technical University of Dresden, Germany*

18:00–18:10 L13*

COFFEE PHENOLICS DEGRADATION AND FORMATION OF BIOACTIVE METABOLITES BY HUMAN COLONIC MICROFLORA

***Iziar-Amaia Ludwig**, University of Navarra, Pamplona, Spain*

18:10–18:20 L14*

OPTIMIZATION OF THE FORMATION OF VINYLDITHIINS, A THERAPEUTICAL COMPOUND FROM GARLIC

***Bérénice Dethier**, University of Liege (Gembloux Agro-Bio Tech), Gembloux, Belgium*

18:30–19:30 Conference restaurant / Conference hall

Conference Welcome Drink (Masaryk College Conference Centre)

* Young scientist presentation

THURSDAY, November 15, 2012

9:00–10:30 Conference hall

ORAL SESSION 3:
Strategies to Improve Food Quality and Safety II

Chairpersons: *Peter Schieberle and Karel Cejpek*

9:00–9:30 **L15**

NOVEL TECHNOLOGIES TO REDUCE PROCESSING CONTAMINANTS AND SOME OTHER UNDESIRABLE FOOD COMPONENTS

***Jana Hajslova**, Institute of Chemical Technology, Prague, Czech Republic*

9:30–9:50 **L16**

ACRYLAMIDE FORMATION IN FOODS: ROLE OF COMPOSITION AND PROCESSING

***Vural Gökmen**, Hacettepe University, Ankara, Turkey*

9:50–10:10 **L17**

FURAN IN HEAT-PROCESSED FOODS: FIVE-YEARS FILED MONITORING, REDUCTION AND RISK ASSESSMENT STUDY IN KOREA

***Kwang-Geun Lee**, Dongguk University, Seoul, Korea*

10:10–10:30 **L18**

FORMATION OF AROMA-ACTIVE COMPOUNDS AS WELL AS TOXICOLOGICALLY RELEVANT STYRENE DURING THE PRODUCTION OF WHEAT BEER

***Michael Granvogl**, German Research Centre of Food Chemistry, Freising, Germany*

10:30–11:00 Conference restaurant / Conference hall

Coffee Break / EXHIBITION

11:00–12:30 Conference hall

ORAL SESSION 4:
Biologically-Active Constituents of Foods and Food Raw Materials II

Chairpersons: *Erich Leitner and Francisco Morales*

11:00–11:30 **L19**

INGREDIENTS FROM LOW VALUE BY-PRODUCTS OF SELECTED FOOD MANUFACTURING INDUSTRIES

***Jennifer Ames**, University of the West of England, Bristol, UK*

11:30–11:50 **L20**

CHEMICAL AND FUNCTIONAL CHARACTERISTICS OF FIBER ISOLATED FROM HAZELNUT SKINS: NEW PREBIOTIC INGREDIENT FROM BY-PRODUCTS?

***Rosa Montella**, Dipartimento di Scienze del Farmaco and DFB (Drug and Food Biotechnology) Center, Universite degli Studi del Piemonte Orientale "A. Avogadro", Novara, Italy*

11:50–12:00 **L21***

IMPACT OF TEMPERATURE AND WATER ACTIVITY ON ENZYMATIC AND NON-ENZYMATIC REACTIONS DEVELOPMENT IN RECONSTITUTED DRIED MANGO

***Emilie Korbel**, CIRAD, UMR QualiSud, Montpellier, France*

12:00–12:20 **L22**

CONTROLLING MAILLARD REACTION BY REACTANTS ENCAPSULATION: SODIUM CHLORIDE IN BISCUITS

***Alberto Fiore**, Department of Food Science, University of Napoli "Federico II", Napoli, Italy*

12:20–12:30 **L23***

THE POTENTIAL OF AMBIENT MASS SPECTROMETRY FOR RAPID MONITORING OF MEAT AND FISH FRESHNESS

***Hana Daňhelová**, Institute of Chemical Technology, Prague, Czech Republic*

12:30–13:30 Conference restaurant

Lunch

* Young scientist presentation

THURSDAY, November 15, 2012

13:30–15:30 Gallery & Meeting room		**POSTER SESSION / EXHIBITION**
15:30–16:00 Conference restaurant / Gallery & Meeting rooms / Conference hall		**Coffee Break / POSTER SESSION / EXHIBITION**
16:00–18:00 Conference hall		**ORAL SESSION 5:** **Compounds Associated with Nutritional and Sensory Quality of Foods I** Chairpersons: *Vieno Piironen and Marek Doležal*
16:00–16:20	**L24**	**OXIDATION OF TRIACYLGLYCEROLS IN PRESENCE OF CAROTENOIDS - IDENTIFICATION OF OXIDIZED SPECIES BY LC-MS** ***Michael Murkovic**, Graz University of Technology, Graz, Austria*
16:20–16:30	**L25***	**QUANTITATIVE AND QUALITATIVE ANALYSIS OF HIGH MOLECULAR COMPOUNDS IN VEGETABLE OILS FORMED UNDER HIGH TEMPERATURES IN THE ABSENCE OF OXYGEN** ***Klara Cihelkova**, Institute of Chemical Technology, Prague, Czech Republic*
16:30–16:50	**L26**	**IMPACT OF INTRINSIC AND EXTRINSIC PARAMETERS ON THE FORMATION OF TOXIC ALDEHYDES IN FOODS** ***Antonios Papastergiadis**, Ghent University, Belgium*
16:50–17:00	**L27***	**STUDIES ON THE FORMATION PATHWAY OF ACROLEIN** ***Alice Ewert**, German Research Center for Food Chemistry*
17:00–17:20	**L28**	**ISOTHERMAL KINETICS OF MALONDIALDEHYDE CONTENT CHANGES IN CHICKEN MEATS** ***Stéphanie Roux**, CIRAD, Saint-Denis, La Réunion, France*
17:20–17:50	**L29**	**AMINO ACID DEGRADATIONS PRODUCED BY LIPID OXIDATION PRODUCTS** ***Francisco J. Hidalgo**, Instituto de la Grasa, CSIC, Seville, Spain*
From 20:00		**Conference Dinner** **(Café & Restaurant Slavia, historical centre of Prague)**

Registration for dinner at the registration desk, until Wednesday, November 14, 16:00

* Young scientist presentation

FRIDAY, November 16, 2012

9:00–10:40
Conference hall

ORAL SESSION 6:
Compounds Associated with Nutritional and Sensory Quality of Foods II

Chairpersons: *Michael Murkovic and Vural Gökmen*

9:00–9:30 **L30**

DISCOVERY, SENSORY ACTIVITY AND TASTE RECEPTOR ACTIVATION OF THERMALLY GENERATED TASTE MODULATORS IN FOODS

Thomas Hofmann & Barbara Suess, *Technische Universität München, Germany*

9:30–10:00 **L31**

NOVEL APPROACHES IN INDUSTRIAL RESEARCH TARGETING QUALITY ATTRIBUTES OF EXTRUDED FOOD PRODUCTS

Tomas Davidek*, Nestle PTC Orbe, Switzerland*

10:00–10:30 **L32**

THE STRECKER DEGRADATION: AN „OLD“ REACTION WITH NEW IMPACT ON FOOD AROMA

Peter Schieberle*, Technische Universität München, Germany*

10:30–10:40 **L33***

IMPROVEMENT OF THE AROMA OF THE GLUTEN-FREE BREAD BY AROMA-ACTIVE MALT PREPARATIONS

Gabriela Ratz*, German Research Center for Food Chemistry, Freising, Germany*

10:40–11:10
Conference restaurant / Conference hall

Brunch / EXHIBITION

11:10–12:30
Conference hall

ORAL SESSION 7:
Compounds Associated with Nutritional and Sensory Quality of Foods III

Chairpersons: *Jenny Ames and Francisco Hidalgo*

11:10–11:40 **L34**

GLYCATION COMPOUNDS IN FOODS: FORMATION, METABOLIC TRANSIT, FUNCTIONAL CONSEQUENCES

Thomas Henle*, Technische Universität Dresden, Germany*

11:40–12:00 **L35**

GASTROINTESTINAL DIGESTION AND EPITHELIAL TRANSPORT OF PYRRALINE

Michael Hellwig*, Technische Universität Dresden, Dresden, Germany*

12:00–12:10 **L36***

AN ACTIVE ROLE OF SELECTED DIETARY POLYPHENOLS IN CARAMELIZATION AND PROTEIN GLYCATION MODEL SYSTEMS

Xinchen Zhang*, The University of Hong Kong, Hong Kong*

12:10–12:30 **L37**

MELANOIDINS EXHIBIT PRO-OXIDATIVE EFFECTS ON ISOLATED AND CELLULAR DNA AFTER COMPLEXATION OF METAL IONS

Bettina Cämmerer*, University of Technology, Institute of Food Technology and Food Chemistry, Berlin, Germany*

12:30–13:00

FINAL DISCUSSION PANEL

Panellists: *Scientific Committee*

13:00–13:30

CLOSING ADDRESS

Jana Hajšlová, Chair of the CRF 2012 Scientific Committee

CRF 2012 poster award
Announcement of the next CRF event

* Young scientist presentation

POSTER SESSION

THURSDAY, November 15, 2012

13:30–15:30	**POSTER SESSION**
	BIOLOGICALLY-ACTIVE CONSTITUENTS OF FOODS AND FOOD RAW MATERIALS
	CHEMISTRY BEHIND NOVEL FOODS
	COMPOUNDS ASSOCIATED WITH NUTRITIONAL AND SENSORY QUALITY OF FOODS
	STRATEGIES TO IMPROVE FOOD QUALITY AND SAFETY

BIOLOGICALLY-ACTIVE CONSTITUENTS OF FOODS AND FOOD RAW MATERIALS

A-1 **INVESTIGATION OF TRANS-RESVERATROL AND STILBENE DERIVATIVE TDPA IN HUNGARIAN WINES USING HPLC AND LMS**
Zoltan Papai, Agnes Bona, Gabor Maasz, Janos Schmidt, Robert Ohmacht, Laszlo Mark

A-2 **EFFECT OF HIGH HYDROSTATIC PRESSURE ON THE ACTIVITY OF CATHEPSINS B AND D OF MACKEREL (*SCOMBEROMORUS MACULATUS*) AND HORSE MACKEREL (*TRACHURUS TRACHURUS*)**
Liliana Fidalgo, Jorge A. Saraiva, Santiago P. Aubourg, Manuel Vázquez, J. Antonio Torres

A-3 **ANTIOXIDANT AND ANTIGENOTOXIC EFFECTS OF SPENT COFFEE EXTRACTS IN HUMAN CELLS**
Jimena Bravo, Leire Arbillaga, Maria Paz de Peńa, Concepción Cid

A-4 **DETERMINATION OF ANTIOXIDANT CAPACITY AND REGENERATION BEHAVIOR OF BIOACTIVE COMPOUNDS BOUND TO INSOLUBLE DIETARY FIBERS**
Ecem Evrim Çelik, Vural Gökmen

A-5 **IMPACT OF EXTRACTION METHODS, TYPE AND STORAGE ON THE PHENOLIC COMPOSITION AND ANTIOXIDANT POTENCY OF CHOCOLATE PRODUCTS**
Antia Orphanides, Vlasios Goulas, Anastasia Shantona, Photis Papademas

A-6 **EFFECT OF DRYING METHOD ON THE NUTRACEUTICAL CONTENT AND ANTIOXIDANT CAPACITY OF CYPRUS SPEARMINT (*MENTHA VIRIDIS*)**
Antia Orphanides, Vlasios Goulas, Vassilis Gekas

A-7 **CHANGES OF ANTIOXIDANT ACTIVITY IN HONEY AFTER HEAT TREATMENT**
Goran Saric, Ksenija Markovic, Marina Krpan, Nikola Major, Mirjana Hruskar, Nada Vahcic

A-8 **PHENOLIC COMPOSITION AND SENSORY PROPERTIES OF CIDERS PRODUCED FROM LATVIAN APPLES**
Rita Riekstina-Dolge, Zanda Kruma, Fredijs Dimins, Daina Karklina

A-9 **BIOAVAILABILITY OF TRYPTOPHAN-CONTAINING PEPTIDES IN HUMAN BLOOD**
Susanne Kaiser, Steffi Rudolph, Diana Lunow, Melanie Martin, Thomas Henle

A-10 **CRUDE PHENOLIC EXTRACT EFFECTS *IN VITRO* AND THEIR IMPACT IN A RAW SAUSAGES STORAGE TEST. PRELIMINARY FINDINGS**
Stefania Balzan, Luca Fasolato, Filomena Montemurro, Barbara Cardazzo, Lisa Carraro, Agrese Taticchi, Maurizio Servili, Enrico Novell

A-11 **BIOACTIVE COMPOUNDS IN LATVIAN BARLEY BEER**
Ilona Dabina-Bicka, Daina Karklina, Zanda Kruma, Fredijs Dimins

A-12 **CHANGES IN PHENOLIC CONTENT AND ANTIOXIDANT ACTIVITY OF FREEZED SPINACH, PEA AND SWEETCORN IN RELATION TO THE STORAGE PERIOD**
Daniel Bajčan, Ján Tomáš, Gabriela Uhlířová, Július Árvay, Pavol Trebichalský, Radovan Stanovič

A-13 **CHOSEN ANTIOXIDANT AND SENSORY PROPERTIES AND THEIR MUTUAL RELATIONS OF SLOVAK RED WINES – CABERNET SAUVIGNON**
Daniel Bajčan, Vladimír Šimanský, Tomáš Tóth, Mária Timoracká, Ľuboš Harangozo

A-14 **ALKYLRESORCINOLS CONTENT IN PEARLED NAKED AND HULLED BARLEY FRACTIONS**
Matteo Bordiga, Monica Locatelli, Rosa Montella, Jean Daniel Coisson, Valentina Sovrani, Massimo Blandino, Valentina Scarpino, Amedeo Reyneri

A-15 **THERAPEUTICAL COMPOUNDS IN GARLIC OIL: PRODUCTION AND EVOLUTION AFTER PREPARATION**
Bérénice Dethier

A-16 **ANTIOXIDANT ACTIVITIES OF LEMON BALM (*MELISSA OFFICINALIS* L., LAMIACEAE)**
Diana Chrpová, Iva Roubíčková, Vojtěch Ilko, Lenka Kouřimská, Monika Sabolová, Jan Pánek

A-17 **ANTIOXIDATION ACTIVITY OF LAMIACEAE HERBS GROWN UNDER ORGANIC FARMING CONDITIONS**
Monika Sabolová, Lenka Kouřimská, Jan Pánek

A-18 ***IN VITRO* CHEMOPREVENTIVE ACTIVITIES OF OLIVE OIL PHENOLIC COMPOUNDS**
Roberto Fabiani, Patrizia Rosignoli, Raffaela Fuccelli, Maria Vittoria Sepporta, Maurizio Servili, Guido Morozzi

A-19 **CULTIVAR INFLUENCE ON TOTAL POLYPHENOL AND RUTIN CONTENTS AND TOTAL ANTIOXIDANT CAPACITY IN BUCKWHEAT, AMARANTH AND QUINOA SEEDS**
Alena Vollmannová, Eva Margitanová, TomášTóth, Dana Urminská, Tatiana Bojňanská

A-20 **BIOACTIVE COMPOUNDS OF CRANBERRY FRUITS IN THE FIRST AND THE SECOND HARVEST**
Alena Vollmannová, Lívia Krížová, Zuzana Poláková, Ján Daniel, Michal Medvecký

A-21 **THE RUTIN DISLOCATION IN DIFFERENT ANATOMY PLANT PARTS AND DIFFERENT GROWTH PHASES OF SELECTED AMARANTH CULTIVARS**
Mária Timoracká, Judita Bystrická, Alena Vollmannová, Zuzana Poláková, Ľubomir Harangozo

A-22 **CHANGES OF POLYPHENOLIC SUBSTANCES IN THE ANATOMICAL PARTS OF BUCKWHEAT (*FAGOPYRUM ESCULENTUM MOENCH*) DURING DIFFERENT GROWTH PHASES**
Judita Bystrická, Alena Vollmannová, Janette Musilová, Tomáš Tóth, Iveta Čičová

A-23 **TOTAL POLYPHENOLS CONTENT IN SELECTED CULTIVARS OF STRAWBERRIES IN RELATION TO CONTENTS OF Cd AND Pb IN SOIL**
Pavol Trebichalský, Daniel Bajčan, Ján Tomáš, Janette Musilová, Ľuboš Harangozo

A-24 **DYNAMICS OF QUERCETIN FORMATION IN ONION (*ALLIUM CEPA* L.) DURING VEGETATION**
Judita Bystrická, Janette Musilová, Ján Tomáš, Mária Timoracká, Július Árvay

A-25 **EFFECT OF FERTILIZATION ON CHANGES OF THE POLYPHENOL CONTENT AND ANTIOXIDANT ACTIVITY IN POTATOE TUBERS (*SOLANUM TUBEROSUM* L.)**
Janette Musilová, Jaromír Lachman, Zuzana Poláková, Peter Kováčik, Diana Hrabovská

A-26 **DYNAMICS IN ANTIOXIDANT ACTIVITY IN THE ANATOMICAL PARTS OF SELECTED CULTIVARS OF BUCKWHEAT IN DIFFERENT GROWTH PHASES**
Janette Musilová, Alena Vollmannová, Judita Bystrická, Mária Timoracká, Iveta Čičová

A-27 **CHANGES OCCURING IN WEIGHT, TEXTURE, TOTAL SOLUBLE SOLIDS AND NATURAL ANTIOXIDANTS (CAROTENOIDS AND TOCOPHEROLS) COMPOSITION OF CANTALUPE MELONS DURING SHORT STORAGE**
Dragan Žnidarčič, Helena Sircelj, Nina Kacjan Maršić

A-28 **MUSTARD PHENOLICS (PROFILE AND ANTIOXIDANT POTENTIAL)**
Neda Nićiforović, Mihaela Skrt, Nataša Poklar Ulrih, Helena Abramovič

A-29 **EFFECT OF ADSORBENT AND ION EXCHANGE RESIN APPLICATIONS ON TOTAL PHENOLIC CONTENT AND ANTIOXIDANT ACTIVITY OF WHITE AND RED GRAPE JUICES**
Mehmet Akbulut, Hacer Coklar

A-30 **INFLUENCE OF VARIETY AND STORING OF CHICORY (*CICHORIUM INTYBUS* L.) ON THE CONTENT OF TOTAL PHENOLS, ANTIOXIDATIVE POTENTIAL AND FATTY ACIDS**
Lovro Sinkovič, Janez Hribar, Rajko Vidrih

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ORAL SESSIONS

L-1
RESEARCH AND INNOVATION IN THE FOOD INDUSTRY

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The availability of safe, nutritious, affordable and food which contributes to a healthy diet has taken on a new and pressing dimension in the face of an ever growing global population and increasing environmental and sustainability concerns.

In today's globalised food markets, food safety issues are viewed from an increasingly international perspective and more and more emphasis is being placed on reducing wastage, energy and water consumption at all stages of the food supply chain.

Despite these facts, compared to other industries in the EU; the food industry spends the smallest amount on innovation. Less than 1 per cent of total investments in the food industry are donated to R&D, according to latest Data and Trends, a study commissioned by FoodDrinkEurope.

Boosting research and development spending and translating the resulted findings into innovative products provides a platform for the EU to regain a more competitive position.

FoodDrinkEurope has played an important role in promoting R&D in the food industry for example through the establishment of the European Technology Platform 'Food for Life', which brought together academia, food industry operators and many other stakeholders of the food chain in 2005. This European Technology Platform has developed a clear industry-led, strategic research and innovation agenda to address the Grand Challenges in Europe.

The ETP 'Food for Life's first Strategic Research Agenda was published in July 2007. However, due to policy developments in the European Commission, such as the publication of the EU 2020 Flagship Initiatives and the establishment of the Innovation Union, it became necessary to revise and update this Strategic Research Programme to properly address aspects of innovation. The new Strategic Research and Innovation Agenda, published in September 2012, outlines several main goals, including:

(i) Innovation supported by Communication, Training & Technology Transfer,

(ii) Health, Well-Being & Longevity,

(iii) Safe Foods that Consumers Can Trust

(iv) Sustainable and Ethical Production

(v) Food Processing, Packaging and Quality

(vi) Food and Consumers

(vii) Food Chain Management

Research and innovation activities, such as those proposed by ETP 'Food for Life', combine academia, the industry and other stakeholders and set research and innovation priorities for 2020 and the years beyond.

L-2
INDUSTRIAL PRODUCTION OF FRUIT JUICES: CURRENT CHALLENGES FOR FOOD RESEARCH

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Fruits and vegetables represent important constituents of modern nutrition. Campaigns like '5 a day' are generally accepted and promote the consumption of plant material as essential parts of a healthy nutrition. According to the recommendations, one portion out of five may be consumed in form of fruit juice. In Europe, the average amount of fruit juice or nectar consumption in 2011 was about 22 litters per capita and year, whereas the highest amount (35 litters per capita and year) was consumed in Germany (www.fruchtsaft.org). Still, the most frequently consumed fruit juices are apple and orange juice, but nevertheless many products – including juices, nectars and smoothies – have been developed during the last 10 years. The quality of fruit juices is influenced by a number of factors. This paper aims to give a survey how some of these factors may influence the products. The development of new products with characteristics that are different to those from the traditional juices, poses new problems to the producer. Besides the quality of the fruits themselves, changes of the quality can be affected by storage conditions, alterations in formulation or production line, for example. In addition, the global trend to replace the heavy-weighted glass bottle by light-weighted packaging materials, that in most cases show different oxygen permeability to the glass bottle, requires investigations whether new packaging materials are suitable to maintain the quality of fruit juices and nectars. Another problem is the contamination by so-called thermoacidophilic bacteria, which are microorganisms that are resistant to the common temperatures that are applied for fruit juice pasteurization even at the low pH value of fruit juices. These bacteria – or the spores thereof – are able to re-germinate in the product in the shelf. Secondary metabolites may lead to sensory deterioration of the product and may, as a consequence, evoke enormous economic damage to the producer. The development of innovative products by fruit juice industry using for example exotic fruits with low acidity as constituents of fruit juice products, may also be a critical point. A higher pH in the product may create a suitable environment for microorganisms, which would not germinate under common, rather low pH values like in orange or apple juice. Within the last years, so-called near-water products – water-based and flavoured drinks – have gained ground on the market. In this context, juices, lemonades and syrups showing floral, citrus-like flavour like for example elderflower drinks have become popular. Anyway, elderflower syrups that can be bought in supermarkets do often not represent the typical and very pleasant elderflower flavour. The extraction of elderflower flavour is very difficult and may be influenced by various factors. Results from recent investigations will be presented to demonstrate the fragility of the flavour of elder flowers.

Keywords: Fruit juice, sensory quality, storage, packaging, off-flavour

L-3

DEEP ULTRAVIOLET LIGHT-EMITTING DIODES TO ENHANCE PLANT NUTRITIONAL VALUE AND MAINTAIN FRESHNESS AND PHYTOCHEMICAL COMPOSITION DURING POSTHARVEST STORAGE

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Compared to mercury lamps, deep ultraviolet (DUV) light-emitting diodes (LEDs) are uniquely suited as low-moderate power UV sources over the 240–320 nm range because of their small size, low weight, low power consumption, low radiant heat, long lifetime, absence of glass and mercury, and increased efficiency at low temperatures. In addition, peak wavelengths can be chosen to maximize desired responses while minimizing undesired consequences, thus optimizing photobiological efficiency compared to broad spectrum lamps. The application of DUV–LEDs to plant growth scenarios (winter greenhouses) and postharvest cold storage of fruits and vegetables is described. Low levels of solar ultraviolet-B radiation (290–320 nm) typically reduce the synthesis and accumulation in growing plants of polyphenolic compounds such as flavonoids that contribute color, flavor and nutritional value. This can be a problem in greenhouses or at low solar angle. Since few wavelength response curves exist for UV effects on phytochemical composition, the narrow (ca. 10 nm) bandwidth of DUV LEDs was exploited to generate spectral response data on accumulation of flavonoids (i.e., quercetin malonylglucoside and cyanidin malonylglucoside) and phenolic acid esters (chlorogenic acid, chicoric acid and caftaric acid) in green and red leaf lettuce varieties grown under simulated winter greenhouse conditions (low photosynthetically-active radiation, short daylengths, cool temperatures and low ambient UV radiation). Results indicate a relatively narrow wavelength window for enhancement of flavonoids with maximal increases observed around 290 nm using 10 h supplemental exposures (10–40 mW m^{-2}) during a 12 h light period for 7 days starting about 3 weeks after seedling emergence. The longest wavelength tested (307 nm) was much less effective. The shortest wavelength tested (272 nm) inhibited growth and caused aberrant leaf morphology. In addition, plant response to intermittent UV irradiation was determined using a DUV LED lamp module (293±5 nm) designed to irradiate a narrow area spanning the width of a growth rack from a height of 30 cm or more. The module was mounted on a motorized track and moved cyclically along the length of a bench irradiating plants periodically. This intermittent exposure to UV was sufficient to induce several fold increases in flavonoids in leaves of a UV-sensitive line of leaf lettuce. Additional requirements for duration and daily integrals of UV are being tested. Together with wavelength response and irradiance data, these experiments will inform the design of cost-effective DUV LED-based modules and systems to treat plants over large areas, thus maximizing their nutritional value during growth and harvest. DUV–LED technology is also being applied to long-term postharvest treatment of cold-stored strawberry, broccoli, and lettuce. Initial studies indicate the potential to maintain freshness, appearance and desirable phytochemicals.

Keywords: LED, UV, lettuce, strawberry, broccoli

L-4

EFFECT OF A PREVIOUS HIGH HYDROSTATIC PRESSURE TREATMENT ON LIPID DAMAGE OF CHILLED JACK MACKEREL (*TRACHURUS MURPHYI*)

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To retain quality as long as possible, fish technologists and the fish trade have developed different advanced processing systems. Among them, high hydrostatic pressure (HHP) technology has shown to inactivate microbial development and endogenous enzyme activity, thus leading to a shelf-life extension. However, HHP technology has been reported to induce deteriorative changes in fish constituents (namely, proteins and lipids).

The present work was focused on the lipid changes of chilled jack mackerel (*Trachurus murphyi*) that was previously treated with HHP technology. This species was chosen as being an abundant small pelagic one, normally considered as an unconventional source of raw material. As previous HHP treatment, different pressure level and pressure holding time conditions (250, 450 and 550 MPa; 3 and 4 min) were tested and compared to untreated fish (Control). Additionally, fish corresponding to pre– and post-*rigor mortis* (RM) catching conditions were also analysed and compared. The study was addressed to the lipid hydrolysis (free fatty acids, FFA) and oxidative (conjugated dienes and trienes; peroxide value; thiobarbituric acid index; fluorescent and browning compounds) changes. Analyses were carried out on the fish muscle after 0, 2, 6, 10 and 14 days of chilled storage; three replicates (n=3) were considered for each HHP condition in order to achieve the statistical analysis.

A marked lipid hydrolysis inhibition was attained by increasing the pressure applied; this inhibition was especially important at 450– and 550–MPa conditions. Additionally, a pressure holding time of 4 min led to lower FFA values in most cases when compared to samples corresponding to 3–min condition. A higher FFA content was observed in pre-RM samples corresponding to Control and 250–MPa treatments; however, lipid hydrolysis was found higher in post-RM samples when 450–MPa treatment was encountered.

Concerning the lipid oxidation development, a definite effect of HHP conditions could not be concluded. Thus, a higher peroxide formation was observed as a result of increasing the pressure holding time, while an increasing pressure value led to a lower peroxide formation. For interaction compound formation (fluorescence and browning development), a different pattern was found for pre- and post-RM samples when considering the effect of pressure and pressure holding time. Evolution of the different kinds of lipid oxidation compounds is taken into account to establish the effect of HHP conditions in the pre–and post–RM conditions.

Keywords: *Trachurus murphyi*, rigor mortis, high pressure, chilling, lipids hydrolysis and oxidation

Acknowledgement: *To FONDECYT (Chile) for financing the Project 1110782.*

L-5
CHANGES IN TANNIN SOLUBILITY AND MICROSTRUCTURE OF HIGH HYDROSTATIC PRESSURE TREATED PERSIMMON DURING STORAGE AT 4ºC

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Condensed tannins, considered as important bioactive compounds, are largely present in persimmon (*Diospyros kaki* L.f.). These compounds are the polymerized products of flavan-3-ols and flavan-3,4-diols and their solubility may be affected during processing and storage of the fruit. The aim of this work was to study the structural changes in persimmon when it is treated with high hydrostatic pressure (HHP) and stored, and their relationship with the solubility and location of tannins and some physicochemical properties. For this purpose, persimmon was submitted to 200 MPa during 3 and 6 min at 37°C (T1 and T2, respectively) and stored at 4°C for 28 days. The microstructural study was carried out by Low Temperature Scanning Electron Microscopy (Cryo-SEM), Light Microscopy (LM) and Transmission Electron Microscopy (TEM). The physicochemical properties studied were total soluble tannins (TST), total soluble solids (TSS), pH, lightness (L*), firmness and cohesiveness. Samples were analyzed after the HHP treatment and after 7, 14, 21 and 28 days of storage at 4°C. Cryo-SEM micrographs show that HHP causes cell wall and membrane disruption in the persimmon parenchymatic tissue. Some intercellular spaces full of material from inside the cell and insoluble material not only inside but also outside some cells can also be observed. Retraction of the tonoplast and loss of cell turgor is greater as the storage time increases. LM micrographs show precipitated tannins inside and outside the cells and a progressive separation of adjacent cells during the storage time. Degradation of membranes and cell wall material due to HHP treatment and storage can be observed in greater detail by TEM. HHP-treated persimmon experiences the main physicochemical changes after 7 days of storage. A significant ($P<0.05$) decrease in TST and TSS, and a significant ($P<0.05$) increase in pH after 7 days of storage take place in HHP-treated samples. Lightness is affected by both HHP treatment and storage. The greater decrease in lightness takes place after 7 days of storage at 4°C in the HHP-treated samples. Regarding textural properties, samples treated for 3 minutes (T1) showed higher firmness than the rest of the samples during the whole storage period, whereas HHP-treated samples show higher cohesiveness than the control samples. HHP treatments as well as later storage at 4°C affect microstructure, tannin solubility and extractability, and some physicochemical properties of persimmon fruit. These effects vary depending on the treatment conditions and the storage time. The main changes in the physicochemical properties take place during the first 7 days of storage.

Keywords: Persimmon, microstructure, high hydrostatic pressure, tannin, storage

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L-6
FORMATION OF SAFE CONTACT ACTIVE ANTIMICROBIAL SURFACES FOR FOOD PACKAGING

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A "contact active" concept provides a new promising approach for fight against harmful microorganisms. In this approach antimicrobial component is covalently linked to a material surface, while its biocide site remains active. A number of contact active surfaces and polymers have been reported to effectively stop the spread of bacteria and fungi. However, currently their formation still posses various drawbacks. The reported materials are often very specific and require complicated synthesis or surface modifications prior to linkage. For a further development and application a formation of the contact active materials should be synthetically simple and universal to provide their economic viability and enabling extended use. In this work we have applied a straightforward reaction based on silylation to form a series of contact active materials with covalently linked antimicrobial quaternary ammonium salt. The approach does not require pre-synthesis of antimicrobial agent or multistep surface modification. The method can be directly implemented on existent polymers and surfaces with no need for prior monomer modifications. The modified materials were characterized by spectroscopic and analytical methods and their antimicrobial activity was demonstrated. In addition, stability studies were performed to reveal the effectiveness and safety of the linkage of the biocide moiety. The new contact active materials benefit from a number of important advantages. In these materials an antimicrobial agent does not consumed by people, does not released to an environment and can be used in small amounts since the surfaces can reused.

Keywords: Contact Active Surfaces, antimicrobial, food

L-7
WINE – FROM GRAPES TO A MATURED PRODUCT

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"A beverage made of the fermented juice of any of various kinds of grapes, usually containing up to approximately 15 percent alcohol by volume" is the simple definition of wine. Behind this short definition a real complex mixture is hidden. In terms of organic chemistry wines contain compounds out of different chemical classes including carbohydrates, alcohols, aldehydes, esters, acids, proteins and vitamins. In addition there are polyhydroxy aromatic compounds such as tannins, anthocyanins and flavonols which contribute to colour and taste.

We know that in the wine there is an interaction between several hundred substances, all of which contribute towards the ultimate flavor, aroma and structure of the drink, yet it is very difficult to assess the importance of any one of them in isolate [1,2].

So it is evident, that the numbers of chemical reactions during the transformation process from grape to the fermented products and further on in the storage period is enormous.

In this presentation several aspects of reactions in all stages of wine making and production will be discussed.

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L-8
CONVERSION OF HISTAMINE IN HISTAMINOL: A NEW PERSPECTIVE FOR WINE QUALITY AND SAFETY?

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Wine is a source of bioactive compounds, largely investigated and well characterized, like resveratrol, anthocyanins, other antioxidant polyphenols and vasoactive biogenic amines. Histamine, like tyramine and other biogenic amines, often contaminates wine. This toxic compound is considered a critical contaminant of natural origin, produced by some decarboxylase-positive microorganisms (such as *O. Oeni*; *L. Hilgardii*; etc.). Histamine is correlated with histaminic poisoning, and some Countries set limits to histamine in wine, despite Swiss recently removed the limit of 10 mg/kg^{-1} for histamine in wine. Histaminol, a secondary metabolite of histamine [1, 2], was identified for the first time in wine by our Group. We firstly optimized a protocol for the synthesis of histaminol (not commercialized) and then we elucidated and confirmed its chemical structure, using ESI-MS and NMR techniques. Subsequently, using the synthetized histaminol, the biological activity of vasodilatation has been investigated, assessing no vasodilatation in isolated rat aorta, contrary to histamine. We have then validated an HPLC–DAD method useful for the quantification of histaminol in red and white wines following international guidelines [3,4]. A quick method for the sample clean-up, based on SPE C-18 cartridge, was developed. We applied the method to an overview on the histaminol content in 20 commercial samples of Italian wines. Red wines were significantly different from white wines ($p<0.05$), showing the highest level of histaminol. All these analytical approaches should be applied in future to investigate with more precision the formation of histaminol in musts and wines, suggesting the possibility to eliminate histamine (biologically converting it on histaminol), employing selected starters cultures. Finally, considering all these findings, we suggest here a formation hypothesis for histaminol, during the fermentation step.

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Keywords: Histaminol, wine, HPLC-DAD, validation

L-9

THE CASE STUDY OF ACRYLAMIDE FORMATION IN THERMALLY TREATED VEGETABLE IN RELATION TO TOTAL POLYPHENOLS CONTENT AND ANTIOXIDANT CAPACITY

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Formation of acrylamide, which is classified as a probable human carcinogen, in thermally processed foods has been confirmed by many research institutes. The highest concentration of acrylamide was reported mainly for potato and cereal food products such as French fries, potato crisps, breakfast cereals, biscuits, crackers, wafers, ginger bread, soft bread or coffee where indicative values was published by EFSA. Presented study pointed out that acrylamide level in grilled vegetable is also significant and concentrations of this processing contaminant could be up to thousands of μg/kg. Evaluation of this health risk in context to benefit of heat treatment from antioxidant capacity and phenolic compounds content point of view was correlated using multivariate analysis methods (PCA) in the case study for selected seasonal types of vegetable (eggplant, pumpkin and squash) also with the monitoring of amino acid asparagine content as a main precursor of acrylamide formation.

Keywords: Grilled vegetable, acrylamide, polyphenols, antioxidant capacity

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L-10

ANTIOXIDATIVE CAPACITY OF BETA VULGARIS EXTRACTS AND THEIR EFFECT ON THE FORMATION OF HETEROCYCLIC AMINES IN A MODEL SYSTEM

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Some heterocyclic amines (HAs), which are formed mainly via Maillard reactions in meat and fish products containing high amounts of proteins at high temperatures, are considered to be genotoxic carcinogens. There are various ways to reduce the concentration of HAs in the heated products, one of them is by using of plant extracts, particularly possessing strong antioxidant activity. However, the results on the effects of plant extracts on the formation of HAs are rather controversial and every new source of phytochemicals should be tested separately. The aim of this study was to assess the effect of fractions obtained from beetroot extract on the formation of such HAs as 2--amino-3,8-dimethylimidazo[4,5-f]quinoxaline (MeIQx), 2-amino-3 methylimidazo[4,5-f] quinoline (IQ) and 2-amino-1-methyl-6-phenylimidazo[4,5-b]pyridine (PhIP). The HAs were produced in a model system consisting of phenylalanine /glucose/creatinine, which was heated in diethylene glycol at 130°C for 2 h. Beetroot aqueous extract was fractionated using flash chromatography on a glass column containing 50 g Sephadex LH-20, and three different colour fractions were collected: F1 - light red, F2 - dark red and F3 - yellow. The antioxidant activity of these fractions was assessed by the 2,2-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) ($ABTS^{\bullet+}$) free radical cation scavenging and oxygen radical absorbance capacity (ORAC) assays. The antioxidative capacity of F1 was about 2 times higher than that of F2 and F3. The addition of the F1 to the reaction mixture reduced the amounts of PhiP, MeIQx and IQ by approximately 60, 77 and 87%, respectively. The F2 and F3 fractions were less effective or did not inhibit the formation of HAs. The differences in chemical composition of different fractions were investigated by UPLC–TOF–MS. It was found that F1 contained the highest amounts of betanin and vulgaxanthin I. The amounts of minor betanidins, isobetanin and vulgaxanthin II in F1 were also considerably higher than in the two other fractions. It may be concluded that beetroots may be preliminary considered as a potential source of compounds that may inhibit the formation of HAs, however, further fractionation is required to obtain pure beetroot constituents and to determine their individual input in the reaction mechanism of HAs formation. These issues will be also discussed in the presentation.

Keywords: Heterocyclic amines, beta vulgaris, betanidin, vulgaxanthin, antioxidative activity

L-11

INFLUENCE OF BAKING IN DIGESTIBILITY AND BIOACCESIBILITY OF CARBOHYDRATES, POLYPHENOLS AND FORMATION OF MAILLARD COMPOUNDS

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Cereal products and bread are rich sources of complex carbohydrates, phytonutrients and dietary fiber (including resistant starch) [1]. Bread baking is a very complex process that involves many variables, regarding both quality and operating aspects. During the baking process, simultaneous heat and mass transfer occurs within the product producing several physical and chemical changes. However a deeper study focusing on the influence of these changes in digestibility and bioaccesibility of carbohydrates, polyphenols and Maillard compounds is still needed. Different cereal products (wheat flour, toasted bread) were selected for the present study and the content and composition of indigestible fraction [2,3,4] including dietary fiber, resistant protein, resistant starch, extractable and non-extractable polyphenols, and Maillard compounds (mainly melanoidins),were analysed. The preliminary results (flour vs. toasted bread) showed significant changes in digestibility of starch and polyphenols bioaccesibility due to a decrease of 59.1% and 65% of total indigestible fraction and resistant starch respectively. Taking into account that primary skeleton of melanoidin is constituted by carbohydrates (including dietary fiber), polyphenols and proteins [5], the increase in digestibility of carbohydrates is expected to affect to melanoidins formation. Further investigation and detailed analysis of phenolics and Maillard compounds are in progress.

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Keywords: Carbohydrates, polyphenols, bioaccesibility, Maillard, baking

L-12

TRYPTOPHAN-CONTAINING DIPEPTIDES: BIOACTIVE COMPOUNDS IN FOODS WITH A BLOOD PRESSURE LOWERING EFFECT?

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Bioactive peptides have relevance for the development of functional food products. In this context, peptides with a possible blood pressure lowering effect due to inhibition of angiotensin converting enzyme (ACE) are of particular importance. ACE is one of the key enzymes in blood pressure regulation by generating the vasoconstrictor angiotensin-II and degradation of bradykinin, a vasodilatory peptide. In our group, we identified several tryptophan-containing dipeptides as a new class of natural ACE inhibitors [1]. These bioactive peptides are inactive within the sequence of their precursor proteins like α-lactalbumin or lysozyme and can be released by specific enzymatic hydrolysis in vitro [2]. For instance, Ile-Trp (IW, IC_{50}=0.7 µM) and Trp-Leu (WL, IC_{50}=10 µM) originating from the sequence of the α-lactalbumin or Ala-Trp (AW, IC_{50}=20 µM) from lysozyme show competitive, fully reversible ACE inhibition. To exert physiological effects in vivo, it is important that these bioactive peptides pass the gastrointestinal tract, are absorbed and reach the cardiovascular system in an active form. For studies on the bioavailability and effectiveness of the peptides in vivo, a LC–ESI–MS/MS method was developed to quantify the content and to determine the half-life of these dipeptides in human plasma. For this method, the limit of detection was 0.2 nM for IW and 0.3 nM for WL. The limit of quantification was 0.4 nM and 0.6 nM, respectively. This LC–ESI–MS/MS method is capable to investigate the bioavailability of tryptophan-containing, bioactive peptides in human plasma. First pharmacokinetic studies in humans showed that the oral intake of IW resulted in a significant increase of IW concentrations in blood compared to baseline levels. The half-life for IW in vivo was calculated to 0.71±0.03 h. These results demonstrate that IW is to some extent absorbed, reaches the cardiovascular system and can cause an ACE-inhibition in vivo due to its relative good stability. Protein hydrolysates containing tryptophan containing dipeptides could be interesting ingredients for functional food as an effective and natural prevention for hypertension, but also as a good source for the amino acid tryptophan.

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Keywords: Angiotensin converting enzyme (ACE), enzyme inhibitors, bioactive peptides, protein hydrolysate, functional food

L-13
COFFEE PHENOLICS DEGRADATION AND FORMATION OF BIOACTIVE METABOLITES BY HUMAN COLONIC MICROFLORA

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Coffee is one of the most consumed beverages and a rich source of chlorogenic acids, a group of phenolics comprising hydroxycinnamates such as caffeic acid and ferulic acid, linked to quinic acid. However, only ca. 30% of the chlorogenic acids are absorbed in the small intestine, whereas the remaining ca.70% reaches the colon, where they can be fermented by the gut microbiota to a wide range of low-molecular metabolites. These metabolites might play an important role in the biological activity ascribed to phenolics, such as antioxidant, anticancerogenic and antiinflammatory activity at colon level. The aim of this work was to study the effect of the colonic microflora on the breakdown of coffee chlorogenic acids and the formation of metabolites using an in vitro fermentation model. The colonic metabolism of single compounds like caffeoylquinic and caffeic acids has been investigated, but up to our knowledge, this is the first time that coffee brew is used. Previous to the in vitro fermentation, the phenolic compounds present in espresso coffee brew were identified and quantified by HPLC-PDA-MS^3. 0.5 g freeze-dried espresso coffee was incubated with fresh faecal samples from three healthy donors in the presence and absence of glucose. Samples were analyzed by HPLC-PDA-MS^2 at 0 h, 1 h, 2 h, 3 h, 4 h, 6 h and 24 h. A total of 12 chlorogenic acids and 4 chlorogenic acid lactones were found in the espresso coffee brew and at the initial point of faecal fermentation (0 h). Additionally, traces of caffeic and ferulic acids were also found. Generally, the addition of glucose enhanced the degradation of coffee phenolics, although there were significant differences among the individuals. After 3-6h only traces of phenolic compounds initially present in the fermentation medium were found, except for volunteer 2 that showed an inhibited degradation of dicaffeoylquinic acids, 4-caffeoylquinic acid lactone, feruloylquinic acids and their lactones that still remained in significant amounts after 6h. This may be attributed to the well-known different bacterial populations present in the individual faecal microbial communities, harboring specific enzymatic capacities. Caffeic acid was the first metabolite detected during the fermentation but it was degraded after 4–6 h. The major phenolic end-product identified was 3-(3,4-dihydrophenyl)propionic acid (dihydrocaffeic acid), which was still present in significant amounts after 24 h. Also, 3,4-dihydroxybenzoic acid (protocatechuic acid) was detected at low levels during fermentation, and traces of 3-(3-hydroxyphenyl)propionic acid were found at 24 h. In conclusion, coffee chlorogenic acids were rapidly degraded by the colonic microflora. However, low amounts of them, and their degradation products or metabolites, remained for several hours during fermentation. Therefore, the presence of these bioactive compounds might explain some of the health benefits of coffee brews, mainly those in colon, including prebiotic effects.

Keywords: Coffee, chlorogenic acids, gut microflora, colonic metabolism, bioactive compounds

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L-14
OPTIMIZATION OF THE FORMATION OF VINYLDITHIINS, A THERAPEUTICAL COMPOUND FROM GARLIC

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Garlic has been used worldwide for centuries for its taste, but also for its health benefits. The therapeutic compounds are mostly organosulfurs, and can be formed when an enzyme, alliinase, gets in contact with its substrate, alliin. Both are separated in an entire cell, and react when the plant is damaged. The product of the enzymatic reaction, allicin, turns quickly into different molecules with potential beneficial effects. The way garlic is processed has a major influence on the released products. Indeed, an aqueous or oily medium, as far as the temperature, can emphasize the formation of specific compounds. Vinyldithiins are one of the active molecules, and are part of the garlic's organoleptic properties. Their effects against obesity have been recently proven. This study aims to describe the conditions of vinyldithiins formation, as well as its extraction, purification and analyse. Vinyldithiins are mostly produced when garlic is crushed in edible oil, at low temperature. Extraction conditions (garlic/oil ratio, oil and garlic source, temperature and extraction time) were optimized. Analysis of the results were performed by HPLC.Finally, a purification process was set up. These results allow better knowledges on vinyldithiins formation that can be applied in garlic-based nutraceuticals. They might also lead to new uses of garlic in the production of highly valuable compounds.

Keywords: Garlic, vinyldithiin, nutraceutical, oil extraction

L-15
NOVEL TECHNOLOGIES TO REDUCE PROCESSING CONTAMINANTS AND SOME OTHER UNDESIRABLE FOOD COMPONENTS

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Various heat induced reactions take place during frying both in potato chips and respective oil bath. Depending on the composition of potato tubers, not only flavour significant compounds are formed at commonly used frying temperatures around 170–180°C, but also formation of potentially carcinogenic acrylamide occurs through Maillard reaction, specifically if raw material contains higher amounts of sugars. At the same time, unsaturated fatty acids, both free and bound in triacyycerols (TAGs) which are the major components of oils, undergo free radical chain reaction yielding in the initiation phase highly unstable hydroperoxides breakdown of which give origin to a variety of products, such as cyclic peroxides, epoxides, hydroxy-/ oxoderivatives, hydrocarbons and also polymers. Another group of hazardous substances are MCPD formation of which is also associated with processing practices. To minimize the extent of these undesirable reactions, vacuum frying enabling preparation of potato chips at lower temperatures, around 120°C, is seen as a challenging alternative. In our study, two sets of frying experiments were established to compare the quality of obtained potato chips in terms of processing contaminants level, as a well as, sensoric properties.

L-16
ACRYLAMIDE FORMATION IN FOODS: ROLE OF COMPOSITION AND PROCESSING

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Detection of acrylamide levels in processed foods has been an intensive area of research shortly after the discovery of acrylamide in heated foods by the Swedish researchers in April 2002. Several researchers have established that the main pathway of acrylamide formation in foods is linked to the Maillard reaction and, in particular, the amino acid asparagine. Thermally processed foods encompass a vast range of different products with many ingredients, processes, recipes and scales of operation. The resulting acrylamide concentrations in these foods change with great deviations as influenced by product composition and thermal processing conditions. Basic facts on the mechanism of acrylamide formation and factors affecting its concentration in thermally processed foods are overviewed in this presentation. Recent research findings for mitigation of acrylamide by means of recipe and process modifications are discussed. In addition, importance of mechanistic models taking physical and chemical changes into account is exemplified by means of a frying model.

Keywords: Thermal processing, acrylamide formation, modelling

L-17

FURAN IN HEAT-PROCESSED FOODS: FIVE-YEARS FILED MONITORING, REDUCTION AND RISK ASSESSMENT STUDY IN KOREA

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The aim of this presentation is to monitor and assess the risk associated with the presence of furan in various food products consumed in Korea. An optimized analytical method was used for the analysis of furan levels. The optimized solid phase microextraction (SPME) fiber exposure conditions as follows for temperature, time and amount of sample were 50°C, 5 g (ml) and 20 min, respectively. Furan was detected in all food samples tested, at levels ranging from 0.4 ng/g in canned crab to 814 ng/g in ground roasted coffee powder. The furan levels in coffee, canned fish, canned meats, sauce, soup, retort, canned vegetables, baby foods, nutritional/diet drinks, confectionary, biscuits and snack, juice, jams, canned fruit were in ng/g 169, 56.1, 30.1, 21.1, 18.1, 15.6, 10.9, 10.6, 7.1, 5.4, 3.7, 3.2, 2.9, respectively. Furan concentrations in baby food products were between 1–102.5 ng/g. In the baby food in retort packaging, the level of furan was reduced by 15–33% after heating the foods at 50°C without a lid. The levels of furan decreased to 58% in beverage products for babies, after storing them at 4°C for 1 day without a lid. The levels of furan in canned foods such as cereal and vegetable were reduced by 0–52% when they were stored without stirring in a refrigerator at 4°C for 1 day. The total exposure estimate of furan was determined to be 10.6 ng/kg/day (maximum, 20 ng/kg/day) for adults, and 17.4 ng/kg/day (maximum. 84.9 ng/kg/day) for babies. Exposure estimates found in this study are lower than those prescribed by the US FDA.

Keywords: Furan, processed foods, SPME, monitoring, risk assessment

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L-18

FORMATION OF AROMA-ACTIVE COMPOUNDS AS WELL AS TOXICOLOGICALLY RELEVANT STYRENE DURING THE PRODUCTION OF WHEAT BEER

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2-Methoxy-4-vinylphenol and 4-vinylphenol are important compounds for the overall aroma of wheat beer, which are formed from their precursors (phenolic acids: ferulic and *p*-coumaric acid, respectively) after thermal or enzymatic decarboxylation during the brewing process. However, in the same way the food-borne toxicant styrene, which is classified as "possibly carcinogenic to humans" (group 2 B) by the International Agency for Research on Cancer (IARC), is generated from cinnamic acid. Therefore, it is a major challenge for breweries to produce wheat beers with reduced styrene concentrations but not to change in parallel the consumers expected aroma. Thus, the aim of the present study was the characterisation of the most important odorants of wheat beer using the molecular sensory science concept. Thereby, a high value was set on vinyl aromatic compounds and their respective precursors both in commercially available wheat beers and in process intermediates in dependency of the applied yeasts. Quantitation via stable isotope dilution assays (SIDA) and calculation of the respective odour activity values (OAVs) revealed ethanol (OAV=1610), (E)-β-damascenone (325), 3-methylbutyl acetate (231), ethyl 2-methylpropanoate (225), ethyl butanoate (115), and 3-methyl-1-butanol (58) as the most aroma-active compounds. With OAVs of 20 and 11, respectively, also the vinyl aromatic compounds 2-methoxy-4-vinylphenol and 4-vinylphenol were proven as important odorants for the overall aroma of wheat beer. In wheat beers with a more pronounced aroma, the concentrations of the vinyl aromatic compounds were much higher compared to a wheat beer only offering a lower aroma impression. Thereby, differences in concentrations were in good agreement with the sensorial evaluation, e.g., 2020 µg (beer A) and 159 µg (beer B) of 4-vinyl-2-methoxyphenol/mL, 882 µg and 59.8 µg of 4-vinylphenol/mL as well as 1.63 µg and 0.63 µg of 2-methoxyphenol/mL. But, associated with these results, also a higher styrene concentration was analysed in beer A (27.7 µg/mL) in comparison to beer B (1.67 µg/mL). Therefore, the precursors (phenolic acids) as well as the respective decarboxylation products were analysed in all process intermediates (seed, malt, mash, unboiled wort, cast wort, green beer, ready-to-drink beer) to get initial ideas about the most crucial steps for the formation of desired odorants, e.g., 2-methoxy-4-vinylphenol and 4-vinylphenol, and the undesired food-borne toxicant styrene. Brewing experiments proved the key role of the yeast in the formation of different amounts of these decarboxylation products. In conclusion, the study showed a good correlation of the overall aroma of wheat beer with the amounts of vinyl aromatic compounds, whose formation was mostly influenced by applied yeasts during the brewing process.

Keywords: Wheat beer, odorants, food-borne toxicant, styrene, precursor

L-19
INGREDIENTS FROM LOW VALUE BY-PRODUCTS OF SELECTED FOOD MANUFACTURING INDUSTRIES

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The EU food industry generates ca. 620 million tonnes of product each year. About half the biomass used to produce food ends up as waste or by-products. Brewing and shellfish processing are examples. Annually, the EU brewing industry produces ca. 3.4 million tonnes of brewers' spent grain (BSG), accounting for 85% of by-products. Processing of crustaceans leads to 50-70% of the original biomass as residue, equivalent to 328 thousand tonnes each year. Food processing residues have various possible uses: biofuel, biomaterials, animal feed or chemical feedstocks – the latter including feedstocks for food ingredients. This presentation focuses on possible uses as food ingredients of food processing residues from BSG, and chitin from shellfish processing. Major components of BSG are fibre and protein, with lower levels of lipid and ash. Fibre comprises 70% (dry mass basis) of BSG. Arabinoxylans are a component of the fibre and contain bound hydroxycinnamic acids. Enzymic release of these acids gives phenolic acid extracts with functional properties. Tocotrienols are structurally related to tocopherols and possess have anti-carcinogenic, neuroprotective and cholesterogenesis-inhibiting properties. α-Tocotrienol has higher antioxidant activity in biological membranes than α-tocopherol and oil from BSG contains 2-3-fold higher amounts of tocotrienols than tocopherols. BSG oil is reported to decrease serum LDL in humans. Germinated barley foodstuff (GBF) is produced from BSG by removing the cellulose-rich husk. Administration of GBF to patients suffering from ulcerative colitis, can result in significant decreases clinical and endoscopic scores and a significant increase in faecal butyrate. Chitin is a polysaccharide in crustacean shells. The deacylated form, chitosan, is readily produced from chitin. Chitin and chitosan possess various useful properties including the ability to form films and fibres, adsorb metals and coagulate suspended solids. These properties can lead to various food applications. Chitosan-based edible coatings are able to control microbial growth in horticultural produce. Dipping tomatoes in 1% chitosan and 0.1% beeswax prior to inoculation with *E. coli* DH5α inhibited microbial growth during storage at 25°C for 48 h. The ability to adsorb metals can result in the significant inhibition of lipid oxidation in salmon stored at 4°C. Chitin nanofibres prepared from shrimp shells are able to decolorise sugar syrups with an efficiency of over 70%. There are numerous other examples of the potential use of other food processing residues for the production of added value food ingredients with a range of functionalities. Exploitation of these residues has potential economic, environmental and health benefits for the food industry and consumer.

Keywords: Food processing, food waste, brewers' spent grain, chitin, chitosan

L-20

CHEMICAL AND FUNCTIONAL CHARACTERISTICS OF FIBER ISOLATED FROM HAZELNUT SKINS: NEW PREBIOTIC INGREDIENT FROM BY-PRODUCTS?

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In the past few years, there has been an increased interest toward alternative sources of "functional" ingredients that, in addition to basic nutritional action, also promote health, reducing the risk of chronic diseases. Particular attention has been directed towards indigestible carbohydrates that are able to stimulate growth and activity of selected probiotic bacteria in the colon. However, the study of prebiotic oligosaccharides has so far being almost an exclusive focus of animal milks (particularly human milk), with hundreds of articles published on the topic on human milk oligosaccharides. Hazelnut skins spontaneously detach from the seed during or after roasting; their accumulation in the food industry makes them a waste/underutilized by-product. However, it was previously shown that roasted hazelnut skins are a rich source of antioxidant polyphenols [1]. In the present work, hazelnut skins were analytically characterized on their fiber composition, and used as source of functional fractions, tested on some probiotic strains evaluating their prebiotic and cryoprotectant capacity. Hazelnut skins were extracted firstly unmodified, then extracted and fractionated after a multi-enzymatic treatment (arabinase, cellulase, β-glucanase, hemicellulase, and xylanase). The fiber was isolated, removing proteins, peptides, lipids and polyphenols to ensure proper functional testing and characterization by Mass Spectrometry. Both the soluble and insoluble fractions isolated from the hazelnut skin promoted in vitro the growth of probiotic lactobacilli *L. crispatus* and *L. plantarum*. Matrix-Assisted Laser Desorption/Ionization Fourier Transform Ion Cyclotron Resonance Mass Spectrometry (MALDI–FTICR) [2] and Micro-Chip liquid chromatography combined with high-performance Mass Spectrometry (Agilent 6520 Accurate-Mass-Quadrupole-Time-of-Flight) [3] were used to characterize all free oligosaccharides expected from hazelnut skins. The high resolution and mass accuracy of the analytics employed readily yielded precise oligosaccharide composition in terms of constituent monosaccharides (Hex, HexNAc, Fuc, NeuAc, HexA, Xylose) resulting in the identification of over twenty neutral oligosaccharides. Unlike most novel food ingredients in which the basic commodities are in low supply and industrial technologies to isolate them unexplored, this project has already assembled the core technologies to characterize the complex oligosaccharides in a widely available by-product of the food industry. Finally, this study provides new insights and suggestions about health-promoting properties of fiber-oligosaccharides naturally found in hazelnut skins, adding value to an industrial by-product.

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Keywords: Fibre, prebiotic, hazelnut, MALDI-FTICR, oligosaccharides

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L-21

IMPACT OF TEMPERATURE AND WATER ACTIVITY ON ENZYMATIC AND NON-ENZYMATIC REACTIONS DEVELOPMENT IN RECONSTITUTED DRIED MANGO

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Mango fruit (*Mangifera Indica* L.), one the most important tropical fruits, is highly perishable and required therefore to be quickly stabilized. Among the technological solutions, drying appears to be a very promising stabilization solution. The traditional drying of mango can be separated in two phases: the first one carried at ~80°C for 10 to 12 hours makes the water activity (a_w) decline from 0.99 to 0.8. During the second phase, the drying temperature is reduced to 40°C until the a_w reach ~ 0.5. These conditions are highly favorable to enzymatic (1st drying) step and non-enzymatic (Maillard) reactions (2nd drying step). The focus of this work was therefore to highlight the coupled effects of aw and T°C on the development of these classes of reactions all along the process. For a global comprehension of the browning of mango during the drying process, we used lyophilized powder reconstituted to the required water activities and thenafter heated during various times. Several precursors (reducing sugars) and intermediary products of the Maillard reaction (5-hydroxymethyl furfural (5-HMF) and other neo-formed furan- and pyran-derivated compounds) were analyzed to estimate non-enzymatic reaction intensity while polyphenoloxidase (PPO) and peroxidase (POD) were associated to enzymatic browning. We confirm that the highest production of 5-HMF as a marker of the Maillard reaction mainly occurs for water activities of 0.6. Kinetic analysis of Maillard reactions precursors and intermediate products in model mango system permit to identify asparagine and glutamic acid as limiting precursor, and 5-HMF among significant a_w-dependent products. Furoic acid and furfural linked to the ascorbic acid degradation followed the same tendency. The impact of the water activity on the thermal degradation of PPO and POD was proved to be strongly different. After 4h at 40°C, the PPO, POD remaining activity at a_w 0.65 was 22 and 97% respectively while these values being 82 and 64% at 0.98 respectively. As a conclusion, we identified critical points in the drying process making the fruit turn to brown and thus improved our global comprehension of the involved mechanisms. The approach we developed could be useful to help to understand the biochemical mechanisms in complex matrices such as fruit and vegetables.

Keywords: Mango, drying, water activity, Maillard, PPO-POD

L-22

CONTROLLING MAILLARD REACTION BY REACTANTS ENCAPSULATION: SODIUM CHLORIDE IN BISCUITS

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Formation of advanced glycoxidation end-product (AGE) including hydroxymethylfurfural (HMF) and acrylamide have been an intensive area of research in the last decades. The Maillard reaction beside sugar degradation represents the main chemical pathway for AGE synthesis and the presence of some reactants such as sodium chloride may influence the reaction mechanism through the dehydratation of various key intermediates. The two main purposes of this work were to evaluate the specific effect of sodium chloride on HMF formation and to demonstrate the protective effect of encapsulation. NaCl in biscuits is added for sensorial reason so it must be encapsulated as long as possible but in the free form at the very end of the cooking process. To achieve this goal three lipid-based coating materials able to melt during the biscuits such as stearic and palmitic acid blend (SAP), candelilla wax (CanW) and carnuba wax (CarW) were selected.

Thirteen biscuits recipes were prepared in order to highlight the different action of free and encapsulated sodium chloride on mechanism reaction. High pressure liquid chromatography coupled to diode array detection together with liquid chromatography and mass spectrometry were used to show the different behavior of free and encapsulated sodium chloride.

Results demonstrated that NaCl encapsulation significantly reduced the formation of HMF: the more heat resistant is the coating the more pronounced was the effect. The effect of reduction of HMF formation is clearly visible in figure 1 where the capsule of NaCl coated with carnauba wax has an higher effect in HMF reduction.

Sensorial properties of the biscuit were evaluated too and no difference were found on the biscuit with NaCl encapsulated vs the biscuit with regular salt.

This is the first paper showing that the encapsulation of Maillard active reactants can be used as innovative strategies to prevent the formation of potentially harmful compounds in some foods.

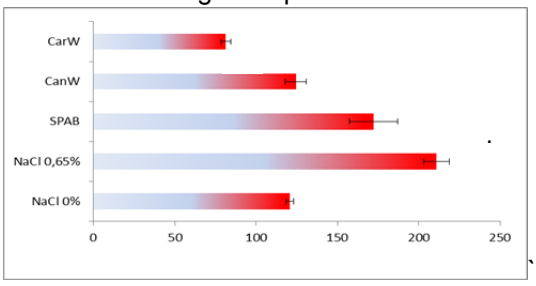

Figure 1: HMF concentration in biscuits prepared with different types of encapsulated NaCl. All samples have the same amount of NaCl (0.5 g). SPAB: encapsulated NaCl with stearic / palmitic acid blend coating; CanW: encapsulated NaCl with candelilla wax coating; CarW: encapsulated NaCl with carnauba wax coating. The significant differences among HMF content were determined by Anova analysis and Duncan's multiple range test ($p \leq 0.05$).

Keywords: Encapsulation, HMF, Maillard reaction, NaCl

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L-23

THE POTENTIAL OF AMBIENT MASS SPECTROMETRY FOR RAPID MONITORING OF MEAT AND FISH FRESHNESS

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Meat as well as fish are foods very sensitive to storage conditions. An inappropriate handling can lead not only to the negatively perceived organoleptic properties, but also to extensive microbial contamination, the products of which may pose a serious health risk to consumers. In this respect, biogenic amines represent an important indicator of meat freshness. Traditionally, the analysis of biogenic amines includes reversed phase high-performance liquid chromatography (HPLC) with fluorescence detection using derivatisation with dansyl chloride or -phthalaldehyde (OPA). The disadvantages of such method include a long analysis time and derivatisation. In this work, the potential of ambient mass spectrometry was evaluated for rapid determination (no chromatographic separation, no derivatisation) of biogenic amines and other markers to indicate the changes in meat and fish muscle caused by storage conditions. For this purpose, a direct analysis in real time (DART) ion source coupled to high-resolution mass spectrometry (MS) with an orbitrap analyser was used. The data generated under the conditions of DART–orbitrap-MS were also compared with a method employing hydrophilic interaction chromatography (HILIC) coupled to high-resolution mass spectrometry. In addition to this target analytes approach, the potential of (non-target) metabolomic fingerprinting for the assessment of meat quality will be outlined.

Keywords: DART, ambient mass spectrometry, biogenic amines, meat, HILIC

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L-24
OXIDATION OF TRIACYLGLYCEROLS IN PRESENCE OF CAROTENOIDS - IDENTIFICATION OF OXIDIZED SPECIES BY LC-MS

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For the analysis of the oxidation of triacylglycerols (TAGs) HPLC-ESI-MS was used. The oxidation experiments were carried out at 110°C in presence of abundant oxygen. In presence of β-carotene the peroxide value and other oxidation products from the TAGs increased quicker compared to the carotenoids free TAG mixture indicating a pro-oxidative effect of β-carotene under these conditions. Among the oxidized species of the TAGs mono-hydroperoxides, bis-hydroperoxides, epoxy-epidioxides, and epoxides were the major compounds identified.

Keywords: Oxidized triacylglycerols, carotenoids

Acknowledgement: Higher Education Commission of Pakistan

L-25

QUANTITATIVE AND QUALITATIVE ANALYSIS OF HIGH MOLECULAR COMPOUNDS IN VEGETABLE OILS FORMED UNDER HIGH TEMPERATURES IN THE ABSENCE OF OXYGEN

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This study simulates temperature conditions of vegetable oil refining at the final stage – deodorisation or physical refining. High temperatures up to about 250 °C in the absence of oxygen lead especially to cis-trans and positional isomerization of double bonds in polyenoic fatty acids and subsequently to triglyceride polymerization with formation of high molecular compounds without an oxygen according to the principle of Diels-Alder reaction. The presence of trans-fatty acids and polymer products in vegetable oils and products made from them is undesirable for health and therefore the producers try to keep the content of *trans* fatty acids in refined oils under 1%. Standard sunflower oil and sunflower oil with admixture of canola oil were heated at 240 and 260°C in the argon atmosphere in order to investigate the process of polymerization. The content of polymer products was determined by HPSEC/ELSD method and for qualitative analysis HPSEC with APCI–MS detection was used. The content of polymers increased with heating time and temperature while amount of origin triglyceride monomers decreased. The linear correlation between the polymer and conjugated linoleic acid content, as the confirmation of Diels-Alder reaction mechanism, was found. Usage of HPSEC/APCI–MS analysis allowed to detect three types of high molecular compounds formed under the heating of oils in the absence of oxygen. Based on the structures of new polymeric compounds generated in heated oils the thermal hydrolytic degradation of ester bonds in triglycerides was showed and thus diglycerides or monoglycerides as the products of hydrolysis are primarily involved in the process of polymerization.

Keywords: Polymerization, triglyceride, conjugated linoleic acid, HPSEC, APCI–MS

L-26
IMPACT OF INTRINSIC AND EXTRINSIC PARAMETERS ON THE FORMATION OF TOXIC ALDEHYDES IN FOODS

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Foods enriched with poly-unsaturated fatty acids are susceptible to lipid oxidation which subsequently leads to the formation of toxic aldehydes such as malondialdehyde, 4-hydroxynonenal, 4-hydroxyhexenal, acrolein and crotonaldehyde. To evaluate the impact of intrinsic parameters on the formation of these oxidation products, experiments were carried out in buffered model systems (Oil, in buffer pH 7) containing different levels of poly-unsaturated fatty acids and simulating foods such as enriched milk, cream, sauces and dressings. Based on this model, the impact of extrinsic factors such as chilled storage (4°C) at oxygen levels of 0.5, 1, 5, 21 and 70% as well as the effect of light were evaluated. In addition to the toxic aldehydes, the content of conjugated di-, and trienes was also monitored along with the decrease of tocopherol content in the oil fraction.

Keywords: PUFA, lipid oxidation, unsaturated aldehydes

L-27
STUDIES ON THE FORMATION PATHWAY OF ACROLEIN

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Acrolein (2-propenal) is a highly reactive and volatile α, ß-unsaturated aldehyde, which is formed during the combustion of fossil fuels and during the heating of cooking oils. Although its presence in heated edible fats and oils is of special interest due to its high toxicological relevance, data about the formation of acrolein on a molecular basis are scarcely available. Up to now, two major hypotheses about acrolein formation from triglycerides are discussed: on the one hand, acrolein may be released by partial hydrolysis of triglycerides, followed by a dehydration step of glycerol to acrolein [1, 2]. On the other hand, formation pathways starting from free fatty acids including oxidative lipid degradation have been suggested [3, 4]. In fact, recent studies on the generation of acrolein during the heating of different fats and oils demonstrated that acrolein formation is closely linked to the content of unsaturated fatty acids [5]. However, the key intermediates of acrolein generation were not characterised so far and therefore, the chemical reactions remained largely hypothetical. Thus, the objective of this study was to clarify the mechanism of acrolein formation from triglycerides. Therefore, synthesised precursors, e.g., partially [^{13}C]-labelled triglycerides as well as fatty acid hydroperoxides were used for model systems. To get deeper insights into the formation pathway, the effect of antioxidants on acrolein generation during heat-processing was investigated in additional studies.

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Keywords: Acrolein, labelling studies, triglycerides, formation pathway, fatty acid hydroperoxides

L-28
ISOTHERMAL KINETICS OF MALONDIALDEHYDE CONTENT CHANGES IN CHICKEN MEATS

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Lipid oxidation is a classic parameter of meat quality evaluation, mostly when the composition of the meat is modified, even by modifying the animal's diet or by adding food supplements during post-slaughter meat processing. Malondialdehyde (MDA) is commonly accepted as the major lipid oxidation product in meat and considered as a terminal accumulating compound. It is generally measured together with the other coloured aldehydes resulting from lipid oxidation, with a global methodology called TBARS (thiobarbituric acid reactive substances) index. This index is expressed as equivalent MDA, considering MDA as the preponderant compound by way of a calibration using 1,1,3,3-tetramethoxypropane (TMP) or 1,1,3,3-tetraethoxypropane (TEP). For more precision, MDA is sometimes directly quantified by high performance liquid chromatography (HPLC). The TBARS index or MDA content are mostly measured on fresh meat, but several studies have focused on their changes during meat processing (storage, cooking or other processes). This study set out to characterize changes in MDA content during meat cooking. For this purpose, small samples (1.5 g) of ground chicken thighs were conditioned in plastic bags to form thin films of less than 0.5 mm. The plastic bags were then immersed in a water bath at different temperatures (50, 70 and 100°C) for different immersion times (from 0 to 30 min). Due to the very thin layer of meat, the temperature was considered as constant from the immersion time to the cooling time in an ice bath. MDA content was measured by HPLC. Different meat samples enriched with omega 3 poly-unsaturated fatty acids (transferred in the meat from a dietary flax seeds supplementation) were used to determine the differences in MDA content depending on the vitamin E and xanthophylls' contents in the diets administered to the chickens. The results showed that the appearance kinetics of MDA depended on the heating temperature. At 70°C, three phases were observed in MDA content change: first an increasing phase, then a plateau phase and finally a decreasing phase. At 100°C, the three phases also appeared but the results were more dispersed, probably because of contradictory MDA appearance and disappearance reactions at the same time. Finally, at 50°C, no modification on the MDA content was observed. So, the best temperature for studying MDA content changes during meat cooking was 70°C. It was selected to study differences between the meat sources. The diet composition had no effect on the kinetics of MDA content changes but it greatly affected the MDA contents. The highest MDA contents were determined in meats from chickens reared without a vitamin E dietary supplement.

Keywords: Oxidation, omega 3, chicken meat, malondialdehyde, kinetics

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L-29
AMINO ACID DEGRADATIONS PRODUCED BY LIPID OXIDATION PRODUCTS

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Nonenzymatic browning has a great impact on sensory and nutritional properties of foods. Thus, it plays a major role in the development of brown color and the formation of taste and flavor compounds during food processing. On the other hand, loss of essential amino acids and formation of toxicological suspect compounds are also produced. A significant number of these changes are related to amino acid degradation reactions. Although they are, at present, mostly considered to be produced by carbonyl-amine reactions initiated by carbohydrates, recent studies have pointed out that these reactions can also be produced by lipid-derived reactive carbonyl compounds. This lecture will review the present knowledge of the pathways involved in amino acid degradation by lipid oxidation products. Thus, the ability of lipid oxidation products to convert amino acids into Strecker aldehydes, alpha-keto acids, and biogenic amines will be discussed. In addition, interconversions among these products as well as their transformation into shorter aldehydes and olefins will also be considered. The effect of processing conditions on the different reaction mechanisms will be discussed in an attempt to find out the most appropriate reaction conditions that favor the formation of the most desirable compounds at the same time that production of compounds with deleterious properties is avoided.

Keywords: Maillard reaction, lipid oxidation, amino acid degradation, flavor compounds, acrylamide

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L-30

DISCOVERY, SENSORY ACTIVITY AND TASTE RECEPTOR ACTIVATION OF THERMALLY GENERATED TASTE MODULATORS IN FOODS

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The identification and characterization of thermally modified food ingredients represents a key discipline in food chemistry throughout the past decades. Apart from the degradation of valuable constituents, also the generation of previously unknown taste active and/or taste modulating compounds constitutes a promising field of research.

In recent years, the activity guided fractionation of food led to the isolation and identification of a variety of food-borne taste modulators, but as the isolation of tastants from the complex food matrix represents a challenging task, the utilization of simplified food model systems is of growing interest and successfully applied [1–4]. The presentation will highlight some recent success in the discovery of taste modulating substances by means of targeted model reactions, the evaluation of their sensory and taste receptor activity, and the validation of their natural occurrence in foods.

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Keywords: Maillard reaction, taste, taste enhancer, taste modulator

L-31
NOVEL APPROACHES IN INDUSTRIAL RESEARCH TARGETING QUALITY ATTRIBUTES OF EXTRUDED FOOD PRODUCTS

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Extrusion cooking has gained considerable importance in food industry for manufacturing numerous food items including cereal products. It permits in one processing step to perform several operations such as mixing, transporting, cooking and texturing. However, desirable flavour characteristics associated with conventionally cooked cereals do not develop to the same extent during extrusion cooking. Consequently, the extruded food products are generally inferior in flavour as compared to those obtained by conventional thermal processing. Extrusion temperature, moisture, pH and composition of the feedstock were shown to have the highest effect on the volatiles formed. Unfortunately, only limited information is available on the formation of odour-active compounds. The use of specific precursors and ingredients is of particular interest to achieve flavour modulation. However, knowledge has to be built-up concerning the effect of precursors and extrusion parameters on flavour development and on other key product attributes such as colour and texture, while mitigating the formation of undesirable compounds. This review will focus on holistic and targeted approaches resulting in a better insight on the role of ingredients and process parameters to obtain desirable food quality attributes. Experiments are based on *(i)* the experimental design approach and *(ii)* using labeled precursors to elucidate the formation mechanisms of defined key odorants with caramel, buttery, and roasty notes by applying the carbohydrate module labelling (CAMOLA) method.

Keywords: Maillard reaction, extrusion cooking, quality attributes, experimental design, formation mechanisms

L-32

THE STRECKER DEGRADATION: AN „OLD“ REACTION WITH NEW IMPACT ON FOOD AROMA

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Since its discovery about 150 years ago in a model system, the thermally induced Strecker degradation of alpha amino acids, is well confirmed today as an important source of aroma-active aldehydes in foods. The generally accepted reaction pathway runs *via* a decarboxylation/ transamination of the amino acid, preferentially initiated by alpha dicarbonyls, which commonly stem from carbohydrate degradation. Recently, it has been shown that also Strecker acids and Strecker amines are generated via the same degradation path from the respective parent amino acids. In the early sixties of the past century, studies on cocoa aroma were among the first reports on the importance of Strecker aldehydes in food flavor, and because nearly all raw materials used in food production contain free amino acids, it is obvious that Strecker aldehydes were later on characterized as key odorants in many other processed foods, such as meat, coffee or nuts. In the lecture, results of our recent investigation on aroma formation in cocoa beans during fermentation and roasting, will be presented showing (*i)* that Strecker aldehydes are already formed during fermentation, and also that the well-known binary reaction of amino acids and dicarbonyls is not the most effective source in the formation of Strecker aldehydes during the cocoa roasting process. Very recently, transient intermediates of the Strecker reaction could been identified in roasted cocoa beans as well as in other thermally processed foods, which are able to release Strecker aldehydes upon contact with saliva during eating. The structures of these new Strecker intermediates will be presented opening a new avenue to improve the overall aroma of comfort foods, such as chocolate, by in-mouth chemistry.

L-33
IMPROVEMENT OF THE AROMA OF THE GLUTEN-FREE BREAD BY AROMA-ACTIVE MALT PREPARATIONS

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Celiac disease (CD) is one of the most frequent food intolerances worldwide, caused by in-gestion of the storage proteins (= gluten) of wheat, rye, barley and possibly oats. The prevalence is about 1 % of the population. CD is characterized by small intestinal mucosal injury and malabsorption in genetically predisposed individuals. The current essential treatment is a strict lifelong withdrawal of gluten from the diet allowing CD-patients to live without symptoms. CD-patients have to consume gluten-free food products, which are made from gluten-free cereals and pseudocereals. Although such gluten-free bread products are commercially available, one major shortcoming is that they have a poor aroma as compared to their gluten-containing counterparts made from wheat. Therefore, the aim of this study was to produce gluten-free bread with an aroma quality comparable to conventional wheat bread. Grains of different cereals (corn, sorghum, millet) and pseudocereals (buckwheat) were germinated and kiln-dried. Kiln drying conditions were optimized to yield malt preparations with the best overall aroma impression as determined by sensory tests. A malt preparation from corn had the best aroma in comparison with other malts and was selected for further experiments. First, important key aroma compounds as 2-acetyl-1-pyrroline, 4-hydroxy-2,5-dimethylfuran-3(2*H*)-one, 3-(methylsulfanyl)propanal, and 4-vinyl-2-methoxyphenol were identified in corn malt by means of aroma extraction dilution analysis. Beside volatile compounds potential precursors of odorants such as reducing sugars and free amino acids were quantified. The corn malt preparation was added to a gluten-free recipe, bread was baked and the selected compounds were quantified by stable isotope dilution analysis. A gluten-free bread without addition of malt was used as a control. Beside the compounds mentioned above the gluten-free bread contained acetic acid and 2-/3-methyl butyric acid as important aroma compounds. The quantitative data showed that by addition of corn malt the amounts of the selected aroma compounds increased in the final bread in comparison to the control bread. This increase was higher than it was expected from the concentrations present in the malt. Thus, it could be concluded that these aroma compounds were generated during baking from odorless precursors in the malt. The contribution of the aroma compounds incorporated from the malt was only a small percentage of the newly generated amounts. Finally, sensory tests (aroma profile, triangle test) were carried out, which confirmed that the addition of suitable malt preparations to a gluten-free recipe positively affects the aroma of the bread. The results will be discussed with special emphasis on aroma compounds and the transfer of precursors from malt into bread during breadmaking.

Keywords: Gluten-free, malt preparation, bread, aroma, celiac disease

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L-34
GLYCATION COMPOUNDS IN FOODS: FORMATION, METABOLIC TRANSIT, FUNCTIONAL CONSEQUENCES

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The Maillard reaction, also referred to as glycation or nonenzymatic browning, is of particular importance for the quality of heated or stored foods. Besides desirable flavour and colour, reaction products are formed which may have a negative impact on the nutritional quality. Our studies during the last years primarily aimed to investigate glycation reactions of proteins. Based on profound analytical techniques, we and others identified and quantified several Maillard reaction products (MRPs) of lysine and arginine in various food systems. Depending on the diet, a daily intake ranging between 500 and 1000 mg for Amadori compounds and 20 to 50 mg for amino acid derivatives originating from advanced stages (“AGEs”), such as pyrraline, N-epsilon-carboxymethylllysine (CML) or pentosidine, can be estimated. Main dietary sources for peptide-bound glycation compounds are bakery products, pasta, coffee and, to a lesser extent, milk products, whereas roasted meat does not contain appreciable amounts of MRPs. Thus, vegetarians generally take up significantly higher amounts of glycation compounds compared to carnivores. In balance studies with human subjects, we found that certain AGEs (e.g. pyrraline) are nearly completely resorbed during digestion and excreted in the urine, whereas Amadori products are not resorbed and excreted. On a molecular level, differences in MRP bioavailability revealed in balance experiments could be attributed to the handling of MRP peptides by the intestinal peptide transporter. Maybe due to a predominating “risk-oriented” perspective, very little knowledge is available about the influence of posttranslational changes occurring in the course of the Maillard reaction and resulting consequences for the technofunctionality of food proteins, i.e. gelation, foaming, and emulsifying properties. Studies in our laboratory have shown that the denaturation reactions of proteins such as beta-lactoglobulin from milk of ovalbumin from egg-white directly depend on the extent of lysine modification. Denaturation temperature increases due to covalent attachment of carbohydrates, which significantly affects the heat stability of globular proteins. Furthermore, the emulsifying properties of whey proteins can be improved by glycation. Knowledge about the relationship between structure and function of glycated food proteins is of particular importance for the use of corresponding proteins as ingredients in complex food systems. The extent of glycation, therefore, should be taken into account not only as parameter for the nutritional properties, but also as a tool to select raw materials for special purposes and/or to functionalize proteins as functional ingredients for “tailor-made” products.

Keywords: Glycation, Maillard reaction, AGEs

L-35
GASTROINTESTINAL DIGESTION AND EPITHELIAL TRANSPORT OF PYRRALINE

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Pyrraline is a Maillard reaction product (MRP) resulting from the reaction of 3-deoxyglucosone and lysine residues in proteins. Substantial amounts of MRPs, e.g., 20–40 mg of pyrraline, are ingested with the daily diet [1]. It was shown in balance studies with humans that after ingestion of protein-bound MRPs in normal food, 60–100% of pyrraline can be recovered in the urine, whereas Amadori products like fructoselysine are not renally excreted [2]. On a molecular level, the differences in the apparent bioavailability can be determined by differences (i) in the digestibility of modified proteins and (ii) in the ability of MRPs to pass the intestinal barrier. In the present work, food proteins (casein, β-casein) were selectively enriched with pyrraline and subjected to simulated gastrointestinal digestion in order to prove the formation of pyrraline-containing peptides capable for epithelial transport. After simulated digestion, low-molecular weight peptides were fractionated by semi-preparative GPC (MWCO=250–500 Da). The release of MRPs follows the chemical properties of their side-chains as is known for the proteinogenic amino acids. Approximately 25% of pyrraline is released into small peptides (di- and tripeptides) as expected from the hydrophobic nature of its side-chain. The occurrence of defined pyrraline dipeptides after digestion of modified β-casein was verified by HPLC–ESI–MS. In order to measure if MRPs can pass the epithelium, we used Caco-2 cells grown on permeable polycarbonate membranes in “Transwell” chambers which allow separate sampling from an apical and a basolateral compartment. The experiments were performed with free and dipeptide-bound MRPs with alanine as the second amino acid [3]. Neither free MRPs nor dipeptides containing fructoselysine were transported through the cells. When other MRP dipeptides were applied to the apical compartment, however, the free modified amino acids could be detected in the basolateral compartment in concentrations pointing to active apical transport. We conclude that alanyl peptides are transported into the cells and hydrolysed. Free MRPs can then leave the cell, most probably by basolateral diffusion. Beyond Ala-Pyrr and Pyrr-Ala, significant transport rates were measured also for Ser-Pyrr, Glu-Pyrr and Pyrr-Glu. Interestingly, Pyrr-Glu was also recovered in the basolateral compartment in intact form, suggesting the passage of intact MRP-dipeptides into the circulation.

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Keywords: Pyrraline, Maillard reaction, gastrointestinal digestion, epithelial transport

L-36
AN ACTIVE ROLE OF SELECTED DIETARY POLYPHENOLS IN CARAMELIZATION AND PROTEIN GLYCATION MODEL SYSTEMS

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Caramelization and Maillard reaction are two of the major groups of nonenzymatic reactions leading to food browning during thermal processing and storage. The complicated reactions not only contribute to the formation of food color and flavor but are responsible for generating hundreds of products either beneficial or harmful to human health. Dietary polyphenols are nowadays gaining a lot of attention due to their antioxidative activity which may contribute to health benefits associated with a lower risk of cardiovascular disease and cancer. Besides, more and more research indicates that polyphenols can actively participate in the reactions between sugar and protein by trapping the reaction intermediates. This study focuses on six kinds of polyphenols (phloretin, naringenin, quercetin, epicatechin, chlorogenic acid, rosmarinic acid) by investigating their impacts on either fructose caramelization or casein glycation induced by glucose in thermal models. In presence or absence of polyphenols (10 mM/1 mM), caramel was prepared by heating fructose (0.05M/40%) in phosphate buffer (0.1M, pH=7/10) while Maillard reaction product was prepared by heating glucose (400 mM) with casein(3.2%) in phosphate buffer. It was found that polyphenol addition could bring about shift in all three chromaticness coordinates L, a, b and correspondingly Chroma value and E index of caramelization and Maillard reaction products. Browning was elucidated to be a cooperative phenomenon resulting from caramelization, Maillard reaction and chemical oxidation of polyphenols as affected by pH. The overall antioxidant capacity of final product was not simply sum of that of polyphenol and caramel or Maillard reaction products but a result of chemical interactions among sugar, protein, polyphenol, and their thermal transformation products. LC–MS analysis yielded evidence on extensive consumption of phloretin and naringenin during caramelization via forming adducts with sugar fragments. Further analysis found out that dietary polyphenols seemed not to affect caramelization extent in the sense of sugar content but significantly influence the amount of two caramelization intermediates hydroxymethylfurfural and furfural depending on both polyphenol category and pH. As for glycation model, polyphenols strongly inhibited the formation of fluorescent advanced glycation products by up to 80%. The inhibition was attributable to polyphenols' effects on the quantity of available free amino groups, extent of sugar incorporation into protein backbone as well as the amount of dicarbonyls such as methylglyoxal and glyoxal. Our research added to the efforts of controlling caramelization and Maillard reaction by dietary polyphenols under thermal conditions comparable to realistic food processing and provided insights into proposing dietary polyphenols as functional food ingredients to lower the amount of heat-induced contaminants in food.

Keywords: Polyphenol, caramelization, glycation

L-37
MELANOIDINS EXHIBIT PRO-OXIDATIVE EFFECTS ON ISOLATED AND CELLULAR DNA AFTER COMPLEXATION OF METAL IONS

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Melanoidins as final reaction products of the Maillard reaction between reducing carbohydrates and amino compounds are components in many heat treated food systems. Their antioxidant activity is well described and may be related to their metal chelating properties. Considering the great number of redox-regulated processes it can be expected that such melanoidin properties are also of great interest in the field of nutrition. Nevertheless the knowledge about mechanisms and possible impact of heat treated food on biological systems is still poor. The aim of the study was to evaluate the impact of metal complexation by melanoidins on their antioxidant properties (TEAC assay, ESR, phenanthroline assay). Furthermore the effect on isolated DNA was investigated by the PM2 assay and in cellular systems by alkaline unwinding using the human colon carcinoma cell line HCT-116. Melanoidins were prepared from D-glucose and different L-amino acids. The high molecular weight fraction was isolated by dialysis (MWCO 12–14 kDa) and charged with metal ions (Cu^{2+}, Fe^{2+}). The amount of metals incorporated into the melanoidin was determined by AAS. ROS (H_2O_2) formation by melanoidins was detected by FOX assay and HPLC/ECD. Independent of the amino acid composition pure melanoidins did not influence cell viability quantified by determination of cell number and colony forming ability. Only negligible DNA damage could be detected on isolated phage DNA after incubation, possibly caused by small amounts of hydrogen peroxide formed by melanoidins. By using melanoidin-cupric ion-complexes a considerable DNA strand breaking activity was determined also in HCT-116 cells, which was again amplified in an oxidative milieu. Since Cu^{2+} ion normally does not provoke the formation of reactive oxygen species (ROS) via Fenton-type reaction, the obtained results have to be attributed to copper complexing melanoidins. In consequence of their reducing ability Cu^{2+} is reduced to the redox-active Cu^{+} ion, which subsequently initiates further redox cycling, leading to the generation of ROS probably responsible for DNA strand breaks. The investigation shows that antioxidant properties of melanoidins in food as well as the possible negative impact of ROS formation in biological systems may be the consequence of the same mechanism.

Keywords: Melanoidins, metal chelating, DNA strand breaks, Fenton reaction, pro-oxidative action

POSTER SESSIONS

Biologically-active constituents of foods and food raw materials

A-1
INVESTIGATION OF TRANS-RESVERATROL AND STILBENE DERIVATIVE TDPA IN HUNGARIAN WINES USING HPLC AND LC-MS

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Plant polyphenols are naturally occurring secondary plant metabolites, synthesized in response to environmental stress factors. As being antioxidants and free-radical scavengers they serve as essential components of the human diet. Among polyphenols well studied representatives is the trans-resveratrol. Trans-resveratrol has been shown to modulate the metabolism of lipids, inhibit the oxidation of low density lipoproteins, reduce platelet aggregation is known to have anti-inflammatory, and has antitumor, cardio- and vasoprotective effects which plays a crucial role in the prevention of chronic cardiovascular and tumorous diseases. In the present study, we investigate trans-resveratrol and the oxidative derivative of trans-resveratrol with a triple bond at the centre of the molecule: 3,4',5-trihidroxy-diphenylacetylene (TDPA) contain in wines. TDPA was first discovered by our research group. Twelve Hungarian wines were analyzed using HPLC–DAD and MS detection. The wines were from Villány wine region representing different wineries from 2008 to 2010 vintage years. Our results show that trans-resveratrol and 3,4',5-trihidroxy-diphenylacetylene content is mainly dependent on variety and vintage year.

Keywords: Resveratrol, LC–MS, HPLC, wine, antioxidant

A-2

EFFECT OF HIGH HYDROSTATIC PRESSURE ON THE ACTIVITY OF CATHEPSINS B AND D OF MACKEREL (*SCOMBEROMORUS MACULATUS*) AND HORSE MACKEREL (*TRACHURUS TRACHURUS*)

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Several modifications occur in the muscle of postmortem fish. Such changes are not only related to microbial spoilage, but are also due to the activity of endogenous enzymes. The main changes of fish muscle proteins are due to proteolysis by endogenous proteases activity, like cathepsins. Among other effects, the catalytic action of these enzymes is responsible for the softening of the muscle tissue. High hydrostatic pressure (HHP) is a non-thermal technology of growing interest for the processing and preservation of foods. HHP has the ability to inactivate microorganisms and affect enzymes activity, increasing food shelf life. In addition HHP processing results in a better retention of nutritional and organoleptic characteristics over traditional thermal processing, allowing to maintain to a large extent the raw (fresh-like) characteristics of foods. This work aimed to study the effect of HHP treatments on the activity of cathepsins B and D in the muscle of two species: mackerel (*Scomberomorus maculatus*) and horse mackerel (*Trachurus trachurus*). Fresh fishes were treated with 150, 300 and 450 MPa pressures during 2.5 and 5 min. Control samples were performed subjecting samples only to pressure come-up and release time (0 min of pressurization holding time) and no HPP treatment (untreated samples). Results showed that HHP affects the enzymatic activity of the cathepsins. For cathepsin B in mackerel, the main effect was observed for higher pressures (450 MPa), being the enzymatic activity decreased by about 25%. However, for the same enzyme in horse mackerel, low pressures (150 MPa) caused a reduction of enzyme activity of 60%. In the case of the cathepsin D, the behavior is similar in both species, with pressure at 300 MPa causing an increase of activity up to 2-fold, while at higher pressures, 450 MPa, caused no changes in the activity (compared to the untreated samples). These results for cathepsin D point for the occurrence of a possible facilitated cathepsin D extractability at 300 MPa, while at 400 MPa pressure caused denaturation might become the relevant effect. The pressure holding times studied showed in general no relevant effects. In conclusion, HHP processing of mackerel has potential to control endogenous enzymes activity in fish muscle, depending of the levels of pressures used, being this interesting to improve fish storage, in what concerns the effects caused by cathepsins.

Keywords: High pressure, mackerel, horse mackerel, cathepsins B and D

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A-3
ANTIOXIDANT AND ANTIGENOTOXIC EFFECTS OF SPENT COFFEE EXTRACTS IN HUMAN CELLS

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Spent coffee that is produced in tons by restaurants and cafeterias, and by consumers at domestic levels, could be a good opportunity to have an important source of natural antioxidants. Spent coffee grounds obtained from the preparation of coffee brews (Arabica from Guatemala and Robusta from Vietnam) with the most common coffeemakers (filter, espresso, plunger and mocha) have antioxidant capacity (Folin-Ciocalteau, ABTS and DPPH) because the presence of relevant amounts of caffeoylquinic acids, mainly dicaffeoylquinic acids, caffeine and melanoidins, with the exception of those obtained from mocha coffeemaker. For this reason and because these antioxidant assays do not reflect the cellular conditions, the aim of the present study was to evaluate the ability of the most antioxidant spent coffee extracts (Arabica filter and Robusta espresso) to protect from oxidation and DNA damage in the human cell line HeLa at short (2 h) and long (24 h) exposure times. Spent coffee extracts were obtained in a filter coffeemaker (24 g spent coffee/400 mL water). Firstly, in order to select non cytotoxic concentrations (≥80% survival), cell viability was measured using MTT test. The selected non cytotoxic concentrations were checked for not being able to produce neither an intracellular increase of reactive oxygen species (ROS) (dichlorofluorescein assay), nor DNA strand breaks (SBs) or oxidative DNA damage (comet assay). Then, to evaluate their antioxidant and antigenotoxic capacities, spent coffee extracts were added to the cells 2 and 24 hours prior to the induction of the oxidative insult. HeLa cells were treated with 500 μM H_2O_2 for 10 min to increase ROS level and induce SBs, and with 1 μM Ro (photosensitizer) plus visible light (10 min) to induce purine oxidation (8-oxoguanine). Both spent coffee extracts were able to significantly reduce the increase of ROS level induced by H_2O_2 at short and long exposure time. Furthermore, DNA strand breaks were also reduced with the pre-treatment with the extracts, but only Robusta espresso spent coffee extract at the highest concentration tested after 24 h was able to reduce the FPG-sensitive sites in the comet assay. Traditionally, the antioxidant capacity of coffee has been attributed to caffeoylquinic acids and melanoidins, however in this study, spent coffee extracts with lower amounts of these compounds were more effective in protection against induced oxidative stress and DNA damage. These findings suggest that other phenolic and non phenolic bioactive compounds, such as caffeine and other Maillard reaction products (MRPs), might contribute to the high antioxidant activity of Vietnam espresso spent coffee extract. In conclusion, this study demonstrates that spent coffee extracts protect from oxidation and DNA damage in human cells. Therefore, spent coffee can be considered as an added-value by-product because is a good source of bioactive compounds.

Keywords: Spent coffee, antioxidant, caffeoylquinic acids, antigenotoxicity, comet assay

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A-4

DETERMINATION OF ANTIOXIDANT CAPACITY AND REGENERATION BEHAVIOR OF BIOACTIVE COMPOUNDS BOUND TO INSOLUBLE DIETARY FIBERS

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Research about health benefits of antioxidant compounds and dietary fiber (DF) has received significant attention in the last decades. These two are commonly found together in food matrices. But interaction between DF and other antioxidants have not been studied well in literature. It is evident that DF affects bioavailibility of antioxidants. The principal physiological effect of DF in the small intestine is to reduce the rate (and in some cases the extent) of nutrients' or antioxidants' release. So all the non-absorbed phenolics and DF reach to colon together in intact form and make an antioxidant environment there. This study investigated whether antioxidants bound to dietary fiber could be regenerated or not, *in vitro*. Number of food matrices rich in insoluble DF and antioxidants such as coffee and bread crust melanoidins, and cocoa powder, dried fruits, cereal brans, nut skins, etc. were analyzed for DF bound antioxidant capacity by means of the QUENCHER procedure. DF rich sample was washed out sequentially by using water, methanol and ethanol, or their mixtures to remove the soluble forms of antioxidants and fibers. Fat rich samples were first defatted by hexane. The insoluble residue was used to measure the DF bound antioxidant capacity using ABTS radical cation as the probe. The antioxidant capacity was expressed in mmol TEAC/kg DF. In order to determine the regeneration efficiency of the initially measured antioxidant capacity, the residue after measurement was washed three times with water. Then it was treated with ascorbic acid under certain conditions. After remaining ascorbic acid was removed by washing with water, the residue was measured again for its antioxidant capacity. The regeneration process was repeated three times as described above. The results revealed that DF bound antioxidant capacity *in vitro* can be significantly regenerated by another antioxidant compound as exemplified here for ascorbic acid. The DFs from different food sources differed significantly in their regeneration capacity. For example, the antioxidant capacity of DF from peanut skin could be regenerated by 50.5% while DF from pistachio skin by 21.5% after the first regeneration step. It was found that significant levels of antioxidant capacity could be observed *in vitro* after three steps of regeneration of the samples by ascorbic acid. These results are considered to be meaningful for the behavior of bound antioxidants *in vivo* during the digestion process in gastrointestinal tract.

Keywords: Antioxidant capacity, bound antioxidants, insoluble dietary fibers, regeneration

A-5
IMPACT OF EXTRACTION METHODS, TYPE AND STORAGE ON THE PHENOLIC COMPOSITION AND ANTIOXIDANT POTENCY OF CHOCOLATE PRODUCTS

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The chocolate products, besides being favourite sweets among consumers, also present high amounts of polyphenols with health-promoting properties. Polyphenols in chocolate products (*Theobroma cacao* L.) can be classified into three main groups: flavan-3-ols (37%), anthocyanins (4%) and proanthocyanidins (58%) [1]. Polyphenolic content of chocolates is affected by several factors such as genetic factors, pre-harvest factors, processing and storage. The analytical methodology for determination of polyphenols is also crucial for the estimated phenolic content of chocolates [2]. The objective of this study was to compare three different methods commonly used for extracting polyphenols from chocolates (ultrasonic assisted extraction, maceration and Soxtec extraction) using different solvents (70% acetone, 70% methanol, and 70% ethanol). The evaluation of their efficacy was performed in the terms of phenolic content and antioxidant potency. The determination of phenolic content was performed with the employment of spectroscopic assays and the antioxidant potency was evaluated using *in vitro* assays (DPPH, FRAP). Results showed that the superiority of maceration and ultrasonic assisted extraction with 70% acetone (1855.8 and 1946.3 mg gallic acid/100 g chocolate respectively) against other tested extraction methods and (i.e. 1559.4 mg gallic acid/100 g chocolate using Soxtec extraction with 70% ethanol). The antioxidant potency and the classification of polyphenols to the hydrocinnamic acids, flavonols and anthocyanins demonstrated also similar trend. Finally, the ultrasonic assisted extraction with 70% acetone was preferred. The optimized methodology was further applied to analyze phenolic composition of three types of chocolate "Flava" Chocolate, "Flava" Chocolate enriched with Royal Jelly, and "Normal" Chocolate. In addition, the changes in phenolic composition during storage for 40 days at 18°C were also monitored. Results showed that "Flava" Chocolates had significantly higher phenolic content than “Normal” (728.1±95.0 and 728.5±167.2 against 356.7±56.4 mg gallic acid/100 g chocolate) and their antioxidant capacity was most potent (2146.3±245.8 and 2073.3±406.4 against 1155.6±193.2 mg Trolox/100g chocolate). Furthermore, no significant changes were found in phenolic content of chocolates during storage period. In summary, the choice of extraction and type of chocolate affects the phenolic content and the antioxidant potency of chocolate, while the amounts of phenolic compounds remained constant during storage for 40 days.

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Keywords: Extraction, polyphenols, storage, *Theobroma cacao* L.

A-6

EFFECT OF DRYING METHOD ON THE NUTRACEUTICAL CONTENT AND ANTIOXIDANT CAPACITY OF CYPRUS SPEARMINT (*MENTHA VIRIDIS*)

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Herbs contain an inexhaustible source of bioactive compounds that has attracted the scientific interest of the biotechnology, cosmetics, pharmaceutical and food industry. Extensive studies have linked the consumption of herbs and other natural products rich in biophenols with protection against plethora of chronic diseases. Drying is considered as a critical factor for the storage and the merchantability of herbs, but it is also accompanied with the loss of bioactive compounds. Researchers have studied the effect of the drying method on various herbs[1,2] and have focused mainly the effect of drying method on the essential oil content, in an attempt to optimize drying methods. In addition, previous studies demonstrated that each herb requires the optimization of its drying procedure. The aim of this study was to compare the effect of five drying methods namely convection oven drying (CD), freeze-drying (FD), microwave drying (MD) and air drying with sun exposure (SUD) and without (SHD) on the nutraceutical content (total phenolics and hydroxycinnamic acid derivatives) as well as the antioxidant capacity which was determined by in vitro assays (FRAP, DPPH) of dried Mentha viridis focusing on the nutraceutical content of final product rather than the essential oil, Results showed the superiority of freeze-drying of mint for all parameters studied, whereas convection oven drying and microwave drying gave the lowest nutraceutical content and antioxidant potential. In particular, freeze drying of mint produced material that had the highest total phenolics (FD: 3995.3±559.8 >> SUD:2451.1±189.0 = SHD: 2142.1±544.2 >> CO:1765.4±186.8 = MD: 1740.9±64.6 gallic acid/100g mint, P=0.05) and the highest antioxidant capacity (FD:11267.5±167.8 >> SUD:5161.5±256.6 >> SHD:4788.1±670.2 ≥ MD: 4601.6±158.8 ≥ CO: 4394.7±196.8 mg Trolox/100 g mint). This superiority of freeze drying could be attributed to the fact that freeze-drying procedure enhances the extractability of biophenols since ice crystals formed within the sample matrix can rapture cell structure, which allows exit of cellular components and access of solvent[3]. To conclude, it is indicated that added energy methods, like convection and microwave oven drying, result to a great loss of compounds, whereas milder methods and especially freeze-drying give end-products with higher amounts of phenols and antioxidant potency.

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Keywords: Drying, phenolics, freeze-dryer, herb

A-7
CHANGES OF ANTIOXIDANT ACTIVITY IN HONEY AFTER HEAT TREATMENT

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Heat treated or otherwise processed food is heavily represented in today's diet. Those technological processes and treatments more or less cause the changes in its nutritional value, chemical composition, and consequently, the antioxidant activity of food. Honey is no exception since it is often used as a sweetener in hot drinks and pastries, biscuits and other confectionery products, which are more or less thermally processed. In this study it was determined how antioxidant activity and total phenolic content of honey change after being subjected to high temperature. Antioxidant activity was determined using two methods – FRAP (ferric reducing antioxidant power) and DPPH (1,1-diphenyl-2-picrylhydrazyl) methods. The research was done on 31 samples of acacia honey and 8 samples of chestnut honey. All measurements were made at two temperatures - at 23°C (room temperature) and after 5 minutes of heating at 95°C. The antioxidant activity of acacia honey measured by FRAP method decreased in 16 samples after thermal treatment. Average reduction in all samples was 33.1% (55.05 to 37.78 μm Fe (II) 10% honey solution (HS)). FRAP values in the other 15 samples increased in average for 39.3%, (37.10 to 58.77 μm Fe (II) 10% HS). FRAP values measured in chestnut honey decreased in all samples on average of 12.1% (321.48 to 279.35 μm Fe (II) 10% HS). DPPH values increased in 23 samples of acacia honey after heating with an average reduction of 41.9% (96.81 to 172.38 mg/mL), where this increase means a decrease in antioxidant activity. Antioxidant activity increased in 8 acacia honey samples in average for 30.5% (151.35 to 104.72 mg/mL). In 4 chestnut honey samples antioxidant activity decreased by 19.5%, while in 4 samples it an average increase of 8.9%. The total phenolic content decreased after heating in 15 acacia honey samples on average by 36.4% and increased in 16 samples by an average of 39.3%. In chestnut honey the total phenolic content decreased in 3 samples on average by 20.2% and increased in 5 samples by an average of 9.4%. Results show uneven changes of antioxidant activity and total phenolic content between individual samples, i.e. in some samples antioxidant activity decreased after heating, while in others it increased. The same applies to the total phenolic content. A statistical analysis of the results (t-test) showed no statistically significant differences between the results measured at two different temperatures (P-value>0.05) in all three methods used, and in both types of honey. The only statistically significant difference (P-value <0.05) was observed in DPPH method measured in acacia honey. According to the obtained results, it can be concluded that thermal treatment of honey at 95°C does not cause the significant change of its antioxidant activity and total phenolic content, which is true for both investigated honey types.

Keywords: Honey, heat treatment, antioxidant activity, phenolics

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A-8
PHENOLIC COMPOSITION AND SENSORY PROPERTIES OF CIDERS PRODUCED FROM LATVIAN APPLES

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Polyphenol compounds are very important components of cider, who response for the colour and the balance of bitterness and astringency. The polyphenolic profile of apples and apple drinks is influenced by several factors: apple variety, climate, maturity, technological processes applied. This paper reports the influence of apple variety on the phenolic compounds and sensory properties of cider. Fermentation of twelve apple variety juices was performed in laboratories of Latvian University of Agriculture, Faculty of Food Technology. Apple juices were fermented with *Saccharomyces cerevisiae* yeasts '71B-1122' (Lalvin, Canada). Total phenol content (TPC) was determined according to the Folin-Ciocalteu spectrophotometric method and results were expressed as gallic acid equivalents. Individual phenolic compounds were determined using high performance liquid chromatography (HPLC). Trained panellists (finished basic course of sensory evaluation) evaluated the intensity of sensory properties, namely, clarity, apple, fruit and yeast aroma, apple, yeast, sour, astringent and bitter taste using line scale. The content of total phenols varied from 29 to 269 mg L^{-1} for ciders depending on used apple variety. The highest TPC was detected in ciders made from crab apple varieties 'Hyslop', Balsgard' and 'Ouakers Beaty'. In this work, special attention is drawn to the use of culinary apples for cider production. The apple varieties 'Lietuvas Pepins', 'Antonovka', 'Auksis' are popular commercially grown varieties in Latvia and are used as a culinary apples for juice production and comparing culinary apples, the highest TPC in variety 'Antonovka' apple cider was detected. Eleven phenols were determined in tested samples, and chlorogenic acid is the dominant phenolic in cider. Correlation analysis was performed to determine interactions between sensory properties and polyphenol content. Very weak correlation between TPC and bitterness were observed although other investigations showed that in cider bitter taste is associated with the presence of acrolein combined to polyphenols. Strong correlation between astringency and TPC was observed and in cider astringency are due to the polyphenols especially procyanidins which are polymers of catechins.

Keywords: Apple variety, cider, phenolic compounds, sensory properties

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A-9
BIOAVAILABILITY OF TRYPTOPHAN-CONTAINING PEPTIDES IN HUMAN BLOOD

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Bioactive peptides have relevance for the development of functional food products. In this context, peptides with a possible blood pressure lowering effect due to inhibition of angiotensin converting enzyme (ACE) are of particular importance. ACE is one of the key enzymes in blood pressure regulation by generating the vasoconstrictor angiotensin-II and degradation of bradykinin, a vasodilatory peptide. In our group, we identified several tryptophan-containing dipeptides as a new class of natural ACE inhibitors [1]. These bioactive peptides are inactive within the sequence of their precursor proteins like α-lactalbumin and can be released by specific enzymatic hydrolysis in vitro [2]. For instance, Ile-Trp (IW, IC_{50}=0.7 µM) and Trp-Leu (WL, IC_{50}=10 µM) originating from the sequence of the α-lactalbumin show a good ACE inhibition *in vitro*. To exert physiological effects in vivo, it is important that these bioactive peptides reach the cardiovascular system in an active form. For investigations on the bioavailability and effectiveness of the peptides in vivo, pharmacokinetic studies were designed. A single defined dose of either the peptides IW/WL or nothing (placebo) were orally given to overall 6 normotensive subjects (3 female, 3 male, age 28–56 years, BMI 20–25 kg/m^2). Before (at least 2.5 h) and during the study (4 h) participants were not permitted to eat, but to drink any blood pressure ineffective beverage. Dipeptides in human plasma were quantified with a LC-ESI-MS/MS method. An oral intake of 100 mg IW or WL resulted in a significant increase of concentrations for both dipeptides in plasma compared to baseline levels (cPlacebo IW=0.8±0.2 nM and cPlacebo WL=4.8±1.0 nM for WL). Maximal concentrations were reached at t_{max}=30 min with c_{max} IW=13.5±9.9 nM and c_{max} WL=30.5±0.7 nM respectively. The half-life in vivo was calculated to 13.5±1.8 min for IW and 24.6±1.5 min for WL with a higher area under the curve (AUC) value for WL (AUCWL=34.8±1.5 nM•h) compared to IW (AUCIW=7.0±1.9 nM•h). These results demonstrate that IW and WL are to some extent absorbed, reach the cardiovascular system and can cause an ACE-inhibition in vivo due to relative good stabilities. Protein hydrolysates with tryptophan containing dipeptides could be interesting ingredients for functional food as an effective and natural prevention for hypertension, but also as a good source for the amino acid tryptophan.

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Keywords: Tryptophan-containing bioactive peptides, ACE-inhibition, pharmacokinetic studies

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A-10

CRUDE PHENOLIC EXTRACT EFFECTS *IN VITRO* AND THEIR IMPACT IN A RAW SAUSAGES STORAGE TEST. PRELIMINARY FINDINGS

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The vegetation waters (VW) could be considered as additional resources for the virgin olive oil (VOO) industry. In fact several phenolic compounds occurring in VW, having the same chemical characteristics of VOO phenols, possess many biological properties that include antioxidant and antimicrobial activity and, for this reason, may be used in food industry as natural preservative. The aim of the study was to assess:a) the in vitro effect of the Crude Phenolic Extract (CPE) obtained by ultrafiltration methods from VW on the growth of several food-borne strains and starter cultures; b) the overall impact of their inclusion in the raw sausages during a storage test. Antibacterial activity (0.375÷12 mg/mL CPE final concentrations) was based on the agar diffusion assay in comparison with the MBC (Minimum Bactericidal Concentration; microtitre assay). The microbial genera tested were: *Staphylococcus* (n.5), *Listeria* (n.4), *Escherichia* (n.2), *Salmonella* (n.1), *Pseudomonas* (n.3), *Lactobacillus* (n.2) and *Pediococcus* (n.1). Fresh pork raw sausages (shoulder and belly cuts, 1.5% NaCl, without) were manufactured, according to local recipe to obtain 3 experimental batch: C control, LL 0.75 g/kg and HL 1.5 g/kg of CPE. The two levels of polyphenol inclusion were based on the results provided by the antibacterial activity linked with the sensory analysis (e.g., lowest level of perceived flavours with antibacterial activity).The sausages were submitted to microbiological analysis in order to evaluate the total aerobic mesophilic and psycrotrophic flora, lactic acid bacteria (LAB), the Micrococcaceae, *Staphylococcus* spp., *Pseudomonas* spp., yeast and mould. The change of micro flora composition during a storage period of 15 days (4.0±0.5°C) was evaluated, furthermore pH, water activity (Aw) and NaCl content were measured. The main in vitro results indicated that S. aureus and L. monocytogens showed the lowest level of resistance to CPE (1.5–3 mg/mL) for both MBC and agar diffusion assay. In contrast the Gram negative strains were unaffected by the tested doses on agar assay whereas the MBCs ranged between 6 to 12 mg/mL.Starter cultures were dramatically reduced on growth (e.g. S. xylosus; MBC 0.75-1.5 mg/mL).The effect of polyphenol in fresh sausages was also evaluated demonstrating a relevant impact in the resident microflora.CPE inclusion influenced the development of microbial numbers, especially the Micrococcaceaeand Staphylococcus spp. counts, during 15 days at 4°C. LAB counts and psycrotrophic flora were affected only in the HL sausages. During the shelf life NaCl varied from 1.5 to 2.2%, meanwhile Aw decreased from 0.970 to 0.945 for C and LL and to 0.938 in HL; pH ranged from 5.9 to 5.7 for C meanwhile for LL and HL the acidification was not appreciable. CPE delayed microbial growth without influenced on sensorial characteristic. Preventing lipid oxidation in raw and cooked sausages should be evaluated.

Keywords: Phenolic compounds, fresh raw sausage, ingredients, microbiology, vegetation waters

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A-11
BIOACTIVE COMPOUNDS IN LATVIAN BARLEY BEER

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Beer is a complex mixture; over 400 different compounds have been characterized in beer which, in addition, contains macromolecules such as proteins, nucleic acids, carbohydrates and lipids. Some of the constituents of beer are derived from the raw material and survive the brewing process unchanged. Others are the results of chemical and biochemical transformation of the raw material during malting, mashing boiling, fermentation and conditioning. Together all these constituents make up the character of beer. Significant health promoting benefits have been attributed to its bioactive secondary metabolites such as phenolics. Polyphenols and phenolic acids present in beer are natural antioxidants, capable of delaying, retarding preventing oxidation processes. The aim of current research is to characterize the bioactive compounds of Latvian barley beer such as phenolic acids and flavanols. Different lager type beers produced in Latvia were analysed in experiment. The total phenolic content (TFC) was determined by spectrophotometer according to the Folin-Ciocalteu colorometric method with some modifications. Total phenolics were expressed as gallic acid equivalents (mg GAE g^{-1} dry weight). Individual phenolic compounds were determined using high performance liquid chromatography (HPLC). The antioxidant potential of beer is analyzed by the 2,2-diphenyl-1-picrylhydrazyl (DPPH) assays. The final results were expressed as micromoles of Trolox equivalents (TE) per gram of dry weight (μmol TE g^{-1} DW). Mainly dark beers contained higher TFC (320.8–863.6 mg GE L^{-1}) than light beers (300.9–475.2 mg GE L^{-1}). High performance liquid chromatography (HPLC) is being used increasingly to resolve the mixture of phenol components in beer. Eleven phenols were determined in tested samples, and gallic, ferulic and caffeic acids and catechin are the dominant phenolics in beer. All beer samples exhibited strong DPPH radical scavenging activity. For light beer it ranged from 441.3 to 1064.2 μmol TE L^{-1}, but for dark beer higher results were obtained – 726.2–1748.7 μmol TE L^{-1}. Antiradical activity of dark beer are also due to products of Maillard reaction, that develops during kilning and wort boiling process, and gives similar colour reaction than phenols.

Keywords: Beer, phenolic, antiradical activity

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A-12

CHANGES IN PHENOLIC CONTENT AND ANTIOXIDANT ACTIVITY OF FREEZED SPINACH, PEA AND SWEETCORN IN RELATION TO THE STORAGE PERIOD

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This work evaluates changes in content of total polyphenols (TP) and antioxidant activity (AA) of freezed vegetables (spinach and pea) and sweetcorn in relation to the storage period. Total polyphenol content in analysed samples was determined by spectrophotometric method using Folin-Ciocalteau reagent. The amount of total polyphenols was expressed as gallic acid equivalent in mg GA per kg of fresh material. Antioxidant activity in analysed samples was determined using the stable free radical DPPH (2,2-diphenyl-1-picrylhydrazyl) by spectrophotometric method. Both, the total phenolic content and antioxidant activity, were analysed in freezed vegetable samples monthly and the changes were monitored during storage at -18°C for 10 months. Freezing had different influence on the levels of TP content and AA in individual analyzed samples. The greatest decrease in AA levels during the entire period of freezing (10 months) was recorded in spinach (79.4%), while the lowest decrease was observed in pea (26.8%). Relatively the significant decrease in AA was also found in corn (62.7%). Contrary, the greatest decrease of TP content throughout the period of freezing was found in pea (62.0%), and lowest decrease of TP content was recorded in corn (only 5.0%). The TP content in spinach decreased after 10 months of storage at 43.1%. The continuous decrease in AA during the storage period (10 months) was found at spinach and corn. The pea, AA first 2 months of growth, then its level fell steadily after 10 months. For spinach, sweetcorn and pea, TP content first 2 months of growth rates, then we recorded the continuous decrease of TP content. While the sweetcorn was this very slight decrease, but in pea and spinach quite significant. Among the factors which can affect the levels of antioxidant activity and total phenolic compounds in food samples could be the species, size and texture of vegetables, the prepared form of the samples and mainly the conditions of storage (e.g. time, temperature).

Keywords: Phenolic compounds, antioxidant activity, vegetables, freezing

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A-13
CHOSEN ANTIOXIDANT AND SENSORY PROPERTIES AND THEIR MUTUAL RELATIONS OF SLOVAK RED WINES – CABERNET SAUVIGNON

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Chosen antioxidant and sensory properties as content of total polyphenols, content of total anthocyanins, antioxidant activity and wine colour density in brand red wines – Cabernet Sauvignon, originating from different Slovak wine regions were determined. Polyphenolic antioxidants of the red wines are very effective in preventing cancer and cardiovascular diseases. Total polyphenol (TP) content was determined with phenol Folin-Ciocalteau´s reagent, the content of total anthocyanins (TA) was determined by pH differential method and antioxidant activity (AA) using DPPH stable free radical by spectrophotometric methods. The wine colour density (WCD) was determined also spectrometrically as the sum of absorbance at 420 nm and 520 nm. The determined total polyphenol contents in observed wines were within the interval 1218–3365 mg gallic acid.L^{-1} (average TP content was 2341 mg gallic acid.L^{-1}), total anthocyanin contents 68.6–421.2 mg.L^{-1} (average TA content was 193.9 mg.L^{-1}). Determined values of antioxidant activity were within the interval 69.0–84.2% (average AA was 79.0%) and wine colour density 0.756–2.782 (average WCD was 1.32). The statistical evaluation of the obtained results confirm very weak negative correlation between total polyphenol content, resp. total anthocyanin content and antioxidant activity (r=-0.181, resp r=-0.207). The positive correlation of total polyphenol content and total anthocyanin content was stronger (r=0.413). The statistical evaluation of the obtained results confirmed middle positive correlation between total polyphenol content, resp. total anthocyanin content and wine colour desnsity (r=0.637, resp. r=0.497). The negative correlation of the antioxidant activity and wine colour density was stronger (r=-0.702).

Keywords: Polyphenol, anthocyanin, antioxidant activity, red wine, Cabernet Sauvignon

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A-14

ALKYLRESORCINOLS CONTENT IN PEARLED NAKED AND HULLED BARLEY FRACTIONS

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5-n-alkylresorcinols (ARs), are an important group of phenolic compounds that occur in bacteria, algae, fungi, animals and higher plants, consisting of a phenolic ring with two hydroxyl groups in the meta position, and an odd numbered alkyl chain at position 5 [1]. ARs are involved in multiple biological activities. Among the cereal grass species, the bran fractions of rye, wheat, triticale and barley contain high levels of saturated ARs homologues, including C15:0, C17:0, C19:0, C21:0, C23:0 and C25:0 [2]. The aim of this work was to investigate the content and the composition of ARs in different barley kernel pearled fractions, obtained from progressive pearling process. Three barley varieties, Mona (naked), Ketos (hulled and six-row barley), and Trasimeno (hulled and two-row barley), were collected from an experimental field. Concerning Mona, starting from unprocessed grain, kernels were initially pearled to remove 5% of the original grain weight (first fraction: 0–5%). The pearling process was then continued until 5–10%, 10–15%, 15–20%, 20–25% fractions, plus a residual 75% of the kernel (25–100%), were collected. Concerning Ketos and Trasimeno, the pearling process yielded 0–5%, 5–10%, 10–15%, 15–20%, 20–25%, 25–30%, 30–35%, 35–40% fractions, plus a residual 60% of the kernel (40–100%). ARs were extracted with ethyl acetate from flour samples and the trimethylsilyl ether derivates were prepared by adding the silylating reagent. ARs were then analyzed by gas chromatography (GC) [2]. Individual compounds were quantified using methyl behenate as internal standard. ARs content in barley has been shown to achieve approximately 100 µg/g of dry matter in Mona (naked cv) and 60/65 µg/g in Ketos and Trasimeno (hulled cv). Even though the total AR content varies both within and between cereal species, the relative homologue composition appears rather constant within species. Different amounts and distribution of these compounds within the barley kernel were highlighted, showing a general progressive decrease from the external to the internal endosperm layers. For the cv. Mona (naked variety), the ARs content significantly decreased at each successive pearling step from the outer (0-5%) fraction towards the inner layers, while for Ketos and Trasimeno varieties (hulled varieties) the highest ARs content was observed for the 10–15% fraction, whereas 0–5 and 5–10% fractions, that contain mainly the hull portions, resulted in a lower concentration. The relative composition of ARs in both whole kernel and progressive pearling fractions was quite constant; the AR 25:0 was predominant together with AR 21:0, accounting for about the 50% (mean value of the three cultivars) of the total ARs content. The other ARs identified (C17:0, C19:0, C23:0) were present in minor concentrations.

[1] Kozubek and Tyman (1999) Chemical Review 99: 1–25
[2] Ross et al. (2003) Food Chem 51: 4111–4118

Keywords: Alkylresorcinols, barley, pearling

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A-15
THERAPEUTICAL COMPOUNDS IN GARLIC OIL: PRODUCTION AND EVOLUTION AFTER PREPARATION

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Allyl sulfides, the main constituents of garlic oil, have been studied in the past decades for their effects against cancer. Since cancers are not all healed, ways to prevent it, especially with natural products that could be included in food preparation, are more than welcome. The synthesis and separation of allyl sulfides can provide larger amount of allyl sulfide than the extraction in order to make biological tests. Furthermore, the improvement of the synthesis conditions helps the understanding of the formation of allyl sulfide while garlic is processed. Finally, the study of the products allows a deeper understanding of the conversion between the allyl sulfides. A parallel can then be done with the conservation conditions of garlic-based products. The first part of this work describes the synthesis optimization using the design of experiment. The reaction of allyl bromide and sodium disulfide was carried under different conditions: the reaction duration, the heating (in microwave oven or in oil bath, at different temperature), the presence of catalyst, and the agitation were assessed. In the second part of the study, extractions are performed on garlic. The composition of the extracts is followed during time under the conditions established in the first part. The results give an optimized way to synthesize allyl sulfides, as much as an idea of the reactions happening on them in garlic preparations under various conditions.

Keywords: Garlic, allyl sulfides, design of experiment

A-16
ANTIOXIDANT ACTIVITIES OF LEMON BALM (*MELISSA OFFICINALIS* L., LAMIACEAE)

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Lemon balm (*Melissa officinalis* L.), is a perennial herb of the family Lamiaceae, native to southern Europe and the Mediterranean region. Lemon balm (tops with leaves) is used as a medicinal plant (subjected, controlled and approved by Czech state pharmaceutical authority – SÚKL) for the treatment of psychovegetative conditioned neurosis, sleep disorders, and functional disorders of the digestive system. Its culinary use as an aromatic spice is therefore very limited due to these effects. Leaves contain compounds with significant antioxidant potential. Non-volatile fraction contains some active flavonoids, e.g. luteolin, caffeic acid derivatives etc., volatile essential oil contains some terpene aldehydes and terpene alcohols. Activity of aqueous extracts of both fresh and dried herbs (simulating their use as medicinal plant for the preparation of infusion and activity in organism) was measured by using DPPH radical scavenging method. Total phenolics content was determined by using the Folin-Ciocalteu reagent method. Antioxidant activity of dried herbs in fat-containing food material (pork lard) was tested by the Schaal test based on the monitoring of the course of fat oxidation gravimetrically with free oxygen access in the dark at 60°C and by adapted Schaal test in thin layer of fat. Lemon balm leaves show very significant antioxidant activities in both systems. Activities in relevant concentration range are comparable or higher than α-tocopherol and synthetic antioxidant BHT used as positive standards.

Keywords: Medicinal plant, Lemon balm, Lamiaceae, natural antioxidants, antioxidant activity

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A-17
ANTIOXIDATION ACTIVITY OF LAMIACEAE HERBS GROWN UNDER ORGANIC FARMING CONDITIONS

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Lamiaceae herbs produce varieties of secondary metabolites which can be effectively used as food antioxidants. Expanding production and interest in organic farming also brings the question comparing the antioxidant activity of these plants grown under different fertilization methods. Antioxidant potential of selected widely used culinary herbs, two oregano varieties (*Origanum vulgare* L. and *Origanum heracleoticum* L.) and mint (*Mentha spicata* L.) from organic and conventional farming was therefore tested. Activity of aqueous extracts (simulating common culinary treatment and activity in organism) was measured by using DPPH radical scavenging method, total phenolics content was determined by using the Folin-Ciocalteu reagent method. Antioxidant activity of both fresh and dried herbs in fat-containing food material was tested by the Schaal test based on the monitoring of the course of fat oxidation gravimetrically with free oxygen access in the dark at 60°C. Essential oils components which play an important role in antioxidant activity of herbs were determined by GC–MS analysis. No statistically significant higher antioxidant activity of herbs from organic farming than their conventional form was found. Disruption of plant tissues during drying process caused a greater release of active substances during their extraction and therefore higher antioxidant activity of dried herbs aqueous extract than extracts from fresh herbs. The activity of herb extracts was different in hydrophilic and lipophilic matrices. Mint was the best antioxidant in an aqueous medium, while the Greek oregano (*Origanum heracleoticum* L.) was ideal for containing fat medium. Its considerable antioxidant effect was because of high content of carvacrol and thymol in its essential oil.

Keywords: Antioxidant activity, organic farming, oregano, Greek oregano, mint

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A-18
IN VITRO CHEMOPREVENTIVE ACTIVITIES OF OLIVE OIL PHENOLIC COMPOUNDS

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The chemopreventive properties of olive oil have been recently attributed to the presence of secoiridoid phenolic compounds. The phenolic composition of olive oil is complex and include hydroxytyrosol (3,4-dihydroxyphenylethanol: 3,4-DHPEA), tyrosol (hydroxyphenylethanol: p-HPEA), the dialdehydic form of elenoic acid linked to either hydroxytyrosol (3,4-DHPEA-EDA) or tyrosol (pHPEA-EDA, oleocanthal), oleuropein aglycon (3,4-DHPEA-EA) and lignans. 3,4-DHPEA and other olive oil phenolic compounds have been deeply investigated in the past years showing both anti-proliferative/pro-apoptotic activities on tumor cell lines [1,2] and oxidative DNA damage preventive capacity on human lymphocytes [3]. In this communication we present our recent findings on the following aspects of olive oil chemopreventive activities: i) anti-inflammatory properties of 3,4-DHPEA on human monocytes; ii) anti-proliferative and pro-differentiation activities of pinoresinol (the most abundant lignan present in olive oil) on human promyelocytic HL60 leukemia cells; iii) anti-genotoxic effect of a complex olive oil phenolic extract on styrene oxide-induced DNA damage in human lymphocytes. The main results indicate that: i) 3,4-DHPEA (100 μM) was able to prevent LPS-mediated COX2 induction on human monocytes both at mRNA (67%, $p<0.05$) and protein levels (46%, $p<0.05$). These effects were coupled to a significant reduction of PGE2 accumulation in the culture medium (71%, $p<0.05$). ii) Pinoresinol efficiently reduced the HL60 growth at a concentration as low as 1 μM (IC50%=8 μM at 96h). This effect was associated to a low percentage of apoptosis at 48 h of incubation (4.4±1.4% at 100 μM vs 0.6±0.3% in the control, $p<0.05$) and to an increment of the cells in the G0/G1 phase of the cell cycle (58±1.5% at 100 μM vs 31±3% in the control, $p<0.05$). The HL60 acquired the ability to undergo the “respiratory burst” and expressed the surface antigen CD11b indicating a more differentiated phenotype after treatment with pinoresinol. iii) A complex mixture of olive oil phenolic extract (OOPE) at 1 μg/mL exerted a preventive activity against the DNA damage induced by styrene oxide (25 μM) on human lymphocytes (40% and 63% at 2 and 24 h of incubation, respectively) as measured by the comet assay (SCGE: single cell gel electrophoresis). This effect was in part mediated by an antioxidant activity since the OOPE was able to reduce both the amount of oxidized bases in DNA (Endo III and FPG sensitive sites) and the concentration of intracellular peroxides induced by styrene oxide treatment. All together, our “in vitro” data give further support to the hypothesis that olive oil phenols may play a role in the cancer prevention properties of the Mediterranean diet.

[1] Fabiani R, De Bartolomeo A, Rosignoli P, et al. J. Nutr. 2006, 136: 614–9
[2] Fabiani R, Rosignoli P, De Bartolomeo A, et al. J. Nutr. 2008, 138: 42–8
[3] Fabiani R, Rosignoli P, De Bartolomeo A, et al. J. Nutr. 2008, 138: 1411–6

Keywords: Olive oil phenols, secoiridoids, lignans, chemoprevention

A-19

CULTIVAR INFLUENCE ON TOTAL POLYPHENOL AND RUTIN CONTENTS AND TOTAL ANTIOXIDANT CAPACITY IN BUCKWHEAT, AMARANTH AND QUINOA SEEDS

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Five cultivars from each of three types of pseudocereals i.e. buckwheat, amaranth and quinoa were studied from the view of the total polyphenol and rutin content as well as the total antioxidant capacity of seeds. Spectrophotometric method was used for the determination of the total polyphenol content (using Folin-Ciocalteau reagent) and the total antioxidant capacity (using DPPH). Rutin content in pseudocereal seeds was determined by HPLC. The determined total polyphenol content in seeds of buckwheat cultivars was in range from 15,874 $mg.kg^{-1}$ (Spačinská) to 71359 $mg.kg^{-1}$ (Bamby). Based on the data of total antioxidant capacity of buckwheat seeds it can be created the following order of studied cultivars: Madawska > Bamby > Spačinská > Aiva > Siva. Rutin content in buckwheat samples was in interval 8722 $mg.kg^{-1}$ (Madawska) – 17,125 $mg.kg^{-1}$ (Bamby). In seeds of studied amaranth cultivars the determined total polyphenol content was in range from 1381 $mg.kg^{-1}$ (Rawa) to 2870 $mg.kg^{-1}$ (Annapurna). The following order of studied amaranth cultivars can be created from the view of the total antioxidant capacity of seeds: Oscar Blanco > Annapurna > Rawa > Koniz > Golden Giant. Rutin content in amaranth samples was in interval 310 $mg.kg^{-1}$ (Oscar Blanco) – 508 $mg.kg^{-1}$ (Annapurna). The total polyphenol content in quinoa seeds ranged from 459 $mg.kg^{-1}$ to 1,839 $mg.kg^{-1}$. Based on values of the total antioxidant capacity of seeds the sudied quinoa cultivars can be ranked as follows: Ccankolla > Yulai > Carmen > Temuco > Quinua. Rutin content in quinoa samples was in interval 170 $mg.kg^{-1}$ (Ccankolla) – 368 $mg.kg^{-1}$ (Quinua). The presented results confirmed the statistically significant influence of cultivar on total polyphenol and rutin content as well as on the total antioxidant capacity of pseudocereal seeds.

Keywords: Pseudocereals, polyphenols, rutin

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A-20
BIOACTIVE COMPOUNDS OF CRANBERRY FRUITS IN THE FIRST AND THE SECOND HARVEST

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Cranberry (*Vaccinium vitis-idaea* L.) is one of few kinds of fruit, those selected cultivars give the harvest twice during the vegetation. The aim of this work was to compare the contents of bioactive components of cranberry fruits (macro-and microelements content, contents of hazardous metals, total polyphenols and anthocyanins) as well as the total antioxidant activity of cranberries obtained in the first and second harvest during one growing season. Samples of three cranberry cultivars able to give yield twice in one vegetation period (Ida, Koralle Bielawski Runo) were obtained from research breeding station in Kriva in Orava. Spectrophotometric method was used for the determination of the total polyphenol content (using Folin-Ciocalteau reagent), total anthocyanin content and the total antioxidant capacity (using DPPH). The contents of macro- and microelements and risky metals were in cranberries samples determined by AAS method. The average contents of macroelements (Mg, K, Na), microelements (Fe, Cr, Cu, Zn) and hazardous metals (Cd, Pb) were 492.4; 6326.4; 5.7; 12.0; 0.4; 2.7; 6.6; 0.1 and 0.4 $mg.kg^{-1}$ DM respectively. The content of Mg and K was higher in cranberries from the second harvest in comparison with the first harvest by 0.2–8% and 0.9–4% respectively as well as the content of Cr, Cu and Zn by 12–200%, 57–325% and 92–267% respectively. On other hand, the content of Cd and Pb in cranberries from the second harvest were lower in comparison with the first harvest by 11–42% and 14–83% respectively. The average values of the total content of anthocyanins and polyphenols were 540.0 and 2261.9 $mg.kg^{-1}$ FM respectively. The statistically significant differences between the first and the second harvest (P-value $2.42.10^{-7}$ and $2.06.10^{-25}$ respectively) were confirmed in values of total anthycyanin as well as polyphenol content, but in values of total antioxidant capacity there were no statistically significant differences (P-value 0.09).

Keywords: Cranberries, polyphenols, anthocyanins, antioxidant capacity

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A-21

THE RUTIN DISLOCATION IN DIFFERENT ANATOMY PLANT PARTS AND DIFFERENT GROWTH PHASES OF SELECTED AMARANTH CULTIVARS

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Amaranth is considered as a pseudocereal with high nutritive value (an excellent aminoacidic balance, a high protein content and other nutritive important compounds, p.e. flavonoid rutin). An HPLC DAD method working at 365 nm was applied for the determination of rutin in stems, leaves and pods of amaranth plants during vegetation period. Five cultivars were analysed: 1008, Olpir, Burgundy, Elephant Head a Plainsman cultivars. The content of rutin was evaluated in growth phase I (formation of buds), in phase II (at the beginning of flowering), in phase III (full blossoming) and in phase IV (full ripeness). When evaluating all four anatomical parts of amaranth we can state that rutin has the dominant position (other flavonoids were under detection limit). The gained results suggest the variety dependence, as well as the growth phase influence on forming of this compound. It has been identificated no statistically significant difference in rutin content in stems of amaranth between the growth phases, but the leaves of all tested samples show the highest value which was observed in IV. growth phase compared with I. and II. growth phase. The amaranth flowers contained about half the amount of rutin compared with amaranth leaves. Maximal increase of rutin content was manifested in each variety in phase IV, i.e. at the end of the vegetation period. On the basis rutin results in last phase (IV. phase) can be created the following order of anatomy parts: seeds (261.46–551.83 $mg.kg^{-1}$) > leaves (10.46–34.42 $mg.kg^{-1}$) > stems (1.61–3.13 $mg.kg^{-1}$). Among the tested anatomical parts (stems, leaves, flowers) was also found statistically significant differences. The most suitable variety from the tested ones was Burgundy from the standpoint of crops using as functional foodstuffs.

Keywords: Total polyphenolics, amaranth, anatomy parts

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A-22

CHANGES OF POLYPHENOLIC SUBSTANCES IN THE ANATOMICAL PARTS OF BUCKWHEAT (*FAGOPYRUM ESCULENTUM* MOENCH) DURING DIFFERENT GROWTH PHASES

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In this study the changes of total polyphenolics in different anatomical parts (stems, leaves, flowers and seeds) of common buckwheat (*Fagopyrum esculentum Moench.*) during vegetation period were analysed. The content of total polyphenolics was evaluated in growth phase I. (formation of buds), in phase II. (at the beginning of flowering), in phase III. (full blossoming) and in phase IV. (full ripeness). The total polyphenolics content was assessed spectrophotometrically using Folin-Ciocalteau assay on Shimadzu UV-1800. In all growth phases (GP) the stems and leaves were evaluated and statistically significant differences in polyphenolics content between these two parts were confirmed. Statistically significant differences ($P<0.01$) in polyphenolic content (in II. and III. GP) between stems and leaves; and between stems and flowers were found. The differences between leaves and flowers remained insignificant ($P>0.05$). In flowers an average of 13.8 times higher and in leaves 6 times higher concentration of polyphenolics in comparison with stems was measured. All anatomical parts in III. GP were analysed. In this phase the content of polyphenolics in common buckwheat was following: flowers>leaves>achene>stems. In flowers an average of 11.9 times higher, in leaves 8.3 times higher and in achenes 5.9 times higher contents of polyphenolics in comparison with stems were found. In III. and IV. GP (leaves, achenes, stems) the leaves contained in average 20 times higher and achenes 5.6 times higher polyphenolics compounds than stems.

Keywords: Buckwheat, total polyphenolics, anatomical parts

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A-23

TOTAL POLYPHENOLS CONTENT IN SELECTED CULTIVARS OF STRAWBERRIES IN RELATION TO CONTENTS OF Cd AND Pb IN SOIL

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The key aim of our study was to define the correlation of total polyphenols contents in strawberries grown in the parcels with exceeded levels of (pseudo-total) Cd and mobile Pb in soil. Strawberries were grown in rows long 200 m, the distance of the plants was 0.35 m and inter-row distance was 1 m. Following fertilizing was applied - at the beginning of vegetation period were the doses of elements (in $kg.ha^{-1}$): N48,2+P78,3+K66,3 +Mg7,2, afterwards for long time these elements were applied in liquid irrigation in amount: N46,5+P34,8+K118,7. Heavy metals in soil were analyzed by the method of atomic absorption spectroscopy and total contents of polyphenols spectrophotometrically. The highest content of all monitored parameters (pseudo-total and mobile Cd and Pb, as well as the amount of total polyphenols) was evaluated in Azia cultivar, in other cultivars there was no correlation compliance in varietal order among them to mentioned parameters. There was statistically significant positive relation of total polyphenols contents in Anthea cultivar to levels of all monitored forms of metals in soil and contrary, in Clery cultivar there was absolute non-correlation of total polyphenols contents on levels of monitored heavy metals in soil (mainly of mobile contents of Cd and Pb contents). Contents of mobile forms of individual heavy metals positively correlated with total polyphenols (except of Clery and Azia cultivars). Varietal relation on levels of total polyphenols was confirmed in experiment prior to relation of these organic compounds on the presence of monitored heavy metals in soil.

Keywords: Strawberries, fertilization, polyphenols

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A-24
DYNAMICS OF QUERCETIN FORMATION IN ONION (*ALLIUM CEPA* L.) DURING VEGETATION

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Onion bulbs (*Allium cepa* L.) are good sources of flavonoids. The aim of this study was to analyse the changes in dynamics of quercetin formation in three varieties of onions (white, yellow, red) during vegetation period. Quercetin content was determined after acid hydrolysis (1.2 M HCl in 50% aqueous methanol) by high-performance liquid chromatography (HPLC). The content of total phenolics was determined using Folin-Ciocalteu reagent (FCR) according to Lachman et al. (2003). The content of polyphenols in onion ranged from 2892.5±342.5 to 6051.5±267.4 mg.kg^{-1} and the content of quercetin ranged from 52.44±3.79 to 280.72±1.01 mg.kg^{-1} in fresh matter. The highest content of polyphenols and quercetin was found in red variety. According to statistical analysis the dynamic of quercetin formation in all varieties had statistically moderate ($P<0.05$) increasing tendency. Increasing content of polyphenols was accompanied with slight increase of quercetin, but the differences remained insignificant ($P>0.05$).

Keywords: Onion, quercetin, total polyphenolics

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A-25

EFFECT OF FERTILIZATION ON CHANGES OF THE POLYPHENOL CONTENT AND ANTIOXIDANT ACTIVITY IN POTATOE TUBERS (*SOLANUM TUBEROSUM* L.)

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Varietal dependency as well as agroenvironmental conditions belong to factors that are the most important because they affect the amount of polyphenol compounds in potatoes. Agroenvironmental conditions include: method of cultivation, weather conditions and impact of the site. The aim of this thesis was to assess the impact of nitrogen applied in a form of granulated vermicompost in graded doses: 0–40–80–120–160–240 kg $N.ha^{-1}$ on the content of polyphenols (TP) and also antioxidant activity (AOA) which relates with this polyphenols (TP). Content of TP which was determined in dry weight of potato tubers was decreasing in the first (control) variant and the fifth variant (399.21–331.09–187.46–97.34–70.40 $mg.kg^{-1}$ DW). In the sixth variant there is the higher amount of applied nitrogen (240 kg $N.ha^{-1}$), and the amount of TP was increasing because it was 1.9 times greater (135.56 $mg.kg^{-1}$ DW) than in the previous variant. We determined differences in TP and these differences are statistically proven, depending on content of applied nitrogen (polynomial function of second degree). Also the strong statistical relationship between the content of TP and the content of TAC (sign. F: $3.24.10^{-10}$) was confirmed. The highest value of AOA was in the first variant. From the first to the fifth variant (7.62–4.84%), the value of AOA was decreasing and in the sixth variant this value increased to 6.31%.

Keywords: Potatoes, polyphenols, antioxidant capacity, nitrogen fertilization

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A-26
DYNAMICS IN ANTIOXIDANT ACTIVITY IN THE ANATOMICAL PARTS OF SELECTED CULTIVARS OF BUCKWHEAT IN DIFFERENT GROWTH PHASES

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Buckwheat, which was once one of the basic components of the human diet, is now used mainly as alternative crop-plant, despite a rich nutrition value. Inter alia it is a source of substances with antioxidant properties that have important beneficial effects on the human health. In this paper the antioxidant activity (AOA) and its dynamics in fourth anatomical parts of plant (stems, leaves, flowers, achenes) in fourth growth phases (GP I–IV: buds formation, beginning of flowering, full flowering, full maturity) in six cultivars of buckwheat (*Fagopyrum exculentum*): Pyra, Spacinska, Kasho, Jana C1, Hrusowska, Emka was compared. The highest values of AOA measured in particular growth phases were in GP I in stems of variety Jana C1 (68.29%) and in leaves cultivar Spacinska (88.65%); in GP II in stems of cv. Jana C1 (71.64%) and in leaves of variety cv. Spacinska (88.65%). In GP III all the anatomical parts of buckwheat were available, the highest values of AOA were measured in stems of cv. Jana C1 (75.95%), in leaves of cv. Kasho (90.12%), in flowers of cv. Jana C1 (93.17%) and in achenes of Hrusowska (88.88%). In the last analysed growth phase (GP IV) the highest AOA was measured in stems of Emka cv. (88.36%), in leaves of Kasho variety (91.55%) and in achenes of cv. Hrusowska (90.47%). Stems and leaves of cv. Pyra showed the highest increase in AOA between GP I to GP IV (stems: 1.53 times, leaves: 1.34 times). In GP III the highest AOA was determined in flowers and the order among the cultivars was as follows: Spacinska (90.94%) – Pyra (91.16%) – Hrusowska (91.73%) – Kasho (92.01%) – Emka (92.38%) – Jana C1 (93.17%). Significant differences were found in AOA among particular anatomical parts of plant within the growth phases. In addition to GP III significant differences in AOA among monitored cultivars were confirmed. For statistical evaluation the program Statgraphic was used. Differences were compared for statistical significance at the level $P<0.05$.

Keywords: Buckwheat, cultivar, growth phase, plant part, antioxidant activity

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A-27

CHANGES OCCURING IN WEIGHT, TEXTURE, TOTAL SOLUBLE SOLIDS AND NATURAL ANTIOXIDANTS (CAROTENOIDS AND TOCOPHEROLS) COMPOSITION OF CANTALUPE MELONS DURING SHORT STORAGE

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The objective of this study was to compare the effects of storage conditions on the cantalupe melons (*Cucumis melo* L. *subsp. melo var. cantalupensins Naudin*) cv. 'Chianti'. Fruits were harvested at horticultural maturity and evaluated for quality traits during 14 days of storage at three different temperatures i.e., 2, 10 and 18°C with 85–95% relative humidity. At the beginning and at each storing interval friuts were weighed and flesh samples were taken for determination of the texture, total soluble solids (TSS), carotenoid components (β-carotene, lutein and lycopene) and isomers' composition of tocopherols (α-, γ- and δ). The results showed that the above mentioned parameters can be used as indicator for the quality atributes of melons. The weight loss of fruits slightly increased during the stored period studied, as well as insignificant differences between the temperatures. Whereas significant texture was lost more rapidly in the samples stored at 18ºC and 10°C than those stored at 2°C. TSS were also affected by storage time and temperature. The TSS content of fruit at 2°C increased and then remained constant over storage. At higher temperatures and at every stage of storage time TSS increased as storage time increased. The changes in the natural antioxidant components of fruits flesh during storage were investigated quantitatively by means of a HPLC technique. The predominant carotenoid in all samples was β-carotene. The carotenoids components were increased and then decreased with the time; however, the decrease processes were delayed by low temperature. The alpha form was the predominant tocopherol fraction. The level of tocopherol isomers significantly (α-tocopherol) and gradualy (γ- and δ-tocopherol) increased during the 7 days, but after 1st week of storing for all isomers a significant decrease was measured. High temperature storage at 18°C in comparison to 10°C and 2°C promoted γ- and 48; δ- tocopherol level.

Keywords: Cantalupe melons, storage, weight loss, carotenoids, tocopherols

A-28
MUSTARD PHENOLICS (PROFILE AND ANTIOXIDANT POTENTIAL)

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White mustard (*Sinapis alba*) is an interesting oilseed crop that belongs to the Brassicaceae family. Seeds of this oilcrop contain around 30% oil. Mustard oil is characterised by high content of polyunsaturated fatty acids (linoleic acid and linolenic acid). Oils rich in these fatty acids could be used in formulation of pharmaceuticals and nutraceuticals, which are industrial applications with a growing market. In recent years these oils have received attention also as a potential source of natural antioxidants (phenolics). Our interest in mustard was inspired by the recent search for antioxidants from natural sources. In the present investigation mustard phenolics were determined and quantified as well as their antioxidant potential was evaluated. Phenolic compounds were extracted with 80% methanol, and their concentration determined spectrophotometrically by the Folin-Ciocalteu method. Qualitative analysis of the phenolic compounds present in the extract was carried out by high pressure liquid chromatography. To describe their antioxidant properties, the effectiveness in scavenging free radicals and the reducing power were assayed. The results showed that the extract has a significant antioxidant effect when tested by each method. In general, the antioxidant activity of the extract was comparable with antioxidants that are widely used in food, such as butylated hydroxytoluene and L-ascorbic acid. Therefore, also mustard could be treated as a novel nutrient with promising antioxidant properties used in food and pharmaceutical industry.

Keywords: Mustard, phenolic compounds, HPLC, antioxidant activity

A-29

EFFECT OF ADSORBENT AND ION EXCHANGE RESIN APPLICATIONS ON TOTAL PHENOLIC CONTENT AND ANTIOXIDANT ACTIVITY OF WHITE AND RED GRAPE JUICES

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Resin application is widely used in fruit juice industry for reduction sugar content, deacidification, decolorization, demineralization and debittering. The aim of this study is to determine effects on total phenolic and antioxidant activity of grape juices of resins used to control the color of the white and red grape juices and to purify them. For this purpose, activated carbon, Dowex® 50Wx8-100 and Amberlite® XAD-16 were applied to the white and red grape juices. Total phenolic and antioxidant activity (DPPH, ABTS and FRAP) analyses were performed in control and resin applied grape juices. Total phenolic content of white grape juice was found as 2.28 g GAE/kg dry weight. Total phenolic contents of white grape juices were decreased to 14.00, 14.00 and 23.24%, respectively, by activated carbon, Dowex® 50Wx8-100 and Amberlite® XAD-16 applications. Similar to white grape juice, the most reduction in the total phenolic content of the red grape juice were obtained by Amberlite® XAD-16 application. The antioxidant activities of both grape juices were also decreased by resin application According to the obtained results, the lowest antioxidant activity values were observed in white grape juices applied to Dowex® 50Wx8-100, and in red grape juices applied to Amberlite® XAD-16.

Keywords: Grape juice, antioxidant activity, phenolic matter, adsorbent and ion exchange resins

A-30

INFLUENCE OF VARIETY AND STORING OF CHICORY (*CICHORIUM INTYBUS* L.) ON THE CONTENT OF TOTAL PHENOLS, ANTIOXIDATIVE POTENTIAL AND FATTY ACIDS

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Chicory (*Cichorium intybus* L.) has a long history of herbal use and is especially of great value due to its tonic effects upon digestive tract. It is popular in Meditteranean contries and is mainly consumed during winter time. In the present study, some nutritionally important ingredients like the content of total polyphenols, fatty acids and antioxidative potential (AOP) in external and internal leaves of different varieties of chicory were investigated. We analyzed the red varieties Leonardo, Trevisio, Mesola, Verona, stained variety Castelfranco, sweet varieties Jupiter, Uranus, Mercurius and red headed variety Chioggia. Chicories have been stored at a temperature of 0.1–0.8°C and relative humidity between 90 and 95%. According to results, variety and leaves (external, internal) influenced significantly the content of total polyphenols and antioxidative potential, while storing influenced the antioxidative potential only. Outer leaves have significantly higher AOP and higher content of total polyphenols. Chicory contains from 100 to 700 mg/100 g total fatty acids. The highest ratio (60%) is represented by linolenic acid, followed by linoleic (30%), palmitic (15%) and oleic (1.5%).

Keywords: *Cichorium intybus* L., storing, total polyphenols, antioxidative potential, fatty acids

A-31

INFLUENCE OF GAMMA IRRADIATION ON TOTAL PHENOLIC AND RESVERATROL CONTENT OF GRAPES AND RAISINS

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Grapes (*Vitis vinifera*) are among the fruits with the highest phenolic compounds content. Public interest on phenolic compounds is increasing due to their beneficial effects on human health, specially their antioxidant activity. Grapes are extremely susceptible to chilling injury, mechanical damage and microorganisms' presence. Treatment of food by specific ionizing radiations to improve microbiological safety and storability has been extensively applied. In this paper, gamma radiation influence on total phenolic compounds and resveratrol content of Benitaka cultivar grapes and raisin were studied in a radiation a dose range of 0–3.0 kGy. Total phenolic compounds were determined by spectrophotometric method while resveratrol analysis was performed by GC–MS after derivarization. Benitaka grapes showed a decrease in total phenolic content with increasing radiation doses. On the other hand, raisin samples showed the same range of total phenolic content despite the radiation dose applied. Storage time had no negative effect in phenolic content both to Benitaka grapes and raisins. The total resveratrol content in analyzed grapes samples increased with increasing radiation doses. Raisin samples showed a negligible resveratrol change due to irradiation treatment.

Keywords: Grapes, raisins, phenolic compounds, resveratrol, irradiation

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MARKERS OF VIRGIN OLIVE OILS SHELF-LIFE: ANALYSIS OF OXIDATION AND ACID HYDROLYSIS PRODUCTS OF PHENOLIC COMPOUNDS

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Phenolic compounds of virgin olive oil (VOO) have been associated with the shelf life and bitter taste of oil and are also related with beneficial health effects in part due to its antioxidant activity [1,2]. Phenolic alcohols, like hydroxytyrosol and tyrosol, phenolic acids, flavonoids, lignans and secoiridoids, are the most important class of phenolic compounds found in VOO. Secoiridoids are the prevalent compounds in VOO and include aglycones of oleuropein and ligstroside derivatives such as the dialdehydic form of decarboxymethyl elenolic acid linked to hydroxytyrosol or tyrosol (3,4-DHPEA-EDA or p-HPEA-EDA), oleuropein aglycone (3,4-DHPEA-EA) and ligstroside aglycone (p-HPEA-EA). These substances are aglycones of secoiridoid glucosides contained in the olive fruit, originated during the oil mechanical extraction process by hydrolysis of oleuropein, dimethyloleuropein and ligstroside. These reactions are catalysed by endogenous β-glucosidases [2]. Oxygen and light are two well-known oxidation-producing factors. Moreover, olive oil acidity exerts a great influence on oil stability as reported by Brenes et al. [1] as main changes in phenolic compounds present in VOO during storage were associated with the acid hydrolysis of the secoiridoid aglycones giving rise to an increase hydroxytyrosol and tyrosol levels. Oxidation products of hydroxytyrosol and tyrosol were evaluated by Di Maio et al. in order to find analytical indicators that could be used both as molecular markers of VOO "freshness" [2]. This work aimed to study the effect of other factors besides oxidation on secoiridoids of VOO. So the objectives of this study were to evaluate 1) the effect of oil acidity and air exposure on phenolic compounds of VOO and 2) the formation of new compounds that could be used as markers of VOO stability and indicators of its shelf life. The study was focused on secoiridoid compounds and for that aim a VOO was studied. For comparison purposes a refined olive oil with equivalent levels of hydroxytyrosol and tyrosol (compared with VOO) was also prepared as well as a refined olive oil with a phenolic extract obtained from the studied VOO. VOO and the refined olive oils prepared were submitted to induced conditions of oxidation (air exposure) and acidity (addition of 1% oleic acid). Phenolic extracts were then prepared and analysed by LC-ESI-MS in order to evaluate the degradation of the target compounds and their oxidation and acid hydrolysis products.

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Keywords: Olive oil, shelf life, phenolic compounds, oxidation, hydrolysis

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STUDY OF THE ANTIOXIDANT CAPACITY, ANTHOCYANIN CONTENT AND PHENOLIC COMPOSITION OF BLUEBERRIES (*VACCINIUM CORYMBOSUM* L.) FROM FOUR DIFFERENT CULTIVARS

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Antioxidants and phenolics have attracted the interest of the scientific community due to their potential health promoting characteristics. Properties that are frequently supported by the epidemiological evidence gathered concerning the consumption of foodstuffs rich in these classes of compounds. Blueberries are a good example of a fruit that is both rich in phenolics and possesses a considerable antioxidant capacity. In recent decades, Portuguese blueberry production has increased greatly with several new blueberry producers appearing every year. Considering the amount of factors that can affect the overall content of the fruits, either intrinsic (e.g. cultivar) or extrinsic (e.g. solar exposure, water availability) it is important to further understand the characteristics of blueberries, particularly when considering the valorization of this particular fruit. Considering all of the above, the properties of *Vaccinium corymbosum* blueberry fruits, from four different cultivars (Bluecrop, Duke, Goldtraube and Ozarkblue) and two different producers (each from a different area of Sever do Vouga), were studied for their antioxidant capacity, anthocyanin content and phenolic content and profile. To do so, several different methods were employed: *(I)* ABTS radical cation for the determination of the total antioxidant capacity. *(II)* The Folin-Ciocalteu reagent was used to assess the total phenolic content. *(III)* The differential pH was used to assess the total monomeric anthocyanins and *(IV)* determination of phenolic compounds by HPLC-DAD. Overall, the total phenolic content of the assayed blueberries ranged from 295.2±10.4 to 455.0± 25.6 mg of gallic acid equivalent per 100 g of fruit, with significant differences being found between both producers considered for the cultivars assayed, the only exception being Goldtraube. When considering the total anthocyanins, a range of 79.5±5.6 to 238.7±13.4 mg of cyanidin-3-glucoside equivalent per 100 g of fruit was observed, with Goldtraube registering the highest values and Ozarkblue the lowest (ca. 230 and 80 mg of cyanidin-3-glucoside equivalent per 100 g, respectively). Contrary to what was observed for the total phenolics, statistically significant differences were only registered for the Duke cultivar. The HPLC analysis of these extracts demonstrated that the overall anthocyanin profile varied quantitatively and qualitatively depending on the cultivar in question. As for the total antioxidant capacity a range from 546.3±46.3 to 781.0±77.4 mg of ascorbic acid equivalent per 100 g were found with, Duke and Goldtraube registering the highest values observed. These cultivars were also the only ones for which the producer had no impact upon the final values.

Keywords: Blueberry, anthocyanin, phenolic content, antioxidant capacity, cultivars

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INTERMEDIARY BREAKDOWN PRODUCTS FORMED BY THE THERMALLY INDUCED DEGRADATION OF ALIPHATIC GLUCOSINOLATES

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Glucosinolates (GSL) are secondary plant metabolites that occur especially in Brassica species such as cabbage, broccoli or mustard [1]. GSL and the enzymatically formed hydrolysis products – e.g. isothiocyanates (ITC) and nitriles – are of particular interest in food research because of their alleged anticarcinogenic effects. Thermal processes such as cooking and canning degrade GSL levels and form ITCs and nitriles [2]. The aim of this study was the investigation of thermal degradation pathways of aliphatic glucosinolates, e. g. sinigrin (allyl glucosinolate), in model systems. Various parameters such as pH value, temperature, iron ions and water content were studied to identify factors influencing the breakdown and the pathways of the degradation of the glucosinolate. GSL-breakdown as well as formation of volatile breakdown products (nitrile and ITC) and non-volatile products, e.g. D-thioglucose and D-glucose, were followed by HPLC-DAD and GC-FID. The breakdown products formed by thermally induce degradation of glucosinolates are dependent on the water content of the sample, on the concentration of iron(II) and on the pH value. In aqueous medium (pH 5.3) sinigrin releases D-glucose and the ITC. Basic medium reduce the thermal stability of the glucosinolate as well as of the ITC, as reported previously [3, 4]. Iron(II) reduces, especially in presence of additional vitamin C, the glucosinolate concentration and nitrile and D–thioglucose will be formed. Under dry conditions the desulfo-glucosinolate was identified for the first time as an intermediary breakdown product in the thermal degradation of glucosinolates.

[1] Fahey et al., Phytochemistry 2001, 56, 5–51
[2] Hanschen et al., J. Agric. Food Chem. 2012, 60, 2231–2241
[3] Hanschen et al., Food Chem. 2012, 130, 1–8
[4] Hanschen et al., J. Agric. Food Chem. 2012, doi: 10.1021/jf301718g

Keywords: Glucosinolate, thermal degradation, desulfoglucosinolate, isothiocyanate

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A-35
QUALITY ASSESSMENT OF COMMON PANDORA (*PAGELLUS ERYTHRINUS*) DURING STORAGE ON ICE MADE FROM CITRUS EXTRACTS

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In this study, the effects of ice with citrus extracts on the quality of common pandora (*Pagellus erythrinus*) were investigated during the storage. Extracts were prepared from albedo and flavedo tissues of two different citrus species (grapefruit and bitter orange) using ethanol. Concentrated citrus extracts were diluted with distilled water (1/100 w/v) before making of ice. Treatment groups were defined as grape fruit albedo (GA), grapefruit flavedo (GF), bitter orange albedo (BOA) and bitter orange flavedo (BOF) and traditional ice treatment was regarded as control group. The ice cubes were spread on each layer of fishes, then stored in a refrigerator at 0°C. The initial pH value was determined as 6.51 and showed a regular increase in control, GA and GF, while this value indicated fluctuations in BOA and BOF during the storage period. TVB-N levels of bitter orange treatment groups were recorded lower than the other groups reaching to 25.11±0.02 mg/100g at the end of the storage. The TMA-N values of bitter orange treatments were found to be lower than that of control and grapefruit. TBA value significantly increased in control; however the oxidation was suppressed in the citrus extracts treatment groups, especially in BOF treatment. The results showed that bitter orange albedo (BOA) and bitter orange flavedo (BOF) in combination with ice storage have more effectiveness in controlling the biochemical indices in common pandora.

Keywords: Pagellus erythrinus, common pandora, grapefruit, bitter orange, ice storage

A-36
INHIBITION OF LIPID OXIDATION IN ANCHOVY OIL BY CITRUS EXTRACTS DURING STORAGE

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The antioxidant effects of the citrus peel extracts on the oxidation of anchovy (Engraulis encrasicholus) lipids were assessed. The albedo and flavedo tissues of grapefruit and bitter orange were extracted in ethanol. 0.1 g of each citrus extract was dissolved in 100 ml anchovy oil, stirred and stored at 25°C in an incubator. The measurements were performed at 10-day intervals. The highest total phenolic content was found 7.93±0.12g GAE/100g in grapefruit albedo extract, whereas the antioxidant activity of bitter orange flavedo was recorded as 0.371±0.036 μM TEAC. The conjugated-diene values remained at low levels in flavedo tissues of both citrus fruit treatments. The p-Av value of bitter orange flavedo was 25.27±1.99 on the 40th day of the storage, whereas the p-Av measurements of control samples and the treatments with albodo tissues were high. TBARS formation increased during storage and reached over 5 mg MDA/kg in all samples, except bitter orange albedo (4.81±0.28 mg MDA/kg) and flavedo (4.42±0.07 mg MDA/kg) treatments. The UV spectrum of the samples supported the results of primary and secondary oxidation measurements. The citrus plant extracts stabilized the rate of oxidation owing to their antioxidant activity. Citrus peels can be replaced with synthetic protective agents in industry. Use of citrus extracts will also provide utilization of by-products of citrus processing.

Keywords: Lipid oxidation, grapefruit, bitter orange, anchovy

A-37
INCORPORATION OF STRAWBERRY INTO YOGHURT: EFFECTS ON THE PHYTOCHEMICAL COMPOSITION

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Yogurt has high nutritional value as source of calcium, protein, and provides the beneficial effects of living bacteria. Fruit preparations can be added to yogurt to create new products and combine the nutritional value of dairy and fruit matrices. Interactions of plant phenolics with proteins may lead to the formation of soluble or insoluble complexes. These interactions may have a detrimental effect on the in vivo bioavailability of both phenolics and proteins. The aims of this study were to establish evaluate the protein profiles of yogurt before and after the addition of strawberry and to assess the antioxidant properties and phytochemical of the fruit yogurt, in order to evaluate the possible interaction between protein and phenolic compounds therein. Industrial strawberry preparates containing 50% of fruit, 23% sucrose, 8% glucose-fructose syrup, starch (2%) were incorporated in natural yogurt and kept during 28 days at 2ºC. Extracts were obtained with methanol: formic acid (9:1 v/v) and stored at -20°C for 1 h to facilitate protein precipitation. Extracts was centrifuged and supernatant filtered with 3 kDa membrane. Total antioxidant activity was assessed by the ABTS method, total phenolics by Folin Ciocalteu's method, and total anthocyanins by pH-differential method. Individual phenolics and anthocyanins were analysed by HPLC-DAD and proteins profile were analyzed by FPLC, SDS-PAGE and Urea-PAGE. An immediate decrease in total antioxidant activity and total phenolics was observed after addition of fruit preparate to yogurt. Antioxidant activity, decrease from 0.84±0.08 to 0.65±0.06 mg ascorbic acid equivalents/g fw. Total phenolics decrease from 1.14±0.05 to 0.98±0.03 mg gallic acid equivalents/g fw and anthocyanins did not change significantly (0.060±0.008 to 0.067±0.017 mg pelargonidin-3-glucoside/g fw). After 28 days at 2°C, the antioxidant activity decrease 18%, total phenolics 11% and anthocyanins 25%. Ellagic acid decreased 20%, while (+)-catechin, (-)-epicatechin, rutin and kaempferol increased 7, 5, 18 and 12%, respectively. Anthocyanins decreased by 18, 48 and 21% for cyanidin-3-glucoside, pelargonidin-3-glucoside and pelargonidin-3-rutinoside, respectively, during the 28-day shelf-life period. (+)-Catechin, (-)-epicatechin, rutin and pelargonidin-3-glucoside were always present in yogurt in lower concentration than in the original fruit (accounted for dilution effects), suggesting strong interaction of these phenolics with the dairy matrix. The only soluble protein detected was alfa-lactalbumin present at 0.22 mg/mL, which decrease 47% when fruit is added. This strong reduction suggests an immediate formation of complexes upon incorporation of strawberry preparate. Free alfa-lactalbumin continued to decrease (48%) during shelf-life, being less available to absorption. These results suggest that interactions between strawberry and yogurt components may affect nutritional availability.

Keywords: Antioxidant activity, yogurt, protein-phenolic complexes

A-38

PRODUCTION AND CHARACTERIZATION OF PHENOLIC-RICH, AQUEOUS EXTRACTS OF DRY FRUIT AND LEAVES FROM VACCINIUM CORYMBOSUM L.

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In plants, phenolic compounds are frequently found as secondary metabolites and, considering the myriad of functions they perform (from structural support to their involvement in the plants reproductive and defense mechanisms), they are essential to plants physiology. There has been a considerable amount of epidemiological data relating the consumption of fruit and vegetables to lower risks of developing several pathologies (e.g. cardiovascular and neurodegenerative diseases), considering the high amount of compounds with a phenolic nature present in these foodstuffs, assuming that they are, at least partially, responsible for the health promoting properties observed. Vaccinium corymbosum, particularly its fruits (blueberries), are known for both their vast antioxidant and phenolic content, as such their use as a base to develop new foodstuffs or even ingredients with some functionality may be of particular interest, especially when considering their pleasing organoleptic characteristics. Considering all of the above, and taking into account the global popularity of tea and other infusions, this work aimed to produce tea-like extracts (infused and boiled), using dry fruits and leaves of *Vaccinium corymbosum*, with the highest antioxidant, phenolic and anthocyanins (for fruits) contents and to characterize their chemical composition. To do so, several different methods were employed: *(I)* ABTS radical cation for the determination of the total antioxidant capacity. *(II)* The Folin-Ciocalteu reagent was used to assess the total phenolic content. *(III)* The differential pH was used to assess the total monomeric anthocyanins. *(IV)* High Performance Liquid Chromatography was used to identify / quantify the main compounds present in the selected extracts. Our results showed that, for fruit extracts, higher extraction times lead to a lower yield of total anthocyanins though the same was not observed for infused or boiled leaf extracts. Overall, leaf extracts proved to possess a higher phenolic and antioxidant content than fruit extracts. The identification of the compounds present in the extracts showed that chlorogenic acid was present in all extracts, frequently being the most abundant compound. Additionally, p-coumaric and caffeic acid, cyanidin-3-galactoside and quercetin-3-galactoside were found in leaf extracts while, peonidin-3-glucoside and galactosides of malvidin and delphinidin were found in fruit extracts as relevant compounds.

Keywords: Phenolic content, antioxidant capacity, blueberry fruit, blueberry leaf

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A-39
SUPERCRITICAL FLUID EXTRACTION OF BETA-CAROTENE FROM TOMATO PASTE

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Supercritical fluid extraction (SFE) is a physical technique which uses supercritical fluids to separate a component from a mixture. The basic principle of extraction by SFE is based on the variable solubility of a solute in a solvent depending on both temperature and pressure. While most frequently applied supercritical fluid is carbon dioxide, modifiers (co-solvents) such as lower alcohols are used to enhance extraction efficiency. The aim of this work was to optimize the process of extraction of beta-carotene from tomato paste. The temperature of the extractor was set up in the range of 30–70°C and the pressure of the extraction fluid (with 10% ethanol as co-solvent) varied from 220–350 bar. The total amounts of beta-carotene in tomato paste and extracts were determined using HPLC. The most effective experimental arrangement resulted in the extraction of 60% of the total amount of beta-carotene. The parameters of the extraction process of the beta-carotene from tomato paste as well as some practical hints with regard to the extraction by SFE are discussed.

Keywords: Tomato, beta-carotene, supercritical fluid extraction

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A-40
FAT SOLUBLE VITAMIN IN CHEESE OF DIFFERENT COMPOSITIONS DURING RIPENING

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Milk fat is considered to be a significant source of vitamin A but a poor source of vitamins D and K. Vitamin contents in sheep milk are usually higher that in cow's and goat's milk, except for carotene. Goat and sheep milks have higher amounts of vitamin A than cow's milk. The composition of milk affects the composition in fat-soluble vitamins of cheeses to a significant extent. The variability in the fat-soluble components of cheese mainly depends on the conditions of milk production. The type of cheese-making technology does not significantly influence the cheese contents of vitamins A and E, apart from aspects related to the fat content of the cheese. Among the factors involved in milk production, the animal species is also likely to significantly influence the nutritional characteristics of cheese. The aim of the present study was to investigate the composition in vitamins A and E of cheeses made from the milk of ewes, goats and cows. A total of 84 cheeses of known composition were elaborated and controlled to determine the influence of different factors: e.g. variable proportions of cow´s, ewe´s and goat´s milk, seasonality (winter/summer), and evolution along the course of ripening. Cow's cheese showed a higher concentration of vitamin A than cheeses made from ewe and goat milk. Regarding vitamin E, the highest concentrations were found in ewe's cheese. The variable proportions from the different species did not vary in either the amount of vitamin A or that of vitamin E in the cheeses. Seasonality was seen to have a significant effect on the concentration of vitamin A. The cheeses elaborated with summer milk exhibited a significant increase in the amount of both vitamins during the first month of ripening, The cheeses made from milk collected in winter showed great variability in the amount of vitamins present in them. Ripening had a significant effect on the concentration of vitamin A. For the milk of the three species studied the concentrations of vitamin A increased up to the second month of ripening in the cheeses made with milk from ewes and goats; by contrast, the cheeses made with cow's milk ahowed a decreased concentration of vitamin E along the ripening period.

Keywords: Vitamin A, vitamin E, cheese ripening

A-41
CHANGES OF VITAMIN A AND E CONTENT IN SHEEP MILK DURING LACTATION

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Milk is one of the basic components of human nutrition. It has the function of nutrition, protection and detoxification. The sheep's milk, whose production increases in the recent years in the CR, is much more digestible than the cow's milk because the fat in it is scattered in small fat balls. Due to better digestibility of sheep's milk proteins it is an important part of the diet of people with allergies to cow's milk. Milk generally is characterized by relatively low content of vitamin A and E but it is consumed in various forms frequently and therefore it is one of their most important resources. Due to the higher average fat content in the sheep's milk (7%) we expect even higher proportion of lipophilic vitamins in the sheep's milk compared to the cow's milk and the goat's milk. The changes of the contents of vitamins A and E in sheep's milk of different breeds during the lactation period have been monitored during the years 2011–2012. Ration for animals consisted of pasture, hay and silage. The supplement diet consisted of a mixture of mashed grain and mineral licks (BIOSAXON). Pool sheep milk samples were collected directly on the farm once a month from April to September. The content of vitamin A and various forms of vitamin E (α-tocopherol and tocotrienol, γ-tocopherol and tocotrienol) were determined by HPLC. Both vitamins were extracted with hexane after previous saponification. Diode array detector (λ=325 nm) for the determination of vitamin A and fluorimetric detector (λex=292 nm, λem=330 nm) for determination of vitamin E) were used. In 2011, sheep's milk of combined breed Romanov sheep was analyzed. The vitamin A content was gradually decreasing during lactation. Statistically, the highest content of vitamin A was detected in early lactation in April, 0.79 mg/kg, the lowest in August, 0.46 mg/kg. The average amount of vitamin A was 0.61 mg/kg. In opposite to the content of vitamin A the total content of vitamin E during lactation increased sharply to July then declined and in September again slightly increased. Statistically, the highest content of vitamin E was recorded in July (3.64 mg/kg) and the lowest content (1.91 mg/kg) was determined in August. The average value of vitamin E in sheep milk was 2.84 mg/kg. The higher contents of both vitamin A and E in the sheep's milk in comparison to the cow's milk were tentatively confirmed. The milk of sheep breeds (East Friesian sheep, Lacaune sheep) is being further analyzed this year the final results from both years will be published as a poster at this conference.

Keywords: Vitamins, sheep, milk, lactation, HPLC

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A-42
ANTIOXIDANT ACTIVITY OF CAFFEIC ACID AND ITS ALKYL ESTERS IN OIL-IN-WATER EMULSION

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Addition of antioxidant to systems with lipids decreases lipid oxidation rate. Lipid oxidation rate depends on type and concentration of antioxidant, type of lipid system and storage conditions. Caffeic acid is naturally occurred polar antioxidant. Alkyl esters of caffeic acid are more lipophilic compounds which are more soluble in nonpolar systems (bulk oil). They affect positively lipid oxidation rate. In this experiment methyl, ethyl, propyl and hexyl esters of caffeic acids were prepared and added (concentration 3 $mmol.kg^{-1}$) to polar medium of oil-in-water emulsion (oil phase consisted of methyl esters of fatty acids of sunflower oil) and nonpolar medium of methyl esters of fatty acids of sunflower oil. Samples under oxygen atmosphere were stored 10 weeks at 20°C and extent of lipid oxidation (peroxide value, conjugated diens content, volatile secondary oxidation products content) and oxidation stability were determined. Caffeic acid showed the lowest activity of all used antioxidants in nonpolar system. Generally, this compound dissolved poorly in methyl esters of fatty acids of sunflower oil. Ethyl and propyl ester of caffeic acid had the highest antioxidant activity. Antioxidant activity of alkyl esters of caffeic acid decreased with increasing alkyl chain, for example hexyl ester of caffeic acid showed lower activity than ethyl and propyl esters of caffeic acid. In polar system hexyl ester of caffeic acid was effective in first phase of oxidation (formation of hydroperoxides) but formation of secondary products (hexanal) was accelerated. The rate of decomposition of hydroperoxides is faster than their formation. Generally, alkyl esters of caffeic acid were very active antioxidants in oil-in-water emulsions.

Keywords: Antioxidant, caffeic acid, alkyl esters of caffeic acid

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A-43

DETERMINATION OF THE EFFECT OF VARIETY ON THE CONTENT OF CAROTENOIDS AND TOCOPHEROLS IN PEELED BUCKWHEAT (FAGOPYRUM) ACHENES

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Buckwheat (Fagopyrum) is a genus of plants, which is widely used in human diet, especially in Asian countries and Ukraine. Recently we have experienced in the EU the transition from production agriculture to the so-called sustainable agriculture and buckwheat appears as one of the attractive crops in crop rotation. In addition, its consumption in the human diet steadily increases due to promotion of healthy eating and nutrition. Buckwheat contains many compounds which possess favourably affects on the human body. Among compounds with significant positive effects on the human organism, this study focused on carotenoids and tocols, which are major lipophilic antioxidants. Antioxidants are substances that are involved in protecting cells against free radicals. This protection is based in their so-called ability to quench free radicals and thus protect cells from oxidation and destruction processes. In the present study, levels of carotenoids and tocopherols in buckwheat that came from the exact field trial set up in 2010. For the research a total of 10 varieties were selected for: Aelita, Ballada, Botansoba, Jana, Kara-Dag, Krupinka, Monori, Pyra, Spačinská 1 and sp. 01Z5000056. The content of individual carotenoids was determined by HPLC-UV-DAD and content of tocopherols and tocotrienols by HPLC-ESI/MS/MS. The measured values were statistically analyzed using analysis of variance (ANOVA). The results of statistical analysis of buckwheat showed that there is a significant difference between varieties in their ability to synthesize carotenoids and tocols. In the peeled achenes average content of carotenoids has been calculated as 2.00 μg/g dry weight in 2010. Among carotenoids contained mainly lutein was present (90.00%), and other carotenoids neoxanthin, violaxanthin and zeaxanthin were present only in lesser amounts (5.38%, 3.05% and 1.57%, respectively). Varieties with high levels of carotenoids were sp.01Z5000056 and Monori. On the contrary, the Jana and Krupinka varieties distinguished with a low average content of carotenoids and violaxanthin and neoxanthin were not in them detected. The average content of tocopherol was 80.27 μg/g fresh weight in 2010. From the total content of tocopherols, γ-tocopherol represented major proportions (90.58%) and in minor amount has been contained δ-tocopherol (6.21%), while β-tocopherol and tocotrienols were not detected. Varieties with a high content of tocopherols are the Kara-Dag, Spačinská 1 and Jana. Conversely the varieties Monori and Ballad were characterized as varieties with a low average content of tocopherols.

Keywords: HPLC-UV-DAD, HPLC-ESI/MS/MS, AOA

A-44

A MATHEMATICAL MODEL FOR THE CHARACTERIZATION AND OBJECTIVE COMPARISON OF ANTIOXIDANT ACTIVITIES PRODUCED BY THE COPPER-INDUCED OXIDATIVE LDL METHOD

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The copper-induced oxidative LDL method is particularly suitable for studying an early stage of the oxidation process and the relationships between LDL oxidizability against the effects of prooxidants and antioxidants on both, in vivo and in vitro experiments. The available data about the involvement of pro- and anti-oxidants in the kinetics of LDL oxidation are abundant, but often only semi-quantitative conclusions can be achieved, due to the difficulty of verifying a detailed kinetic model in a process with the complexity of the LDL oxidation. Under these conditions, the experimental designs are frequently intuitive, and many of the abundant bibliographical data in this regard are incomplete from a kinetic point of view, preventing the full characterization of the relevant regularities of the response. The pseudo-mechanistic model that we propose is based on three consecutive accumulative Weibull's functions, and it represents a formal transfer from the field of the dose-response relationships. It allows to include the effects of any number of pro- or anti-oxidant concentrations allowing to evaluate the response simultaneously. Its application is simple: it provides parametric estimates, which characterize the oxidative process and the pro- or anti-oxidant activities; it facilitates rigorous comparisons between the effects of distinct compounds in the LDL from different patients; reduces the sensitivity to the experimental error; and its mathematical form constitutes a useful orientation to prepare more economic and efficient trial designs. The model was assayed, firstly, using the kinetic simulation of the oxidative process, and finally, it was applied to a variety of experimental data from other authors, obtaining highly satisfactory results in all cases.

Keywords: Copper-induced oxidative LDL method, mathematical modelling, dose-response relationships, prooxidant and antioxidant effects, kinetic simulation

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A-45
A MATHEMATICAL APPROACH TO EVALUATE THE KINETIC ANTIOXIDANT ACTIVITY RESPONSES: THE β-CAROTENE METHOD AS A CASE STUDY

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The β-carotene (βC) bleaching assay, a common method for evaluating antioxidant activity, has been widely criticized due to its low reproducibility, problematic quantification, complex reagent preparation and interference of different factors (temperature, pH, solvents and metals). The βC method operates on a system of lipid micelles in an aqueous environment, which could constitute an acceptable model for many foods and even some biological systems, but it is not a universal environment. Additionally when kinetic models are disregarded and measures are performed at a single time, this sensitivity seems to require a complex and perhaps impractical standardization. In this work we develop a highly reproducible procedure for microplate assay, evaluate the critical points of the method and propose a kinetic model for quantifying both antioxidant and pro-oxidant activities. The proposed kinetic model produces stable and meaningful characterizations, and the microplate assay provides an appropriate tool for ensuring that sample series with a large number of items can be now simultaneously assessed. The application of these resources produced very consistent results, which provided robust and meaningful criteria to compare in detail the characteristics of several well-known commercial antioxidants, as well as several predictable pro-oxidants.

Keywords: Antioxidant activity, β-carotene method, mathematical modelling, kinetic response

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A-46
LOSSES OF TOCOPHEROLS DURING PAN FRYING

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In literature and similar sources, there is sufficiency of reliable results documenting content of tocopherols in different food raw materials and unprocessed foodstuffs, but results describing this for heat-treated foodstuffs are not enough and existing data should be used with caution. Therefore, losses of tocopherols during culinary food treatment were started to study. In the present part of this project, the losses of tocopherols during frying of bread were quantified. To assess factors affecting the losses, different fats and oils (differing in fatty acids composition and initial tocopherols content) were used for the frying and some operating parameters (as temperature, time of heating of oil before the frying etc.) were varied.

Keywords: Tocopherols, frying, losses

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A-47
BIOLOGICALLY ACTIVE COMPOUNDS IN POTATOES FROM ORGANIC AND CONVENTIONAL FARMING

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Potatoes (*Solanum tuberosum*) are one of the world's major agricultural crops and their production in organic farming systems increase significantly. Potatoes serve as major, inexpensive, and available low-fat food source providing energy, high-quality protein, fibre, and vitamins. Potatoes produce biologically active plant secondary metabolites, which may have both adverse and beneficial effects in the diet. Contaminant content (pesticide residues, heavy metals, nitrates, etc.) is significantly lower in organically grown products, however there is a risk of natural toxic compounds appearance (e.g. alkaloids), which are naturally created by plants against diseases and vermin. The aim of this project was an investigation of the influence of farming conditions on the potato quality. Potato varieties Katka and Finka were grown at two different localities in Czech Republic. To determine a possible inter-annual variability, the experiments were performed on crops from five consecutive years (2008–2012). The monitored parameters included positive biologically active compounds (vitamin C, chlorogenic acid, amino acids, sugars; and also starch and dry weight) and also toxic secondary metabolites (glycoalkaloids α–solanine and α–chaconine and calystegines A3, B2 and B4). High performance liquid chromatography coupled with conventional detectors was used for determination of vitamin C, chlorogenic acid, sugars or amino acids. For determination of toxic glycoalkaloids and calystegines ultra-high performance liquid chromatography coupled with tandem mass spectrometer was used. The levels of targeted biologically active compounds depend mainly on the variety and climatic conditions (year of farming), the type of a cultivation system did not play a significant role. The currently investigated natural toxic alkaloids calystegines were found at higher levels than the monitored and legislatively controlled natural toxicants glycoalkaloides (MRL 200 mg/kg).

Keywords: Potatoes, biologically active compounds, organic farming, UPLC–MS/MS

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A-48
MONITORING OF PHYTOESTROGENS METABOLITE EQUOL IN MILK AND MILK PRODUCTS

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Phytoestogens are plant-derived compounds, which commonly occur in wide range of foodstuffs. Phytoestrogens can exert biological activity by interacting with the mammalian hormone system, mimicking the effect of mammalian steroidal estrogens. An important group of these compounds are phytoestogenic isoflavones (daidzein, genistein and glycitein), which can be found in legumes, especially soybeans. Red clover also incorporates isoflavones abundantly, mostly formononetin and biochanin A. According to epidemiological studies, phytoestrogen rich diet, as in Southeast Asian countries, appears to diminish chronic diseases such as breast and prostate cancers, cardiovascular disease and osteoporosis. Less than half of the human population are so-called equol producers. This means that the gut microflora of such people is able to convert isoflavones precursors into equol. Equol, because of its similar chemical structure to estrogen and its ability to human estrogen receptors, is of considerable interest as a possible risk or protective factor for reproductive cancers. Other mammalian species, such as cows, are thought to invariably have intestinal bacteria capable of metabolizing daidzein to equol. Target phytoestrogens were monitored in 12 milk samples and 22 milk products (cheese and yogurt) from conventional and organic farming. Phytoestrogens were released from bound forms by enzymatic hydrolysis and analysed as free aglycones (dadzein, genistein, and glycitein) together with dadzein metabolite equol. Ultra-high performance liquid chromatography coupled with tandem mass spectrometer (UHPLC–MS/MS) was used for determination of these compounds. The eguol content in milk, yogurt, and cheese was in the range of 17–73 ng/mL, 51–410 ng/mL, and 37–1200 ng/g, respectively.

Keywords: Phytoestrogens, metabolism, equol, UPLC–MS/MS

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A-49
CHANGES IN REDUCING POWER DURING PROCESSING OF CEREALS AND VEGETABLES

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HPLC with amperometric detection (HPLC–ECD) was used to determine reducing power of processed cereal (coffee surrogates, bread crumb and crust, roux, crackers and biscuits) and vegetable products (baked potatoes, French fries and chips, tomato paste and ketchup, fried onion, etc.). The main objective was to evaluate the effect of selected technological and culinary processes on reducing power of wide range of foods of plant origin by comparing the differences in the profile and amount of electrochemically active compounds in prepared foods and their raw materials. Changes in the composition of electrochemically active substances and an increase of the total reducing power in processed foods can be due to the release of phenolic and other reducing compounds from insoluble matrices, transformation of phenolic compounds and the Maillard reaction. We focused on the assessment of participation of the process-induced compounds on reducing power and the characterization of the compounds with significant electrochemical activity. Aqueous and methanol extracts of boiled foods, e.g. potatoes and tomato paste, were only slightly higher (up to 1.5 times) in electrochemical reducing capacity than the respective raw materials. On the other hand, frying, baking and especially roasting were the processes at which the compounds possessing reducing power were intensely formed, mostly via the Maillard reaction. While the electrochemical capacity of raw materials did not exceed 1.8 g BHA equivalents (BHAE)/kg, the highest values – 8–20 g BHAE/kg – were found for coffee surrogates containing roasted cereals. The reducing power in most cereal and potato products was partly due to the presence of a Maillard-derived 2,3-dihydro-3,5-dihydroxy-6-methyl-(4*H*)-pyran-4-one (DDMP), which was responsible for 3–43% electrochemical capacity of the foods. Reducing power of the cereal and vegetable products determined by the amperometric method was confirmed by the comparison with well correlated results of DPPH radical scavenging assay.

Keywords: Cereals, potatoes, reducing power, Maillard reaction, 2,3-dihydro-3,5-dihydroxy-6-methyl-(4*H*)-pyran-4-one

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A-50
IMPACT OF THE ROASTING PROCESS ON ANTIOXIDANT COMPOUNDS IN COFFEE

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Coffee is one of the most popular drinks worldwide, with a consumption of almost 7 million tons in 2010. Several studies have investigated the impact of coffee consumption on health showing interesting benefic effects on Alzheimer's, Parkinson's and diabetes. Part of the bioactivity in coffee can be related to its antioxidant potential, due to the presence of phenolic compounds, mostly chlorogenic acids. In this study we investigated the effect of the roasting process on coffee's phenolic content, and the related radical scavenging capacity. Therefore, we roasted green coffee at 200°C, 225°C or 235°C with durations ranging from 0 to 30 min, resulting respectively in light, medium or dark roasted coffees. Phenolic compounds in a methanolic extract, obtained by accelerated solvent extraction, were analyzed by HPLC, with on-line post-column radical scavenging detection based on $ABTS^{\bullet+}$ decolorisation. Phenolic composition evolved from 6 native radical scavenging compounds in green coffee, up to 16 antioxidant compounds of which 10 are newly formed during the roasting process in roasted coffees. The 6 native compounds were identified as 5'-O-, 3'-O- and 4'-O-caffeolyquinic acid (CQA), and 3',4'-O-, 4',5'-O- and 3',5'-O-dicaffeoylquinic acid, by means of 1H NMR and HRMS analysis after purification. In short roasted coffees, 5'-CQA and 4'-CQA related radical scavenging capacity (expressed in Trolox Equivalent Antioxidant Capacity: TEAC) increase before their decline in medium and dark coffees. The other four native compounds show a progressive decline of their corresponding TEAC values in light medium and dark coffee. This could mean that degradation of the latter four can lead the neoformation of 5'-CQA and 4'-CQA. These reactions go either through loss of a caffeic acid moiety from a dicaffeoylquinic acid, and or isomerization from a caffeoylquinic acid, resulting in a caffeic acid migration from one position to another on the quinic acid moiety. Further, four of the newly formed active compounds could be identified by ^{1}H and ^{13}C NMR analysis as two caffeoylquinolactones (5'-O-Caffeoyl quinide and 4'-O-caffeoyl quinide), and two feruloylquinic acids (3'-O-Feruloylquinic acid and 5'-O-Feruloylquinic acid). These results suggest that water loss followed by formation of an intramolecular bond, or methylation occur during the roasting process. The formation of these compounds follows a general pattern: each reaching a maximum TEAC value in light or medium roasted coffee, before their decline in darker ones. The neoformated radical scavenging compounds do not completely compensate the loss of total antioxidant capacity in coffee extracts related to the native phenolic degradation. Indeed, the total antioxidant capacity in coffee extracts declines during the roasting process along with temperature and duration. Although, the higher roasting temperature is, the lower is the remaining radical scavenging capacity.

Keywords: Chlorogenic acids, radical scavengers, ABTS, coffee, polyphenols

A-51
DEVELOPMENT OF A RAPID IMMUNOCHEMICAL TEST FOR DETECTION OF STEROIDAL ANABOLIC NANDROLONE

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19-Nortestosterone (17β-Hydroxy-19-norandrost-4-en-3-one), also named nandrolone, is an anabolic steroid (a muscle-building chemical) which occurs naturally in the human body, but only in tiny quantities. It is very similar in structure to the male hormone testosterone, and has many of the same effects in terms of increasing muscle mass. Ergogenic uses for this steroid in sports, racing, and bodybuilding are controversial because of its adverse effects and the potential to gain an advantage conventionally considered "cheating." Its use is referred to as doping and banned by all major sporting organizations. Traditional methods for analysis of anabolic steroids (e.g. LC/MS, GC/MS), although sensitive, are expensive and usually require specialized instrumentation and highly skilled personnel. Moreover, pretreatment procedures of any samples involve numerous extraction steps that are time-consuming and unsuitable for routine analysis of a large number of samples or on-site determinations. Compared with instrumental methods, immunoassays are portable and cost-effective, with adequate sensitivity, high selectivity, and a simple sample extraction process. Therefore, immunochemical techniques have become popular and are increasingly considered as alternative/complementary methods for residue analysis. The aim of this study was to develop an enzyme-linked immunosorbent assay (ELISA) for the rapid detection of steroid nortestosterone and its metabolite residues. For construction of indirect competitive tests polyclonal antibodies and the nortestosterone-ovalbumin conjugate were used. Under optimal experimental conditions, the most sensitive assay achieved IC50 and limit of detection values of 6.41 and 0.003 ng/mL, respectively, when it was run in 0.01 M phosphate-buffered saline containing 0.1% gelatin. The developed system was tested for cross-reactivity with several endogenous steroids. Expect for cross-reactivity with another frequently abused steroid testosterone (23%), other interference to the assay was negligible (<0.1%). We conclude that after the validation for particular matrices (e.g. food complements, biological materials), the method will be a useful tool for analysis of 19-nortestosterone, especially in low volume and low concentration samples.

Keywords: 19-nortestosterone, anabolic steroid, immunosorbent assay

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A-52

IS TYPE OF VARIETY, PRODUCTION METHOD AND SO_2 LEVEL AFFECTING ANTIOXIDATIVE PROPERTIES OF WINE?

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Antioxidative effect of natural products and compounds is one of the most thoroughly studied biological activities. Grape wine holds high position among the strongest natural antioxidants. Many health effects of grape wine, such as reduced risk of cardiovascular diseases, are assigned to its free radical scavenging ability. Hence, interest is now turned to which factors can affect antioxidative potential of wine. Oxygen Radical Absorbance Capacity (ORAC), 2,2-diphenyl-1-picrylhydrazyl radical scavenging capacity assay (DPPH) and content of total sulphites (colorimetric assay) were measured in wines manufactured in Georgia, Czech Republic and Western Europe (France, Austria and Italy). Wines from Saperavi belonged to the most active radical scavengers among red wines exhibiting values at 52.2 mmol TE/L in ORAC and 1.5 μL/mL in DPPH assay (mean values). Other tested red varieties (Rulandské modré, Pinot Noir, Cabernet Sauvignon, Cabernet Moravia) showed values at 48.0, 45.7, 44.2 and 35.6 mmol TE/L (ORAC) respectively and 2.4, 2.5, 2.7, 3.5 μL/mL (DPPH) respectively (mean values). Except for Saperavi, high variability of antioxidative activity within varieties was observed. White wines (Rkatsiteli, Mtsvane, Chardonnay, Sauvignon) showed activity in range of 6.8–2.5 mmol TE/L for ORAC and 14.0–109.2 μL/mL for DPPH. Nevertheless, Kakhetian Nobile (mix of Rkatsiteli, Mtsvane, Khikvi) showed outstanding activity among white wines (22.6 mmol TE/L [ORAC], EC_{50}=2.5 μL/mL [DPPH]). This wine was prepared by traditional Georgian (Khakhetian) method, which means fermented with pomace. Sulphite content varied from 0–73.2 in red wines and 87.0–202.0 mg SO_2/L in white wines. Saperavi wines had one of the lowest SO_2 content (0 – 26.7 mg SO_2/L) from all samples. Solution of 400 mg SO_2/L exhibited activity 0.06 mmol TE/L in ORAC and EC_{50}=223.5 mg/L in DPPH assay. In conclusion, Saperavi wines from Georgia had one of the highest antioxidative activities from all tested wines. Therefore, type of variety seems to be important factor affecting antioxidative potential of grapes and their products. In general, red wines are significantly stronger free radical scavengers then white wines. Results suggest that fermentation process can improve antioxidative ability of white wines. Positive correlation between variety and fermentation process on antioxidative effect was also described in other studies. Sulphites added to wine have no significant effect on free radical scavenging activity. Sulphites added to wine have no significant effect on free radical scavenging activity. This is the first report of this phenomenon in ORAC and DPPH models.

Keywords: Antioxidative activity, DPPH, ORAC, saperavi, wine

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A-53

FORMATION OF BIOGENIC AMINES DURING DRYING AND RIPENING OF TRADITIONAL DRY FERMENTED SAUSAGE *PETROVSKÁ KLOBÁSA* PRODUCED IN PROVINCE OF VOJVODINA (NORTHERN SERBIA)

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Petrovská klobása is a traditional dry fermented sausage from Northern Serbia. Formation of nine biogenic amines during drying and ripening of Petrovská klobása in traditional room and industrial ripening chamber, produced from hot deboned and cold meat, were determined using HPLC. Separation of dansyl chloride derivatizated amines was completed in 8 minutes. Analyses were performed by HPLC–DAD on Eclipse XDB–C18 column. Spermin was determined in all analyzed samples, while histamine, serotonin and spermidine were not detected in any samples. At the end of drying phenilyethylamine was the prevaling amine in the first (51.6 mg/kg) and tryptamine in second (38.1 mg/kg) and third (28.7 mg/kg) sausage group, while at the end of ripening tryptamine was the prevailing biogenic amine in all sausage groups (133, 121 and 39.8 mg/kg). Total level of biogenic amines in all investigated sausages did not exceed 174 at the end of drying and 238 mg/kg at the end of ripening.

Keywords: Biogenic amines, traditional dry fermented sausage, Petrovská klobása

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A-54
STUDY ON DOMESTIC BOILED COW MILK: ALLERGENIC AND NUTRITIONAL PROPERTIES

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Since some years in many European countries, including Italy, consumers have the possibility to get fresh milk local produced directly from automatic distributors spread in their neighbourhood. By the law, the farmers producing the 'raw' milk are obliged to suggest their customers to boil it before consumption. Milk boiling is essential to avoid *Escherichia coli* infection that may cause the hemolytic-uremic syndrome (SEU), which is a major cause of acute renal failure in children. This study intended to evaluate the effects of common domestic boiling treatments on both the nutritional and allergenic properties of milk. Aliquots of the raw milk were heated either on hotplates or in the microwave oven. The samples were analysed by native 1-D electrophoresis to compare the protein patterns of raw milk with both treated milk samples. All proteins were identified by N-terminal amino acid sequencing. To evaluate changes in protein immune reactivity due to the heat treatments, immunoblotting experiments were performed. Utilizing the IgEs from the serum of 20 milk-allergic patients it was observed that the most reacting proteins are α-S1 casein, β-casein and β-lactoglobulin. Aggregates of β-lactoglobulin were identified in a band at the gel-well level in both treated milk samples but no immunologic reactions was observed for these bands probably because the immuno reactive epitopes are masked by the aggregation phenomenum. Moreover, possible changes in nutritional properties of boiled milk were investigated by the evaluation of total available lysine content (modified OPA method), lactoperoxidase activity and ε-fructosil lysine formation. Our results show that boiled milk presents a different migration protein pattern whereas this does not drive to significant changes in the immunoreactions pattern.

Keywords: Boiled milk, native electrophoresis, immunoblotting, β-lactoglobulinavailable lysine

A-55
DETERMINATION OF WHEY PROTEINS WITH ALLERGENIC POTENTIAL IN DIFFERENT TYPES OF MILK

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The aim of this work was to introduce method for detection of main whey proteins with allergenic potential and to determinate their amounts in different types of milk. The major whey proteins with allergenic potential are α-lactalbumin and β-lactoglobulin. Genetic variants of these whey proteins were detected also. Samples of cow´s milk, goat´s milk and sheep´s milk were collected from farms and milk bars in the Czech Republic from April to June 2010. These 40 samples of each milk type were analyzed by means of reversed phase high-performance liquid chromatography on liquid chromatograph Alliance 2695 with PDA detector 2996. The separation was performed on C18 column X Bridge^{TM}, 150×3.0 mm, 3.5 μm. Gradient elution was used. Mobile phase contained water, acetonitrile and trifluoroacetic acid. Average values of the determined proteins were following: cow´s milk contained 1.16±0.10 g.L^{-1} α-lactalbumin and 4.10±0.25 g.L^{-1} β-lactoglobulin, goat´s milk 1.27±0.34 g.L^{-1} α-lactalbumin and 3.07±0.49 g.L^{-1} β-lactoglobulin and sheep´s milk 0.95±0.38 g.L^{-1} α-lactalbumin and 5.97±1.54 g.L^{-1} β-lactoglobulin, respectively. Overall statistically highly significant difference in α-lactalbumin and β-lactoglobulin of cow's, goat's and sheep's milk was evaluated by Wilcoxon pair test ($P<0.01$). This method is suitable for whey proteins determination in milk.

Keywords: α-lactalbumin, β-lactoglobulin, cow´s milk, goat´s milk, sheep´s milk, RP-HPLC

Acknowledgement: *This work was supported by IGA VFU Brno 72/2010/FVHE of the Czech Republic.*

A-56

EVALUATION OF AUTHENTICITY AND BOTANIC ORIGIN OF BUCKWHEAT HONEYS ON THE BASIS OF THEIR PHYSIOCHEMICAL, PHENOLIC AND VOLATILE COMPOSITION DATA

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Honey is a remarkably complex natural liquid produced by bees from plants nectars, plant secretions and excretions of plant sucking insect. Its composition depend strongly on the plant species, although external factors also play a role, such as seasonal, environmental factors and processing conditions. Honey consists of a supersaturated solution of sugars, of which the main contributors are fructose (38%) and glucose (31%). A wide range of minor constituents is also present in honey, many of which, including phenolic compounds, are known to have antioxidant proprieties. The presence of this substances highlight the role of honey as a nutritional source of natural antioxidants with a wide range of biological effects, such as anti-bacterial, anti-inflammatory, anti-thrombotic, but also anticancer and anticarcinogenic proprieties. Buckwheat honey is a dark honey produced by bees that collect pollen and nectar from the little pink flowers of the buckwheat plant (Fagopyrum esculentum Möench, Polygonaceae), popularly recognized as an excellent honey source and a selective antioxidant and hypolipidaemic nutrient food. In literature have been reported the high antioxidant proprieties of the buckwheat honey, as well as its antibacterial activity. Considering the several health benefits of this honey, the principal aim of this work was the characterization of some buckwheat honeys. Ten buckwheat honey samples, from different collection place, were characterized on the basis of their pollen, physicochemical, phenolic and volatile composition data. Electrical conductivity, optical rotation, pH and sugar composition were the physicochemical parameters with higher discrimination, showing the presence of three blended honeys. Honey volatiles, analysed by solid phase microextraction (SPME) and gas chromatography mass-spectrometry (GC/MS), revealed more than 100 volatile compounds, most of them present in all honey samples even though with quantitative variation. Besides many furfural derivates, 3-methylbutanoc acid was the main volatile compound found in most of the samples. The presence of 2- and 3-methylbutanal, pheynalcetaldehyde, among with 3- methylbutanoic acid confirmed the typical buckwheat aroma of some studied samples and corroborated physiochemical data. HPLC-MS phenolic profile did not show significant differences cross the honeys and p-coumaric and p-hydroxybenzoic acid were found to be the most abundant compounds in all buckwheat honeys. While melissopalynological analysis remains nowadays as the only technique which allows a direct botanical source characterization, physicochemical parameters afford quantitative results, allowing an approximate estimation of the kind of honey and the possible presence of honey blends. Also volatile and phenolic compounds function as important role in honey characterization and improves our knowledge about honey as a source of antioxidants.

Keywords: buckwheat honey; pollen and physiochemical parameters; volatile compounds sugar composition; phenolic compound.

Chemistry behind novel foods

B-1
NATURAL TOMATO SOLID CONTENT EVALUATION IN TOMATO PRODUCTS

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Twenty years of experiences with the determination of natural tomato soluble solid (NTSS) is discussed and results of analyses are summarized. The robustness of the markers, the chemical changes during the processing and storage of tomato products are evaluated. The recent regression equations for the most common chemical markers updated according to the properties of recently used raw materials are formulated (Pyrrolidon carboxylic acid: NTSS=(0.0328×PCA (at 100°Bx)+5.93)×Rf/100, Formol number: NTSS=(0.176×FN (at 100°Bx)-1.778) x Rf/100, Potassium: NTSS=(0.0233×K (at 100°Bx)+4.97)xRf/100, Malic acid: NTSS=(0.0776×MALIC (at 100°Bx)+8.88)×Rf/100 and citric acid: NTSS=(0.00916×CITRIC (at 100°Bx)-0.997)xRf/100). The often interpretation failures are summarized and the possibilities of chemometric and omics approaches and general use of real time analyses in tomato product authentication are discussed on the own preliminary analytical results of the limited set of tomato samples.

Keywords: Tomato products, natural tomato solid, authenticity

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B-2

EFFECT OF RELATIVE HUMIDITY ON STABILITY OF ENCAPSULATED SUNFLOWER OIL

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Lipid oxidation is the major reaction leading to deterioration of foods containing unsaturated lipids. One approach to stabilize unsaturated oils is to encapsulate them by spray-drying to limit the contact between lipids and oxygen. The aim of this work was to determine the effect of relative humidity (RH) on stability of encapsulated polyunsaturated oils by studying primary and secondary oxidation products. As a simple model, sunflower oil was encapsulated in a maltodextrin-sodium caseinate matrix by spray-drying. An emulsion containing refined sunflower oil (30% of dry matter), Na-caseinate (3% of dry matter), maltodextrin DE 22.2 (67% of dry matter), and water was spray-dried to powders. The dried emulsion was stored under five RHs (~0, 11, 33, 54, and 75%) at 22°C in the dark for 29 weeks, and studied at certain time points for hydroperoxides by peroxide values (PV) and volatile compounds by solid-phase micro extraction (SPME) with GC–MS. At RH ~0,11 and 33% the encapsulated oil started to oxidize without any induction period. The highest PV was obtained in dried emulsions at RH ~0%, followed by RH 11, 33 and 54%. Based on the PVs, the dried emulsion was most stable against oxidation at RH 75%. At this RH, the maltodextrin-sodium caseinate matrix collapsed and the powder particles stuck together. This increased the length of the diffusion path of oxygen across the matrix. The main volatile secondary oxidation products formed by decomposition of hydroperoxides at all RHs were hexanal, hexanoic acid and nonanal. The main formation of volatiles in powders occurred after 13 weeks of storage. At this time also noticeable differences between dried emulsions stored at different RH were found. The greatest number and content of volatiles was found in powders stored at RH~0 and 11%. At theses RHs and also at RH 54% a further oxidation of hexanal to hexanoic acid was noticed. In these powders a decrease in hexanal and an increase of hexanoic acid was observed after 25 weeks. The lowest amount of volatiles was detected in powders stored at RH 75%. An unexpectedly low content of volatiles was found in the dried emulsion at RH 33%. That might be explained by limited decomposition of hydroperoxides caused by reactions with sodium caseinate and water induced changes in the structure of the maltodextrin-sodium caseinate matrix. In conclusion the significant effect of RH on the stability of encapsulated sunflower oil could be assigned to physical changes of the matrix and chemical reaction with matrix proteins.

Keywords: Spray-dried emulsion, lipid oxidation, relative humidity, SPME

B-3
APPLICATION OF MATHEMATICAL METHODS FOR DEVELOPING NEW FOODSTUFF

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Foodstuff should have certain physical, chemical and organoleptic characteristics which will present to the consumer these products in the best look. Characteristics of foodstuff are connected with various chemical reactions which occur between initial components of a compounding in a preparation time of finished goods. Interaction of components of foodstuff among themselves, and also their influence on such most widespread characteristics of foodstuff, as acidity, pH, viscosity, hardness, a mass fraction of soluble solids, a mass fraction of fat and so is known further. The ratio of prescription components plays defining role on values of these characteristics. For definition of necessary ratios of components it is possible to use mathematical methods which allow to work out the mathematical equations on everyone, from defining for this product, to characteristics. All equations can be reduced in uniform system. Values of characteristics demanded for this foodstuff should be criterion function of the decision of the specified system of the equations. Thus, absolutely diverse indicators of foodstuff mathematically communicate. Such approach provides the producer with the closest compounding of finished goods, to that compounding which satisfies the consumer. In system of the equations it is also possible to add and ratios which regulate process parameters of production and, therefore, to connect them mathematically with prescription characteristics of foodstuff.

Keywords: Foodstuff, components, mathematics, system

B-4
PREDICTIVE DETERIORATION MODEL OF TOFU DURING STORAGE PERIODS

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Tofu has been consumed by Asians and has attracted interest in other parts of world because of the biological effects. However tofu is perishable and easily influenced by storage conditions. The aim of this study was to evaluate the effect of storage temperature on preservation of tofu and quantify concentration of microorganism and organic acids during storage period. For the analysis of microorganism and organic acids, two types of tofu were used; packed tofu and unpacked tofu. Each type of tofu was divided into three groups according to temperature and storage period. The first was stored in a refrigerator at 4°C and another was stored in a thermo-hygrostat controlled temperature at 10°C for 15 days. The other was stored in an incubator at 25°C for 7 days. Measured microorganism was calculated after incubation at 37°C for 48 h. Using high performance liquid chromatography, total seven kinds of organic acids were determinated; phytic acid, oxalic acid, citric acid, quinic acid, lactic acid, formic acid and acetic acid. The concentration of microorganism in packed tofu stored at 4°C was increased up to log 7.31 CFU/g after 15 days. The concentration of microorganism in packed tofu stored 10°C and 25°C were increased to log 8.91 CFU/g and log 8.69 CFU/g after 12 days and 2 days respectively. In contrast with packed tofu, unpacked tofu was contaminated with microorganism (log 3.76 CFU/g) from the beginning storage. The concentration of microorganism in unpacked tofu stored 4°C was reached maximum value of log 8.33 CFU/g after 9 days. The concentration of microorganism in unpacked tofu stored 10°C and 25°C were increased to log 8.95 CFU/g and 9.57 CFU/g after 6 days and 4 days respectively. After reaching maximum concentration, these values remain constant for the rest of the storage period. Quinic acid, lactic acid and acetic acid from packed tofu were not detected during initial storage period, while these from unpacked tofu were detected up to 5044 mg/L, 67 mg/L and 71 mg/L respectively. Oxalic acid and citric acid from both packed and unpacked tofu were decreased during storage days. Lactic acid from packed tofu was not detected, whereas the concentration of that from unpacked tofu was 67 mg/L during initial storage period. the highest concentration of lactic acid was observed as 20 mg/L, 422 mg/L and 331 mg/L from packed tofu stored at 4, 10 and 25°C after 15, 9 and 3 days respectively. After reaching maximum concentration, these values were decreased. Among measured organic acids, it was considered that only lactic acid has correlation with the concentration of microorganism. The concentration of microorganism was proportional to the concentration of lactic acid. The correlation coefficient between concentration of lactic acid and microorganism was from 0.7838 to 0.997. These results show that lactic acid is product can indicate freshness and quality of tofu.

Keywords: Tofu, organic acid, predictive deterioration model

B-5
ANALYSIS OF ATP DEGRADATION PRODUCTS AS FRESHNESS INDICATORS IN FLAT FISH DURING STORAGE

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Fish freshness was evaluated by visual observation of fish, physical and sensory characteristics and a variety of way on the complementary relationship. The K value is a quality index for fish quality assessment based on nucleotide changes. When generating ATP – degradation products from deterioration of fish, according to concentration of xanthine oxidase which is turn hypoxanthine into xanthine, optical freshness indicator was confirmed by color development with Nitroblue Tetrazolium (NBT). Therefore the aim of this study is to determine the freshness of fish by correlation of K value, amount of total aerobic bacteria and concentration of xanthine oxidase. Flatfish was obtained from local retail shop in Korea. Flatfish was stored at 4°C before analysis. ATP degradation products during storage such as hypoxanthine, ATP, ADP, AMP, IMP and inosine were analyzed using by High performance liquid chromatography. Deterioration of fish was determined by K value which is defined as the ratio of the sum of inosine and hypoxanthine to the sum of the ATP degradation products expressed as a percentage. Color development was carried out by reacting xanthine oxidase with Nitroblue Tetrazolium (NBT) using enzyme – linked immunosorbent assay (ELISA) measured at 575nm. The concentration of microorganism in flat fish stored at 4°C was increased up to log 7.20 CFU/g after 15 days from log 4.03 CFU/g at 0 day. The range for K value of flatfish stored at 4°C gradually increased from 11.42±0.06% to 31±0.09% at 0 day and 14 days respectively. The main changes arised in IMP and hypoxanthine, whereas ATP, ADP, AMP and inosine roughly constant in very low concentration during the 14 day storage period. Especially, the concentration of IMP was inversely proportional to hypoxanthine. Xanthine oxidase variation was detected based on xanthine oxidase standard curve which was analyzed by Nitroblue Tetrazolium (NBT). The result of absorbance for NBT solution at stored 4°C was 0.3027±0.0023 to 0.3237±0.0106 at 0 day and 14 days respectively. It was shown that the color change as an freshness indicator. The color of blank NBT solution was yellow but during storage days, the color changed to purple. This color change is the result of xanthine oxidase concentration. The brightness of purple is proportional to concentration of xanthine oxidase. The K value was proportional to the concentration of microorganism and color change of NBT solution. The correlation coefficient between concentration of microorganism and K value was 0.9309 and between K value and color change of NBT solution was 0.8097. On the basis of these results, K value and color change of NBT solution can be a freshness indicator.

Keywords: K value, xanthine oxidase, NBT solution

B-6

PREPARATION AND CHARACTERIZATION OF ORGANIC MICRO- AND NANOPARTICLES WITH ENCAPSULATED CAFFEINE

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Caffeine is a bitter xanthine alkaloid that acts as a stimulant drug. Caffeine is found in varying quantities in the seeds, leaves, and fruit of some plants, where it acts as a natural pesticide that paralyzes and kills certain insects feeding on the plants. It is most commonly consumed by humans in infusions extracted from the seed of the coffee plant and the leaves of the tea bush, as well as from various foods and drinks containing products derived from the kola nut. Other sources include yerba maté, guarana berries etc.. In humans, caffeine acts as a central nervous system stimulant, temporarily warding off drowsiness and restoring alertness. It is the world's most widely consumed psychoactive drug, but, unlike many other psychoactive substances, it is both legal and unregulated in nearly all parts of the world. Beverages containing caffeine, such as coffee, tea, soft drinks, and energy drinks, enjoy great popularity. Caffeine is toxic at sufficiently high doses, but ordinary consumption poses few known health risks, even when carried on for years — there may be a modest protective effect against some diseases, including certain types of cancer. Some people experience sleep disruption if they consume caffeine, especially during the evening hours, but others show little disturbance and the effect of caffeine on sleep is highly variable. There are some tendencies to regulated release of caffeine from beverages into human organism. One of this ways is encapsulation of caffeine into edible organic micro- and nanoparticles with controlled release. This study is focused on possibilities of encapsulation caffeine in micro- and nanoparticles. Five different methods were used for preparation of organic particles with encapsulated caffeine. Caffeine was packaged into liposomes (ethanol injection, thin layer evaporation) and polysaccharide particles (chitosan/alginate or chitosan). Encapsulation's effectiveness was determined by HPLC/PDA. Characterization of size, distribution and stability of particles was done by dynamic light scattering and electron microscopy. Analytical centrifugation was used to measurement of sedimentation velocity and stability of the prepared particles. The particles were exposed to the artificial stomach and intestinal juices and bile acids. Particle stability and amount of released caffeine in model physiological conditions was monitored. Caffeine containing particles were added in several soft drinks to determine particles amount when turbidity occurred. It can be concluded that mainly liposomes prepared by thin layer evaporation and chitosan particles could be a suitable alternative for addition of caffeine into stimulation beverages. Encapsulated form of caffeine enables controlled release of caffeine in digestive system.

Keywords: Caffeine, encapsulation, dynamic light scattering, analytical centrifugation

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B-7

NUTRITIONAL VALUE OF PEA LEAVES: THE NEW BABY LEAF VEGETABLE IN READY-TO-EAT SALADS

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Peas (*Pisum sativum*) are among the most consumed vegetables worldwide, with a registered global production of 15 million tonnes in 2010 (FAO, 2012). It is normally consumed as a seed food, and is known as being a good source of proteins, vitamins and minerals. The consumption of leaves of the pea plants is not as common as eating the peas. However, pea leaves have been recently introduced in the market as a new baby leaf, also known as pea shoots. They are harvested in a very early maturation stage (within 2 to 4 weeks), when the leaves and tendrils are tender, crispy and have an intense pea flavour. This is a new green vegetable that can be cultivated during all year. The producers defend that is a very versatile leaf that can be eaten raw in salads, or cooked with others ingredients. Pea leaves packed solely or in mixtures with other green leafy vegetables are convenient products. These packages of ready-to-eat vegetables are an option for those who want to include fresh vegetables in the diet, but have little time to dispend in their meal preparation. Green leafy vegetables are an essential item in a balanced diet due to their richness in vitamins, minerals and others antioxidant phytochemicals, and also low content of fat and carbohydrates. The nutritional composition of green peas is extensively studied and available in nutritional tables. For the pea leaves, besides some nutritional allegations off being rich in vitamin C and A, made mostly by producers, the scientific data available is scarce. Due to this lack of information, the objective of this work was to determine the nutritional profile of pea baby leaves. Moisture (934.01, AOAC), protein (920.54 AOAC), fat (920.39 AOAC), ashes (942.05 AOAC), total dietary fiber (985.29 AOAC) and carbohydrates (by difference) contents of pea leaves were determined by official methods recommended by Association of Official Analytical Chemists. Several free forms of water-soluble vitamins (C, B1, B2, B3, B5, B6 and B9) and fat- soluble vitamins (A and E) were assessed by HPLC-MS/MS and HPLC-DAD methods. Mineral composition (P, K, Na, Ca, Mg, Fe, Zn, Mn and Cu) was evaluated by a high-resolution continuum source atomic absorption spectrometry method after microwave digestion of the samples. The pea leaves showed a high water content (91.5%) and low fat content (0.3%). Protein, ashes, total dietary fiber and carbohydrates content was 3.3%, 0.8%, 2.1% and 1.9%, respectively. The vitamin analyses revealed a higher content of vitamin C and A (154.0±2.8 mg/100 and 14.4±0.1 mg/100), than the others vitamins studied. In their mineral composition, this leaves showed some distinctive results in their micromineral composition, especially in Zn (0.48±0.01 mg/100g) and Cu (0.13±0.01 mg/100g) content, showing higher values than other green leafy vegetables. In an overall evaluation, this new baby-leaf appears to be a good choice to the consumer, adding nutritional value and diversity of flavours to their salads.

Keywords: Pea leaf, nutritional composition, ready-to-eat salads, vitamins and minerals

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B-8

IMPACT OF FREEZING ON NUTRITIONAL AND CHEMICAL COMPOSITION OF SOME LESS KNOWN SELECTED FRESH FISHES IN IRAN

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The study was designed to investigate the effect of duration of frozen storage on the proximate profile of fishes lizadussmieri, sparidae, sciaenide and platycephalidae. The fishes were subjected to sixty days of frozen storage. Protein decreases with increasing duration of frozen storage; with the fresh samples (not frozen) having the highest protein content (13.02±0.09%). while the least (10.13±0.06) was recorded for fish samples that were frozen for sixty days. Similar results were obtained for the fat content where the highest fat content (0.25±0.20%) was recorded for the fresh samples and the least value was recorded for those stored for sixty days. Ash content and moisture content also decreased during storage. The most susceptible fish to protein loss during frozen storage was fish sparidae (13.02%-12.74%) respectively. Frozen storage lead to a loss of nutrient quality in Iranian fishes during the processing.

Keywords: Frozen period, nutritive quality, fishes lizadussmieri, sparidae, sciaenide and platycephalidae.

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B-9

A PREPARATION OF STABLE POWDER OF HEALTH BENEFICIAL MARINE XANTHOPHYLL FUCOXANTHIN WITH POLYSACCHARIDE FUCOIDAN

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Brown sea alga *Laminaria japonica* includes two kinds of health beneficial ingredients, xanthophyll fucoxanthin and polysulfated polysaccharide fucoidan. In human, dietary fucoxanthin was incorporated into blood after deacetylation to fucoxanthinol in the intestinal epithelium and shows the pharmaceutical parameters of 4 h for T_{max}, 44.2 nmol/L for C_{max}, 7.0 h for $t_{1/2}$, and 664 nmol/L · h for AUC(∞), and then mostly excreted through urine until 25 h after the dose. No side effects of fucoxanthin were detected when such a large amount of fucoxanthin 70 mg/kg body weight/day that stained mouse organs to fucoxanthin color orange was dosed for 3 months. Fucoxanthin significantly prolonged life span of mice fed on a strong carcinogen benzepyrene-containing diet, and further suppressed colon, skin, and spontaneous hepatic cancers. In colon cancer cell line WiDr, fucoxanthinol induced a cell cycle arrest at G_0/G_1 phase through stimulating expression of p21 protein which inhibited phosphorylation of Rb protein and then suppressed a release of transcription factor E2F to promote the cell cycle. In leukemia cell HL-60, fucoxanthinol facilitated DISC formation of FADD with Fas and elevated the activity of caspase 8, and resulted in inducing apoptosis. Also, fucoxanthin has been reported to possess an anti-obesity activity and anti-osteoporosis activity. However, the health beneficial fucoxanthin is very unstable and easily oxidized to degraded products during the storage. Another ingredient fucoidan was not digested and not incorporated into body, and was excreted in feces 32 h after the dose when determined with the monoclonal antibody. The dietary fucoidan exhibited anti-thrombosis activity through acting on intestinal epithelial cells and stimulating expression of mRNAs of NOX1 and DUOX2, and then reduced the de-aggregation time after platelet was occluded in human. Then, fucoxanthin extracted with alcohol from *L. japonica* was wrapped with the extract residue fucoidan after removing undesirab le factors such as salts and arsenic. The resulted fine powder was stable to oxidation, and 90% of fucoxanthin remained unchanged in the powder after one-year storage at 25°C. The powder was able to be used for processing to various types of food.

Keywords: Fucoxanthin, fucoidan, stable powder, anti-tomorigenesis, anti-thrombosis

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B-10
DETERMINATION OF FLAVONOIDS IN DIVERSE *CAPSICUM ANNUUM* L.

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Capsicum annuum L. is popular fruit which includes carotenoids, vitamin C, hydroxycinnamates, capsaicinoids, and flavonoids. The flavonoids are phenolic compounds that are secondary metabolites, antioxidants. However, not much analysis has been paid for flavonoids in *Capsicum annuum* L. The aim of this study carry out separation and structure determination of flavonoids in diverse varieties of *Capsicum annuum* L. Samples were extracted 70% MeOH and clean up employed SPE method on hydrophobic C_{18} functionalized silica gel. The structure of flavonoids were identified into each samples using UPLC–ESI–MS/MS. We compared with each mass fragments and characterized the derivatized C- or O-glycoside on an aglycon.

Keywords: Flavonoid, *Capsicum annuum* L, polyphenol

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Compounds associated with nutritional and sensory quality of foods

C-1
DEVELOPMENT OF A METHOD FOR THE ANALYSIS OF THE VOLATILE COMPOUNDS OF ALMOND BEVERAGES

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Almond (*Prunus dulcis*) beverages are considered a good alternative to cows milk due to its promising human health benefits. The development of milk substitutes has become a necessary practice due to a section of the population that is lactose intolerant or is allergic to the proteins in milk. Almond beverages are often processed by ultra-high temperature processing (UHT) for its preservation. The technological treatments can generate volatiles that may influence the flavor of almond beverages. This may affect consumer acceptability of the product. Headspace solid-phase microextraction gas chromatography-mass spectrometry (HS–SPME GC-MS) was used to analyze the volatile profile of almond beverages. The experimental parameters of fiber coating (PDMS/DVB, CAR/PDMS, PA, and DVB/CAR/PDMS), stirring rate (60–1,600 rpm), extraction time (15–90 min.), sample volume (2 mL or 4 mL), sample temperature (40°C or 60°C), and ionic strength were studied (salt or no salt). A 2mL almond beverage aliquot was selected in the presence of salt at saturation levels, heating of the sample was carried out at 60°C for 60 minutes with a stirring rate of 700 rpm using a DVB/CAR/PDMS fiber. Over one hundred compounds were tentatively identified. Most volatiles belong to the aldehyde, ketone, and alcohol functional groups. Furans and pyrazines were detected in lower numbers. Six commercial UHT-treated almond beverages were analyzed from the market, along with pilot plant processed product. A comparison of the volatile profile of six commercial almond beverages was conducted. Few differences were found between the samples.

Keywords: Headspace–solid phase microextraction, gas chromatogtaphy–mass spectrometry, volatile compounds, almond beverage

C-2

γ-GLUTAMYLTRANSPEPTIDASE-CATALYZED CHEMOENZYMATIC SYNTHESIS OF NATURALLY OCCURRING FLAVOUR ENHANCERS

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The use of flavour enhancers in the food industry could be beneficial for several reasons: they ensure homogeneity of the final products, reduce costs for condiments and favor consumer's acceptance. On the other hand, the consumer's attention for convenient, minimally-processed, nutritious, healthy, yet tasty food prompts the food industry to an accurate choice of the ingredients. In this scenario, naturally occurring kokumi substances could play an important role. Kokumi is a Japanese term that refers to mouthfulness, thickness and long-lasting savory sensations. Kokumi substances are represented mainly by γ-glutamyl derivatives of amino acids. They are nearly tasteless for themselves, but they elicit a strong taste sensation, expecially in conjunction with protein-rich food [2]. In vegetables of the genus Allium, kokumi substances were identified in γ-glutamyl derivatives of *S*-alkyl and *S*-alkenyl cysteines and their *S*-oxides [3]. There is a number of difficulties connected with the supplying of these materials. Isolation from natural sources is laborious, and their content in vegetables varies with cultivation and storage. In addition, upon crushing the plant, they are enzymatically degraded. The chemical synthesis is not economical, due to the need of protection/deprotection steps. We exploited recently the enzymatic synthesis at the laboratory scale of the γ-glutamyl derivatives of *S*-allyl cysteine, S-methyl cysteine and methionine, catalyzed by a commercially available mammalian GGT [4]. In this communication we present the use of a purified, home-made bacterial GGT from a GRAS organism suited to food processing, for the enzymatic synthesis of flavour enhancers with kokumi properties found in garlic and other plants of the genus Allium.

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Keywords: γ-glutamyltranspeptidase, S-substituted cysteines, flavour enhancers, enzymatic synthesis

C-3
FACTORS INFLUENCING CHANGES OF FLAVOR OF FRUIT FLAVORED SOFT DRINK

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The different type of sensory defects can be found in the non-carbonated soft drinks. Identifying the causes of the sensory defect is often problematic; defect, among others, may be caused by microbial contamination. A current problem in the manufacture of soft drinks is contamination with bacteria of genus *Asaia* sp. (bacteria of the acetic acid fermentation). These bacteria are able to grow at low pH values (pH<3), which corresponds to the pH of finished soft drinks. But unfortunately, the threshold perception of specific substances responsible for non-standard (defective) smell is in some cases lower than the detection limit of the analytical method. The aim of study is to evaluate the effect of model beverages contaminated by bacteria *Asaia* sp. on sensory, physical and chemical properties of the fruit flavored soft drink during storage (e.g. formation of sensory active metabolites, degradation of components of drink, pH changes, formation of turbidity, etc.).

Keywords: Soft drinks, fruit flavor, Asaia sp., SPME, sensory defects

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C-4

SYNTHESIS OF THE MENTHOL BY CYCLISATION OF THE CITRONELLAL CATALYZED BY THE TRICHLOROBISMUTH (III)

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The menthol is widely obtained by means of culture of the plant " Mentha arvenis " in China today. However, it is produced by synthesis in Germany, in United States, and in Japan [1]. It finds its use in numerous consumer goods as cigarettes, chewing gum, toothpaste, pharmaceutical products1 etc. Several processes of synthesis of the menthol were used [1]. We cyclise the citronellal isopulegol by thermal method, microwaves, by using reactives such as the active carbon, the silica gel, the sulphuric acid, the boric acid with or without aluminium oxyde, the acetic anhydride, Nickel of Raney etc. For us, we used the triflate of bismuth [2] in the syntheses of this molecule. The isolated products are characterized by the various spectral techniques. A study of optimization of the yield is realized.

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Keywords: Citronellal, trichloroBismuth (III), menthol

C-5

ANALYSIS OF VOLATILE COMPOUNDS OF SEED OILS FROM DIFFERENT POPPY VARIETIES (*PAPAVER SOMNIFERUM* L.) FOR FOOD PURPOSES

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The Slovak Republic has a unique position in development of new varieties of poppy plant which are dominant at planting areas of the Czech Republic, the worldwide largest poppy grower for use this commodity in food industry. Nowadays, the objective is to select a new material with improved seed quality parameters compared to varieties currently used in cultivation practice and submit it to corporate or national seed testing. Research is focused on the collection and evaluation of local land varieties appropriate for food use, as well as on a comprehensive analysis of poppy cultivars breeding and growing in Slovakia. Apart from evaluation of seeds in terms of content of total lipids with emphasis on unsaturated fatty acids, content of fibre, micro and macro elements, heavy metals, secondary metabolites other than morphinane type of alkaloids and their biological activity, considerable attention is devoted to study of volatile aroma-active compounds of poppy seed oils which constitute their overall flavour. The methods of GC/MS and GC/FID were used for study of volatiles in poppy seed oils of 8 different varieties introduced in the Document of registered varieties in SR for the year 2011. Plant breeding and production of poppy seeds for research purposes were provided by Research and Breeding Station in Malý Sariš of Plant Production Research Center in Piešťany. The aim of this work was study of qualitative and quantitative differences in profiles of volatiles distinguishing both individual poppy varieties one from other, and blue-seed varieties (Maraton, Opal, Gerlach, Lazur, Malsar, Major and Bergam) from the white-seed variety Albin. Volatiles of poppy seed oils were trapped by SPME method and analysed by GC using columns of different polarities. On the grounds of obtained results from analysis of blue-seed varieties is evident that variety Gerlach showed the weakest qualitative and quantitative profiles of volatiles. On the other hand the variety Malsar is the richest in qualitative aspect, as well as quantitative evaluation of volatile compounds. Majority volatiles in view of average relative contents in declining order are: 1-hexanal, 1-hexanol, 1-pentanal, 1-pentanol, hexanoic acid, 2-pentyl furan, *E*-2-octenal. Concerning the assessment of the white-seed variety Albin, it excels in both qualitative and quantitative evaluation of analysed samples poppy varieties. The qualitative profile comprises more than 40 volatiles, which are jointly responsible for the overall flavour of poppy seeds, strongly reminding the walnut aroma in this case. Dominant volatile constituents of this variety from quantitative point of view are: 1-hexanal, 2-pentyl furan, *E*-2-octenal, hexanoic acid, 1-pentanal, *E*-2-heptenal, 1-pentanol, 1-octen-3-ol, 1-hexanol. We expect that the original results will increase competitiveness of Slovak commodities and interest in poppy plants cultivation, which will have a positive impact on the economy of planting subjects.

Keywords: Poppy seeds, GC, volatile organic compounds, flavour

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C-6
QUALITATIVE COMPARISON OF YOGURTS WITH SUGAR AND STEVIOL GLYCOSIDES

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Nowadays, many consumers are searching for the products with the lowest calorific value, reduced or eliminated sugar content. These encourages scientists to do research into the use of natural sweeteners as substitutes for sugar and synthetic sweeteners. These can be glycosides which are extracted from the leaves of *Stevia rebaudiana*. The study was undertaken to compare the physicochemical, microbiological and organoleptic quality of yogurt with sugar and *Stevia rebaudiana* extract. Yogurts were produced from skimmed milk with 12% of solid content, using yogurt culture, with the addition of sugar or a preparation containing 95% steviol glycosides in its composition. The research was conducted in two stages. The first stage involved production of natural yogurts and the ones with 5% of sugar content or Stevia preparation at the following concentrations: 0.05, 0.1 and 0.15 g/L. As a result, it was found that the most beneficial properties could be traced in the samples containing 0.1g/L steviol glycosides. At the second stage of the study, yogurts with 2% sugar and 0.1 g/L Stevia extract were produced. After 7 and 21 days of storage at 6±1°C the following properties of yogurt were determined: composition, pH value, syneresis, apparent viscosity, number of yogurt bacteria, texture analysis and organoleptic assessment. On the basis of the results obtained in the first stage, it can be stated that Stevia preparation did not affect the process of fermentation or differentiation of pH values, either in natural yogurt products (4.46) or in those with sweetening additives (4.44–4.50). The results obtained in stage II, indicate that syneresis of yogurt curd with sugar and Stevia was similar (23.98% and 24.06% respectively). The syneresis process was intensifying during storage. The apparent viscosity of yogurts with steviosides, at the shear rate of 12.5 s^{-1}, after 7 days of storage, was higher than that of yogurts with sugar by 0.35 Pas. After 21 days, the difference was much smaller, dropping down to 0.1 Pas. Steviol glycosides did not limit the growth or survival of yogurt bacteria. On the last day of storage, the number of lactobacilli was 19 milion cfu and the number of streptococci was 240 milion cfu. The yogurts with Stevia glycosides were characterised by the higher curd consistency values and viscosity index than samples with sugar, namely by 13.18% and 6.7%, respectively. The firmness and cohesiveness of curd was similar. The same correlations were found after 21 days of storage. The difference in organoleptic quality of yogurt with Stevia and that with sugar concerned mainly the sensing of sweet taste, which was more intense in the samples with steviol glycosides. The results of the study demonstrate that the characteristic features of yogurts, with the evaluated sweetening additives, were similar and that *Stevia rebaudiana* glycosides can be used as substitute for sugar in the production of sweetened yogurts.

Keywords: Yogurt, steviol glycoside, organoleptic properties

C-7
NEW METHOD TO DETECT ESSENTIAL OILS RELEASE LOADED IN ORDERED MESOPOROUS SILICA MATERIALS

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Specific method to detect thymol in the presence of specific enzyme and the development of “in vitro” validated controlled release systems based on biocompatible mesoporous silica materials loaded with thymol and capped with saccharides acting as molecular gates in order to evaluate the stability of the materials in the food has been created. Thymol is rapidly evaporated losing its antioxidant activity, for this reason it is interesting to create systems to protect thymol. The mesoporous silica materials loaded with thymol and capped with saccharides are new systems able to protect the thymol evaporation and allow the thymol release in a controlled manner. This material inhibits the thymol release due to the creation of a saccharide network blocking the thymol inside the mesoporous. The presence of pancreatin hydrolyzes the saccharides and allows the thymol release. To detect thymol release Folin-Ciocalteu reagent method has been applied. This reagent determines the content of total phenolics and shows high absorbance at 760 nm when the materials are put in contact with enzyme solution. On the other hand, the solid without enzyme solution doesn't show any absorbance. It has been demonstrated that the attachment of a hydrolyzed starch derivative as a gatekeeper on the surface of mesoporous silica materials provides a suitable method for the design of mesoporous systems able to deliver the entrapped guest in the presence of suitable enzymes. Whereas the capped solids showed “zero release”, the same solids in water in the presence of pancreatin released the cargo in a controlled fashion due to the enzyme-induced hydrolysis of the glycosidic bonds in the anchored saccharides. This system seems to have a potential application to protect essential oils which could be for example applied to certain fungi and used as antifungal agent.

Keywords: Thymol, mesoporous silica materials, controlled release, pancreatin

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C-8
MAIZE BREAD "BROA": ARE TRADITIONAL PORTUGUESE MAIZE VARIETIES RESPONSIBLE FOR DIFFERENT FLAVORS?

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Maize (*Zea mays*) arrived in Portugal in the sixteenth century from America and underwent a natural and human selection process [1]. Most Portuguese traditional varieties were selected with the final purpose of bread production [2]. The traditional maize bread, "broa", is mostly consumed in the northern and central regions of Portugal. Traditionally, maize flour (50 to 80%) is prepared with hot water, rye and wheat flours, yeast and leavened dough from a late broa. Then, the dough is baked at 250°C during arround 40 min [2]. This traditional bread making process produces a type of bread very appreciated by its sensory characteristics [2,3]. Several chemical reactions are involved in the fermentation and baking 'broa' process. The formation of 'broa' flavour depends of crumb and crust formation. Bread crust browning always been associated with Maillard reaction [4] between proteins and carbohydrates.The most important chemical groups responsible for wheat bread aroma include alcohols, aldehydes, esters, ketones, acids, pyrazines, pyrrolines, furans, hydrocarbons and lactones [6]. In what concerns maize, the few studies about aroma volatile compounds were related with freshly cooked sweet corn, canned corn, popcorn, corn tortilla and taco shell [5,7,8]. To our knowledge there is a lack of information about volatile compounds in Traditional Portuguese varieties of maize and respective maize bread. Within SOLIBAM "Strategies for Organic and Low-input Integrated Breeding and Management" project, funded by 7th Framework Programme (FP7) we want to valorize Portuguese broa produced with traditional maize varieties, through characterization of quality parameters which may increase the acceptability of the product by consumers. With this objective, we studied the volatile compounds responsible for traditional Portuguese maize bread aroma, through analysis of 12 different varieties of Portuguese maize flour and the corresponding maize bread, by Solid Phase MicroExtraction (SPME) and Gas Chromatography-Mass Spectrometry (GC–MS). The maize bread made with commercial hybrid maize flour was used as a reference to establish the comparison with the traditional ones.

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Keywords: Maize, bread, flavor, characterization, SPME-GC-MS

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C-9

EVALUATION OF THE EFFECT OF ORGANIC PRODUCTION SYSTEM ON THE VOLATILE ORGANIC COMPOUNDS PROFILE OF EWE'S CHEESE MILK

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The volatile organic compound (VOC) profile of cheese is affected by the milk composition, which is strongly related to the rearing system and ewe's diet. Modifications of the fatty acid profile may exert changes in lipid-derived volatile compounds arising from lipid oxidation. Previous works have shown that the fatty acid composition of ewe's milk varies owing to the production system, and milk from sheep raised under organic production system show higher proportion of polyunsaturated fatty acids than those under conventional production (Revilla et al., 2009) because organic farming regulations restrict the use of concentrates in the ewe's diet. Taking into account these previous results the aim of this work was to study the effect of organic production system on the volatile organic compound profile of ewe's cheese and to follow its evolution during the maturation process. Bulk tank ewe's milks from flocks of two local breeds (Castellana and Churra) and two production systems (organic and conventional) from the same geographical area were used to manufacture the cheeses in accordance with the Regulatory Board of the D.O. Queso Zamorano. Samples were taken after 3 and 9 months of maturation. Homogeneizated and grinded samples were heated at 40ºC and purged with helium. Volatile compounds were concentrated in a Tenax/Charcoal trap and then desorbed using helium flux. The volatile compounds were separated and detected in a gas chromatograph coupled with a mass detector operating in the scan mode. Peak identification was by comparison of retention times with authentic standards and quantification was carried out by the sum of the abundance of all the ions (TIC), with reference to the cyclohexanone which was added as the internal standard. The number of volatile compounds found and identified by GC–MS was different for each production system and organic cheeses had a higher number of volatiles and the 1-butanol only was detected in organic cheeses. Organic cheeses showed higher levels of almost of the alcohols, except ethanol, at both maturation points. Alcohols confer a sweet odour or fruity sensation but ethanol gives a dry or dusty aroma. Conventional cheeses showed higher quantities of ketones that are related with hay, butter or sweet odour. However organic cheeses showed higher values of 2-butanone that is characterized by a fruity aroma. Finally, organic cheeses showed higher levels of acetic and butyric acid but the difference was statistically significant only at the 3rd month of maturation.

Keywords: Odour profile, organic, ewe, cheese, maturation

C-10
FATTY ACID PROFILE OF MILK FAT IN THE LOCAL DAIRY PRODUCTS FROM NORTHEASTERN POLAND

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Nowadays, consumers are searching for the products that offer them some unique healthful and sensory properties, standing out from the products available on the market. Among such products one can count those made by the local producers of raw materials coming from the region. Quality control, and related to this evaluation of the authenticity of the products manufactured by the regional producers, in this case, is particularly important. An important factor in determining the quality of local dairy products such as butter, cheese, milk is fatty acid profile. It is characteristic for the local dairy production to use the raw materials from a variety of animals including sheep, goat. Nutritionists point out the benefits of dairy products based on goat and sheep's milk due to the beneficial fatty acid profile and valuable content of macro-and micronutrients. The aim of this study was to determine the fatty acid profile of cheese and tvorog milk fat obtained from the local producers from the region of northeastern Poland. The material consisted of cheese (n=14) and tvorog samples (n=11), made from cow, goat and sheep's milk. Methyl esters of fatty acids in milk fat were prepared with the use of IDF Standard 1999. Separation and quantitative determination of fatty acids was performed by the gas chromatography method using HP 6890 gas chromatograph with a flame ionization detector (FID). Differences were found in the percentage share of each fatty acid group depending on the type of milk from which the cheese was produced. The milk fat of cheese made from cow's milk had a higher content of monounsaturated and polyunsaturated fatty acids as compared with the milk fat of cheese produced from other types of milk. The percentage share of mono- and polyunsaturated fatty acids of cheese made from cow's milk ranged respectively: 67.8, 25.1 and 3.2%. Tvorogs, made from goat's milk, had higher content of saturated and polyunsaturated 72.07% and 2.88% fatty acid as compared to cow's milk products. It has been demonstrated that in the fat of tvorog made from cow's milk is about 23.81% of monounsaturated fatty acids.

Keywords: Cheeses, fatty acids, local production

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C-11

CONCENTRATING n-3 PUFA FROM CRUDE AND REFINED COMMERCIAL SALMON OIL: EFFECT OF DIFFERENT REACTION FACTORS

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Marine species have attracted considerable attention as a source of high amounts of important nutritional components to the human diet. Thus, fish oils are a readily available source of long-chain PUFA, especially those of the n-3 series, mainly *cis*-5,8,11,14,17-eicosapentaenoic acid (EPA) and *cis*-4,7,10,13,16,19-docosahexaenoic acid (DHA). Several methods have been reported for concentrating PUFA in marine oils, with varied yields. Among them, urea complexation has been applied extensively, as allowing handling of large quantities of materials in simple equipments and being a relatively inexpensive method. In the present research, urea complexation was used to concentrate n-3 PUFA from crude (raw) and refined commercial salmon oils. The experimental procedure included salmon oil saponification, free fatty acid (FFA) collection, formation of urea-FFA inclusion complexes, and extraction of free n-3 PUFA. Fatty acid composition was analysed after conversion into fatty acid methyl esters (FAME) and further analysis by gas-liquid chromatography. Different factors such as reaction time, temperature reaction, urea-FFA ratio and stirring extent were taken into account. Characterisation of starting crude and refined salmon oils was carried out by assessment of FFA, conjugated dienes and trienes, peroxide value, anisidine value, moisture and impurities, unsaponifiable matter, iodine value and colour (L*, a*, b*) values. As a result, differences between crude and refined salmon oil could be observed. Thus, crude oil provided higher typical odour, viscosity and suspension particle values. Concerning chemical analyses, crude salmon oil showed a higher FFA content and iodine value. Related to physical colour assessment, refined salmon oil showed lower a* (redness loss) and b* (yellowness loss) values when compared to its counterpart crude oil. Related to n-3 PUFA concentration, a high yield of n-3 PUFA recovering could be observed in all cases, which confirmed salmon oil to be a profitable source of such highly valuable constituents. Factors such as temperature of reaction and urea-FFA ratio showed to be markedly significant in order to achieve a higher value concentration.

Keywords: Salmon oil, crude and refined, n-3 PUFA, urea complexation, reaction factors

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C-12

FORMATION OF LACTONES BY *LACTOCOCCUS LACTIS* SUBSP. *LACTIS* VAR. DIACETYLACTIS DURING FERMENTATION IN CREAM

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Lactones are potent flavour compounds contributing to creamy, fruity and coconut-like notes in milk products. Although the chemical formation of lactones in milk products from hydroxy-fatty acid triglycerides is accepted [1,2], their production by lactic acid bacteria (LAB) and the metabolic pathways are up to date uncertain [3,4]. The aim of our study was to investigate the microbial formation of lactones in cream with and without addition of hydroxy-fatty acids (HFA), supposed precursors of lactones [4]. The strain of *Lc. lactis* subsp. lactis var. diacetylactis FAM18027 was selected from 65 strains of different LAB species, for its ability to develop buttery and fruity aroma notes in cream after fermentation. Full fat cream was fermented by FAM18027 at 30°C for 24 h, with and without HFA addition. Furthermore, the cream was incubated without strain addition in the same conditions in presence or absence of HFA. The formation of lactones was evaluated by head-space solid phase microextraction-gas chromatography-mass spectrometry (HS–SPME–GC–MS). δ-Octalactone and δ-decalactone were the main volatile lactones found in the samples. GC–MS analyses revealed an increase of the signals of these two lactones after 4 h of fermentation already. In cream samples fermented with addition of LAB for 24 h, the δ-octalactone and δ-decalactone signals were about five times higher than in samples incubated without LAB over the same period of time. Addition of HFA seemed to slow down the fermentation, which resulted in a slower acidification and less important lactone formation. As a conclusion, LAB strains such as FAM18027 may be used to increase the flavour of fermented cream and sour cream butter.

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Keywords: Lactones, hydroxy fatty acids, lactic acid bacteria, cream, GC-MS

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C-13
HYDROPEROXIDES IN UNSAPONIFIABLE MATTER – A KEY IN ANALYSIS AND REACTION RATE DETERMINATION

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Oxidation of minor components present in the unsaponifiable matter is similar to autooxidation reactions of free or ester-bound fatty acids. Its rate and the progress are strongly influenced by the lipid matrix composition. Sterols, tocopherols, tocotrienols and pigments are isolated in unsaponifiable matter after alkaline saponification. Under the alkaline conditions hydroperoxides are quickly transformed into stable sterol oxidation products with oxo, hydroxy or epoxy group. Stabilization of intermediates or selective transformation of hydroperoxides is necessary for their accurate determination or for the kinetic studies. It was developed method for the determination of hydroperoxides in unsaponifiable matter that involves reducing of hydroperoxides by organophosphorus compounds (i.e. triphenyl phosphine, triethyl phosphite) and transformation of present oxo-derivatives into appropriate oximes. Without the stabilization of target compounds will be their determination incorrect. GC–MS and GC–FID was used for the qualification and quantification of primary and secondary oxidation products of unsaponifiables. The next aim of the study was trapping of radicals and their stabilization with special probes. Pure primary and secondary sterol oxidation products were synthesized and characterized by NMR, MS, IR, UV and elementary analysis.

Keywords: Steryl hydroperoxide, GC–MS, triphenyl phosphine, unssaponifiable matter

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C-14
ROASTING EFFECT ON CHEMICAL COMPOSITION OF SOUR CHERRY KERNEL OIL

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Sour cherry seed arises as a waste material during processing of the fruits into juices. Owing to its high oil content up to 20%, it can be utilized to produce sour cherry seed kernel oil. Roasting the seeds prior to oil extraction not only increases oil efficiency but also affects physicochemical properties of oil. This study aimed to investigate the effect of various roasting conditions on oil composition. The seeds were roasted in a temperature-controlled oven at 160°C for 0 (control), 10, 20, 30, and 40 min. Oil extraction was performed by hexane. After removing hexane phase by evaporation, the resulting oil samples were analyzed for fatty acid composition, tocopherols, antioxidant capacity, total phenolic compounds (TPC) and hydroxymethylfurfural (HMF). In general, the results revealed that the oil from sour cherry seed is a rich source of bioactive compounds, namely tocopherols (up to 428.62 mg/L in toto) and polyunsaturated fatty acids (up to 45%). Roasting was found to cause an increase of HMF and TPC whereas total tocopherol content decreased as roasting times increased. In addition to that the roasting at different times did not indicate any significant differences in the fatty acids compositions of extracted oils. Overall the results suggest that a mild roasting treatment of sour cherry seed is useful to increase the concentrations of certain bioactive compounds in oil.

Keywords: Sour cherry kernel oil, roasting, tocopherol, fatty acid composition

C-15

DART-HRMS TECHNIQUE FOR COMPARING THE QUALITY OF VACUUM AND CONVENTIONAL FRIED OIL

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Deep fat frying is a very common and popular process to prepare food with a desirable flavor, color and crispy texture. However, the quality of oil during frying is affected by many chemical reactions like oxidation, polymerization and hydrolysis. As lipids oxidize desirable and undesirable (volatile and nonvolatile) compounds are formed and change the flavor stability and quality, color and texture and nutritional quality of food. While some of these compounds improve the flavor other can affect its nutritional value and induce adverse health effects upon ingestion in the human gastrointestinal tract. Considering the need to minimize thermal oxidation and polymerization, vacuum deep-fat frying process represents a conceivable option, as it enables frying at lower temperature and moreover improves the fried product safety and quality. Direct analysis in real time (DART) hyphenated to the (ultra)high resolution mass spectrometry (HRMS) was used to monitor oxidation dynamics in oil during vacuum and conventional frying. DART–HRMS enabled rapid fingerprinting of triacylglycerols (TAGs), sterols, fatty acids and their oxidation products. To visualize the differences between vacuum and convenional heated oil samples, multivariate statistical analysis (principal component analysis, PCA) was used. TAGs polymers were determined by an independent technique (HP–SEC–RID) applying and internal normalization method

Keywords: Oxidation, oil, vacuum frying, conventional frying, direct analysis in real time, mass spectrometry

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C-16

EFFECT OF LACTIC ACID BACTERIA FERMENTATION ON LIPIDIC AND PHENOLIC PROFILES OF TOFU

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Soy foods have long been a staple of the human diet in Asia and tofu may be the most popular food made of soy worldwide. Tofu is prepared from soymilk curd and it can also be further processed (grilled, frozen, or fermented) modifying taste, texture and end-uses compared to traditional tofu. Fermentation also prevent undesired microbial and chemical reactions improving the stability and the quality of the product. Moreover, in addition to be natural, nutritious and safe, epidemiological studies have shown that fermented soybean products exhibit healthy effects. In fact, soybean and its products contains biologically active compounds including isoflavones, essential fatty acids, sterols, phospholipids and tocopherols which may contribute to prevent chronic diseases such as hormone-dependent cancers, cardiovascular diseases and osteoporosis. Due to the influence of processing on the bioactive compounds content of soy food products, soybean, traditional tofu and tofu obtained by soymilk fermentation were studied by GC–FID and HPLC coupled with fluorimetric, light scattering and electrospray ionization mass spectrometry detector, in order to analyse their lipidic profile (fatty acids, tocopherols and sterols) and isoflavones content and composition. In particular, for isoflavones analysis, a new method was set up. Fermentation was performed using specific strains of lactic acid bacteria (LAB), such as *Lactobacillus casei* and *Lactobacillus acidophilus*. Fatty acids profile of soybean and tofu samples were very similar, even in term of trans fatty acids and omega-3 fatty acids, so fermentation cannot differentiate tofu from soybean by fatty acid composition. The most abundant fatty acid was linoleic acid, followed by oleic acid, palmitic acid and α-linolenic acid, as reported by other authors. The processing conditions did not significantly affect neither tocopherol composition of tofu (traditional and fermented) compared to soybean and in all products the main tocopherol was γ-tocopherol followed by δ-tocopherol and α-tocopherol. As concerning phytosterol content, ß-sitosterol was the main sterol found with the phytosterol composition that was similar in soybean and traditional and fermented tofu. The main effects of fermentation were on isoflavone profile: in fact, significantly differences were seen in fermented tofu isoflavones content compared to soybean and to traditional tofu. In particular, fifteen isoflavones were identified as aglycones and glycosides: the aglycone forms were significantly higher in fermented tofu compared to the traditional one. These results suggest that fermentation by LAB cannot discriminate the fatty acid, tocopherol and phytosterol composition of fermented tofu from the traditional one and from soybean, but it plays an essential role not only on shelf-life extend, but also in the increase of the content of aglycone isoflavones with important biologically implications on human health.

Keywords: Tofu, fermentation, isoflavones, lipids, soybean

C-17
OXYSTEROLS IN FOOD – METHODOLOGY

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Cholesterol is widely distributed in foods of animal origin, and is susceptible to oxidation to form cholesterol oxidation products (COPs) during storage and culinary treatment in the presence of oxygen. Numerous studies have shown that COPs may possess biological effects such as atherogenicity, cytotoxicity, mutagenicity, carcinogenicity, cell membrane damage, pro-inflammatory effects and inhibition of cholesterol biosynthesis. Normally, content of cholesterol is determined by GC/FID or GC/MS in fat sample after saponification and isolation of unsaponificable matter. The problem with this method can be possible oxidation of cholesterol to COPs or decomposition of some COPs during hot saponification. Therefore, the method for separation of cholesterol and COPs from fat matrix on activated silica gel column was adapted. Content and chemical composition of cholesterol and COPs were determined by GC–MS (quadrupole) after derivatization by bis-(trimethylsilyl)-trifluoroacetamide in pyridine. Basic performance characteristics of this method (specificity, limit of quantification, precision-repeatability and accuracy-recovery) were determined for cholesterol. Pork lard performed to oxidation at various conditions and various ranges was used as the basic analytical material. Typical products of cholesterol autoxidation, 7β-hydroxycholesterol and 7-oxocholesterol were identified in various concentrations. Besides that, another product of non-enzymatic oxidation, 25-hydroxycholesterol was identified in significant concentration. The method could be used for the monitoring of COPs occurrence during various types of culinary treated meat products.

Keywords: Cholesterol, oxysterols, fat, GC–MS

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C-18
INFLUENCE OF SAMPLE PROCESSING ON VALUES OF LIPOXYGENASE ACTIVITY IN BARLEY GRAIN

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Lipoxygenases (LOXs) are members of a large enzyme family that catalyze oxygenation of free polyunsaturated fatty acids into diverse hydroperoxide compounds, collectively called oxylipins. Lipoxygenases (LOX, linoleate:oxygen oxidoreductase, EC 1.13.11.1 2) are non-heme iron-containing dioxygenases that catalyze hydroperoxidation of fatty acids by introduction of molecular oxygen into the cis,cis-1,4-pentadiene structure of its substrate. In plants, the hydroperoxide fatty acids, (mainly derived from linolenic and linoleic acids) are further metabolized into physiologically active lipid-breakdown products. Therefore, LOXs are implicated in different aspects of plant physiology, although their biological functions are not yet fully understood. In barley and malt, LOX activity directly influences final sensory quality of beer. The objective of this work is to characterize influence of sample processing on lipoxygenase activity in spring malting barley grain. 21 different varieties of spring barley were enrolled into this study. Barley varieties were bred to at Agrotest fyto, Ltd. LOX activity was measured spectrophotometrically at 234 nm using linoleic acid as substrate. For activity determination crucial is grain homogenization mode and reaction mixture temperature. Disintegration of barley grains should be performed immediately before analysis. About 3 hours after milling standard deviation of LOX^{-1} activity in dry homogenate increased for several times. During 48 h of grain powder storage LOX^{-1} activity decreased to about 40–50%, after 90 days only traces of activity were found in most of samples. Reaction mixture should be tempered to 20–25°C (laboratory temperature). At 8°C substantially lower activity was found in all samples when compared with 24°C.Detailed description of stability of water-loss dry grain homogenates was not available yet.

Keywords: Lipoxygenase, barley, enzyme activity, sample preparation

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C-19
QUALITY OF REFINED OILS USED IN FRYING FROZEN POTATO STICKS

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Frying is a widespread procedure for preparation of food, because is a rapid, easy and inexpensive method of food preparation. The quality of fried foods depends upon the quality of the frying oil and thus it is of prime importance to maintain and protect the quality of the frying medium. In fact, frying oils, used continuously and repeatedly at high temperatures, are subject to a series of degradation reactions. Therefore, it is necessary to examine some major changes which occur during deep fat frying. This work aimed to study the quality of refined sunflower, grape seed and olive pomace oils, after successive reuses in frying pre-fried frozen potato sticks. The oils used were kindly donated by Parceria de Azeites, S.A., Torres Novas and the pre-fried frozen potato sticks were a gift from Frijobel, S.A., Penela and produced by Farm Frites International B.V., Holanda. The experiments of frying included 3 days of work and in each day were performed 5 frying cycles. In the first cycle of the day, oil was heated for 20 min, the potato sticks were fried at 180°C for 7 min and the oil was cooled until temperature reached 50°C. Oil was reheated for 15 min and used in the next frying cycle. The quality parameters, acidity, peroxide value, fatty acid profile, viscosity and color were analyzed according to AOAC methods and Portuguese Regulations. A rapid test kit commonly used in the restoration context was used for total polar compounds evaluation. The peroxide value, acidity, free fatty acids and viscosity of the oils increased through the frying cycles performed as it was expected due to thermal oxidation, polymerization, and hydrolysis of the oil. The olive pomace oil, in fresh and after subsequent re-use, had a percentage of unsaturated fatty acids significantly lower (83.8%, 80.2%) than those of grape seed (88.6%, 84.7%) and sunflower (88.8%, 84.7%) oils, indicating a greater oxidative stability. In addition, it was observed a change to a darker color of the oils and a loss of their transparency. Also, the results obtained with the test kit indicated an increase in the total polar compounds from 5% in the fresh oils to 17–23% after the frying cycles performed. However, all the oils studied showed total polar compounds values beneath the discard limit for frying oils ruled by the international legislation (24–27%).

Keywords: Frying oils, sunflower oil, grapeseed oil, pomace olive oil, deep fat frying

C-20

STERYL HYDROPEROXIDES – KEY INTERMEDIATES SIMILAR TO LIFESPAN OF MAYFLIES

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Steryl hydroperoxides like adults of mayflies exist only for a very short time. Hydroperoxides – primary oxidation products of sterols as one-day flies undergo metamorphosis, which is accompanied by formation of hydroperoxyl or alkoxyl radicals. These compounds are very reactive and are quickly transformed into stable sterol oxidation products. In the case of cholesterol these products are 7-oxocholesterol, epimeric 7-hydroxycholesterol and 5,6-epoxycholesterol and cholestane-3β,5α,6β-triol. Sterol oxidation is similar to autooxidation reactions of free or ester-bound fatty acids. This reaction is started by homolytic cleavage of a C-H bond in the initiation phase. The bond dissociation enthalpy necessary for the homolytic cleavage of a C-H bond is smaller in case of methylene group in the bis-allylic position of unsaturated fatty acids (326–334 kJ/mol – prof. N.A. Porter, 272 kJ/mol – prof. J. Velisek) in comparison with the position 7 of the B ring of the cholesterol (368 kJ/mol – prof. N.A. Porter). In the presence of excelent H-atom donors (tocopherols, phenolic acid derivatives, carotenoids) is formation of kinetic products prefered. For example formation of unconjugated hydroperoxides in the case of fatty acids or hydroperoxides in the position 5 on the B ring of cholesterol. Hydroperoxides are generally more polar and more mobile, which is closely connected with their behaviour in cytoplasmatic membrane of eukaryotic cells. In our experiments we determined the influence of temperature, lipid matrix of model systems, fatty acid moiety ester-bound to cholesterol, and an air flow on the oxidation rate of cholesterol. Kinetic data were evaluated in programms Origin, Sigma plot, Era and Statistica.

Keywords: Steryl hydroperoxide, GC/MS, triphenyl phosphine, model system

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C-21
CHANGES OF LIPIDS DURING THE FRYING WITH NATURAL ANTIOXIDANTS

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Varieties of chemical reactions take place in lipids during the frying, which decrease their nutritional and hygienic value. The addition of antioxidants can slow down the oxidation rate and natural antioxidants are usually better accepted by consumers than synthetic compounds. Inhibition of pork lard, rapeseed and sunflower oils oxidation during the frying of French fries with *Lamiaceae* herbs as natural antioxidants source was monitored and compared with BHT and propyl gallate. Frying medium was first used once and then repeatedly with each antioxidant. Antioxidant activity was monitored by the Schaal oven test, HPLC analysis of non-volatile oxidation products and triacylglycerol polymers, conjugated dienes measurement and fatty acids profile determination by GC. Sensory analysis of French fries was used to evaluate the effect of repeated frying on their quality. Analytical determination of active substances in herbs was carried out as well. *Salvia officinalis* L. and *Origanum heracleoticum* L. were proved as statistically significant antioxidants comparable with propyl gallate. *Origanum vulgare* L. did not exhibit so high antioxidant activity. The major effect on fatty acids profile of lard was found in oleic acid changes. Rancidity and bitter taste of French fries positively correlated with increasing number of fryings.

Keywords: Natural antioxidants, frying, Lamiaceae, antioxidant activity, sensory analysis

C-22

COMPARATIVE EFFECTS OF ELECTRON BEAM AND GAMMA RADIATION ON THE TRIACYLGLYCEROL PROFILES OF PORTUGUESE CHESTNUTS (*CASTANEA SATIVA* MILL.)

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Chestnut fruits (*Castanea sativa*) global production is continuously growing; Portugal produces about 20 000 ton per year, being 75% of this production concentrated in the Trás-os-Montes region [1]. In order to reach new markets, effective conservation technologies are mandatory, especially since the European Union banned methyl bromide for allegedly being toxic to operators and a severe environment pollutant [2]. Other conservation methods like heat treatment still lack efficiency [3], allowing other treatments like gamma [4] and electron beam irradiation [5] gaining interest as an alternative for food processing. In previous studies of our research group, the effects of gamma and e-beam irradiation doses on the chemical composition and bioactivity of chestnuts were evaluated. Despite the usefulness of those studies, the influence of irradiation on the triacylglycerol composition of chestnut remains undone. Herein, the effects on chestnut triacylglycerol profiles in fresh and stored samples of gamma and e-beam irradiation were studied. The results were classified trough an analysis of variance and a stepwise based linear discrimination analysis as a supervised classification technique, in order to understand the observed changes. Independently of radiation type, samples irradiated with upper doses showed higher modifications in their triacylglycerol profile. These differences classified the assayed samples in different groups trough the applied linear discriminant analysis, allowing identification of irradiated samples.

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Keywords: Chestnut fruits, *Castanea sativa*, gamma and e-beam irradiation, storage effects, linear discriminant analysis

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C-23
FATTY ACIDS OF DIFFERENT SHRIMP SPECIES CAUGHT FROM THE GULF OF ANTALYA, MEDITERRANEAN SEA

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The aim of this study was to investigate fatty acid profiles of different shrimp species caught from deep water and shallow water. The shrimp species investigated in the study were *Aristeus antennatus, Aristeomorpha foliacea, Plesionica martia, Parapenaeus longirostris, Plesionica edwardsi* from deep water and *Metapenaeus monoceros, Penaeus semisulcatus, Penaeus kerathurus, Penaeus japonicus* from shallow water. Fatty acid compositions of shrimps varied. The main fatty acids were C18:1n9, C16:0, C25:6n3, C22:5n3, C18:0. Saturated, monounsaturated and polyunsaturated fatty acid contents of *P. longirostris*, *P. edwardsi* and *M. monoceros* were markedly different, respectively. The ratio of n6/n3 of the edible tissue of *M. monoceros* was found 0.795, whereas this value was 0.152 in *A. foliacea*. Among the species studied, the highest DHA+EPA value was found for P. kerathurus. The levels of PUFAs of shallow water shrimps (ranging from 33.44% to 42.77%) were found higher than those of deep water shrimps (ranging from 29.68% to 33.95%). Marine animals in the upper water layers gain nutrition through phytoplankton which provides n-3 PUFA depending on solar energy. Shallow water shrimp species provide satisfying amount of PUFA.

Keywords: Shrimp, fatty acids, PUFA, trawl net

C-24

EFFECT OF ESSENTIAL OILS ADDITION AT DIFFERENT DOSES TO THE EWE'S DIET ON MILK QUALITY

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A relatively new topic in ruminant nutrition is the study of dietary additives capable of modulating rumen fermentation in order to improve nutrient utilization by rumen microorganisms. Essential oils (EO) are secondary compounds obtained from several parts of plants which have antimicrobial properties that may modify rumen microbial fermentation, without using antibiotics. This has been especially pronounced in products that are marketed as organic, where there is a need to find safe and effective replacements for chemical preservatives and antibiotics. However, the modification of ruminal activity may change the milk characteristics. Few studies have been published on effects of EO or their constituents on milk production and composition of milk. Several studies observed no changes in DM intake, milk production, and milk components of dairy cows. However, an increase of the concentration of conjugated linoleic acid (CLA) when a high concentration of MEO was added to dairy cows has been reported. Taking into account the scarce data about this topic in organic dairy sheep the aim of this work was to study the effect of thyme essential oil (EO) addition at different levels to the organic ewe's diet. To do that two trials were carried out. In the first trial a total of forty animals in organic production system and at the same stage of lactation stage were divided into two equal groups: control and treated groups. The control ewe's diet (a pasture of fresh oats ad libitum) was supplemented (maximum 30% of the ration) with a mixture certified by the "Organic Agriculture Council of Castile Leon" and 70 ml of Morea®. The treated group received 5 mg of thyme EO per day and animal dissolved in the Morea®. Milk samples were taken on 0, 10, 20 and 30 days of treatment. In the second trial the control group was fed with the control diet as previously described and the treated group received 100 mg of thyme EO per day and animal. Milk was analysed for pH, total nitrogen, titratable acidity, total solids, lactose and fat. Fatty acids were determined according with the method. Milk lipids were extracted and the fatty acids were methylated and analysed by gas chromatography according with the method described by Revilla et al. (2009) The results showed an increase in total fat content in the treated group milk samples without changes in the lactose and protein contents or density values. Milk samples from animals with EO-supplemented diet showed slightly lower pH and higher acidity values than control milk samples at the beginning of treatment. There was also an increase in the total polyphenol content sampling but no significant increase of antioxidant activity was observed. Regarding the fatty acid profile a decrease of short-chain fatty acids and an increase in the monounsaturated fatty acids were observed

Keywords: Thymol, antioxidant activity, fatty acids, phenolic composition

C-25
IMPACT OF CULINARY OLIVE OIL TREATMENT ON SOME QUALITATIVE PARAMETERS

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The study has examined effect of some common culinary treatments and shelf of olive oil on variation of chlorophylls, carotenoids and alpha-tocopherol contents. The extra virgin oil samples of Spanish provenance were subjected to microwave heating using different microwave power at continuous and discontinuous profiles, exposure in boiling salt or vinegar aqueous solutions, heating in oven at 130°C, as well as exposure to UV radiation at continuous and discontinuous profiles. Visible and fluorescence spectrometry were used to follow variations of the mentioned components. It was found the changes of individual parameters under continuous heating or UV irradiation profiles were not identical with those under discontinuous exposure at the same summary exposure time. The alpha-tocopherol was more sensible to the UV radiation and heating than the chlorophylls and carotenoids. The vinegar aqueous medium affected each from the followed component more destructively than the salt aqueous solution. Trends of the chlorophyll and carotenoid changes were very similar in contrast to the alpha-tocopherol however, each of the oils showed individual development. Relation of intensity ratio of 443/518 nm peaks in the fluorescence spectrum with profile of the chlorophylls and carotenoids variations in the oil associated with development of fatty acid oxidation products was verified.

Keywords: Olive oil, chlorphylls, carotenoids, alpha-tocopherol

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C-26

EFFECT OF MICROWAVE THAWING ON MICROSTRUCTURE AND PHYSICOCHEMICAL STABILITY OF LOW FAT WHITE SAUCES MADE WITH SOY PROTEIN

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The microstructural and physicochemical stability of white sauces made with soy protein and modified waxy corn starch was evaluated after subjecting them to a freezing-thawing process by conventional or microwave oven. Microstructure of sauces revealed a structured matrix of soy protein and starch polymers where fat globules and swollen starch granules remain dispersed. Both thawing methods affected fat globule size and morphology but did not affect the starch granules. The SDS-PAGE analysis did not show apparent changes between sauces thawed by both methods. Moreover, a similar pattern was found in the soy protein isolate used as a raw material indicating that this type of protein was also stable to the cooking process. There were no significant differences ($P>0.05$) in the reducing power of the sauces regardless the thawing method used. However, the acidity index and the k232 parameter were significantly higher ($P<0.05$) in conventional-thawed sauces. Finally, syneresis was negligible and no significant differences ($P>0.05$) were found among different frozen storage periods. In conclusion, the formulation of the sauce is appropriate to develop low fat, vegetarian meals which can be subjected to frozen storage and microwave reheating.

Keywords: Starch, soy protein isolate, microwave heating, CLSM, SDS-PAGE

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C-27

THE IMPACT OF CONTINUOUS PURIFICATION PROCESS ON THE QUALITY OF FRYING OIL

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Deep-fat frying may lead to formation of both desirable and undesirable flavour compounds through various hydrolytic and oxidation processes. In addition to sensorial changes, also nutritive value / safety of heat treated oils is deteriorated due to formation of polymers and other harmful products. To remove these compounds, thus preserve good quality of fried foods, circulation of oil bath through relevant filters is employed by food industry. In this study, we tested in a close cooperation with filter producer, the efficiency of continuous purification process during hamburgers deep-fat frying. The palm oil in continuous fryer (operated with or without filter) palm was held at the temperature 180°C. 20 oil samples were taken in two hour intervals. In addition to degradation changes in levels of 3-MCPD esters which are processing contaminants typically occurring in palm oil were monitored, too

The highest concentrations of 3-MCPD esters were found in fresh frying fat. The decrease of 3-MCPD levels was observed during frying cycle. The filter had no significant effect on levels of 3-MCPD esters. Moreover, its use significantly increased content of both polymerized triacylglycerols and total polar compounds. It can be due to sorption of antioxidant agents (tocopherols, phospolipides) on filter.

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C-28

UTILIZATION OF DART-ORBITRAP-MS TECHNIQUE FOR STUDY OF OXIDATIVE CHANGES IN FISH AND COD LIVER OIL

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Fish oil and cod liver oil are popular naturally-derived nutritional supplements because of their high content of essential omega-3 fatty acids, represented mainly by polyunsaturated acids like eicosapentaenic acid (EPA) and docosahexaenic acid (DHA). These polyunsaturated fatty acids (PUFAs) cannot be synthesized in human body, and therefore, they need to be consumed as part of a diet, or through respective supplements. However, PUFAs are susceptible to oxidative spoilage, in the presence of air oxygen, radical reaction – autooxidation takes place. In the first step, hydroperoxides are formed, and these are subsequently converted into a broad range of molecules. Hydroperoxides of PUFAs formed by autooxidation are very unstable and break down into a wide variety of volatile sensory active compounds as well as nonvolatile products like oxo-, hydroxy- and epoxy- acids (with both the same or shorter chain). The aim of this study was to monitor potential oxidative changes in fish oil used in food supplements by means of non-target fingerprinting strategy. For this purpose, ambient mass spectrometry technique employing Direct Analysis in Real Time (DART) ion source coupled with High Resolution Mass Spectrometer (HR–MS) with orbitrap mass analyzer was used. This technique provides very rapid measurement; one sample could be measured in several second. Both the polar and nonpolar fractions were measured. Typically the polar fraction of fish oil (extracted by aqueous methanol) contains free fatty acids and, after oxidation, polar oxidation products and the nonpolar fraction consists mainly of triacylglycerols. The effect of both heating and long-term storage of cod liver oil as well as the effect of antioxidant addition was well documented by this technique. Markers of oxidation products were found especially in heated oil, and, at the same time, a relative decrease of PUFAs was observed.

Keywords: Fish oil, cod liver oil, food supplement, non-target fingerprinting, DART–MS

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C-29
N-TERMINAL MAILLARD REACTION OF PEPTIDES UNDER BAKING CONDITIONS

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Heat induced reactions of sugar compounds with side chains of amino acids is a well known reaction resulting in a variety of Maillard Products. These products can be used for the evaluation of process induced protein modification in food [1]. Beside side chain derivatization, the N-terminus of proteins can undergo the same reactions [2]. The focus of the presented study is to analyze the N-terminal derivatization of dipeptides with N-terminal valine, namely Val-Leu, Val-His, Val-Ser, Val-Tyr or Val-Glu, under typical baking conditions. We want to show how the amino acid sequence can effect the reactivity of the N-terminal amino function by analyzing Amadori rearangement products (ARP), 2(1*H*)-pyrazinons and carboxyalkylpeptides [2,3]. For this, 5 µmol glucose and 5 µmol peptide were diluted in 1 ml 0.1 M phosphate buffer. The pH value was adjusted to 5.6 and 0.5 g of cellulose was added. To simulate “crumb”, samples were heatet at 100°C in sealed tubes, and for “crust”, samples after lyophilisation were heatet at 120°C in open tubes for up to 40 min, respectively. For quantification of glycation compounds, the “crust” and “crumb” samples were hydrolyzed with 6 N HCl to convert ARPs to furoylmethylvaline, which was analyzed by RP–HPLC–UV. Glyoxal derived 2(1*H*)-pyrazinons and carboxymethylpeptides were analyzed by liquid chromatography with tandem mass spectrometry directly from solutions of the samples. For the ARP quantified as furoylmethylvaline all peptides showed an equal reactivity. Depending on the reaction time, the maximum values were between 3 and 5 mmol/mol peptide for crust samples and 150 and 250 µmol/mol peptide for crumb samples. Carboxymethylvalylpeptides in the crust samples were found over a wide range of 6 µmol/mol peptide for Val Leu and 286 µmol/mol peptide for Val-Glu. In the crumb samples, similar amounts between 100 (Val-Leu) and 300 (Val-His and Val-Glu) µmol/mol peptide could be estimated. 2(1H)-pyrazinons in crust samples ranged from 160 (Val-His) to 880 (Val-Tyr) µmol/mol peptide. In crumb samples creation of 2(1*H*)-pyrazinons were found exclusively for Val His (6 µmol/mol peptide) and Val-Ser (65 µmol/mol peptide). Comparing the sum of glyoxal derived N-terminal AGEs (carboxymethylvalylpeptides and 2(1*H*)-pyrazinons), all peptides showed higher reactivity under crust conditions (higher temperature, lower water activity). Significant differences in the creation of 2(1*H*)-pyrazinons comparing crust and crumb conditions were observed. The reactivity of the N-terminal amino function of peptides for the formation of advanced glycation endproducts (AGE) was remarkably influenced by the amino acid sequence.

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Keywords: 2(1*H*)-pyrazinon, carboxymethylvaline, N-terminal advanced glycation endproducts, Amadori rearangement products, peptide

C-30
RAPID DETERMINATION OF 5-HYDROXYMETHYLFURFURAL BY DART

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The DART (direct analysis in real time) is novel technique with wide potential of a fast and screening analysis. The determination of 5-hydroxymethylfurfural (5-HMF) as a typical temperature marker of food by DART was optimised. The validation parameters of analytical method were determined. The quantification of 5-HMF was provided by stable isotope-labeled 5-HMF standard prepared from glucose. The formation of 5-HMF from saccharides, as a potential source of overestimation of results, was evaluated. The 44 real samples (honey, caramel) and the 50 model samples of heated honey were measured. The possibilities of detection of heated samples of honey by DART were confirmed. The HPLC and DART/TOF–MS method of determination of 5-HMF was compared. The correlation equation between HPLC and DART was: DART=1.028 HPLC+0.2134, R^2=0.9557. The DART/TOF–MS method seems to be efficient and fast tool for determination of 5-HMF in various food matrices – caramel, honey etc.

Keywords: DART/TOF–MS, 5-HMF, HPLC, honey, caramel

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C-31

AQUEOUS POMEGRANATE SEED EXTRACT PROTECTS HUMAN HEPATOMA HEPG2 AGAINST OXIDATIVE STRESS INDUCED BY tert-BUTYL HYDROPEROXIDE

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The search for Advanced glycation end-products (AGEs) formation inhibitors has recently received much attention and certain by-products of plant extracts have been evaluated for their antiglycation potential. AGEs are a group of complex and heterogeneous products formed in the advanced stage of the Maillard reaction in foods and glycation in living bodies. AGEs are involved in the development of several health disorders such as diabetes and its complications [1], atherosclerosis [2], Alzheimer's disease and normal aging [3]. Recently, our group has identified a significant antiglycative activity in aqueous extracts of pomegranate seeds (*Punica granatum*). The aim of this study is to investigate the protective effect of aqueous pomegranate seed extract (PSE) on toxicity and oxidative stress induced by *tert*-butyl hydroperoxide (t-BOOH) in HepG2 cell lines before to be used as ingredient in foods and pharmacological preparations. Human hepatoma HepG2 cells initially isolated from a liver biopsy in a 15-year old Caucasian male. This cell line was grown in a humidified incubator containing 5% CO_2 and 95% air at 37°C. They were grown in DMEM F-12 medium, supplemented with 2.5% Biowhitaker fetal bovine serum [4]. Cell viability was determined by using the Crystal Violet assay. Cytotoxicity was determined by the lactate dehydrogenase (LDH) leakage from cells. Cellular reactive oxygen species (ROS) were quantified by the dichlorofluorescein (DCFH) assay [4]. Pretreatment of HepG2 in the range of 0.01 to 0.1 mg/mL of PSE completely prevented LDH leakage from the cells. Reactive oxygen species generation induced by t-BOOH was significantly reduced when cells were pretreated for 20 h with 0.001 to 0.1 mg/mL PSE. It is concluded that treatment of HepG2 cells in culture with the aqueous extracts of pomegranate seeds strongly protects the cells against an oxidative insult, increasing cell viability as well.

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Keywords: Pomegranate, glycation, reactive oxygen species, oxidative stress, cell viability

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C-32

ON THE ROLE OF AMADORI-REARRANGEMENT PRODUCTS IN THE GENERATION OF STRECKER ALDEHYDES AND STRECKER AMINES DURING THERMAL FOOD PROCESSING

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Strecker aldehydes, formed by an oxidative transamination/decarboxylation reaction of alpha amino acids with alpha dicarbonyl compounds are well-known contributors to the overall aroma of processed foods. Recently, it has also been shown that via the same reaction pathway also the respective Strecker amines can be formed from the parent amino acids [1]. In addition, it has earlier been observed that Strecker aldehydes are not only formed by degradation of the respective amino acid, but also from the respective Amadori-Rearrangement Products (ARPs) [2]. However, up to now reliable quantitative correlations between the amounts of precursors present in the raw material and the yields of Strecker aldehydes and amines formed after processing are lacking. Therefore, the aim of this study was to quantitate Strecker aldehydes and Strecker amines [3] as well as the respective amino acids and ARPs in several foods before and after a thermal treatment. For this purpose, in particular a new method for the quantitation of ARPs was developed. Methylpropanal, 2-methylbutanal, 3-methylbutanal, 3-(methylthio)propionaldehyde, phenylacetaldehyde and *p*-hydroxyphenylacetaldehyde as well as the respective amines were quantitated by means of Stable Isotope Dilution Assays (SIDA) in unroasted and roasted cocoa; green and roasted coffee; barley malt, wheat malt, wheat beer; pepper and pepper powder as well as in tomatoes and tomato powder. The results will be discussed with special emphasis on the influence of the processing conditions on precursor degradation and formation of the volatile reaction products.

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Keywords: Amadori-Rearrangement Product (ARP), Strecker aldehyde, Strecker amine

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DEVELOPMENT OF A METHOD FOR THE QUANTITATION OF AMADORI COMPOUNDS IN FOODS USING A STABLE ISOTOPE DILUTION ASSAY AND LC-MS/MS ANALYSIS

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Amadori rearrangement products (ARPs) are relatively stable intermediates formed during the Maillard reaction. They are precursors, inter alia of aroma-relevant Strecker aldehydes. For example, 3-methylbutanal is formed from ARP-leucine. Furthermore, some ARPs show gustatory properties, e.g. the umami-tasting fructosyl-glutamate. For the analysis of ARPs, particularly HPLC with post column derivatisation [1], cation exchange chromatography [2], and gas chromatographic analysis of their oximes [1] have been suggested. However, these methods either include time-consuming pre-separation and derivatisation steps or require special analytical equipment. In most cases, quantitation was achieved by external calibration, thus losses during the workup were not considered. The aim of the work presented here was, therefore, to develop a new, direct, and rapid LC–MS/MS method for the quantitation of ARPs based on a stable isotope dilution approach. The method should be focused on aroma-related ARPs and also be applicable to foods with rather low ARP concentrations, e.g. roasted coffee. As a first step, ARP-valine, ARP-leucine, ARP-isoleucine, ARP-methionine, ARP-phenylalanine, ARP-tyrosine and ARP-histidine were synthesised as reference substances. Quantitative NMR was applied to determine their purity. Then, the isotopically labelled analogues were synthesised from [$^{13}C_6$]-glucose to be used as internal standards. These standards were added to different kinds of food and methanolic extracts were analysed by LC–MS/MS after SPE purification. All considered ARPs and [$^{13}C_6$]-ARPs were separated using their molecular ions, except for the leucine and isoleucine derivatives. ARP-leucine and ARP-isoleucine were at first determined as sum and then their ratio was calculated from the intensities of selected daughter ions. The method showed an excellent peak to noise ratio with low matrix interferences and a good reproducibility even for food low in ARPs. Thus, LC–MS/MS analysis in combination with a stable isotope dilution assay is suggested as a general approach for the fast and accurate quantitation of aroma-relevant Amadori compounds in foods.

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Keywords: Amadori, SIDA, LC–MS/MS, quantitation, food

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C-34

CYTOTOXIC AND OXIDATIVE EFFECTS OF NUTRITIONAL RELEVANT MELANOIDINS IN H4IIE HEPATOMA CELLS

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Melanoidins are polymeric brown compounds formed in the last stage of the Maillard reaction, which are present in some widely consumed foods such as coffee, bread, cocoa and beer. Apart from their contribution to visible features certain biological effects such as antimicrobial and antihypertensive activities were shown [1]. While a number of in vitro and cell culture studies show antioxidative activities of early and late maillard products, our very recent data indicate prooxidative properties of melanoidins on isolated and cellular DNA particularly after metal ion binding [2]. The aim of the present work were to study pro- and antioxidative effects of melanoidins generated from D-Glu/L-Ala heated mixtures with and without redox-active metal ions in H4IIE hepatoma cells. According to our results, the pure melanoidins did not influence cell viability. Even though TEAC-assay revealed antioxidative properties, no protection of cultured H4IIE cells from exposure to H_2O_2 was observed. Furthermore, incubation with melanoidins charged with metal ions (Cu^{2+}, Fe^{2+}) even induced the formation of reactive oxygen species analyzed by DCF assay. Consequently, metabolic activity and colony forming ability of the cells were reduced. These findings suggest that nutritional relevant melanoidins posses both antioxidative as well as prooxidative properties particularly after complexation of redox-active metals. Further investigations are needed to clarify mechanisms involved in generation of ROS and subsequent cellular response pathways.

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Keywords: Maillard, melanoidin, ROS, cytotoxicity, hepatoma cells

C-35
METABOLIC TRANSIT OF DIETARY METHYLGLYOXAL

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Recently, methylglyoxal (MGO) was identified as the compound being responsible for the pronounced antibacterial activity of manuka honey [1], which has been linked to several medicinal properties, in particular wound healing. The methylglyoxal content of manuka honeys varies from 40 and 760 mg/kg and correlates with its antibacterial activity [1,2]. The observation that methylglyoxal showed cytotoxic properties in in vitro experiments [3] raises the questions concerning possible health risks resulting from methylgloxyal ingested with the daily diet. At present, virtually no information about the metabolic transit and bioavailabilty of dietary methylglyoxal is available. In the present study, "metabolic transit" is defined as the urinary excretion of the compound and its metabolites following uptake via foods, because this implies absorption from the intestine. Possible metabolic pathways of MGO during digestion are enzymatic detoxification to less reactive compounds like D-lactate and formation of advanced glycation endproducts (AGEs) with alimentary or physiological amino acids/proteins [4,5]. Urinary excretion of MGO following consumption of manuka honey with defined content of the dicarbonyl compound was investigated. Determination of MGO in urine was performed with GC–MS after derivatization with *O*-(2,3,4,5,6-pentafluorobenzyl)-hydroxylamine hydrochloride (limit of detection: 10 nM) and D-lactate was quantified enzymatically (limit of detection: 5 µM). The metabolic transit of MGO was studied within a 4 day dietary recall with 4 healthy volunteers who collected their 24 h urine. Following a raw food diet virtually free from MGO and other glycation compounds, a defined amount MGO (0.5 mmol, in manuka honey) was administered in the morning of day 2. On day 3 and 4, the raw food diet was continued. Renal excretion was between 0.1 and 0.4 µmol/day for MGO and between 35 and 280 µmol/day for D-lactate. No influence on excretion of both compounds was observed following administration of MGO. To investigate the stability of methylglyoxal under physiological conditions, a simulated in vitro gastrointestinal digestion was performed with MGO-containing honey [6]. After eight-hour in vitro digestion, only 5–20% of the initial methylglyoxal was recovered, but no formation of D- and L-lactate could be observed. These findings indicate that dietary MGO is rapidly degraded during the digestive process, is not absorbed from the intestine and, therefore, exerts no influence on the MGO level in vivo.

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Keywords: Methylglyoxal, D-lactat, metabolic transit, in vitro gastrointestinal digestion, manuka honey

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C-36

MODIFICATION OF THE ε-AMINO GROUP OF LYSINE BY *trans*-2-HEPTENAL - A PRODUCT OF LIPID PEROXIDATION OF HEATED PEANUT OIL

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Peanuts are a common food and serve as ingredients for many foods, being a rich source of protein (25%) and fat (48%) [1]. About 85% of the fatty acids are unsaturated [2]. In Europe and the US, peanuts are mostly consumed roasted. During the roasting process, several chemical reactions occur, among which glycation and lipid peroxidation are of particular importance. Recently, we have shown that up to 30–50% of lysine is modified as a result of peanut roasting [3]. Pyrraline proved to be the most important glycation compound, reaching concentrations up to 38.5 mmol/mol lysine. However, only about one tenth of the totally observed lysine modification could be explained by the formation of known Maillard reaction products. We assumed that reactions between proteins and carbonyl compounds originating from lipid peroxidation play an important role for lysine derivatization in peanuts. Following heating of peanut oil in laboratory scale (20 min at 170°C), volatile carbonyl compounds as potential precursors for lysine modification were analyzed in the headspace by GC–MS (EI). The most important compounds were hexanal, 2-heptenal and nonanal. After incubation of *N*-α-acetyl-L-lysine as a model for peptide-bound lysine with trans-2-heptenal at 75°C for 4 h, two main UV-active compounds were detected after acid hydrolysis by LC–ESI–MS/MS, showing characteristic fragmentation patterns of pyridinium derivatives [4]. Thus trans-2-heptenal is one of the most important secondary products of lipid peroxidation of peanut oil and able to modify the ε-amino group of lysine. Hence we conclude that lipid peroxidation leads to the formation of reactive compounds which are not only important as aromatics and flavourings, but which are able to modify amino acids, contributing to functional changes and a decrease in the nutritional value of proteins.

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Keywords: Lipid peroxidation, trans-2-heptenal, lysine derivatization, pyridinium derivative

C-37

EFFECTS OF CONSUMPTION OF MAILLARD REACTION PRODUCTS ON DIGESTIBILITY OF DAMAGED AND UNDAMAGED NITROGEN IN YOUNG AND ADULT RATS

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Introduction: Maillard reaction (MR) may cause degradation of nutritional protein quality by the destruction of essential amino acids or by a reduction in their availability. The deterioration of the protein digestibility do not seem to be only caused by the participation of amino groups in the MR, but the presence of Maillard reaction products (MRP) could also affect the digestibility of unaltered proteins. The purpose of this study was to investigate the effects of the consumption of MRP from glucose-lysine model system heated 150°C – 90 min on digestibility and net protein utilization (NPU) in young and adult rats.

Material and methods: Equimolar mixture of glucose-lysine-HCl (GL) (40% moisture) were heated in open recipients in an oven at 150°C for 90 min to obtain the GL90 model system. The GL90 sample was added to the AIN-93G diet (Control diet) to reach a final concentration of 3%. Thirty weanling Wistar rats were randomly distributed into three groups (10 animals per group) and one of them was sacrificed by anaesthesia overdose at day 0 to analyse their initial nitrogen body content. The remaining groups were assigned to one of the dietary treatments. The intake was monitored during the whole experimental period. To investigate the nitrogen balance in different stages of growth, in the 3 and the 12-week faeces and urines were collected as 1-week pool. At the end of the experimental period, animals were sacrificed and a global nitrogen balance was carried out by analysing the carcasses. Nitrogen in diets, faeces and urine as well as in carcass was determined by using a Kjeldahl procedure.

Main results: Despite the nitrogen intake was similar in both groups, the faecal excretion markedly increased in GL90 group in the third-week balance (20.46 vs. 41.49 for Control and GL90, respectively) and in the last week balance (22.29 vs. 34.64 for Control and GL90, respectively). Although the absorption was unmodified, the digestibility was lower in rats fed GL90 diet compared with controls in both week-balances. The NPU was also unaffected after consuming GL90 diet in both periods, in the 3 and 12-week balances. In the same line, the retention and the NPU in the whole assay did not vary after consumption of diet containing model MRP from GL90.

Conclusion: Intake of assayed MRP decreases protein digestibility due to a higher nitrogen faecal excretion. In young rats it is clearly stated that the major faecal nitrogen compared with the Control group exceeds the total amount of ingested nitrogen coming from MRP, demonstrating that browning products compromise the digestibility of undamaged nitrogen. The same action is suspected in adult rats, but the result is quantitatively less clear, so that in this case the effect of MRP intake on unaltered nitrogen was moderate.

Keywords: Maillard reaction products, nitrogen, digestibility, net protein utilization

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STUDIES ON THE FORMATION AND SENSORY ACTIVITY OF MAILLARD-DERIVED GUANOSINE 5'-MONOPHOSPHATE DERIVATIVES

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Since decades the Maillard reaction, or non-enzymatic browning, is known to be a major contributor to the development of colour, aroma and taste of thermally treated foods. While numerous studies on the heat induced transformation of precursor molecules to desirable aroma compounds were published, knowledge on the Maillard induced transformation or de novo synthesis of taste active and/or modulating molecules remain fragmentary. Recently, the analysis of a commercially available yeast extract combined with targeted Maillard model reactions revealed the heat induced interaction of guanosine 5'-monophosphate (5'-GMP) with reducing carbohydrates to result in a series of N-glycated 5'-GMP reaction products [1,2]. Further investigations implied short chained, reactive sugar degradation species to be capable to crosslink the nucleotide backbone with additional amino compounds. All obtained products obtained mainly from the reaction of aliphatic amines with dihydroxyaceton and 5'-GMP were, after purification, investigated for their umami taste enhancing activities by means of human psychophysical experiments as well as in vitro taste receptor studies [3]. These investigations clearly demonstrate the umami enhancing activity of glycated 5'-GMP derivatives not only being strongly dependent on the chemical structure of the substituent in N2 position, but also on the stereochemistry of the glycation moiety.

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Keywords: Maillard reaction, 5'-GMP, umami, taste receptors

C-39

CHANGES IN REDUCING ACTIVITY OF REDUCTONES AND PHENOLIC ACIDS BY MEANS OF REACTIONS WITH MAILLARD CARBONYL INTERMEDIATES

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The Maillard reaction (MR), transformation of reducing saccharides in the presence of amino compounds, belongs to the most important chemical reactions in food during processing and storage. MR results in brown pigments, volatile compounds, flavoring matter, etc., including compounds with highly reductive properties which can contribute to the stabilization of food against oxidative deterioration. Reducing power arising in Maillard reaction systems is attributed mainly to reductones and compounds with sufficiently acidic methylene moiety. In addition to oxidation of these reducing compounds, they may be transformed also by non-oxidative processes. There are several kinds of reactive carbonyl intermediates within the Maillard reaction that have a potential to form addition products with the simple reductones and methylene-active compounds. However, to date a little information is known about the transformations and particularly the possible carryover of reducing capability to the consecutive Maillard products. Participation of naturally occurring phenolic reductants in the reactions with the reactive carbonyls is also expected as a part of non-enzymatic browning reaction. In this way, phenolic compounds can significantly suppress the formation of substances which are responsible for flavor generation in foods and can reduce the negative effects of reactive carbonyl Maillard intermediates *in vivo* (carbonyl stress). This work was focused on the evaluation of reducing activity during reactions of principal low molecular non-nitrogen Maillard reducing intermediates (norfuraneol and 2,3-dihydro-3,5-dihydroxy-6-methyl-(4*H*)-pyran-4-one) in binary mixtures with reactive carbonyl Maillard intermediates (furan-2-carbaldehyde, 5-hydroxymethylfuran-2-carbaldehyde and pyrrol-2-carbaldehyde), products of lipid oxidation and derivatives of cinnamic acid (ferulic acid, *p*-coumaric acid, caffeic acid, chlorogenic acid and sinapic acid). The formation of active products in binary mixtures of the phenolic acids with reactive α-dicarbonyl and α-hydroxycarbonyl intermediates of the MR and lipid oxidation was also investigated. HPLC with amperometric detection and redox potential measurements were used for the assessment of reducing power. Some electrochemically active products were identified. Optimum conditions were found for the best yield of the target products in binary mixtures. Investigation of the effect of various parameters such as pH and water activity was carried out.

Keywords: Reductones, α-dicarbonyls, hydroxycinnamic acids, reducing power, Maillard reaction

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C-40

HPLC DETERMINATION OF SUGARS IN READY TO EAT FRUIT SALAD EXPOSED TO LOW-DOSE IONIZING RADIATION

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The demand for minimal processing food becomes increasingly popularly. As consumer lifestyle and trends move towards convenience and quality foods seem to have led to an increased desire for ready-to-eat. Processed by radiation, fruit salad provides the consumer a safety, quality and practicality product. Ionizing radiation used in low doses is effective to maintain the quality of commercially processed food, reducing the microbiological load without compromising the nutritional and sensory values. The purpose of this study is the determination of sugars in fruit based salads exposed low-dose ionizing radiation: 0.5 kGy, 1.0 kGy, 1.5 kGy and 2.0 kGy using high-performance liquid chromatography (HPLC) methods to identify and quantify these compounds.

Keywords: Fruit salad, food processed, HPLC, ionizing radiation

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EVALUATION OF PEACH PALM IN NATURA MINIMALLY PROCESSED BY IONIZING RADIATION

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The peach palm is from Amazon Region native and the economic potential of the species lies in the fruit and palm heart. The peach palm has characteristics as sweet favor, quality, precocity, rusticity and tillering. The food irradiation is effective in reducing microorganisms, is considered a versatile and effective treatment for preservation and is known as a good method protect against pathogens agents in food. Its use combined with minimal processing could increase the quality, self life and safety of minimally processed vegetables. The aim of this study was to evaluate the physical characteristics as color and texture of the peach palm in natura minimally processed, subjected to ionizing radiation of Electron beam accelerator and multipurpose 60Co irradiator.

Keywords: Peach palm, food irradiation

C-42

"FROM ANCIENT CROPS MATERIALS AND PRODUCTS FOR THE FUTURE" (VELICA PROJECT)

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The VeLiCa project is aimed at making again gainful the growing of flax and hemp in Regione Lombardia (Northern Italy) where it used to be widespread at the beginning of the 20th century. This target is being pursued by the exploitation of all the parts of the plant to make different products with high-added value according to the "biorefinery" concept [1]. Within this frame, our group is currently investigating both oil and protein content from hempseed (*Cannabis sativa* L.) because of their rising interest in food industry. To this aim, we are developing "green processes" based on enzyme-catalyzed reactions and environmentally friendly solvents. On one hand, hemp oil is a well-known source of linoleic acid (LA) and alpha-linolenic acid (ALA), which are omega-6 and omega-3 essential fatty acids (FFAs), respectively. Due to the health benefits of these FFAs [2], there is a great demand for pure and concentrated extracts of those compounds. We have thus studied the enrichment of hemp oil with LA and ALA by using selective lipase-catalyzed hydrolysis. The two classical hydrolytic biphasic systems consisting of either oil/water or oil/isooctane/buffer [3] were compared with a new homogeneous one based on oil and tert-butanol/water. The new system is easier handling and appeared superior to literature protocols. The fatty acid composition of the hydrolyzed hempseed oil was determined by an analytical method based on SPE of the produced FFAs and GC analysis of the corresponding fatty acid methyl esters (FAMEs). When Pseudomonas cepacia lipase was used as the biocatalyst, LA content reached 68% after 24 h. On the other hand, protein content of hempseed, which is comparable to soy's one, has been reported to be far more digestible and nutritional in all essential amino acids [4]. Chemical and/or enzymatic hydrolysis of vegetable raw materials rich in proteins can produce hydrolyzed vegetable proteins (HVPs) containing bioactive peptides with nutraceutical properties [5]. Defatted hempseed cake was thus hydrolyzed by using two food grade protease cocktails, Flavourzyme and Umamizyme. Hempseed protein hydrolyzates were subjected to ultrafiltration and the antioxidant activity of the fractions <10 kDa was measured by the DPPH radical-scavenging assay and the ferrozine test. A few samples displayed a remarkable Fe^{2+} chelating ability and were found to exert radical scavenging properties comparable to those of glutathione. Not less important, hempseed cake, that is a usually discarded waste, is here upgraded as a valuable source of peptides for human nutrition. These results suggest that enzymatic hydrolysis of hempseed oil and proteins can be used to produce high added-value products and to set up new productive chains.

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Keywords: Hemp, essential fatty acids, hydrolyzed vegetable proteins, enzyme-catalyzed reactions, biorefinery

C-43

INVESTIGATION ON THE HEAT STABILITY AND THE pH-DRIVEN CHANGES IN THE PROTEIN PROFILE OF AN ITALIAN WHITE WINE

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In the recent years, there is a great interest concerning the relation between wine pH and the formation of turbidity in unfined white wines. Actually, a haze in bottled wine can reduce or invalidate its commercial value, and winemakers typically perform fining treatments to avoid wine turbidity. In the present study, the protein profile of the Erbaluce wine, one typical Italian white wine, was analyzed in relation to the haze-forming tendency and pH variation. A pH range from 3.00–3.60 was obtained experimentally and the corresponding heat stability was analyzed. The results indicated increased heat stability at lower pH, which was indicated by a lack of haze formation even above 70°C. The increase in pH was accompanied by a progressive shift in the haze formation from 50–60°C to 70–80°C. The proteins extracted from the wine samples at different pHs were separated by SDS-PAGE and identified by mass spectrometry analyses. Proteins derived from both yeast and grape tissues were present in the wine extracts. The statistical analysis of band intensities revealed that the solubility of certain yeast and grape proteins was modified by the pH shift. Interestingly, most of the proteins were glycosylated inducing the hypothesis that the sugar chain may have an influence on the differential solubility of wine proteins in response to pH. This finding could encourage researchers to consider wine to be a more complex model fluid with respect to those currently taken into account in wine heat stability studies.

Keywords: Wine proteins, glycoproteins, haze formation; heat stability, wine pH

C-44

THE STUDY OF BEER QUALITY PRODUCED USING MALT OF KOSOVA, AND COMPARISON OF RESULTS WITH BEER PRODUCED FROM MALT ORIGINATING IN REGIONAL COUNTRIES

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More comprehensive application as large savings in the industry as well as improving the quality of study quality beer in question is aimed at optimizing the production process based on evaluation of malts produced in regions of Europe, was reflected in improved the properties of beer in the brewery "Birra Peja" – Kosovo. This study has included all of the beer production chain, in close cooperation with the production staff of the laboratory and factory. Produced malts were studied in four European countries and the Ukraine, Croatia, Serbia and Kosovo, and Kosovo malts mixtures with that of Ukraine and Croatia in the proportion 30:70, and beers produced by these malts. Analyzes were carried beer produced by these malts in all stages of production as chemical and microbiological and sensory and the tasting. Production and beer tasting for this study was done in two intervals of two groups for tasting; group "Birra Peja" and the Agricultural University of Tirana. For this study are serving and consulting and laboratory staff working brewery "Union", which tests have served as reference for this work. Chemical and microbiological tests are done based on the methods under the European Beer Convention (EBC) and MEBAK. The analyzes and assessments made for the quality of beer have come to the conclusion that beer malts produced by Croatian origin is that which corresponds to better quality beer which produce even now.

Keywords: Malt, beer, production technology, EBC, MEBAK.

C-45
CHOLESTEROL, A ROBUST MARKER OF EGG CONTENT IN EGG PASTA?

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Pasta made from the common wheat (*Triticum aestivum*) often contains also the eggs, which improves its nutritional value, physical and organoleptic properties and are understood as the characteristic of traditional product. Eggs addition also enhances the price, which may bring some unscrupulous or irresponsible manufacturers to put into the product less eggs than it is declared on the label. For the official control proposes, whether the sample contain declared amount of egg solids, the methodology based on the cholesterol, as the characteristic marker of egg yolk, is comonly used. The aim of the project was to evaluate the robustness of the cholesterol as the marker of egg content in egg pasta from the point of a) natural variability b) processing technology. Dried eggs, samples taken during the manufacturing process and commercial egg pastas were analysed within the study. The results of standard method of cholesterol determination (based on the hydrolysis, saponification and extraction step prior to GC–FID analysis) were compared with direct ionisation in real time method (DART/TOF–MS). This technique was found to be fast, semi quantitative alternative for the rapid indication of unsatisfactory or out lied samples of raw materials or finished products, requiring only the step of proper dilution in solvent, filtration and internal standard addition (5α-cholestane or isotope labelled cholesterol-2,2,3,4,4,6-d6).

Keywords: Egg pasta, cholesterol, egg content

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C-46

CHANGES IN PHYSICO-CHEMICAL, MICROBIOLOGICAL, SENSORY ATTRIBUTES AND VOLATILE COMPOUNDS PROFILE DURING RIPENING OF DRY FERMENTED SAUSAGES WITH STRAWBERRY LEAF EXTRACT

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The objective of this work was to evaluate the feasibility of using strawberry (Fragaria × ananassa) leaves extract (SLE) in dry fermented sausages, to enrich them with healthy plant compounds and to increase stability by inhibiting oxidation reactions. The effect of dried extract additives on lipid oxidation (PV and TBARS values), pH, moisture, water activity, fatty acids composition, sensorial (colour and odour) and microbiological analysis as well as the profile of volatile compounds of dry-fermented sausage “Salchichón” were monitored during the ripening period. PV and TBARS values, showing effectiveness of the natural antioxidants from SLE, not exposed significant differences between samples. SLE had only slight effect on microbiological characteristics. It was observed that SLE the most influenced sausage volatile profile, pH value and sensorial properties. The main identified volatile compounds were those derived from the added spices. The effect of time and SLE amount on the profile of volatile compounds was estimated and it was found that SLE slightly modified the volatile profile of sausages, especially in the content of limonene, camphor and some aldehydes. The pH was slightly lower in the samples with higher SLE concentration (0.5%). CIELab system was used to measure color changes and it was observed that higher extract concentration reduced a* value while increased b* value. Sensory analysis of final product showed that SLE imparts recognizable herbal odour, however it did not reduce overall product acceptability. It may be concluded that addition of SLE to dry fermented sausages has no negative effect upon the ripening process, but the influence of SLE on lipid oxidation is not deduced.

Keywords: Dry fermented sausage, salchichón, strawberry leaf, fragaria × ananassa, ripening

C-47

EFFECT OF INFUSION CONDITIONS ON FREE AMINO ACIDS PROFILE OF GREEN AND BLACK TEA

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This study aimed to investigate free amino acid profiles of black and green tea samples. Hydrophilic interaction liquid chromatography coupled to tandem mass spectrometry was used to analyze free amino acids in tea without any pre-column or post-column derivatization. A total 20 amino acids (18 proteinogenic and 2 non-proteinogenic) were determined in black and green tea samples. Sum of the free amino acids within tea infusion after 2 min of brewing process was 220 mg/L and 211 mg/L for black and green tea, respectively. It linearly increased reaching to 311.2 mg/L and 277 mg/L for black and green tea, respectively, within 15 min of brewing at 85°C. Leaching rates were differed significantly for different individual amino acids. In general, hydrophilic amino acids leached into infusion faster than hydrophobic amino acids. Approximately 30% of total free amino acids in tea infusions after 15 min of brewing was theanine.

Keywords: Tea, infusion, amino acid profile

C-48

CAUSES AND POSSIBLE WAYS OF PREVENTION OF THE FORMATION OF WHITE SPOTS ON THE SALAMIS AND SAUSAGES

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The causes of the most common surface defect of salamis and sausages (fermented or heat treated and dried), the formation of white spots on the casing surface, are described. The white spots are formed by crystallization of salts present in the sausage or salami (calcium phosphates, calcium lactate, sodium chloride mainly), which are able to pass through the casing, if the casing itself and the surface of the product is wet. The defect takes place in the case of condensation of water vapour on the product surface when the storage temperatures are not stable. The model experiments to evaluate the effect of the properties (water and water vapour permeability) of casing (natural casings, collagen or cellulose casings) were done, the correlation between the tendency to the white spot formation and permeability has been studied. The possible way of the prevention of these defects are summarised.

Keywords: Salamis, sausages, surface defects, casings, white spots

C-49
QUALITY OF TRADITIONAL AND CONVENTIONAL POLISH HAM

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Consumer's buying decisions have been more frequently related to seeking safe food of acknowledged origin. Legal regulations in force in the European Union facilitate the protection of original agricultural products and food characteristics in respect of its original place and traditional production technology. Product quality determined by its production technology, processing and raw materials should be the most important attribute of a regional product and its positive image. Protection of regional and traditional products is particular importance now that the globalization of food production has caused the food market to be dominated by mass products whose quality parameters are frequently below expectations. This study presents the results of a study carried out as a part of a research project into a comprehensive evaluation of the quality of local and regional products available in the province of Warmia and Mazury (north-eastern Poland) on the basis of health value and consumer opinions. The aim of this fragment study was to determine the differences in the content of protein, lipids, iron, phosphorus, Σ DDT and γ-HCH in ham from small local manufacturers and large producers. The research material was selected on the basis of previously conducted studies that focused on the identification of meat products which, in the opinion of the producers, demonstrated exceptional quality, resulting mainly from the traditional method of production and from raw materials (of local origin).The study was carried out on pork ham produced by: a) small-scale producers that labelled their products with the sign "Culinary Heritage Warmia Mazury Powiśle", b) small-scale producers that declared the use of local raw materials and methods for producing traditional products; c) large plants that manufactured products with names related to the following terms: "rural", "traditional. Proteins concentration was determined using Kjeldahl method. Lipids were extracted from the material with a mixture of ethers, in accordance with the Schmidt-Bondzyński-Ratzlaff procedure. Iron and phosphorus content in the mineral residue were determined using flame atomic absorption spectroscopy. Measurements were carried out with the use of atomic absorption spectrometer Unicam 939 Solar – Great Britain, equipped with an Optimus data station, background correction (deuterium discharge lamp) and an appropriate cathode lamp. The identification and quantitative determination of the organochlorine pesticides were carried out with gas chromatography using a PYE Unicam 4600 apparatus with an EC detector. The study showed that hams produced by small, local enterprises contained more lipids and significantly less γ-HCH as compared to the smoked hams produced by large meat processing plants.

Keywords: Quality, ham, traditional food, conventional food

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C-50

CHEMICAL HAZARDS IN MEAT – SEGMENTATION OF CONSUMERS IN TERMS OF PERCEIVED RISK

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Many studies were carried out aimed at understanding consumer concern about food-related health risks. Food risk perception is what consumers believe would be the amount of risk, if any, they would face when consuming a food product. Several studies proved the importance of the theory of perceived risk for the understanding of how consumers made choices. Fears of illness make consumers perceive a series of possible consequences, such as days off from work and loss in income. Those findings may indicate a lower financial risk tolerance. The objective of this paper was to identify consumer segments based on a detailed analysis of perceived risk components. by the perceived risk types. A personal interview (face to face) was used to poll respondents (group of 1073 consumers from the Warmia and Masuria region, situated in NE Poland). The survey method applied was a quota sampling technique with quotas for age, educational level, and gender of those polled. There were two criteria for selecting a particular respondent for polling: whether or not they were the persons in the household to responsible for shopping and the preparation of meals, and, they had to regularly eat meat. The questionnaire consisted of statements used to measure the probability and importance of loss referred to the perception of health risk, psychological risk, financial risk, product performance risk and time risk. All statements (Probability of loss and Importance of loss) were measured using a 7-point scale from 'very unlikely'(1) and 'very likely'(7) for probability of loss, and 'not at all'(1) and 'very much'(7) for importance of loss. The reliability of the defined scales was assessed using a Cronbach's Alpha. In order to define the relationships between the items of perceived consequent loss, a method of PCA was implemented. In order to explore the existence of specific consumer segments within the survey sample based on their risk perceptions, a method of Cluster Analysis was applied. The study showed that the psychological risk and health risk were major components of the perception of risk resulting from the presence of chemical compounds in meat. Three profiles of segments were distinguished; they significantly differed in the intensity of perceived health and psychological risks: a segment of respondents, who were more aware of health risk, a segment of respondents, who were more sensitive to psychological risk, and an intermediate segment consisting of respondents with an equal sensitiveness to health risk and to psychological risk. No strong relationships were found between the demographic characteristics and the fact of belonging to a specific segment.

Keywords: Risk preception, consumer, food hazards

C-51
PROTEOLYTIC CHANGES IN RIPENED COW, SHEEP AND GOAT CHEESES MADE BY LOCAL PRODUCERS

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Products from rural areas are regarded as being of high quality. The EU legal regulations provide the possibility of protecting original agricultural and food products which are characteristic in terms of their origin and traditional method of production. These include: Protected Designation of Origin (PDO), Protected Geographical Indication (PGI) and Traditional Speciality Guaranteed (TSG). A way to ensure authenticity and high quality of traditional food products is to establish criteria for their registration that will thereafter determine standards for their commercial production. The aim of the study was to determine range of proteolytic changes that take place in ripened cheeses produced by local and mass manufacturers from cow, goat and sheep milk was assessed by determination of nitrogen compounds in the products, such as: total nitrogen, water-soluble nitrogen compounds at pH 4.6, peptide nitrogen content and amino acid nitrogen content. Stock was taken of ripened cheeses made by agritourist farms and dairy micro-companies operating in the region of Warmia and Mazury in Poland. Selected from the indicated products were those with above-standard quality, which results from traditional production methods and the raw materials (of local origin) used in the process. Of the selected rennet cheeses, samples of 15 hard and semi-hard goat and sheep cheeses were taken for analysis. Moreover, hard and semi-hard cheeses from 10 large producers were purchased on the local market (these samples were labelled as conventional). The results indicate that cheese made from sheep milk by local manufacturers contained the largest amounts of all the nitrogen forms under study. The total nitrogen content in them was 5.75%, the content of water-soluble nitrogen compounds at pH 4.6 was 18.27% Ntotal on average, peptide nitrogen was 2.63% Ntotal and amino acid nitrogen was 12.75% Ntotal. Moreover, the study showed that the concentration of individual forms of nitrogen compounds was higher in products made by local manufacturers compared to the same products made by mass manufacturers. Other parameters determined in cheese samples included pH, water, NaCl and fat content, fat content.

Keywords: Cow cheese, goat cheese, sheep cheese, proteolysis, nitrogen compounds

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C-52

INFLUENCE OF TNE ORIGIN ON THE SELECTED QUALITY DETERMINANTS OF THE PORK MEAT PRODUCTS

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Changes in consumer requirements of meat products as well as increased global competition are causing an unprecedented spur in processing and ingredient system developments within the meat manufacturing sector. Consumers demand healthier meat products which contains lower amount of salt, fat, nitrites in general and contain in addition health-promoting bioactive components for example carotenoids, unsaturated fatty acids, and fibers. The aim of the paper was to assess influence of the origin on the selected quality determinants of the pork meat products such as: fat, protein, salt nitrates (V and III), hydroxyproline and L-(+)-glutamic acid content. All samples were tested for the presence of dye such tartrazine (E102), quinoline yellow (E104), sunset yellow (E110), amaranth (E123), ponceau (E124) and indigo (E132). The study was carried out on raw pork, ham and sausage produced by: small-scale producers (meat processing plants) that labelled their products with the sign "Culinary Heritage Warmia Mazury Powiśle" (CHWMP), small-scale producers that declared the use of local raw materials and methods for producing traditional products (L); large plants that manufactured products with names related to the following terms: "rural", "peasant", "traditional", and "for generations". The research material (CHWMP and L) was selected on the basis of previously conducted studies that focused on the identification of meat products which, in the opinion of the producers, demonstrated exceptional quality, resulting mainly from the traditional method of production and from raw materials (local origin). From among catalogued products, a total of 23 samples of raw pork, 29 hams and 23 sausages were taken. Based on the experimental, it was found protein content in samples of meat and ham was similar 20mg/kg on average. Significantly higher protein content was found in sausages produced by large producers (24.73±1.98mg/kg). The fat content was significantly higher in the traditional ham (16.25±14.47mg/kg), compared with the local ham (4.38±2.26mg/kg) and the mass (9.29±5.25mg/kg). The ham samples traditional and local contained significantly higher salt content (respectively 3.31±0.72 and 2.90±0.54mg/kg). In all tested samples were no detected dye compounds. There was no statistically significant differences in hydroxyproline and L-(+)-glutamic acid content between traditional and conventional samples of meat products. Analysis the nitrate (V) and nitrate (III) showed a statistically significant difference in the average contents of these compounds. Significantly higher levels of nitrates contained only traditional ham samples (respectively 12.60±8.08mg NaNO(V)/kg, and 17.53±27.91mg NaNO(III)/kg of product), while there was a large variation in the content of these compounds in the samples.

Keywords: Pork, meat pork products, meat products quality

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C-53
INFLUENCE OF SALTS ON SELECTIVE COAGULATION OF WHEY PROTEINS

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Whey proteins are an important constituent of milk and especially of whey from manufacture of cheese. These proteins have many valuable functionally properties such as foaming and emulsifying ability or formation of gel. They are a very good source of amino acids and they have antibacterial and antiviral properties. Some of the whey proteins are sensitive to salts content in solution. High or low salts content may lead to selective coagulation of these proteins. For this experiment commercial whey protein concentrate was used. From this concentrate was prepared solution containing 7.5 wt % of protein. Subsequently was increased ionic strength by the addition of 7 wt % NaCl and so the part of whey proteins (the grater part of α-lactalbumin, BSA and immunoglobulins) was precipitated. In supernatant remained β-lactoglobulin and caseinomacropeptide (CMP). With this procedure was precipitated 95.1 wt % of α-lactalbumin, 87.2 wt % of immunoglobulins, 100 wt % of BSA, 59.5 wt % of CMP and 31.9 wt % of β-lactoglobulin. If solution of whey protein concentrate was desalinated by electrodialysis, the coagulation of α-lactalbumin and immunoglobulins increased. For the selective coagulation of caseinomacropeptide from supernatant was necessary demineralised this material by electrodialysis and subsequently added ethanol and adjusted pH. This reduction of the ionic strength and the addition of ethanol induced the selective precipitation of caseinomacropeptide (91.4 wt % from the original amount of CMP) and in solution remained β-lactoglobulin with a purity of 91%. This method can be therefore used for the isolation of β-lactoglobulin from whey.

Keywords: Whey protein, electrodialysis, coagulation

C-54

APPLICATION AND COMPARISON OF BACTERIAL STARTER CULTURES PRODUCING EXOPOLYSACCHARIDES IN THE PRODUCTION OF YOGHURT

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Three different commercial bacterial starter cultures, differing in their ability to produce exopolysaccharides (EPS), containing *Streptococcus thermophilus* i *Lactobacillus delbrueckii subsp. bulgaricus* strains were used to study the influence of EPS on the properties of yoghurt. The syneresis, rheological and sensory parameters were measured 1 day, 7 days and 14 days after the production of yoghurts. EPS production was found to have a substantial impact on the texture and sensory characteristics. The obtained results indicate, that the EPS produced by lactic acid bacteria increase the viscosity of products, leading to its thickening. Yoghurts become more viscous and its residence time in the mouth is enhanced. This improves sensation in the mouth and overall sensory acceptance, which only slightly changed during storage. During shearing the structure recovery of the yoghurts is disrupted in the presence of EPS. It is probably caused by the EPS-EPS and EPS-protein interactions, which slow down reconstruction of the original structure. EPS, however, due to their water binding capacity decrease syneresis. The functional properties of EPS from lactic acid bacteria as an alternative to conventional stabilizers, may decrease current production costs of fermented food products.

Keywords: Exopolysaccharides, yoghurt, LAB

C-55

NOVEL QA CLASSIFICATION OF GREEN COFFEE BEANS ACCORDING TO SPECIES AND POST HARVEST TREATMENTS BY NEAR INFRARED SPECTROSCOPY AND MULTIVARIATE CHEMOMETRICS

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This study aims at the evaluation of near infrared spectroscopy (NIR) technology for classification of coffee samples from different species and post harvesting processing. Near infrared spectroscopy (NIR) was used to discriminate between crude coffee varieties. Thirty three green coffee samples, including the two species arabica and robusta, were subjected to different post harvest treatments. All green beans were finely ground and analyzed in the full VIS/NIR spectral range (400–2498 nm) using a near-infrared FOSS NIRSystem 6500 II, equipped with a reflectance detector and spinning cup module. In food industries, NIR technology has become an important method in routine measurements of specific components and quality control. Two classification models were constructed: Soft Independent Modelling Class Analogy (SIMCA) and PLS Discriminant Analysis (PLS–DA). The findings show that NIR spectroscopy, coupled with either SIMCA or PLS-DA multivariate models, can be a useful tool for on-line quality control to differentiate and characterize green coffee of different species and post harvest treatments and in order to identify fraudulent mixtures and to confirm and improve the quality of coffee in the chain.

Keywords: Raw coffee beans, NIR spectroscopy, PLS–DA, SIMCA

C-56

EFFECT OF MODIFIED WHEY PROTEINS ON TEXTURE AND SENSORY QUALITY OF PROCESSED CHEESE

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Processed cheese is dairy product which is widely used in Czech cuisine. One of the possibilities to enhance its health benefits is the incorporation of whey protein hydrolysate as a material with lowered allergenicity and content of peptides with biological activity and prebiotic function. However, it is necessary to characterize effect of this addition on texture and sensory aspects of the products, because the hydrolysis of whey proteins modified its functional properties and generate also hydrophobic peptides with bitter flavour. Whey protein hydrolysate was prepared by enzymatic hydrolysis of reconstituted whey with three different dry matter content, 7, 14 and 21 wt % respectively, using enzyme mixture Promod 439L with additional removing of the hydrophobic peptides using activated carbon Norit CG1. Processed cheese (dry matter content 38 wt %, fat in dry matter content 31 wt %, pH 5.8) was manufactured from Edam cheese (dry matter content 57 wt %, fat in dry matter content 45 wt %), low-fat fresh cheese (dry matter content 18 wt %, fat in dry matter content 0.55 wt %), emulsifying salts and four different types of water phase (tap water, reconstituted whey, whey protein hydrolysate and whey protein hydrolysate after removing of hydrophobic peptides) which constituted 25 wt % of the processed cheese formula. Rheology of products was characterised by dynamic oscillation rheometry in system of two profiled plates, texture by texture profile analysis with puncture method and sensory quality was investigated using ranking and descriptive quantitative analysis with ordinal scale. It was observed that the application of whey protein and modified whey protein solution with 7 wt % of dry matter as the water phase compare to tap water decreased shear complex modulus, yield stress and hardness and enhanced phase shift, adhesiveness and chewiness of processed cheese. The increase of dry matter content from 7 to 21 wt % in protein solution led to the products with higher shear complex modulus, yield stress, hardness and chewiness and lower phase shift and adhesiveness. Processed cheese containing whey proteins after enzymatic hydrolysis showed the decrease in shear complex modulus and yield stress and the increase in phase shift and was softer, better chewable and less adhesive than the samples manufactured with non-modified whey proteins as the water phase. The additional removing of hydrophobic peptides from whey protein hydrolysate intensified these changes in rheological and texture properties of the products because of lowering the nitrogenous substances content. However, the sensory analysis of processed cheese with different water phases did not demonstrate difference in its hedonic quality, therefore could be the removing of nutritionally valuable hydrophobic peptides passed over.

Keywords: Processed cheese, whey protein hydrolysate, texture, sensory analysis

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C-57

NON-TRADITIONAL FORMS OF CEREALS - NATURAL FORTIFICANTS OF NUTRITIONALLY VALUABLE SUBSTANCES

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Cereals and cereal products constitute a staple food for the majority of our population. Cereal grains are only processed for the purposes of human nutrition, namely by milling and subsequent separating of milled fractions according to particle size, mineral content and the kind of the cereal grain used. The present-day milling technologies lead to unwanted heating of grain, removal of bran and the loss of germ, i.e. to substantial qualitative depreciation of the initial cereal grains. In Europe the most widely used flour is plain wheat flour, which is suitable particularly for making white bread, pastry, cakes, wafers and biscuits. The integration of some non-traditional cereals with grains of a higher nutritional value into the formulas especially of bakery products improves the nutritional and sensory quality of these, being the answer to the increasing demands of nutritionists and physicians, as well as consumers. Enough information gained by the study of vegetational, biological and agricultural features and properties, technological qualities and, last but not least, nutritional composition of respective non-traditional cereals is the prerequisite of their successful use. The aim of our work was to obtain information on some nutritionally valuable substances in the grains of non-traditional cereals. The basic food composition and the content of thiamin, riboflavin, niacin, pyridoxine and pantothenic acid were determined in whole-grain flour of three forms of common wheat (*Triticum aestivum* L.) with coloured grain – RU440-6, 'Citrus' and ANK-28B, of two varieties of emmer (*Triticum dicoccum* Schrank) – 'Rudico' and 'Tapioszele', and of two forms of barley (*Hordeum vulgare* L.) – 'AF Lucius' and KM1057. Unconventional wheat and barley forms were grown under standard field conditions at the localities of Kroměříž and Praha-Ruzyně in the years 2010–2011. The highest contents of proteins, ash and vitamin B1 were detected in the emmer variety 'Tapioszele'. The hulless spring barley line KM1057 showed the highest contents of fat, fibre and vitamins B2 and B6. The highest contents of niacin or pantothenic acid and of saccharides were determined in emmer variety 'Rudico' and in common wheat variety 'Citrus'. Based on the content values of observed nutrient factors, the studied non-traditional forms of cereals can be used as targeted fortificants of plain wheat flour.

Keywords: Non-traditional cereals, wheat, emmer, barley, vitamins

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C-58
DISCRIMINANT ANALYSIS OF OLOMOUC CURD CHEESE BY FT-NIR SPECTROSCOPY

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The aim of this study was to use the discriminant analysis of curd cheese during storage by Fourier transform near infrared spectroscopy method (FT–NIRs). Olomouc curd cheese samples were stored at 5°C (n=84) and at 20°C (n=84) during seven weeks. The spectra of samples were measured on the integration sphere in reflectance mode with the use of compressive cell in the spectral range of 10,000–4,000 cm^{-1} with 100 scans. The discriminant analysis is a classification technique which can be used to determine the class or classes of known materials which are most similar to an unknown material by computing the unknown's distance from each class center in Mahalanobis distance units. Ten principal components were used for all the calibration models. Great similarity between the samples stored at 5 and 20°C was found. Twelve samples stored at 20°C for 1 week and 2 samples stored at 20°C for 2 weeks were classified as samples stored at 5°C. The different results were found by comparing of the storage time. 100% variability was described between the spectra scanned in different weeks of storing at 5°C. The same results were obtained for the samples stored at 2°C. Only 1 sample in each class was misclassified. Mahalanobis distance between the sample classes stored for different period was significantly increasing with the length of storage time. Thus, discriminant analysis of Olomouc curd cheese by FT–NIRs is suitable method for the determination of ripening time.

Keywords: FT–NIRs, discriminant analysis, cheese, storage

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C-59

OPTIMIZATION OF SHRIMP SNACK PRODUCTION BY RESPONSE SURFACE METHODOLOGY: SENSORY PROPERTIES

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Snacks are enjoyable food products that are consumed throughout the world and tend to be high calories and fat but low in protein, vitamins, and other nutrients. Extrusion processing is widely used in the food industry for production of variety of fabricated, cooked and shaped products of varying texture. Seafood is an excellent source of good quality animal protein and mineral in the diet of human. In this study, corn flour was fortified with 20 g/100g dried shrimp meat for extrusion cooking. The objectives of this study were to develop a novel snack, enhance the nutritional value of corn flour based snack by using dried shrimp flour and investigate the effect of extrusion conditions, including feed moisture (17, 20, 23%), cooking temperature (110, 130 and 150°C) and screw speed (200, 350 and 500 rpm) on the sensory properties of shrimp snacks by response surface methodology. Shrimp snack extruded at high temperature, high screw speed and moderate feed moisture had the highest score for odour, appearance, flavour, and overall acceptance. Increasing feed moisture at highest temperature decreased odour, whereas increasing feed moisture at lowest temperature first increased and later decreased odour. The maximum appearance sensory score was obtained at highest screw speeds and lowest feed moistures, whereas minimum appearance score was obtained at highest feed moistures and lowest screw speeds. The maximum sensory flavour scores were obtained at highest screw speeds and lowest feed moistures, whereas minimum flavour scores were obtained at lowest screw speeds. Maximum overall acceptance scores were obtained at lowest temperature and highest screw speed, whereas minimum overall acceptance scores were obtained at lowest temperature and lowest screw speed. There was a relatively strong relationship between overall acceptance and other sensory properties. The results indicate that the poor nutrient content of corn based snacks can be enriched without compromising the desired product attributes by addition of shrimp meat.

Keywords: Response Surface Methodology, shrimp snack, extrusion, fortification, sensory properties

C-60

EFFECT OF ALKALINIZATION ON THE CHEMICAL REACTIVITY OF QUINOA PROTEINS (*CHENOPODIUM QUINOA* WILLD.), AND ON FILM FORMATION IN THE ABSENCE OF PLASTICIZERS

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An important factor that affects the reactivity of proteins is pH. It changes a protein's charge and degree of denaturation, and it is possible to obtain unfolded proteins under the effect of pH, exposing functional groups like sulfhydryl and hydrophobic groups and facilitating their association with water, chain-to-chain protein associations, and establishing new covalent bonds such as S-S. Edible protein films have been formed from different sources, and they generally require some degree of chemical changes prior to forming films. The aim of this study was to evaluate the effect of alkalinization on the chemical reactivity of quinoa proteins (QP) and on film formation in the absence of plasticizers. Protein extracts were obtained between pH 8 to 12 and their changes were evaluated by PAGE-SDS, size-exclusion HPLC light scattering, fluorescence spectroscopy, and SH and SS. Film was characterized by FTIR, SEM, tensile strength, barrier, and color. Protein extracts (PE, between pH 8 to 12) showed similar protein profiles, polypeptides with/without 2-ME. It was also found that the 11S protein type of quinoa at 55.0±0.9 kDa. SH-free increases from 2.4±0.4 to 9.8±0.4 µmol/g of protein, under the effect of pH. When proteins are treated with urea SH-exposed can be determined, with values ranging between 9.0±1.4 and 19.2±11.5 µmol/g of protein. SS groups also increased with alkali treatment, from 24.4±3.3 to 44.3±2.8 µmol/g of protein. These results showed an increase of SS formation and SH exposure, because cysteine is deprotonated under alkaline conditions, forming disulfide bonds and increasing SS values. This increase of SS bonds with increasing pH can be a significant factor for the presence of aggregated polypeptides found in high performance size exclusion chromatography. Fluorescence analysis showed that when PEs are treated at extremely alkaline pH (11 and 12), there are substantial changes in the emission maxima and a significant decrease of the fluorescence intensity of PE-11 and PE-12. Th e emission maximum was 344±1 for PE-8; 347±2 for PE-9; 348±2 for PE-10; 354±1 nm for PE-11; and 357±1 nm for PE-12. At pH 11 and 12, fluorescence intensity was lower and λmax shifted to longer wavelengths compared to PE obtained between pH 8 to 10. Alkalinization over pH 10 produces significant denaturation/aggregation/dissociation changes of QP. pH 12 was the condition to form a film (film-12). FTIR showed hydrogen bonds and hydrophobic film interactions. Film-12 had 16.6±3.8% elongation, 15.7±1.1 MPa tensile strength, and water vapor permeability was 5.18±0.38 g mm m-2d-1kPa^{-1}. Film-12 had a brownish color. A high degree of denaturation/aggregation/dissociation of QP structure is required to form a film without plasticizer.

Keywords: Quinoa proteins, alkalinization, chemical reactivity, film formation

Acknowledgement: *Innova Corfo, Cód.07CT9PUT-06*

C-61

CLASSIFICATION OF SPECIES AND POST HARVEST TREATMENT OF RAW COFFEE USING NEARED INFRARED SPECTROSCOPY AND MULTIVARIATE CALIBRATION

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This work aims at using near infrared spectroscopy (NIR) technology in the field of classification of coffee samples from different species and post harvesting processing. Near infrared spectroscopy (NIR) was used to discriminate between arabica and robusta raw coffee varieties. Thirty three green coffee samples, of different species and post harvest treatment were supplied by a multinational company. All green beans were finely ground and analyzed in the full VIS/NIR spectral range (400–2498 nm) using a near-infrared FOSS NIRSystem 6500 II, equipped with a reflectance detector and spinning cup module. In food industries, NIR technology has become an important method in routine measurements of specific components and quality control. Two classification models were constructed: Soft Independent Modelling Class Analogy (SIMCA) and PLS Discriminant Analysis (PLS–DA). The findings show that NIR spectroscopy, coupled with either SIMCA or PLS-DA multivariate models, can be a useful tool for on-line quality control purpose rapid, rather, methodology to differentiate and characterize green coffee of different species and treatment post in order to identify fraudulent mixtures and improve the quality raw coffee chain.

Keywords: Raw coffee beans, NIR spectroscopy, PLS–DA, SIMCA

C-62

LOCALITY INFLUENCE ON THE CHEMICAL COMPOSITION OF SELECTED SOYBEAN CULTIVARS

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The aim of the study was to determine the contents of mineral elements, trace elements, and total polyphenols in the seven varieties of cultivated raw soybeans (*Glycine max* L.) – cv. Bolyi45, Supra, Korada, Belmont, Crystal, Erin, Quito and to elucidate the relationship of these components of soybean grown at five different localities of Slovakia. Heavy metals and mineral elements in soybean samples were analyzed by AAS method. The total polyphenol content was estimated spectrophotometrically using Folin-Ciocalteaus phenol reagent and expressed in mg tannin per kg DM. For information completing the agrochemical characteristics were determined and evaluated within the soil hygiene of soil samples from parcels, on which soybean were cultivated. In many cases, enhanced reference values were defined for background element concentration in soil (Cd), not very markedly, but the total contents of all monitored risky elements did not reach in any case indicative limit value for soil contamination. Soybean proved to be a good source of many minerals and elements. Minerals or heavy metals contents show significant differences between cultivars and localities. The potassium content was found to be higher than those of other minerals in all soybeans, followed by P, Mg, Ca, which contents varied in the ranges 4.35–8.43 $g.kg^{-1}$, 1.89–2.36 $g.kg^{-1}$, 0.74–1.17 $g.kg^{-1}$, dw, respectively. The values show imbalance between the potassium content and other components, but the ratio K:Na, and Ca:P is adequate for human nutrition. The order of the elements levels in all tested soybean seeds was determined as following: Fe>Zn>Mn>Cu>Ni>Pb>Cr ≈ Co>Cd. The risky elements contents, with the exception of cadmium and cuprum (only cv. Korada grown in three localities) and Ni content in cultivars Crystal, Belmont grown in JelSovce), did not exceed a limit for the maximum levels of chosen risk elements in legumes (Food Codex Slovak republic). At the same time, the total polyphenols contents were determined: their concentrations were found to range from 817.65 to 1281.00 mg. kg^{-1}. From quantitative analysis polyphenols, the gained results suggest the variety dependence, but the locality influence on these compounds forming was not significantly confirmed, but results both indicate that the formation of polyphenols is probably genetically determined.

Keywords: Soybean, heavy metals, minerals, polyphenols, locality

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C-63

PHYSICAL CHEMICAL PROPERTIES OF CHICKEN SKIN COLLAGEN IN COMPARISON WITH COLLAGENS FROM OTHER ANIMAL TISSUES

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Collagen is one of the major proteins in the living body. Collagen and gelatin are widely used in the food, pharmaceutical, cosmetic, biomedical materials and leather industries. The main sources of industrial collagen are those from pig and bovine skin and bones. But the outbreaks of mad cow disease have resulted in anxieties amongst users of cattle collagen. Besides, the collagen from pig's skin and bone is not allowed to use in some regions due to religious reasons. In fish-eating and fish-processing countries, carp skin is extensively studied as an alternative source of collagen. Problem with fish collagen applications lies in fact that fish is among the most important allergen sources causing IgE-mediated food hypersensitivity. The objective of this investigation was isolation and determination of physico-chemical and molecular properties of collagen type I obtained from chicken skin tissue as an interesting and cheap alternative source. Chicken skin collagen was analyzed and compared to bovine tendon collagen. Various other skin tissues such as carp, turkey and pork were analyzed at the same time in order to obtain more valuable information for comparison between different species. Hen and 5 years old hen skin collagens were also analyzed to examine modifications in properties during animal growth. Molecular weight of collagen samples was determined by SDS-PAGE. Thermal stability and denaturation temperatures were determined by ultrasonic spectroscopy and viscosimetry. Amino acid composition was analyzed by AAA, trace elements levels were determined by AAS. Effects of incubation with some model denaturation conditions on properties and protein stability were determined in scope of this research. In this work collagen was isolated from chicken skin using modified procedure. After collagen was extracted, chicken skin proved to be vast source of acid soluble collagen type I consisting of typical fractions α1 and α2 chains in ratio 2:1. Predicted yield was 25% and water content was around 67%. Total nitrogen was 14.2%. The proportional lysine content was 7.07%, proline 5.88% and glycine 10.07%. Denaturation temperature was determined at approx. 50°C for chicken, 60°C for hen and 78°C for mature hen collagen. Elementar trace analysis was used for collagen characterization too. Obtained results suggests that waste chicken skin has potential to be an excellent alternative source of raw collagen, which could find it's application not only in food industry but, after further research and processing, as a component of various collagen based biomaterials in biomedicine.

Keywords: Collagen, chicken skin, viscosimetry, ultrasonic spectroscopy, PAGE-SDS

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QUALITY ASSESSMENT OF NATURAL YOGURT AND YOGURT WITH THE TRANSGLUTAMINASE ADDITION

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The study was undertaken to assess the quality of natural yogurt and the one enriched by the addition of transglutaminase (TG). Intra- and inter-molecular polymerization of milk proteins, enzyme-catalyzed, strengthens the protein network of yogurt. It contributes to increased firmness of curd, reduced and evenly distributed pores in the protein network of curd. As a result, it restricts the syneresis and strengthens yogurt resistance to the action of mechanical factors during production and storage. In addition, low-fat yogurts with the addition of TG are characterized by a flavor and texture typical of milk fat products. In the study low-fat yogurts with a standardized composition, produced in one of the Polish dairy plants were subjected to the assessment. Half of them contained transglutaminase preparation Saprovia 100 New (P.M.T. Trading Sp. z o.o., Łódź) with the enzymatic activity of 100 U/g protein. Testing products, without TG (JC) and with the enzyme (JTG) were observed and examined after 1,7,14 and 21 days of storage at 6±1°C. The study involved analysis of the content, pH values, viscosity, syneresis, texture and organoleptic assessment performed by a six-member panel that evaluated taste, structure, consistency and texture in the 0 to 5 scale. On the basis of the obtained results, it can be stated that milk fermentation process with and without the enzyme proceeded in a similar way. The milk acid and pH value content in the testing products and the ones with the TG were alike and amounted to 1.26–1.30% and 4.36–4.42% respectively. Results of marking the water retention capability of a curd demonstrate that products JTG were less susceptible to syneresis than testing ones. The amount of centrifuged whey accounted for 19.77% in the fresh samples and 10.90% in the samples kept for 21 days, and it was 34–42% lower than in the JC. The analysis of the texture parameters indicates that firmness and cohesiveness of the curd and consistency thickness were 33–36% higher in the samples with TG while both types of products were comparable with respect to the value of viscosity index. The values of texture parameters of all samples were intensifying during storage. However, dynamic viscosity of JTG products was determined to be higher than in the case of testing products, both fresh and stored for 21 days. Initial viscosity of the yogurts with TG after 1 day of storage accounted for 16.6 Pas and was 45% higher compared to JC samples. Lengthening of the storage period triggered the viscosity intensification in all of the samples. The organoleptic quality of yogurt with and without TG was similar, however compared to JC, products with the enzyme were characterized by more thicken, homogenous consistency and smooth, creamy texture. Results obtained in the study demonstrated that the catalytic activity of transglutaminase makes it possible to improve the physicochemical features and organoleptic properties of yogurt.

Keywords: Yogurt, transglutaminase

C-65
COMPARISON OF AMINO ACID PROFILES AND SPECTROSCOPIC CHARACTERISTICS OF SOME SPELT FLOURS

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Cereals including spelt wheat play important role in human diet with positively impact on human health. Flours are significant source of bioactive compounds, e.g. essential amino acids, vitamins, minerals and dietary fibre. In addition, it is widely accepted that their benefits follow from the presence of polyphenols with multiple biological effects, including antioxidant activity. The aim of the present work was to evaluate the amino acid profiles and antioxidant activity of selected samples of spelt flour, prepared from grains of two spelt cultivars grown following organic and conventional production practices in different geographical localities and to perform the mutual correlation of their composition and way of flour production/processing. The profile of amino acids comprising twenty free amino acids was determined by HPLC/ESI-MS method in spelt flour aqueous extracts. Besides that, antioxidant and radical-scavenging properties of spelt extracts in 50% ethanol/water solution (*v/v*) were evaluated using $DPPH^{\bullet}$ and $ABTS^{\bullet+}$ assays by EPR and UV–VIS spectroscopy. Basic characteristics of extracts such as the content of total polyphenols (TPC) were evaluated, as well. Multivariate statistical analysis was subsequently applied on the whole dataset of the obtained experimental characteristics in order to assess the influence of the way of production (organic vs. conventional), geographical locality and of spelt cultivar on the monitored characteristics. Besides that, the possibility of flour differentiation according to the previously mentioned characteristics was tested. Results obtained clearly proved that the composition of flour (content and concentration of aminoacids, as well as polyphenols) as well as their radical-scavenging properties is significantly influenced by several factors, including way of farming, geographical origin or varietal composition. Multivariate statistical analysis represents an effective tool for discrimination of spelt flour. By means of canonical discrimination analysis, flour samples were with >89% correctness discriminated according to previously mentioned parameters.

Keywords: Spelt flour, polyphenols, antioxidant capacity, amino acid profile

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CHARACTERIZATION OF GARLIC USING MASS SPECTROMETRY-BASED METABOLOMIC APPROACHES

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Garlic (*Allium sativum* L.) is food ingredient widely used in gastronomy. Besides to be used like food, garlic has long been used in folk medicine with protective and curative purposes. Several investigations indicate that the biological and medical functions of garlic are mainly due to the high content of organo-sulphur compounds. The primary sulphur-containing constituents in both whole vegetables are the *S*-alk(en)yl-L-cysteine sulphoxides (ACSOs), such as alliin, and γ–glutamylcysteines. These compounds are responsible for garlic's characteristic odour and flavour, as well as most of their biological properties. In total, 60,000 tons of garlic are annually imported into the EU, mainly from China (60%), Argentina (30%) and other countries (Russia, Egypt). Recently, the demand for high-quality garlic has prompted grower interest in the EU including also its domestic production. To distinguish garlic of different origin and/or containing different content of biologically active compounds, comprehensive analysis of its constituents is needed. For this purpose, the mass spectrometry-based metabolomic approach, aiming at global analysis of numerous targeted or non-targeted low molecular compounds (metabolites) in a biological sample, has found its application in diverse research areas including plant. Beside of conventional techniques (LC–MS, GC–MS, NMR) used for metabolomic fingerprinting/profiling, ambient desorption ionisation techniques represent a novel solution for direct sample examination in the open atmosphere, with minimal or no sample preparation requirements and remarkably high sample throughput. In this study, two mass spectrometry-based metabolomic approaches were investigated for characterisation of the garlic extracts. In particular, direct analysis in real time ionisation coupled to a high-resolution (orbitrap) mass spectrometer and liquid chromatography–electrospray–high–resolution mass spectrometry with a QTOF mass analyser (LC–ESI–QTOFMS). The acquired data were processed employing multivariate data analysis with the aim to characterise the diverse varieties of garlic in terms of their quality (content of biologically active substances beneficial to health) and the resistance.

Keywords: Garlic, metabolomic, mass spectrometry,S-alk(en)yl-L-cysteine sulphoxides

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C-67

EFFECT OF HIGH-PRESSURE HOMOGENIZATION AT DIFFERENT TEMPERATURE ON CHANGE IN MILK EMULSION AND COLLOIDAL SYSTEM

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The effect of high-pressure homogenization on selected properties of milk standardized to fat content of 3.5%, especially emulsion and colloidal stabilities, were determined in the study. The following parameters were: constans pressure of 120 MPa and different temperatures of 6°C, 20°C, 40°C and 60°C. Milk was also homogenized at the pressure of 20 MPa and temperature of 60°C. The results indicated a rise in milk temperature during the high-pressure homogenization, depending on the milk temperature before the process, max. app. 29°C. The application of high-pressure homogenization contributed to an increase in fat globule size reduction and unification along with an increase in homogenization temperature. Decrease of Sauter mean diameter of milk fat globules and increase of fat area (up to 8-fold) were the results of homogenization in the case of milk at the temperature of 60°C. The rise of fat dispersion degree and their surface as the consequence of homogenization of milk at the temperature of 6°C was app. twofold. Each of the homogenized milk was characterized by the predominant participation (≥90%) of fat globules with a diameter of ≤2μm. Due to the modification of emulsion phase, the reduction of the rate of creaming of milk fat along with a rise at the temperature of the process was observed. In the case of milk high-pressure homogenization at the temperature of 60°C, there was scarcely any separation of fat from milk. Content of free fat in homogenized milk increased along with decrease of process temperature and homogenization of milk at the temperature of 6°C contributed to almost 3-fold increase in comparision with non-homogenized milk. The application of high-pressure homogenization of milk at the temperature of 40°C resulted in similar efficiency in stabilization of emulsion phase (creaming, fat globules diameter) and content of free fat in milk as homogenization at pressure of 20 MPa and temperature of 60°C. Effect of high-pressure homogenization on colloidal stability of milk was connected with reduction of heat stability and rennet coagulation time of milk. Milk resistance to coagulating factors decreased along with a decrease of initial value of process temperature. On the basis of the obtained results, reduction of heat stability and rennet coagulation time of milk upon high-pressure homogenization at the temperature of 6°C amounted to 33% and 25%, respectively. High-pressure homogenization carried out at higher temperature, especially ≥40°C, caused shortening of heat and rennet coagulation time of milk by app. 27% and 13%, respectively. Heat stability as well as rennet coagulation time of milk subjected high-pressure homogenization reached lower values in comparision with the results of homogenization at pressure 20 MPa at the temperature of 60°C. The high-pressure homogenization caused increase of milk viscosity, max. around 18% as a consequence of process realised at the temperature of 40°C.

Keywords: High-pressure homogenization, milk, emulsion stability, colloidal stability

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AN ENZYME SENSOR FOR THE ESTIMATION OF PROCESSING TIME IN DRY-CURED MEATS

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The use of biosensors is being used in the food industry as an alternative to conventional methods to estimate the freshness and bacterial spoilage where some nucleosides have been confirmed as useful chemical indicators. More recently, xanthine has shown the possibility to estimate the time of meat ageing or even time of further processing. In this context, an enzyme sensor employing xanthine oxidase in combination of an oxygen electrode has been developed and optimised to estimate the xanthine content in dry-cured ham and dry-fermented sausages during their processing and correlate such values with time of processing. The enzyme xanthine oxidase was immobilised on a preactivated membrane (Immunodyne ABC) using glutaraldehyde as cross-linking agent. Previous to the immobilisation, the enzymatic solution was filtered in an Ultrafree-CL high-flow Biomax-PB polyethersulfone membrane (100 KDa) in order to concentrate the specific activity. Determination was based on the measurement of the consumed O_2 in a platinum electrode poised at -600 mV versus Ag/AgCl. The reaction was started by the direct injection of the standard or meat extract (40 µL) on the enzymatic membrane. The current decrease up to 60 s or the enzymatic rate at 30 s was used for the detection of xanthine. The enzyme membrane was stable at 4°C for at least two months and could be used up to 30 analyses in a day without significant loss of sensitivity. Data obtained with the biosensor was compared to those from a standard HPLC method and a good correlation was found between both methods. Thus, this study may help to the development of a simple, rapid and low cost method to estimate the time of processing for dry-cured ham and dry-fermented sausages that can be used for quality control in the meat industry.

Keywords: Enzyme sensor, nucleosides, xanthine, dry-cured ham, dry-fermented sausages

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C-69
INFLUENCE OF STORAGE AT 4ºC ON THE STABILITY OF HIGH HYDROSTATIC PRESSURE TREATED ONION

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The application of high hydrostatic pressure (HHP) in food processing allows an improvement in the retention of food quality attributes and nutritional values in comparison with other processing technologies. The aim of this work was to evaluate the effects of refrigerated storage on microstructure and physicochemical properties of HHP-treated onion. Onion was submitted to two different HHP treatments (100 MPa at 50°C and 400 MPa at 25°C, both for 5 min). The microstructural study was carried out by Low Temperature Scanning Electron Microscopy (Cryo-SEM) and Light Microscopy (LM). Some physicochemical properties, such as total soluble phenolics (TSP), soluble protein percentage (SPP) and texture were also studied. HHP-treated samples were analyzed immediately after treatment and after 7, 14, 21 and 28 days of storage at 4°C. HHP treatments affected the permeability of cell walls and membranes, favoring the diffusion of soluble material to the apoplast and producing loss of cell turgor. Storage at 4°C also caused important structural degradation in all the HHP-treated samples. This degradation was higher when treating at 400 MPa at 25°C if compared to 100 MPa at 50°C and led to changes in some physicochemical properties during the first week of storage. TSP and SPP increased after the application of HHP but lower values were obtained during the storage period. Onion samples treated at 400 MPa at 25°C maintained higher texture values during the whole storage period. The differences between the two different HHP-treated samples decreased progressively during storage. Interactions between phenolics and other compounds such as solubilized cell wall material or proteins could explain the decrease in TSP and SPP during storage.

Keywords: Onion, high hydrostatic pressure, microstructure, phenolics, storage

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C-70

EFFECT OF TEMPERATURE AND PRESSURE ON THE MICROSTRUCTURAL AND TEXTURE PROPERTIES OF PERSIMMON

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The application of high hydrostatic pressure (HHP) in food processing could improve the retention of food quality attributes and nutritional values in comparison with other processing technologies. The aim of this work was to study the relationship between the microstructure and texture of persimmon cv. 'Rojo Brillante' submitted to high hydrostatic pressures and pasteurization process. Persimmon was submitted to HHP (200 MPa/25ºC/6 min) and pasteurization (70ºC/15 min). The microstructure study was carried out by Scanning Electron Microscopy at Low Temperature (Cryo-SEM). Firmness, cohesiveness and shear force were also studied. The microstructural changes observed could be related to an improvement in the availability of nutritive compounds, such as tannins, after the HHP treatments. Cryo-SEM micrographs of untreated persimmon showed a compact tissue with rounded cells and intercellular spaces filled with air. HHP treatment caused degradation of cell wall material resulting in separation of adjacent cells and in intercellular spaces filled with soluble material from the cells, due to compression. Pasteurized samples, however, presented less structural degradation and some intercellular spaces were still full of air; a compact mass also appeared inside some cells, which could be attributed to the formation of polymeric substances from the soluble tannins. The effects of the treatments on the microstructure of persimmon caused changes in the textural properties of the samples. HHP-treated samples had lower firmness, cohesiveness and shear force than untreated and pasteurized ones. Part of the precipitated tannins observed in the pasteurized samples could interact with components of the cell wall and contribute to the maintenance of textural properties. HHP-treated samples, despite showing higher degradation and worse textural properties than pasteurized samples, presented better diffusion of soluble compounds throughout the tissue. Therefore, the availability of soluble compounds could be increased using HHP treatment if compared to conventional pasteurization process.

Keywords: Keywords: persimmon, microstructure, HHP, texture, pasteurization.

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C-71

EFFECT OF DIFFERENT RICE STARCHES, INULIN AND SOY PROTEIN ON MICROSTRUCTURAL, PHYSICAL AND SENSORY PROPERTIES OF LOW-FAT, GLUTEN AND LACTOSE WHITE SAUCES

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Approximately 50% of people with celiac disease have lactose intolerance. The typical ingredients of white sauces are milk, oil, wheat flour and salt. Therefore, this kind of sauces cannot be consumed by this sector of the population. Furthermore, the demand of low-fat products is increasing every day. The microstructural, physical, and sensory properties of low-fat sauces made with different rice starches, soy protein, and inulin as a fat replacer were analyzed in order to obtain sauces suitable for celiac and lactose intolerant consumers. Soy protein and inulin could have a protective effect against starch degradation. This fact could be related to the high water binding capacity of inulin and soy protein. Moreover, protein molecules could diffuse into the starch granules and soluble inulin could interact with starch polymers within the granule. Both effects would hinder amylose leaching. Inulin provides better diffusion capacity of gelatinized granules and soy protein-starch granule aggregates than sunflower oil. The diffusion capacity of the continuous phase and the number of gelatinized starch granules and soy protein-starch granule aggregates determine the sauce viscosity and stability. Inulin decreases viscosity in sauces made with modified rice starch sauces and overall it maintains stable viscosity values during the whole refrigerated storage period. The use of soy protein would be suitable to prevent syneresis in modified or native rice starch sauces. In the same way, the replacement of oil by inulin does not affect the sauce stability. Inulin affects L*, Cab*, hab y AE* values in native rice starch sauces, probably because inulin-retrogradated amylose polymers interactions take place. Sauces made with sunflower oil and modified rice starch are best rated by the consumers. However, according to the statistical analyses, the replacement of oil by inulin could be suitable to prepare low-fat, gluten and lactose free white sauces when modified rice starch is used.

Keywords: Inulin, rice starch, soy protein, microstructure, viscosity, stability

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C-72

ELECTRON BEAM RADIATION APPLIED TO PORTUGUESE CHESTNUTS (*CASTANEA SATIVA* MILL.): EVALUATION OF THE EFFECTS ON NUTRITIONAL PARAMETERS

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Chestnuts are a widely consumed nut around the world, being China the biggest producer. Portugal represents 4% of its worldwide production with a gross weight of 22105 tons of chestnut in 2010, and an income of 15 M€. The Trás-os-Montes region produces 75% on the nation's chestnuts being one of its main economic resources. Storage of these nuts is an important step during processing to avoid rotting related to their relatively high metabolic activity. Until 2010 the most common treatment for post-harvest desinfestation was methyl bromide, a widely used fumigant which was banned in the European Union under the Montreal protocol due to its toxicity and deleterious effect on the environment [1]. Other treatments like temperature, high pressure blast, hot water dip and other fumigants still pose quite a few disadvantages [2]. Food irradiation processing has been introduced as a safe and environment friendly alternative for conservation, being used in several products such as meat, fish or fruits [3]. Previous studies of our research group showed that gamma radiation had no major negative effect on the nutritional value of chestnuts; in fact, storage time had a much bigger influence on the chestnut quality [4]. In the present study, we report the effect of another type of radiation post-harvest processing, electron beam, with doses of 0, 0.5, 1, 3 and 6 kGy in the nutritional value of chestnuts (ash, energy, fatty acids, sugars and tocopherols), stored at 4°C during 0, 30 and 60 days. The storage time seemed to reduce fat and energetic values but reported a tendency for higher values of dry matter. Regarding fatty acids, there was a higher detected quantity of C20:2 in non-irradiated samples, and four fatty acids were only detected in trace quantities. The levels of γ-tocopherol decreased during storage time but did not alter its quantity for all the radiation doses, in fact they seemed to be present in higher concentrations in the irradiated samples. Sucrose and total sugars were lower in non -irradiated samples and raffinose was only detected in irradiated samples. Electron beam irradiation seems to be a suitable methodology, since the effects on chemical and nutritional composition are very low, while storage time seems to be quite important in chestnut deterioration.

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Keywords: Chestnut fruits, *Castanea sativa*, e-beam irradiation, nutritional parameters

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C-73
FUNCTIONALITY OF SEVERAL CAKE INGREDIENTS: A COMPREHENSIVE APPROACH

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The physicochemical properties of cakes are largely dependent on batter and cake structure. Therefore, understanding the internal macro and microstructure of bakery products is essential. Bakery products constitute one of the most consumed foods in the world. Among them, cakes are particularly popular and are associated in the consumer's mind with a delicious sponge product with particular organoleptic characteristics. Cake batter is a complex emulsion and foam system. Flour, milk, fat, sugar, egg and leavening agent are the main ingredients used in its elaboration; each ingredient plays an important function in cake structure. For that reason several microstructural techniques were applied in this work to correlate batter and cake structure with the physicochemical properties. Also, image analysis and quantification of relative features were applied as they are the basis of modern food microscopy. The objective of this study was to understand the functionality of oil and inulin (as fat replacer) as structural ingredients of sponge cake, and to correlate the structure with the physicochemical properties. The role of leavening agent was also evaluated as it is an ingredient which contributes to give the proper structure. For this purpose four different formulations including or not oil, leavening agent and inulin were used. Oil played an important role in batter stability, due to its contribution increasing batter viscosity and occluding air during mixing. The addition of leavening agent was crucial to the final height and sponginess of the cakes. When inulin was used as fat replacer, the absence of oil caused a decrease in the stability of the batter, where larger air bubbles were occluded. Inulin dispersed uniformly in the batter creating a competition for water with flour components: gluten was not properly hydrated and some starch granules were not fully incorporated into the matrix. Thus, the development of a continuous network was disrupted and the cake was shorter and softer; it had interconnected air cells in the crumb, and was easily crumbled. Structure studies were decisive to understand the physicochemical properties.

Keywords: Cake, structure, oil, inulin, leaving agent

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C-74

CHEMICAL, MICROSTRUCTURAL AND TECHNOLOGICAL CHARACTERIZATION OF ITALIAN FARRO *TRITICUM TURGIDUM* SS. DICOCCUM

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The aim of this study was to evaluate the microstructural characteristics and differences between two types of Italian farro (*Triticum turgidum* ssp. *Dicoccum*) also known as emmer wheat. The common wheat was used as control. Microstructural studies were conducted using Scanning Electron Microscopy (SEM and CryoSEM). Grains, flours and freeze-dried doughs obtained from farro and wheat flour were observed by SEM, while doughs and gluten were analyzed by CryoSEM. Protein fractions were extracted from flours, and SDS-PAGE of these fractions under denaturing conditions were also studied in order to understand their technological properties. As regards the microstructural characteristics of the grains of hard and soft farro, the observations showed that starch granules were covered by protein in hard farro samples more than in the soft ones. These differences could also be noticed in flours. Starch granules appeared embedded in the protein matrix in doughs made from hard farro, while in soft farro doughs, starch granules appeared looser on the matrix surface. Hard farro gluten was observed as a homogeneous structure showing similarities with common wheat gluten, while the gluten obtained from soft farro appeared to be more heterogeneous. The results obtained by SDS–PAGE regarding the gluten protein fractions (gliadins and glutenins) showed that the gliadin fraction patterns of both farro samples were similar. The common wheat gliadin fraction showed a different pattern in the area ranging from 115 to 55 kDa if compared to farro one. When the glutenin fraction was analyzed by SDS–PAGE, the electrophoretic pattern of hard farro appeared to be similar to that of the common wheat. This could explain the technological behavior of the hard farro, which could be more useful than the soft one for the development of new bakery products.

Keywords: Farro, microstructure, SDS-PAGE, Cryo-SEM, SEM

C-75
BEER FINGERPRINTING BY MALDI-TOF MASS SPECTROMETRY

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Thanks to its rapidity and simplicity, profiling by using Matrix-Assisted Laser Desorption-Ionization – Time of Flight Mass Spectrometry (MALDI–TOFMS) serves as an effective tool for several applications. Based on the selected sample preparation protocol, the method is capable to provide fingerprints reflecting the abundance of diverse classes of compounds. In our work we developed a sample preparation protocol which enables parallel MALDI–TOFMS fingerprinting of maltooligosaccharides and proteins from untreated beer samples. Apart from the influence of used raw materials, the content of these compounds in beer depends on several chemical and enzymatic reactions taking place during the brewing process. For these reasons we examined whether the MALDI–TOFMS profiling allows distinguishing beer samples according to their producer or brand. On a set of 15 beer bottles of the same brand produced in three different breweries we demonstrated that method enables distinguishing the producers on the basis of the cluster analysis of obtained protein profiles. Another proof on the discriminatory abilities of the technique was obtained by analysis of 17 lager beers of different brands, where the method allowed reliable distinguishing of majority of the examined samples. On the basis of obtained results, we propose MALDI–TOFMS profiling of untreated beer samples as a rapid tool for beer brewing technology process monitoring, quality control, and determination of beer authenticity.

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Keywords: Beer, MALDI–TOF mass spectrometry, proteins, maltooligosaccharides

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C-76

EVALUATION AND DEVELOPMENT OF A RESTRUCTURED COHO SALMON (*ONCORHYNCHUS KISUTCH*) PRODUCT HOLDING FUNCTIONAL PROPERTIES

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New trends in food consumption are being developed as a result of actual lifestyle and nutritional demands of consumers. Among them, functional foods have attracted a great attention since in addition to provide an important nutritional value, healthy and beneficial properties to the human body are expected to be supplied in order to reduce chronic and non-spreading diseases. To achieve such objectives, different kinds of natural additives or compounds have been tested. Among them, inulin has been found a prebiotic soluble dietary fibre and non-digestible food ingredient, which selectively stimulates the growth and/or activity of intestinal bacteria; further, citric fibre is reported to provide valuable physiological functions, in addition to technological properties such as increasing the water retention capacity in foods. This study concerns the development and optimisation of a new functional and restructured product based on coho salmon (Oncorhynchu kisutch) which would mean an increased added value. This fish species has recently attracted a great commercial attention as a farmed product including a great content of polyunsaturated fatty acids (namely, docosahexaenoic and eicosapentaenoic acids). To this aim, inulin and citric fibre were added to salmon muscle, being the physical, chemical and sensory parameters analysed. A two-dimensional central composite design consisting of 10 experimental trials with varying concentrations of inulin and citric fibre (0-5 g of each per 18-g-portions of restructured product) was performed. The response variables of the model chosen to be analysed corresponded to the sensory (Quantitative Descriptive Analysis methodology; and overall quality test (colour, odour, texture, smell, taste and), physical (texture, crispness, gaping, expressible moisture, dripping, Hunter L*, a* and b* parameters, cooked yield and water holding capacity) and chemical (pH, water and lipid contents, fatty acid analysis). For the optimisation of the design variables, the RSM (Response Surface Methodology) technology was applied. As a result, the restructured products were optimised with inulin and citric fibre by employing 4.83 g and 2.94 g per portion, respectively. For this optimised product, a desirability score of 0.78 (from a maximum value of 0.8) was attained, being the overall quality value of 87.06 from a 0–100 scale. It is concluded that the addition of such inulin and citric fibre quantities allow a profitable retention of physical, chemical and sensory properties in the proposed restructured salmon product.

Keywords: Oncorhynchus kisutch, functional food, inulin, citric fiber, added value

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C-77

EFFECT OF SEASONAL CHANGES ON THE COMPOSITION OF SUGARS AND FRUCTO-OLIGOSACCHARIDES IN TUBERS OF JERUSALEM ARTICHOKE FROM BALTIC REGION

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The Jerusalem artichoke (*Helianthus tuberosus* L.) (JA) is a member of the same Compositae family as the sunflower (*H. annuus* L.). The plant grows under different pedoclimatic conditions and shows a good frost and drought tolerance as well as a resistance to pests and diseases. The JA is a high biomass yielding plant. Moreover, JA tubers do not contain starch and accumulate carbohydrates as inulin that has a positive physiological effect in humans. Thus, JA tubers can be used for food or animal feed, as a raw material for the industrial production of fructose and fructans [1] or as a source of bifidogenic factors in human nutrition [2]. In the present work we performed a detailed investigation on the composition of soluble carbohydrates and inulin in JA tubers harvested at the end of March after being exposed to frost during winter and during the period of the following vegetative growth and intensive photosynthetic activity in summer. JA was uprooted monthly. Three genetic variants of JA, i.e. Sauliai, Albik and Rubik, which are cultivated in Lithuania, were used for investigation. Qualitative and quantitative determination of sugars and fructo-oligosaccharides was performed by enzymatic assays and HPLC. At the end of March the highest amount of inulin (approximately 20% of wet weight) was found in Albik tubers as compared with ones of other cultivars Sauliai and Rubik. The amount of glucose and fructose was low in tubers of all cultivars and was equal to 0.1–0.2% and 1.0–1.6% of wet weight, respectively. However, during the period of plant growth the amount of fructose increased by 9–15-fold and sharply decreased at the end of June. Senescent tubers of all cultivars contained approximately 1.0% (of wet weight) of fructose. On the contrary, the amount of sucrose decreased gradually from 5.0–5.5% at the end of March to 1.0–1.5% of wet weight of senescent tubers at the end of June. The amount of inulin in senescent tubers of the cultivar Albik decreased by approximately 7-fold. However, in the tubers of other two cultivars the amount of inulin was found to be similar to the one at spring time. Additional experiments are needed to explain and understand this phenomenon. Plant life stages are mirrored in the changes of tuber carbohydrate composition. Further experiments will be performed during the period of tuber developing and maturing. They will lead to the conclusion what is the best genetic variant cultivated in Lithuania and what is the best time to harvest tubers used for obtaining functional food.

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Keywords: *Helianthus tuberosus* L., sugars, fructo-oligosaccarides, inulin, Baltic region

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C-78
NUTRITONALLLY IMPORTANT FATTY ACIDS IN SEEDS OF SMALL FRUITS

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Beside n-3 fatty acids, γ-linolenic (C18:3 n-6) and stearidonic acid (C18:4 n-3) are of particular value to the pharmaceutical and health food industry. The purpose of the present study was to determine the content of total fats and fatty acid composition in seeds of small fruits. 5 cultivars of gooseberry (*Ribes uva-crispa* L.), 10 cultivars of black currant (*Ribes nigrum* L.), 4 cultivars of red currant (*Ribes rubrum* L.) and one cultivar of josta (*Ribes nidigrolaria Bauer*) were analysed. In average the seeds of analysed small fruits contain 18.61% of total fats. Fat from the seeds of small fruits contain 39.1% of linoleic, 18.5% of α-linolenic, 12.8% of oleic, and 10% of γ-linoleic fatty acids. The highest ratio of linoleic (43.3 %) and γ-linolenic (12.3 %) fatty acid was found in the seeds of black currant while that of α-linolenic (28 %) and palmitic (8.5%) fatty acids was found in the seeds of josta. All small fruits also contain stearidonic acid (C18:4 n-3), in the range of 2 to 5%. Results revealed statistically significant differences in fatty acid composition between cultivars as well as between small fruit species. From the nutritional point of view, small fruits represent an important source of α- and γ-linolenic acid as well as stearidonic acid. According to the aterogenic index (0.43), to the ratio polyunsaturated/saturated fatty acids (4.7), and to the ratio of n-6/n-3 (2.5:1) fat from seeds of small fruits fulfils the nutrition recommendations. Due to favourable fatty acid composition seeds of small fruits are interesting new source of nutritionally important fatty acids that can be used as the supplement in everyday nutrition.

Keywords: Small fruits, fatty acid composition, γ-linolenic (C18:3 n-6), stearidonic acid (C18:4 n-3)

C-79
CHEMICAL COMPOSITION OF MEDITERRANEAN HACKBERRY FRUITS (*CELTIS AUSTRALIS* L.)

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Celtis australis, commonly known as the European nettle tree, Mediterranean hackberry, lote tree, or honeyberry, is a deciduous tree that can grow 20 or 25 meters in height. Fruits are seldom used for nutritional purposes. We have determined the nutritional and physico-chemical properties including dry weight, water content, crude fiber, proteins, vitamins, minerals and total phenolic content of ripe hackberry fruits from Istria. Ripe hackberry fruits contain 51.1% of soluble solids, 13.4 of dietary fibers, 4.9 % of proteins, 18.6% of reducing sugars and water. The total amount of phenols determined in fruits was 0.83 mmol/g (chlorogenic acid equivalent). The vitamins found in hackberry fruits were lutein (1.42 µg/g dry mass), β-caroten (5.58 µg/g), zeaksantin (0.74 µg/g), α-tocopherol (150 µg/g), γ-tocopherol (4.9 µg/g) and δ-tocopherol 2.8 (µg/g). The minerals determined in mesocarp were: Zn, Fe, Cr, Na, K, P, Mg, Ca, Pb, Cu, Cd, Ni and Mn. K was the main mineral (1.05 g/100 g), followed by Mg (437 mg/100 g), Ca (354 mg/100 g) and Na (52 mg/100 g). Interestingly, the main mineral in seeds was Ca (34 g/100 g) followed by K (402 mg/100 g) and Mg (194 mg/100 g). Seeds contain 6.7% of total fat. Prevailing fatty acids in the seeds are: linoleic (76.29%), oleic (14.18%), stearic (2.81%) and palmitic (6.72%).

Keywords: Celtis australis, dietary fibers, minerals, fatty acids, total phenols

C-80

NUTRITIONAL AND PHYSICO – CHEMICAL PROPERTIES OF STRAWBERRY TREE (*ARBUTUS UNEDO* L.) FRUITS

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The nutritional and physico – chemical properties of ripe fruits of strawberry tree (*Arbutus unedo* L.) were determined. In ripe strawberry tree fruits we analyzed the water content, ash, crude fat, proteins, total phenols, sugar, and the content of vitamin C. Fruits contain 46.7% of water, 23.5% of soluble solids, 0.48% of ash, 118.61 mg/100 g of potassium, 20.63 mg/100 g of sodium, 36.05 mg/100 g of calcium, 9.66 mg/100 g of magnesium, 1.29 mg/100 g of iron, 19.99 mg/100 g of phosphorus, 0.45 mg/100 g of zinc, <0.99 mg/100 g of manganese, <0.99 mg/100 g of chromium, <0.10 mg/100 g of nickel, <1.32 mg/100 g of lead and <0.10 mg/100 g of cadmium. Among nutritionally important components found in fruits were: total fat (0.43%), proteins (0.82%), total phenols (0.59 g/100 g), fibers (18.5 g/100 g), of which insoluble (14.3 g/100 g) and soluble fiber (4.19 g/100 g), titratable acids (5.1 mg/100 g), glucose (6.2 g/100 g) and fructose (17.2 g/100 g). Ripe fruits contained 271.5 mg/100 g vitamin C, of which 255.3 mg/ 100 g L–ascorbic acid and 16.2 mg /100 g of dehydroascorbic acid

Keywords: Arbutus unedo, ascorbic acid, minerals, fatty acids, total phenols

C-81
ANALYSIS OF SOME FOOD COLORANTS BY HPLC

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This project has focused on separation colorants food by green chromatography and determination of tungsten in water sampler. A green chromatographic method of Tartrazine, Sunset yellow and Allura red as a standard sample used in the separation. The method is based on the modification of a C18 column with 0.025 (*v/v*) Triton X-100 aqueous solution and usage of the same surfactant solution as mobile phase without using any organic solvent modifier. Different range of pH used to control the separation and determined effect of pH on chromatographic separation such as retention time, efficiency, peak width, area under peak and symmetry factor of peak separation. High temperature (70°C) has improved the separation of colorants by using TX-100 at pH 6.4 mobile phase and analysis run was carried out within 13 minutes. However, selected colorants decomposed at 120°C due to instability of food colorants at high temperature. Pure water used as a mobile phase at increasing temperature to reduce viscosity and back pressure and enhance mass transfer thereby reduces analysis time for the separation E110, E102 and E129 and their mixture. However, pure water at 120°C provide spilt in the peak shape indicating instability of food colorants at 120°C. High flow rate 3mL/min at elevated temperature 100°C used to decrease the polarity of mobile phase without using any organic modification. The buffer solution pH=6.4 to carried out the separation within 7 minutes. Limits of detection for Tartrazine, Sunset yellow and Allura red found in method used as 0.4, 7.166, 10 ppm, respectively.

Keywords: Food colorant, effect temperature, green HPLC

C-82

PROTEIN INTERACTIONS AND RHEOLOGICAL PROPERTIES OF SKIM MILK HEATED IN THE PRESENCE OF A REDUCING AGENT

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The manufacture of almost all milk products involves some form of heat treatment. Heating milk above 70°C leads to the exposure of the β-lactoglobulin free thiol group. This will initiate thiol-disulphide exchange reaction with other whey proteins or with caseins that contain disulphide bonds (especially κ-casein). This intermolecular aggregation reaction can play a significant role in determining the functional properties of milk products. In this study, the ratio of free thiol groups to disulphide bonds in milk proteins was altered by adding low levels of a disulphide reducing agent to skim milk prior to heating. This milk was subsequently made into acid gels of which the functional properties were examined. The involvement of milk proteins in disulphide bond formation was also investigated using SDS polyacrylamide gel electrophoresis. Adding low levels of reducing agent to skim milk before heating converted native disulphide-bonded polymeric κ-casein to monomers containing free thiol groups. After heating the milk, the levels of β lactoglobulin and α lactalbumin associated with casein micelles increased. The level of α lactalbumin involved in inter-molecular disulphide bonds also increased. This suggests that the monomeric κ-casein may act as a propagator for the thiol-disulphide exchange reaction involving the casein micelles' surface, leading to high levels the denatured whey proteins associating with the casein micelles. This may increase the levels of proteins that can form bridges from one casein micelle to another and to the serum aggregates during acid gelation. Consequently, the acid gel had higher firmness and higher levels of interconnectivities in the network compared with acid gels prepared from control-heated skim milk.

Keywords: Disulphide bonds, thiol reducing agent, thiol-disulphide exchange reactions, milk proteins, acid gels

Acknowledgement: *Ministry of Science and Innovation (FCDL0810) for funding.*

C-83

THE CONTENT OF SUGARS AND SORBITOL AND THE MATURITY RATIO OF ROWANBERRIES AND THEIR HYBRIDS GROWN IN LATVIA

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The rowanberries (*Sorbus aucuparia* L.) are small orange-red fruits of a rowan tree and belong to the family Rosaceae. These berries have been described as an important source of flavonoids, and their antioxidant activity affects reactive oxygen species and lipid peroxidation; therefore they are suitable for production of health-food products. The ripe wild rowanberries have traditionally been used for jellies and jams, but their use as a food ingredient has been less popular because of their bitter taste. Sweeter and less astringent than wild rowanberries are different cultivars of sweet rowanberries and hybrids with other species. They have been studied little in Latvia; therefore, the aim of the current research was to evaluate the soluble solids content, titratable acidity and sugar content of rowanberries and their hybrids Wild rowanberries *S. aucuparia* and 16 cultivars of sweet rowanberries grown in Latvia were analysed. Analyses were done for fresh and frozen fruits of rowanberry cultivars 'Concentra', 'Krasnaya krupnaya', 'Kubovaya', 'Moravica', 'Nevezhinskaya', 'Rossica', 'Zholtaya', rowanberry × pear hybrids 'Alaya Krupnaya' and 'Titan', rowanberry × aronia hybrids 'Burka' and 'Likernaya', rowanberry × harthorn hybrid 'Granatnaya', *S. aucuparia* var. fructolutea, *S. aucuparia* var. sibirica, *S. arranensis* and *S. hybrida*. All experiments were performed at the Latvia University of Agriculture, Faculty of Food Technology. The content of titratable acids (TA) was detected using standard method ISO 750:1998 by titration with 0.1N NaOH. Soluble solids content (SSC) was analysed using standard method ISO 2173:2003 by digital refractometer RX-5000CX. For determination of the sugars (glucose, fructose and sucrose) and sorbitol content, high performance liquid chromatography was used. Results showed that there are significant differences between varieties ($p<0.05$) for all parameters. TA content varied from 0.44–4.12 g malic acid 100 g^{-1} for fresh rowanberries and 0.40-3.90 g malic acid 100 g^{-1} for frozen rowanberries, SSC – 10.55–20.47°Brix for fresh berries and 9.77–17.00°Brix for frozen rowanberries. The highest ratio between SSC and TA was for *S. arranensis* (44.7) and *S. hybrida* (23.9), while the lowest - *S. aucuparia* var. fructolutea (4.2). The results showed that the rowanberries contain 2.48–3.75 g 100 g^{-1} of fructose, 2.01–3.36 g 100 g^{-1} of glucose and 2.54–7.50 g 100 g^{-1} of sorbitol which was the dominant compound in all investigated berries. After freezing and thawing of rowanberries, content of sugars and sorbitol decreased by 5–23% with some exclusion.

Keywords: Rowanberries, sugars, sorbitol, titratable acids, soluble solids

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C-84
VARIABILITY AND STABILITY OF THE CHARACTERISTIC COMPONENTS OF ARONIA

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Fruits of aronia (*Aronia melanocarpa*, Black chokeberry) have high biological and nutritional value. Besides vitamins, minerals, saccharides and organic acids aronia contains a considerable amount of phenolic compounds with antioxidant effect. Aronia extract is used in the food industry for coloring products due to its high content of anthocyanins. The content of these substances is variable depending on the variety, degree of ripeness and climate conditions. Anthocyanins are also unstable during storage and processing technology. Despite this fact, fingerprint of anthocyanins seems to be an appropriate qualitative indicator of raw material – can be used to detect fruit substitution or undeclared coloring of fruit product by aronia extracts or vice versa of aronia fruit juices and jams. The aim of this work was to verify the stability and variability of characteristic components of aronia. We focused mainly on the total phenolic compounds and major anthocyanins (cyanidin-3-galactoside, cyanidin-3-glucoside, cyanidin-3-arabinoside a cyanidin-3-xyloside), but also other constituents such as malic, citric and isocitric acid, mineral and sugar profile were taken into the consideration. The authentic samples (concentrate, juice, raw and dried fruit of aronia) and products from the market were analysed.

Keywords: Aronia, anthocyanin profile, components, variability, stability

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C-85
CHARACTERISTICS OF THE CZECH GARLIC

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Kitchen Garlic (*Allium sativum* L.) is one of the most popular representatives of the family Liliaceae. Garlic is used for direct consumption, food seasoning, as well as in production of drugs. Garlic contains more than 100 different compounds. The content of each substance varies according to the variety, origin, soil, weather, fertilization treatment. Currently, the Czech growers are trying to return the Czech garlic, a traditional crop in the Czech Republic, to the Czech market and they are trying to expand the cultivation garlic areas. Despite an increased consumer demand for the Czech garlic, the import of garlic from abroad, especially from China, remains an issue of prize, contrary to the quality and proper origin labelling. The aim of the study was to choose and evaluate the chemical characteristic of garlic of Czech origin. In the preliminary study for small set of samples there were evaluated the following basic qualitative characteristics of garlic of different origin (Czech, EU, South America, China): color, size, weight, firmness, soluble solids, moisture, density. Furthermore samples sensory qualities, such as external color, firmness, aroma, taste (pungency intensity), were assessed. The pungency of garlic was also objectively evaluated from the amount of enzymatically produced pyruvate as a result of the alliinase activity on the endogenous substrate (*S*-alk(en)yl-L-cysteine-sulfoxide). The main study was focused on the determination of relevant qualitative parameters for set of garlic samples from different production areas in the Czech Republic (harvest 2012) and assessment of (acceptable) minimum (or maximum) limits of selected qualitative parameters of garlic. There were monitoring (verifying) following parameters of pungency and discernment of aroma and taste: the determination of precursors, i.e. alliin and allicin, by HPLC, spectrophotometric pungency test based on the content of pyruvate, the assessment of the characteristic profile and content of volatile substances by SPME–GC–MS.

Keywords: Czech garlic, alliin, allicin, pungency, volatile substances

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C-86

EFFECT OF HIGH PRESSURE HOMOGENIZATION IN COMBINATION WITH THERMAL PROCESSES IN THE AROMA PROFILE OF TOMATO PUREE

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High Pressure Homogenization (HPH) is a common process in the industrial preparation of tomato products. HPH, and other high shear mixers, can fibrillate fiber strands present in tomato, increasing the water binding capacity and the apparent viscosity and reducing the syneresis of tomato products. However, little is known about the effect of HPH in the volatile profile of tomato products. Thermal treatments are also used in the tomato industry with the aim of modify the viscosity of the final products being Hot Break (HB) and Cold Break (CB) the most widely used. The aim of this study was to investigate the effect of these mechanical and thermal processes and their combination in the volatile profile of tomato products. Principal Component Analysis of the total volatile profile showed that the difference between no homogenized samples and HPH processed samples is reduced with the application of thermal treatments. Results also showed that, although thermal treatments have a higher impact in the volatile profile than the HPH processes, the amount of some volatile products from lipid oxidation and carotenoid degradation was decreased after homogenization.

Keywords: High pressure homogenization, tomato, flavour

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C-87
EVALUATION OF EGG YOLK COLOUR

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Egg yolk colour belongs to the most important quality characteristics of eggs. Consuments and food producers usually claim intesively coloured yolk. The egg yolk colour depends on amount and types of carotenoids which deposit to the yolk from feed. The variability of feed composition causes also the variability of egg yolk colour from pale yellow to dark orange. The aim of work was to compare visual evaluation of yolk colour judged by usual method applying La Roche scale with spectrophotometric measurement by AOAC method of beta carotene determination and by new rapid analyser iCheckTM Egg photometr (Bio Analys). The eggs under consideration were purchased on current Czech market. Eggs have originated from various countries and from various types of breeding including some specialities. Yolk colour varied between values 4–13 of La Roche scale. The carotenoid content expressed as beta carotene measured by AOAC method varied between 11–87 mg.kg$^{-1.}$ The carotenoid content expressed as beta carotene measured by analyser iCheckTM Egg photometr was lower and varied between 7.5–68.5 mg.kg^{-1}. Correlation between the colour hue measured visually and the carotenoid content was not proved. The most dark yolks (value 13) had Slovak eggs from cages which contained 28.3 mg carotenoids in 1 kg of yolk, the most pale yolks (value 6) had Czech bio eggs which contained 20.2 mg carotenoids in 1 kg of yolk. The highest content of carotenoids was found in eggs from home hen raising (72.5 mg carotenoids in 1 kg of yolk) whose colour hue had value 10.

Keywords: Egg yolk, colour of egg yolk, carotenoids

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C-88

ZINC CONTENTS IN THE *LONGISSIMUS DORSI* AND SEMIMEMBRANOSUS MUSCLES FOR FIVE PUREBRED PIGS FROM VOJVODINA (NORTHERN SERBIA)

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The content of zinc (Zn) was investigated in *M. longissimus dorsi and M. semimembranosus* for five purebred pigs (Large White–LW, n=6; Landrace–L, n=6; Duroc–D, n=6; Hampshire–H, n=6; Pietrain–P, n=6), produced in Vojvodina, northern Serbia. Zn was determined by flame atomic absorption spectrometry after mineralization by dry ashing. The difference in the Zn content among the five purebred pigs was not significant in the analysed longissimus dorsi (P=0.493) and semimembranosus (P=0.532) muscles tissues. Muscles had no significant effect on the Zn content (P=0.580). The order of the purebred pigs regarding Zn content in the longissimus dorsi muscle samples in mg/100g was: P (2.47–3.17, on average 2.85) > H (2.52–3.07, on average 2.83) > D (2.50–3.01, on average 2.79) > L (2.24–3.45, on average 2.74) > LW (2.32–2.94, on average 2.57). The average Zn content in all investigated longissimus dorsi muscle samples was 2.75 mg/100g. The order of the purebred pigs regarding Zn content in the semimembranosus muscle samples in mg/100g was: P (2.43–3.24, on average 2.83) > L (2.50–2.95, on average 2.74) > D (2.55–2.79, on average 2.70) > LW (2.59–2.80, on average 2.68) > H (2.32–2.87, on average 2.63). The average Zn content in all investigated semimembranosus muscle samples was 2.72 mg/100g. The Vojvodian pig meat showed similar Zn content compared with the values found in other countries.

Keywords: Pigs, *M. longissimus dorsi*, *M. semimembranosus*, zinc

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C-89

CHANGES OF ANTHOCYANINS CONTENT IN 21 CLONES OF *LONICERA KAMTSCHATICA* (SEVAST.) POJARK FRUIT DURING FREEZING AND THEIR RELATION TO OTHER CHEMICAL CONSTITUENTS

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The aim of study was to determine changes in anthocyanins level in selected Klčov´s clones of *Lonicera kamtschatica* (Sevast.) Pojark during freezing at -18°C for 6 month and found out relationship between anthocyanins and dry mater, sugar, organic acids and vitamin C content by cluster analysis. The results of our experiment show that there were no statistical significant changes in anthocyanins during storage by freezing. 21 clones of Lonicera kamtschatica can be set up into 3 clusters, the first with the highest level of anthocyanins and organic acids (LKL-5, LKL-7, LKL-15, LKL-19, LKL-21, LKL-31, LKL-48, LKL-58, LKL-96, LKL-102 and LKL-103), the second with low anthocyanin and organic acids levels accompanied with high ascorbic acid and sugar accumulation (LKL-3, LKL-16,LKL-54), for the third cluster was typical balanced content between anthocyanins and dry mater content and high content of ascorbic acid with organic acids (LKL-2, LKL-16, LKL-14, LKL-18, LKL-20, LKL-33, LKL-35, LKL-42, LKL-49). Anthocyanins level measured in samples was 5.63-14.22 mg/100g. Anthocyanins displayed a positive correlation with antiradical activity of fruits measured by DPPH method (r=0.58).

Keywords: *Lonicera kamtschatica*, clones, antocyanins, freezing

C-90

SYNTHESIS OF (-)-ISOPULEGOL BY CYCLISATION OF THE CITRONELLAL CATALYSED BY THE TRIFLATE OF BISMUTH (III)

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Several processes of synthesis of the menthol were used. The citronellal cyclised in isopulegol by thermal way, microwaves, by using reactives such as the active carbon, the silica gel, the sulphuric acid, the boric acid with or without aluminum oxide, the anhydride acetic, Ni of Raney etc… It finds use in numerous consumer goods as cigarettes, chewing gum, toothpaste, pharmaceutical products etc. [1]. The menthol is widely obtained by means of culture of the plant "Mentha arvenis" in China today. However, it is produced by synthesis in Germany, in United States, and in Japan [1]. In this work, we exploited the triflate of bismuth [2] in the synthesis of this molecule. The isolated products are characterized by the various spectral techniques. A study of optimization of the yields is realized.

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Keywords: Menthol, citronellal, catalyse

C-91

SCYTALIDIUM THERMOPHILUM CATALASE PHENOL OXIDASE OXIDATION AFFECTS ON BLACK TEA PRODUCTION STEPS

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Phenol oxidases have very wide substrate range and final oxidation products of these substrates are quinones, which are highly reactive molecules and polymerize into brown, red or black water-insoluble compounds. Catalases are antioxidant enzymes that catalyze the conversion of hydrogen peroxide to water and molecular oxygen, serving to protect cells from its toxic effects. In our group's previous studies, a bifunctional catalase from *Scytalidium thermophilum* was shown to possess phenol oxidase activity (Ögel 2006). S. thermophilum catalase phenol oxidase (CATPO) was produced constitutively and in a growth-associated manner. Catechol, caffeic acid, chlorogenic acid and catechin were efficiently oxidized by CATPO. Their products were also analyzed in terms of their antioxidant potentials as well as the initial substrates. The oxidized products had higher antioxidant capacities when compared to pure substances. TEAC and FRAP values were revealed. The potential to employ this bifunctional enzyme in tea production process steps was explored.

Keywords: Tea, antioxidant capacity, oxidation, catalase, phenol oxidase

C-92

COMPARISON OF SELECTED ELEMENTS IN VARIETIES OF POPPY SEEDS GROWN IN TWO REGIONS OF SLOVAKIA

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Opium poppy (*Papaver somniferum* L.) is a traditional crop in Slovakia, which is grown on about 2000 ha (in year 2010). Farmers have cultivated this plant mostly for seeds production for culinary purposes. Slovakia belongs to countries with large-scale poppy cultivation and has registered seven varieties of poppy in the European common catalogue of varieties of agriculture crops. The aim of the present work was to evaluate which varieties of poppy among 15 tested ones (cultivated in two regions of Malý Šariš and Vígľaš) could contain the largest amount of toxic and nutritional elements, and whether it is possible to use these elements for geographic authentication of poppy. Eight elements (toxic-cadmium, mercury, lead as well as minerals- magnesium, potassium, calcium, iron and zinc) were determined in varieties of poppy seeds Bergam, Gerlach, Major, Malsar, Maraton. Opal, Orfeus, Aristo, Buddha, Albin, Racek, Redy, MSZB–3, MS–387 and MS–423, by means of atomic absorption spectrometry on graphite furnace and flame. We found out that level of Hg toxic element was higher in the growing varieties of poppy from Šariš, Cd level was higher in varieties grown in Vígľaš but for Pb could not be estimated any trend level. The lowest amount of Hg was measured in Racek variety and the highest one in Major variety in both growing areas (Šariš – from 0.0063 to 0.0078 mg/kg, Vígľaš – from 0.005 to 0.0068 mg/kg). The lowest amount of Cd showed Bergam variety from both growing areas, but the highest level of Cd was found in Opal variety from Šariš and in Albín variety from Vígľaš (Šariš – from 0.211 to 0.313 mg/kg, Vígľaš - from 0.380 to 0.460 mg/kg). The highest amount of Pb rendered MS 387 variety grown in Šariš (0.690 mg/kg) and in Vígľaš (0.940 mg/kg). The lowest Pb content was estimated in Bergam variety (0.116 mg/kg) from Šariš, and in Racek variety (0.09 mg/kg) from Vígľaš region. The levels of mineral elements Zn and Fe in the poppy varieties grown in the Vígľaš region were higher (from 57.6 to 73.7 mg/kg and from 83.2 to 124 mg/kg, respectively) than in region Šariš (40.1–56.6 mg/kg, 38.8–55.1 mg/kg, respectively). The highest level of these elements contained Albin variety, the smallest ones Bergam and Gerlach varieties. All cultivated varieties were rich in Ca levels (Redy variety contained at least 11535 mg/kg and Albin variety 16 415 mg/kg). Albin variety contained the lowest levels of K in both regions (Šariš – 5663 mg/kg, Vígľaš – 6878 mg/kg). The highest levels of K revealed the Bergam variety from Šariš (7967 mg/kg) and Major (9825 mg/kg) and MSZB 3 variety (9564 mg/kg) from Vígľaš. The levels of Mg were approximately the same in all investigated varieties of poppy (Šariš – from 3133 to 3579 mg/kg, Vígľaš – from 3098 to 3622 mg/kg). The elements Cd, Zn, Fe Hg, Pb, K and Ca have shown as the best elemental markers for regional differentiation of poppy varieties, determined by principal component analysis (PCA).

Keywords: Poppy seeds, AAS, mineral elements, toxic elements

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C-93

THE EFFECT OF RATIONALIZATION OF GROWING SYSTEMS ON INTAKE OF OLIGOBIOGENIC ELEMENTS (K, Fe) INTO BARLEY GRAIN

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The influence of applied inorganic nutrients in soil, the method of tillage (conventional and minimalization) on accumulation of Fe and K into the grain of selected varieties of barley was observed. Small-parcel experiments were carried out on fields with size 14 m^2 in three parallels. Four levels of fertilization were done: 1) control – not fertilized variant 2) N70 + P4,36 + K16,6, 3) N60 + P22,7 + K36, 4) N60 + P22,7 + K36 + Ca25. Oligobiogenic elements (Fe, K) in grain of barley were mineralized by the method of Koppová and their content was assessed by the method of flame AAS at instrument VARIAN 240FS. Content of Fe in dry matter of barley grain in analyzed samples were v interval 34.7–109.0 $g.kg^{-1}$ and of potassium 293.1–658.9 $g.kg^{-1}$. Application of macro elements (N, P, K) into the soil had on the accumulation of Fe into the barley grain reduction effect (beside KM2084 variety) and contrary, in comparison with control variant increased influence on intake of mentioned element in winter varieties (beside variety Malwinta) not dependent on the method of amendments of soil by tillage. Content of Fe in grain of barley in dependence of nitrogen, potassium and phosphorus doses precisely correlated with polynomial function of the 2nd degree - with exception of KM2084 variety where decline of Fe content in grain of spring barley varieties was evaluated in fertilized variants (in comparison to control), as well as decline of this element in grain of reducing doses of N and also increased amounts of P and K in fertilizers only in experimental variants with high variability (above 74%). More significant influence of applied tillage (minimalization) was evaluated in varieties of winter barley, where the analogy (as well as high variability) in varieties Malwinta and Graciosa was observed (with spring varieties of barley on Fe content in grain). Content of another element was not significantly influenced by presence (as well as by level) of applied nutrients (N, P, K) in spring barley, not significant difference in winter barley was only in fertilized variants in variety Malwinta, their amount of K in grain was higher in comparison to control variants in both applied types of tillage. In other two variants of winter barley there could not be evaluated continual proportion in content of K in grain of barley from amount of applied nutrients in soil. As well as the accumulation of Fe into grain of barley, also content of K in winter varieties Malwinta and Graciosa is objectively stated by polynomial function of the 2nd degree: in these variants with applied macro elements higher content of K was evaluated, in classical relation to mentioned parameters higher content of P and K increased content of K into the grain, but addition of $CaCO_3$ in fertilizers affected with contrary – reducing effect.

Keywords: Barley, fertilization, macroelements

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C-94

CONTROLLED PRODUCTION OF Se-ENRICHED BIOMASS BY MICROALGAE *C.SOROKINIANA*

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Selenium is a trace element with important nutritional functions due to the enzymatic activity of selenoproteins which are present in many organisms with exception of fungi and higher plants. As many as 25 selenoproteins are found in human containing selenocystein (Sec) incorporated at the active site where strong nucleophilicity of Se atom is required. Sec is the 21st amino acid incorporated into proteins at the UGA codon site if fallowed with Sec insertion sequence located in the 3'-untranslated region of the translated mRNA. Selenoproteins are integral components of 3 major metabolic systems – antioxidant defense, thyroid hormone metabolism and reproductive performance. Se effect on human health is dose dependent. It is essential at low doses and WHO recommends 40 µg/day per adult. Doses higher than 800 µg/day are toxic. Dietary Se occurs in bread, cereal, meat and poultry. In Europe, Se levels are generally low. Natural food supplements such as Se-enriched yeast and microalgae containing bioavailable organic Se forms are far more suitable and less toxic then Se sodium salts. Se-enriched yeast due to batch-to-batch variability have inconstant selenides composition with up to 85% selenomethionine (SeMet) which can result in Se accumulation to toxic levels in tissue. This study offers new insight in continual cultivation of Se-enriched *C.sorokiniana* in stirred aerated illuminated tank. Steady-state productive phase lasts for weeks providing biomass of constant Se content of up to 70% SeMet accompanied by various other selenides and selenoproteins. Microalgae were grown in a conventional culture medium containing 5–50 mg/L selenate. Constant productivity determined dilution rates increase. Se concentration in the culture medium was analytically monitored for every dilution rate. Viability of the colony was checked by biomass concentration, total chlorophyll content, oxygen production and maximum quantum efficiency of PS II. Although Se exposure slightly lowered culture viability it grew well at all selenate concentrations. Based on productivity and yield on light energy, selenate concentration of 40 mg/L provided best results. Intracellular SeMet content grew linearly with selenate in solution reaching 40 µg/g at 40 mg/L selenate in solution. Fractionation in a 20–80% ammonium sulfate revealed that selenoproteins were present in Se-enriched culture, while control non-Se grown culture was negative. This procedure separated the 50 kDa selenoprotein, apparently similar to subunits of the carbon fixation enzyme. Se-enriched microalgae presents Se source of high bioavailability. High productive, low cost operation without contamination risks provides Se-enriched biomass with economical down-streaming. High biotransformation rate of selenate to intracellular SeMet and selenoproteins presence shows that C. sorokiniana can be successfully employed as the system for expression of selenoproteins.

Keywords: Microalgae, *C. sorokiana*, selenoproteins, Se-enriched biomas

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C-95

EFFECT OF CONCENTRATION BY BOILING AT ATMOSPHERIC PRESSURE ON MINERAL CONTENT OF WHITE AND RED GRAPE JUICES

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It is known that some processes cause changes in physical and chemical properties of foods. Pekmez (molasses), concentrated by boiling in open vessel at atmospheric pressure from sugar-rich fruit and vegetable juices, is a traditional product consumed widely in Turkey. In this works, heat treatment during the concentration by boiling in open vessel at atmospheric pressure was examined the effect on major and minor elements of grape juices. After the white and red grape juice were concentrated in open vessel at atmospheric pressure up to 50, 60 and 70 oBrix, minerals (Ca, B, Cr, Cu, Fe, K, Mg, Mn, Na, Ni, P, Pb, S and Zn) in samples were determined by ICP-AES. The content of Ca, B, Fe, K, Mn, and P increased in both grape juices concentrated up to 50 and 60 oBrix, and decreased in 70 oBrix. The highest decrease was occurred to be 96.83% in Pb of red grape juice during the concentration process. Also, the decrease in Cu, Ni, S and Zn content of the red grape juice was found as 70.39, 69.78, 63.15, and 53.94%, respectively. While, similar to the red grape juice, the most reduction was seen to be 79.23% in Pb of white grape juice concentrated up to 70 oBrix, decrease in the content of Ni, Zn, Fe, Cu and S was determined to be 74.37, 45.30, 34.88, 31.15 and 26.16%, respectively.

Keywords: Grape juice, concentration, mineral content

C-96
CONTENT OF LIPOPHILIC VITAMINS A AND E IN CAPRINE COLOSTRUM

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Colostrum is the first milk secreted after parturition. It contains plenty of immunologically and physiologically active components, including liposoluble vitamins retinol and tocopherol. Both vitamins are essential to ensure a good functionality of the immune system. The role of vitamin E is its antioxidant function. It protects polyunsaturated fatty acids in cell membranes and lipoproteins from oxidation. It is extremely important for newborns which exhibit a greater sensitivity to oxidative damage than adults. Vitamin A plays a critical role in vision, reproduction, cell differentiation as well as growth and development. The aim of this study was to evaluate the total fat soluble vitamins retinol and tocopherol in caprine colostrums by means of ultra-performance liquid chromatography (UPLC). Colostrum samples were obtained from private goat farms of Moravia and Bohemia. There were analyzed a total of 93 samples, each in a parallel setting. Samples were taken during the period February to June 2012. Samples were prepared by saponification with methanolic potassium hydroxide solution. To avoid vitamin losses or degradation during the procedure, antioxidants were added to the sample extraction media. The saponification step was always followed by the liquid-liquid extraction with non polar organic solvent. Determination of final samples was provided by UPLC method with isocratic elution (methanol/water – 98/2 w/w) and UV detection. The wavelength at which the response was greatest was 325 nm for retinol and 295 nm for tocopherol. The actual content of retinol determined in samples of caprine colostrum ranged from 1.54 $mg.L^{-1}$ to 18.46 $mg.L^{-1}$ and of tocoferol from 2.04 $mg.L^{-1}$ to 23.95 $mg.L^{-1}$.

Keywords: Retinol, tocopherol, caprine colostrum, UPLC

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C-97

DETERMINATION OF TARGET VITAMINS IN RAW BOVINE MILK IN THE CZECH REPUBLIC

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Milk is unique animal product secreted by mammary gland during the lactation with high content of natural occurring biological active substances that exert various important roles in metabolism. This study is concerning the content of lipophilic vitamins retinol, tocopherol and hydrophilic vitamin riboflavin in raw cow milk originating from bulk tank milk samples obtained in conventional and ecological dairy farms in Czech and Moravia during the summer 2012. For the separation of target lipophilic vitamins was used saponification with methanolic KOH followed with hexane extraction. Vitamins from this extract were determined by reverse – phase HPLC with UV detection at wavelengths 325 nm for retinol and 205 nm for tocopherol. Acid extraction of proteins at pH 4.6 with HCl is important for riboflavin analysis. The concentration of riboflavin was detected fluorometrically (excitation 450 nm, emission 520 nm). The content of vitamins in milk samples ranged from 820–1280 $\mu g.L^{-1}$ for riboflavin, 1331–1950 $\mu g.L^{-1}$ for retinol and 1056–3059 $\mu g.L^{-1}$ for tocopherol.

Keywords: Retinol, tocopherol, riboflavin, bovine milk, HPLC

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C-98

HYGIENIC QUALITY OF FISH DEPENDING OF CADMIUM (Cd) AND LEAD (Pb) CONTENT IN BOTTOM SEDIMENTS OF WATER RESERVOIRS KOLÍŇANY

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Fish meat is an optimal nutrition, meeting the requirements of rational nutrition. It provides the organism healthy, eupeptic material rich in fully-fledged proteins, minerals and vitamins. Fish meat contains amino acids, unsaturated fatty acids, vitamins B, A, D as well as minerals. The aim of this paperwork was to provide information on Cd and Pb content in biotic and abiotic fish environment: bottom sediments from water reservoirs (WR) Kolíňany, analysis of Cd and Pb content in different parts of Common carp (muscle, liver, kidney). The selected area is located east of the village Kolíňany, Nitra district. Bottom sediments and fish were sampled the 18th of November 2010, 5 samples were taken under the law regulations. Each sample was dried and analysed for Cd, Pb content using the AAS method. Final analytical analysis was performed with the VARIAN AA 240 FS device. The Cd and Pb content in fish body parts were compared with Codex Alimentarius (CA) SR. Bio-Concentration Faktor (BCF) shows the ratio between the concentrations of chemical in an organism through the body, respectively, in specific tissue and concentration of that chemical. The results of the analysis show that the limit values for Cd and Pb in the sediment were not exceeded. The average Cd content in the sediment is 3.306 mg.kg^{-1}, compared with the limit value of 10 mg.kg^{-1}, it represents a 33.0% content. Pb content was 15.12 mg.kg^{-1}, compared with limit value (750 mg.kg^{-1}) it represents 2.1% content. The content of hazardous elements doesn't predict their increasing input to the fish organism, although all hazardous elements have the ability to accumulate in animal tissues. This accumulation is directly proportional to the fish age. There were caught 4 pieces of carps with weight of 1285–2702 g. The tissue was sampled from muscle, liver and kidney and each was determined by Cd and Pb content. The results indicate the accumulations of different hazardous elements in different parts of fish body. Cd content was lowest precisely in the fish muscle (0.0673 mg.kg^{-1}), then its content increased in order liver (0.0721 mg.kg^{-1}) and kidney (0.1342 mg.kg^{-1}). In the fish muscle the highest allowed content for Cd, which is defined in CA SR (0.1 mg.kg^{-1}) has not been exceeded. Similar results were achieved with Pb samples analyses. The highest accumulation of Pb was analysed in kidney (0.6112 mg.kg^{-1}), followed by liver (0.2722 mg.kg^{-1}) and the lowest in fish muscle (0.0328 mg.kg^{-1}). The highest allowed content of Pb (1.0 mg.kg^{-1}) in the fish meat has not been exceeded. Within biotransformation we can determine the order of analyzed apparatuses in terms of the amount of analyzed Cd, Pb content Cd, Pb: kidney>liver>muscle Despite the low content of hazardous elements in the sediment, accumulation in the fish tissues is possible, because the fish are characterized by relatively high in fat, which accumulates these elements highly and fish age causes increasing their content as well.

Keywords: Cadmium, lead, carp, bottom sediment

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C-99
DETERMINATION OF SELENIUM IN DIETARY SUPPLEMENTS BY HR-CS ETAAS

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Selenium (Se) nutritional essentiality was established in 1973, when its role at the active site of glutathione peroxidase was determined. The biological functions of Se are exerted as a component of the 21st amino acid, selenocysteine (Sec), an integral component of the active site of selenoproteins. Important selenoproteins include glutathione peroxidase and thioredoxin reductases that act as antioxidants, protecting cells from oxidative damage and, the iodothyronine deiodinase enzymes that are involved in thyroid hormones metabolism. The recommended daily allowance (RDA) for Se is 55 µg/day for adult men and women and the acceptable upper limit 300 µg/day, although it has been reported that Se intakes above the RDA, might reduce certain types of cancer. The important role of Se in health triggered the development of a plethora of selenium-enriched dietary supplements, followed by a growing popularity of these products among consumers. Control of Se levels in dietary supplements is particularly important due to the narrow gap between essentiality and toxicity of this element. The aim of this work was to optimize and validate a simple and rapid method for total Se determination in dietary supplements by high-resolution continuum source atomic absorption spectrometry (HR-CS AAS) and assess compliance with the labelled values. In this work, 8 dietary supplements containing Se, representing 6 different brands names, purchased in health food stores and supermarkets were evaluated. According to the labels, selenium was present as sodium selenite (n=1), L-selenomethionine (n=3), Se-enriched yeast (n=2), one formulation referred sodium selenium, and one other just selenium, without specifying origin. Samples were prepared by microwave digestion (MARS X) with nitric acid and hydrogen peroxide. Analyses were carried out in a high-resolution continuum source atomic absorption spectrometer ContrAA 700 (Analytik Jena). A mix solution of palladium nitrate and magnesium nitrate was used as matrix modifier. Due to the sample matrix interferences, the standard addition method was found to be the most appropriate. Analytical quality control was achieved by using certified reference material SELM-1. Replicate analyses were in the range of certified values. Intra- and inter-day precision was always better than 10% and low limits of detection and quantification were obtained. HR–CS ETAAS is a simple and rapid method for Se determination in dietary supplements. Se content varied between 23.0 µg and 208.7 µg of Se per capsule/tablet. Sample results are in accordance with the values specified by the manufacturer, ranging between -10 and +13% for the labelled value.

Keywords: Selenium, dietary supplements, microwave digestion, HR–CS ETAAS

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C-100
INTERACTION OF ADDED INORGANIC IRON(II) WITH MILK BABY FORMULA INGREDIENTS

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Addition of iron in milk baby formula is common practice in their production, because the basic raw material does not contain enough of these essential trace elements. Iron(II) sulfate is most often used as inorganic source of iron. Biological availability of iron depends of iron's oxidation number and type of compound used. During production of milk formula in powder form out of solution, changes of added iron(II) out of sulfate happen during the production process and later on during dissolution of powder for preparation of meals. In this work are shown the research results of interactions between inorganic iron and other ingredients in three different domestic milk baby formulas. Distribution of iron in solution and in form of particles, which could be separated by centrifugation, is approx. 30% i.e. 70%, respectively. Iron in sediment is agglomerate of base-iron(III)-proteinate, and in supernatant is iron(II)-ion dominant. Presumption is that biological availability of iron is greater from solution. The results of this examination on study of iron speciation in researched products, studies that are currently taking place, will contribute with evaluation of biological value of added iron.

Keywords: Iron addition, iron form, milk baby formula

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C-101

DETERMINATION OF MACRO AND MICRO MINERAL COMPOSITION OF READY-TO-EAT "BABY LEAF" VEGETABLES BY USING MICROWAVE DIGESTION AND HIGH-RESOLUTION CONTINUUM SOURCE ATOMIC ABSORPTION SPECTROMETRY

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Vegetables have almost all of the essentials nutrients for human metabolism, being it consumption recommended and linked to the promotion of good health. However, it is not always easy to coordinate the busy lifestyle of today's modern society with the time required to prepare a healthy meal with fresh vegetables. Ready-to-eat vegetables packages offer the consumer a nutrient rich fresh product. Producers are usually more concerned with the appearance and safety of fresh-cut products, but fresh vegetables respond to minimal processing, increasing their metabolism, which can lead to nutritional losses. Baby leaf salad is a ready-to-eat product, prepared of young leaves, harvested at very early stage of maturation and in an active metabolic stage. The producers defend that this leaves have a greater stability during shelf life. Nowadays, accurate nutritional information is needed due to its use by public agencies and agricultural industries to promote different traditional fresh products. The basic functions of minerals in biological systems have been known for many years (in the proteins, lipids and carbohydrates metabolism; as main cellular and structural building materials; role in osmotic pressure and acid/base regulation). Leafy vegetables are among the foods that can guarantee the presence of these micronutrients in a healthy diet. The aim of this study was to optimize and validate a method for determining macro- (K, Na, Ca and Mg) and micro- (Fe, Mn, Zn and Cu) mineral composition of eight read-to-eat fresh-cut "baby leaf", which included microwave mineralization of the samples. The samples were: green and ruby red lettuce, wild rocket (from conventional and organic production), swiss chard, watercress, spinach and lamb's lettuce. The mineral content was determined in a high-resolution continuum source atomic absorption spectrometer (HR–CS–AAS), with flame atomization for the quantification of K, Na, Ca, Mg, Fe, Mn and Zn, and with electrothermal atomization for Cu analysis. The determination of the LOD, LOQ, linearity range, intra- and inter-day precision, accuracy of the method and the analysis of a certified reference material (BCR®-679 white cabbage) was done to validate the proposed methodology. The developed methods showed analytical results suitable to the analysis of minerals in several vegetal matrixes, with low LOD and LOQ and high reproducibility for all minerals under study. The recovery assays performed showed good recoveries (92–110%) and the certified values of the reference material were similar with values obtained by this method. The "baby leaf" analysed show a mineral profile similar to the values presented by nutritional tables for the more mature vegetables. The vegetables showed a higher content of K (200–500 mg/100 g) and Ca (65–325 mg/100 g) in the macro mineral composition. Fe was the principal component of their micro mineral fraction (0.5–1.7 mg/100 g).

Keywords: Baby-leaf vegetables, ready-to-eat salads, minerals, microwave digestion, HR–CS–AAS

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C-102

CALCIUM AND MAGNESIUM BIOAVAILABILITY AND TISSUE DISTRIBUTION IN ADULT RATS AFTER MODEL MAILLARD PRODUCTS CONSUMPTION

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Introduction: From the eighties, it was well-established the ability of soluble and insoluble melanoidins, the final Maillard reaction products (MRP), to bind metal cations and form stable complexes. Thus, the absorption and the bioavailability in vivo of Ca, Mg, Fe, Cu and Zn may be compromised by the behaviour of MRP as chelating agents. The purpose of this study was to investigate the effects of the consumption of MRP from glucose-lysine model system heated 150ºC-90 min on calcium and magnesium bioavailability and tissue distribution in adult rats.

Material and methods: Equimolar mixture of glucose-lysine-HCl (GL) (40% moisture) were heated in open recipients in an oven at 150ºC for 90 min to obtain the GL90 model system. The GL90 sample was added to the AIN-93G diet (Control diet) to reach a final concentration of 3%. Thirty-six weanling Wistar rats were randomly distributed into three groups (12 animals per group) and one of them was sacrificed by anaesthesia overdose at day 0 to analyse their initial calcium and magnesium body content. The remaining groups were assigned to one of the dietary treatments. The intake was monitored during the whole experimental period. To investigate the calcium and magnesium balance during 3 months, two different balances were carried out: on one hand, during the 12-week faeces and urines were collected as a pool; on the other hand, at the end of the experimental period, animals were sacrificed and a global calcium and magnesium balance was performed by analysing the carcasses. Calcium and magnesium in diets, faeces, urine and some organs as well as in carcass were determined by using AAE.

Main results: Calcium absorption and retention were stable after consumption of model MRP assayed as well as its bioavailability during last week of experiment or the whole period considered. Consequently none modifications were appreciated in calcium tissue distribution. In the case of magnesium, the last week balance showed a decrease in its faecal excretion leading to higher absorption and retention. So that magnesium digestibility (A/I %), retention efficiency (R/A %) and bioavailability (R/I %) significantly increased after consumption of GL90 diet compared with the Control group (23, 19 and 38%, respectively). However, these modifications should be punctual for the last week of the assay since after the entire experiment the magnesium body content and its retention did not vary, also supported by the stability of magnesium content and concentration in organs.

Conclusion: Although improvements of magnesium digestibility and bioavailability were detected in the last week of the assay, the intake of glucose-lysine model MRP did not modify the calcium or magnesium global bioavailability and tissue distribution. These results are in agreement with previous research of several authors who stated that that calcium should be one of the minerals with least affinity to form MRP complexes, followed by magnesium.

Keywords: Maillard reaction products, magnesium, calcium, bioavailability, tissue distribution

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C-103

VERIFICATION OF SIMPLE METHOD FOR SIMULTANEOUS DETERMINATION OF PIGMENTS IN FRUITS. AN EFFECT OF SONICATION

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Determination of natural pigments requires the use of optimal sample preparation procedure including the appropriate extraction reagent. In our experiments we verified applicability of two developed methods (Nagata and Yamashita, 1992; Yanget al., 1998) for simultaneous determination of chlorophylls (a, b) and carotenoids (lycopene, β-carotene or total carotenoids). The process of sample preparation was slightly modified. The homogenized samples of strawberries, apricots and raspberries were individually weighed and the pigments were extracted with either acetone-hexane (4:6) mixture or acetone (80%) in homogenizer in order to compare the effect of solvent on extraction of the pigments. In other parallel variants following the homogenization, the method of sonication was applied to investigate the effect of ultrasound on the yield of the extracted components. The optical density of each supernatant was measured in the range of wave lengths from 360 to 780 nm. The obtained spectral data were confronted with the previously proposed set of equations designed to calculate the pigment content. The statistic evaluation of the results showed that the combination of mixing and sonication improved extraction of the pigments in both types of solvents. The amount of pigments estimated from the proposed equations in case of apricot and raspberries were comparable to those observed by other authors. However, the results obtained using acetone-hexane extract of strawberries appeared to be problematic in terms of the determination of β-carotene content, giving negative values. This was most probably due to the similarity of the absorbance values of all three pigments. Although the method, originally developed for tomato, was successfully used for determination of the pigments for various plants, our results indicate the limitations in use of the proposed set of equations for plant samples with comparable amounts of studied pigments.

Keywords: Natural pigments, chlorophylls, carotenoids, sonication

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C-104

SELECTED FACTORS INFLUENCING THE ABILITY OF *BIFIDOBACTERIUM* TO FORM BIOGENIC AMINES

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The ability of typical probiotic culture of *Bifidobacterium* to produce biogenic amines could be considered a contrastive feature to the beneficial dietary effect on the human health. The aim of this pilot study was to evaluate the decarboxylase activity of *Bifidobacterium animalis* subsp. *lactis* CCDM 239. Selected factors (pH 4.5 and 5.0; the contents of NaCl 0-2.0% (w/v), glucose and lactose 0-1.0% (w/v)) influencing *in vitro* biogenic amine production were studied. The kinetics of the biogenic amine production under the above mentioned conditions was also monitored. The compositions of cultivation broths were adjusted according to the observed factors (concetrations of NaCl, glucose and lactose; pH values were controlled using NaOH or HCl). The detection of biogenic amine content (histamine, tyramine, phenylethylamine, tryptamine, putrescine, cadaverine, spermine, spermidine) was carried out in the supernatants of inoculated broth (MRS enriched with precursors of biogenic amines: arginine, ornithine, lysine, tyrosine; each at the concentration of 3.0 g/L) after the cultivation (48 h, 37±1°C). For the analysis of biogenic amine contents, HPLC after the pre-column derivatisation with dansyl chloride was used. In most of cases the low concentrations of putrescine and tyramine were monitored (<15 mg/L). Simultaneously, it was found out that the addition of fermentable saccharides and NaCl seemed to have supporting effect on the decarboxylase activity of tested *Bifidobacterium*. The determined concentrations usually cannot themselves cause the toxic effects for health of consumers. Nevertheless, the toxicological reactions can be observed in sensitive and/or weakened individuals already after the ingestion of relatively low biogenic amine concentrations. Therefore, the usage of *Bifidobacterium* combined with other decarboxylase positive bacterial cultures can contribute to the total amounts of biogenic amines in dairy products and subsequently seriously endanger consumers´ health. The next step of the research will be the observation of biogenic amine formation in the real food matrix during cheese production/manufacture of fermented dairy products.

Keywords: Biogenic amines, *Bifidobacterium*, external factors, kinetics of biogenic amine formation, tyramine

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Strategies to improve food quality and safety

D-1
IMPACT OF OHMIC HEATING ON FURAN AND OTHER VOLATILES IN VEGETABLE PUREE

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Processing contaminant furan, classified by IARC as a probable human carcinogen (group 2B) is formed during heating of food. Vegetable based baby food is one of the commodities in which the highest levels of this volatile contaminant appear repetitively, due to used raw materials and also because of usage of heat when the final product is sterilized. Because vegetable puree represents an important component of babies' diet, effective measures should be taken to protect their health. Alternative ways to conventional retorting are tested to minimize furan formation during heat treatment. One of such approaches is ohmic heating that uses electric current to heat the food up. We used head-space solid-phase microextraction coupled to gas chromatography – mass spectrometry (HS–SPME/GC-MS) to analyze furan and volatile fingerprints in vegetable purée treated by conventional retorting method or ohmic heating. The levels of furan in ohmic heated puree were half of those observed in retorted baby food, specifically, the levels were in average 20 against 40 μg/kg. While in the case of ohmic heating the levels of furan seemed to be time independent the amount of furan was slightly increasing with the time of retorting. Terpenes (terpinolene, γ-terpinene, limonene, myrcene and α-pinene) were identified as the most abundant compounds in both types of samples. Evaluation of volatile fingerprints using principal component and linear discriminant analysis resulted in distribution of the samples in two groups, according to the way of sterilization.

Keywords: Furan, volatiles, baby food, SPME–GC/TOFMS

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D-2

MIGRATION OF DIPROPYLENE AND TRIPROPYLENE GLYCOL DIACRYLATES FROM PACKAGING MATERIALS INTO FOOD SIMULANTS

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The dipropylene glycol diacrylate (DPGDA) and tripropylene glycol diacrylate (TPGDA) were found in the packaging materials (films consisting from printed paper laminated with LDPE) in levels up to 0.44 mg/kg and 0.22 mg/kg, respectively. The presence of both solvent residuals caused unpleasant smell of crystalline sugar in stickpacks made of these packaging materials, the content of diacrylates in sugar was up to 0.23 mg/kg. The migration tests into food stimulants (10% and 95% ethanol, 3% acetic acid and olive oil) at 40°C for 10 days were made using GC-MS method for diacrylate determination. The highest migration 0.13 mg/dm^2 of DPGDA was found into 95% ethanol. Besides mentioned outcomes the poster will present *(i)* the results of study of the course of DPDGA and TPGDA migration from two laboratory prepared samples of similar packaging materials into 10% ethanol as well as 95% ethanol and *(ii)* the health risk assessment for consumers of food containing DPDGA and TPGDA.

Keywords: Migration, food packaging, diacrylate, printing

D-3
HEAVY METALS CONTENTS IN SOME FOOD FROM EASTERN ROMANIA

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Heavy metals toxicity is the result of their interactions with the enzymatic systems from the animal cells or some constituents of cells membranes. The interactions of heavy metals with usual elements from diet (Pb, Cd) have an important role in acute and chronic toxicity. Population can be contaminated with heavy metals by ingestion of contaminated or polluted food or water. The concentration of heavy metals in food products is varied, depending on their origin, storage conditions and progressing technologies. The study presents the results obtained in 2011 year of the metals (Pb, Cd, Cu) concentrations from 206 vegetables harvested from rural area on Eastern Romania (53 villages). The Pb and Cd contents were analyzed in vegetable samples [63 tomato,41 cucumber,50 peppers,15 bell pepper,13 carrots,7 eggplant,17 bean pod] and the copper contents was analyzed in 48 tomato samples(treated with copper sulphate) Trace elements concentrations were analyzed by atomic absorption spectrophotometer, using Schimadzu 6300 Atomic absorption spectrometer (AAS) - with graphite furnace atomizer. In all analyzed samples these metals were found. Generally, a wide variation between individual samples was observed The contents of lead to varied between 0.007 mg/kg (cucumbers, bell pepper) and 0.04 mg/kg (pod) and the contents of cadmium to varied from 0.017 mg/kg (tomatoes, peppers) and 0.024 mg/kg (bell pepper). The contents of Copper from tomatoes to varied between 0.0006 mg/kg and 0.003mg/kg. The presence of heavy metals in vegetable samples require monitoring of their, in order to prevent pollution of the environment.

Keywords: Heavy metals, vegetables, Romania

D-4
ASSESSMENT EXPOSURE AT CHEMICALS POLLUTANTS ON FOOD

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Ever since human have become aware that health is inseparably linked to an impact and healthy environment, the control and reduction of pollution have become the focus of worldwide concern. Investigation on possible health and environmental hazards involved have led many industrial countries to restrict or ban the use of chemicals (pesticides, heavy metals) and enforce the tolerance levels for the residues in food and feeds. The aim of the study was to investigate the variation of some chemical pollutants with cancer risk (nitrate/nitrite, heavy metals, pesticides residues) in some food (vegetables, meat, milk, daily diets) from the Romania area, in 2010–2011. The concentration of the nitrates/nitrites and the heavy metals [Cu, Cd, Pb, and Zn] were investigated in 5386 food samples (vegetables (1364 samples), meat and meat products (2743 samples), milk and dairy products (952 samples), total diets (327 samples). The foods were harvested from the Romania area (37 districts), in 2010–2011. In the milk, vegetables, meat and total diets were analyzed the metals (Cd, Pb, Zn, Cu) by absorption spectrophotometer method. The nitrate/nitrite contents were determined by colorimetric method and the pesticides residues by gas-chromatography. In all analyzed samples these chemical pollutants were found. Generally, wide variations between in individual samples were observed. Nitrate/nitrite contents were, generally, in normal limits and the total diets contained quantities below acceptable daily intake. The analysis of results obtained showed that in food was found the heavy metals in varied concentrations but in the admissible limits. The results show that the pesticide residues are present in all food analyzed. The determinations of chemical pollutants in food are important in environmental monitoring for the prevention, control and reduction of pollution as well as for occupational health, legal, decisions and epidemiological studies.

Keywords: Chemical pollutants, food, Romania

D-5
NOVEL APPROACH TO IDENTIFY MELAMINE AND STRUCTURAL ANALOGUES USING RAMAN SPECTROSCOPY

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Melamine and structural analogues have been involved in the contamination of pet food (USA, 2007) and powder infant formula (China, 2008) with the objective of boosting the apparent protein content in feed and food. These events have caused the death of babies and animals and in consequence, a worldwide food security concern. A lot of methods based on different analytical techniques have been developed to detect melamine in different matrices. However there is a need for simple, specific and rapid methods to discourage the adulteration with melamine of feed and food products. The vibrational spectroscopic characteristic of melamine and its structural analogues are presented as an advantage for using Raman spectroscopy as a simple and fast technique to detect these adulterations. In this study a fast screening method for the detection of melamine and its structural analogues cyanuric acid, ammelide and ammeline in soybean meal has been developed. The Raman spectroscopic method developed in this study can detect melamine and cyanuric acid in soybean meal at concentrations as low as 0.1%. This method could easily be extended to other feed or food matrices.

Keywords: Feed analysis, melamine, Raman spectroscopy

D-6
SUMMARY OF SENSORY DEFECTS OF BEVERAGES CAUSED BY MICROORGANISMS

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In the non-alcoholic beverages, particularly in non-carbonated, there are various defects, such as turbidity or sediment, which may be caused by microbial contamination. The aim of this study is to summarize the microbial and sensorial defects in soft drinks which have occurred over the last 10 years. Among the defects which may occur in the beverage, the scent changes are caused by either activity of microorganisms or chemical changes. From samples showing marks of defects microorganisms that cause these defects were isolated. Isolated microorganisms were identified (using PCR techniques). In non-carbonated soft drinks occurred acetic acid bacteria of the genus Asaia, which had been isolated from such non-carbonated drink with strawberry concentrate, Gluconacetobacter bacteria isolated from green tea. These bacteria cause the creation, in particular, of sediments and floating villi in beverages. Another agent causing sensorial changes are bacteria Alicyclobacillus, which isolated from orange and apple juice. These bacteria cause the formation of undesirable odors derived from guaiacol. The molds and yeasts of different species cause the sensory defects of beveragestoo, creating an unacceptable appearance of beverages by growing the mycelium, but they are also capable, under certain circumstances, to produce substances such as 1,3-pentadiene, which causes strong odor and devalues drink by this way.

Keywords: Beverages, sensorial changes, guaiacol, 1,3 pentadiene

D-7
IDENTIFICATION OF POTENTIAL ANTIGENS FROM SUB-FRACTIONATED *CRONOBACTER SAKAZAKII* CELL MEMBRANE

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Bacterial contamination of food and industrial products can pose a threat for potential consumers. The genus Cronobacter represents an opportunistic foodborne pathogen implicated with serious illnesses among neonates and infants following the consumption of powder infant formula. Although, the incidence is low, the high mortality rate forced the World Health Organization to replace genus Cronobacter together with Salmonella into the risk category A of food-borne pathogens. Currently, all the seven species of genus Cronobacter are considered to be pathogenic, albeit epidemiological studies indicate variations in their virulence. This work presents the study of bacterial membrane proteins with their potential antigenic aspects and their consequent usage for the development of immunochemical identification tests. The protocols for the sub-fractionation have been established and the distinct membrane fractions were confirmed via commercial antibodies raised against membrane marker proteins. The sub-fraction protein content was studied using mass spectrometry approaches and the immunogenic properties of isolated proteins were tested via polyclonal antibodies prepared against whole *Cronobacter sakazakii* cells.

Keywords: Cronobacter, membrane proteins, mass spectrometry

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D-8

USE OF OZONE GAS TO LIMIT THE GROWTH OF MOLDS AND YEASTS IN THE ROOM OF MATURATION OF FRESH CHEESES. PRELIMINARY RESULTS

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In the manufacture of fresh cheeses one of the main drawbacks is the mold proliferation on the surface of the product. Therefore, the use of chemical agents with antimicrobial activity is almost mandatory. However, the industry's interest is oriented toward alternatives preferably in line with consumers expectations for food products with a short list of ingredients. In this study, the antimicrobial efficacy of ozone against molds and yeasts that contaminate the rooms of fresh cheese maturation has been tested. The experiment was conducted using 2 adjacent rooms with a volume of 360 m^3 each. Five successive batches of cheese obtained in as many days were considered for experimental purposes. At the end of the salting phase (in brine for 48 h) the cheeses of each batch were equally split between the 2 rooms. The cheeses were settled on plastic shelves located in the opposite side of the door. In one room were placed 4 ozone generators (2 near the door and 2 in the opposite side) operating according to corona discharge with a production capacity of 5 g ozone/h each one. The emission of ozone was continued for 28 consecutive days from 6:00 pm to 4:00 am. The microbial samples were taken by scrubbing 100 cm^2 surface area both for shelves (accurately sanitized before the beginning of the trial) and for cheese. The scrubs were collected the day after the cheese manufacture, at the end of first, second, third and fourth week of maturing. In all cases the collections were taken at 8:00 AM. At the fourth week, microbial analysis was also conducted on 25 g of crust cheese removed for a thickness of about 2 mm. The molds and yeasts cells collected were submitted to grown on DRBCA AGAR BASE and OGYE AGAR BASE at 25°C for 5 days. The scrubs coming from the room used as control showed molds charges that were in the order of magnitude of 1 log in the first sampling time rising up to 3–4 logs after 28 days both for shelves and cheeses. The yeasts showed more variable values in the first sampling time (from 1 to 3 logs) and higher values at the end of the experiment (up to 5 logs). In the room submitted to ozone treatment the microbial behavior was completely different. The molds were almost absent from all the surfaces tested already at the end of the first week of maturing time and this situation persisted until the end. The yeasts showed a linear decrease until disappearing at the end of the third week. In conclusion, the employment of ozone in gaseous form seems to be able to eliminate molds and yeasts both from shelves surface and from the crust of fresh cheeses. That of ozone is a technology relatively easy to use that leaving no residual is also environmental friendly. It was defined as GRAS by the US–FDA and employed as direct food additive. In Italy the Ministry of Health in 1996 recognized the ozone for the treatment of water and air as a tool for the sterilization of confined environments contaminated by viruses, bacteria, spores, molds and mites.

Keywords: Fresh cheese, ozone, mold, yeast

D-9

COMPARING THE GROWTH OF B. CEREUS AND PRODUCTION OF ENTEROTOXIN IN COW´S, GOAT'S AND SHEEP'S MILK

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In goat's, sheep's and cow´s milk inoculated with a strain *of Bacillus cereus*, *B. cereus* growth and diarrhoeal enterotoxin production was tested. Enzyme-linked immunosorbent assay (ELISA) was used for enterotoxin detection, *B. cereus* was count by the plate method in accordance with ČSN ISO 7932. The influence of storage conditions (8°C, 15°C, 22°C) and of the type of milk on monitored parameters was evaluated. The results of these model experiments conclusively linked multiplication of *B. cereus* and subsequent production of diarrhoeal enterotoxin with storage temperature and the type of milk. In raw cow´s, goat´s, and sheep´s milk, the *B. cereus* growth rate and production of diarrhoeal enterotoxin can be suppressed by competitive microflora.

Keywords: *B. cereus*, diarrhoeal enterotoxin, cow´s milk, goat's milk, sheep's milk

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D-10

EVALUATION OF GOAT, SHEEP, COW MILK, AND DAIRY PRODUCTS SAFETY IN TERMS OF STAPHYLOCOCCAL ENTEROTOXINS PRESENCE

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In sheep, goat and cow milk inoculated with a strain of *Staphylococcus aureus*, staphylococcal growth by plate method in accordance with EN ISO 6888-1 on Baird-Parker agar and staphylococcal enterotoxin C production by enzyme-linked immunofluorescent assay (ELFA) was detected. The automated instrument miniVIDAS® was used for this detection. Fresh cheeses were made from the the different milk types. Cheeses were also tested then for staphylococcal enterotoxins presence. The influence of poor storage conditions (15°C, 22°C) and milk type was evaluated with monitored parameters. Results of the model experiments showed the dependence of *S. aureus* multiplication and subsequent production of staphylococcal enterotoxins on the culture / storage temperature and the type of milk. It is noteworthy that in raw milk and in fresh cheese, the *S. aureus* growth rate and production of enterotoxins can be suppressed by competitive microflora. The results of our study support the importance of maintaining the cold chain (at a safe temperature under 8°C) from production through the retail sale to insure for the safety of milk and dairy products.

Keywords: *Staphylococcus aureus*, staphylococcal enterotoxins, milk, ELFA, fresh cheese

Acknowledgement: *The study was supported by research grant MSM 6215712402 Veterinary aspects of food safety and quality and IGA 18/2012/FVHE.*

D-11
THE ISOLATION OF *CRONOBACTER* STRAINS FROM HERBS AND SPICES

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Bacteria from the genus Cronobacter are opportunistic food-borne pathogens belonging to the family Enterobacteriaceae. The illnesses caused by these organisms are rare however life-threatening for neonates and infants. The infections are mostly associated with consummation of contaminated powder infant formula. The members of the genus Cronobacter are ubiquitous microorganisms that have been isolated from a wide range of environmental sources and from various foods, particularly of plant origin and dried material. Its presence in foods raises concern about the safety risks of these foods posed to neonates and immunocompromised consumers. In this study, food samples of herbs and spices (marjoram, caraway, basil, ginger, mustard seed, sesame, pumpkin, instant lentil soup, oregano) have been screened for the presence of *Cronobacter spp*. The enrichment and isolation of *Cronobacter spp.* were performed according to procedures described in the directive ČSN P ISO/TS 22964 (2006) Milk and milk products - Detection of *Enterobacter sakazakii*. Isolated strains have been identified by both biochemical tests and MALDI–TOF–MS on genus Cronobacter level. Cronobacter species identity was confirmed by biochemical characterization using ID 32E test system as well as species targeted PCR system. Eleven samples (73% from tested samples) contained *Cronobacter spp*. The predominant species recovered from the samples was *Cronobacter sakazakii* (n=6). The remaining isolates were identified as *Cronobacter muytjensii* (n=4) and *Cronobacter malonaticus* (n=3). Further, the serotypes of isolated strains were determined.

Keywords: Cronobacter, food

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D-12

THE INFLUENCE OF CULTIVATION MEDIUM ON *CRONOBACTER SAKAZAKII* MASS SPECTRUM QUALITY

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Cronobacter species are known as emerging foodborne pathogens that have been regarded as causative agents of meningitis, septicaemia, and necrotizing enterocolitis in infants, with a mortality rate of 20 to 50%. Thus, rapid diagnostic methods of these bacteria are of a great interest. Matrix-assisted laser desorption/ionization time of flight mass spectrometry (MALDI-TOF-MS) was introduced several years ago as a new method for bacterial identification. The advantages of MALDI–TOF–MS are mainly the rapid analyses, simple sample application procedure, the ability to detect minor differences between strains, and the possibility of direct comparison between strains-characteristic patterns. In order to establish the optimal growth conditions for good quality of measured mass spectra were tested fourth different media: Brain Hearth Infusion Agar (BHA), Tryptone Soya Agar (TSA), Blood Agar Base (sheep, BA) and Enterobacter sakazakii Isolation Agar (ESIA). All media used in this study were solid. This type of medium helps to achieve more reproducible results contrary to liquid one. BHA, TSA and BA are universal media commonly used to grow wide range of bacteria. ESIA on the other hand is the selective medium and its composition is optimized to favour the growth of Cronobacter strains over other microorganism. The mass spectral patterns obtained from cells grown on TSA and BHA were comparable; it was possible to detect the same amount of peaks. TSA showed better ratio of absolute intensity (a. i.) of peaks to the noise level than BHA and achieved higher average a. i. The relative lack of peaks in pattern compositions of cells cultured on BA was detected by contrast to the media BHA and TSA. The lowered number of peaks in total was detected also in spectra from cells grown on ESIA, furthermore the measured intensities were markedly suppressed. This can be associated with *(i)* less rich composition of ESIA in comparison with the other used media (BHA, BA, TSA) and *(ii)* the presence of some selective agents (e.g. sodium chloride, sodium desoxycholate, crystal violet) for inhibition of possible competing microorganism in this medium. TSA medium showed the best properties from tested media and was chosen for usage in further experiments.

Keywords: Cronobacter, MALDI–TOF–MS

Acknowledgement: *Czech Grant Agency: project No. P503/12/P704, Specific University Research: MSMT No. 21/2012*

D-13
CHARACTERIZATION OF DIFFERENT INTERFERING MOLECULES PRODUCED BY CHEESE-ISOLATED LAB STRAINS

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Lactic acid bacteria are used for food improvement in a wide range of fermented foods and some strains can also produce antimicrobial peptides, named bacteriocins, active against food pathogens such as *Listeria monocytogenes* and *Staphyloccus aureus*. The aim of this study was to characterize antimicrobial peptides produced by two *Enterococcus faecium* strains, a *Lactobacillus plantarum*, a *Lactococcus lactis ssp*. cremoris and five *Lactococcus lactis ssp. lactis*. All bacteria were isolated from Italian cheeses. Lactococci and enterococci were grown in M17 broth at 37°C while Lactobacillus plantarum was grown in MRS broth at 30°C. All cultures were performed both in microaerophylia and in presence of O_2 to test whether stress (presence of oxigen) stimulates bacteriocins production.The whole culture and the cell-free supernatant of both strains were tested against pathogens using well-diffusion assay and their antimicrobial activity was shown by the formation of an inhibition zone around the well on the plate. In order to evaluate the time of maximum production of bacteriocins, cultures at different growth phases were tested. Moreover, two different peptide purification protocols were tested to recover a larger amount of bateriocins: 60% ammonium sulphate precipitation and adsorpion/desorption pH dependent of bacteriocins from the cell surface. A biochemical characterization of bacteriocins was performed: molecular size determination by Tricine-SDS-PAGE, thermostability at 70°C, 80°C and 90°C and proteinase k sensitivity. Bacteriocins activity was arbitrary determined by the measure of the inhibition halo around the well; the arbitrary units (AU) of the bacteriocin were determined as the reciprocal of the highest dilution showing inhibition of the indicator strains. Molecular targeting experiments of bacteriocins encoding genes by PCR were performed in order to confirm the bacteriocins identification. This study demonstrated that all lactococcal strains produced nisin A, the two enterecoccus strain produced enterocin A, while the *L. plantarum* was able to inhibit pathogen by means of large amounts of lactic acid.

Keywords: LAB, bacteriocin, *Listeria monocytogenes*, *Staphylococcus aureus*

D-14

PRODUCTION OF HYDROGEN PEROXIDE BY SELECTED STRAINS OF THE LACTOBACILLUS GENUS FROM DIFFERENT SOURCES

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Lactobacilli produce a number of antimicrobial active substances through which they are able to protect food products and human body against invasion and multiplication of pathogenic microorganisms. The antimicrobial substances produced by these bacteria include organic acids (mainly lactic acid), carbon dioxide, hydrogen peroxide, surfactants, diacetyl, bacteriocin-like substances and bacteriocins. Hydrogen peroxide is one of the most active antimicrobial substances produced by lactobacilli. The bactericidal effect of H_2O_2 is based on the formation of oxidation products (e.g. radical $OH^{\bullet}$) that cause changes in cellular DNA of sensitive microorganisms, as well as a strong oxidizing effect on their membrane lipids and cellular proteins. The aim of this work was to compare the production of H_2O_2 for selected species of the *Lactobacillus genus* from different sources. The testing methods were based on qualitative and quantitative detection of H_2O_2 production. For testing of 12 collection lactobacilli strains and four isolates of lactobacilli from vaginal tract of women were used. Detection of H_2O_2 production by selected strains of the *Lactobacillus genus* was qualitatively determined on MRS agar modified by addition of tetramethylbenzidine and horse radish peroxidase (HRP). Concentration of H_2O_2 produced by lactobacilli was determined by spectrophotometric method at 400 nm. Supernatant obtained by centrifugation of fresh lactobacilli and modified by addition of the HRP and o- dianisidine was used for this measurement. H_2O_2 concentration in individual samples was evaluated by use of calibration curve. It was found that all tested lactobacilli produced H_2O_2 at different concentrations. The highest concentrations of H_2O_2 (11.40 $mg.L^{-1}$ phosphate buffer) was produced by vaginal isolate Lbc. crispatus A4 and the lowest concentration by vaginal isolate *Lbc. fermentum* A1 (0.04 $mg{\bullet}L^{-1}$ phosphate buffer). Production of H_2O_2 on the modified agar was observed only for a few strains depending on the way of cultivation. The qualitative method has been proved to be inaccurate. It was found that H_2O_2 production is strain-specific and does not dependent on the origin of the tested lactobacilli strains.

Keywords: Antimicrobial active substances, *Lactobacillus*, hydrogen peroxide

D-15
INFLUENCE OF LACTIC ACID BACTERIA ON PERSISTENT LISTERIAS IN SYNTHETIC AGAR, CURD AND SMEAR RIPENED CHEESES

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Listeria monocytogenes is recognized as an unsafe foodborne pathogen. In dairy industry, particularly high risk present so-called persistent *L. monocytogenes* strains able to create biofilms on inadequately cleaned and disinfected surfaces of producing equipment. Biofilms exhibit higher resistance to antimicrobial substances than planktonic forms of listerias. Moreover, a part of the biofilm could be release and contaminate raw materials, intermediate products or final foodstuffs. One possible way to eliminate listerias in dairy products is the application of lactic acid bacteria (LAB), which display numerous antimicrobial activities. This is mainly due to the production of organic acids, but also of other compounds, such as bacteriocins. LAB and their bacteriocins can be used as safe alternatives to chemical preservatives in foods. Eight bacteriocin-producing LAB strains (species of *Lactococcus lactis*, *Lactobacillus plantarum*, *Pediococcus acidilactici* and *Enterococcus mundtii*) and three bacteriocin-non-producing commercial mesophilic cheese starter cultures were tested for their ability to inhibit/reduce the growth of five persistent (biofilm-forming) and one non-persistent *L. monocytogenes* strains. Firstly, antilisterial activities of LAB were tested in synthetic BHI agar using agar diffusion method. Secondly, selected LAB confirmed as antilisterial active were tested for their activity in two food systems – low fat curd and smear ripened cheeses. Curd and cheeses were intentionally contaminated by individual listerial strains or by mixture of listerias and LAB and stored at 22°C for 21 days, aerobically. During the storage experiment, a total of 6 sampling were performed to determinate the number of present *L. monocytogenes*. Seven of the 11 tested LAB were confirmed as antilisterial active: two lactococci were able to inhibited one non-persistent listeria, five LAB and one cheese culture showed antilisterial activity against some/all persistent listerias. *Ent. mundtii* was confirmed as the most effective in the inhibition of all persistent listerias. Listerias added into the curd were inhibited within 14 days of storage regardless of whether using LAB were added as well. On the other hand, there was no significant inhibition of listerias in any sample even after three-week cultivation with LAB. Results of agar diffusion method confirmed that used LAB strains disposed of a strong biochemical potencial able to inhibit *L. monocytogenes*, including highly unsafe persistent strains. The curd was confirmed as safety foodstuff due to the low pH (<4.0), which did not allow *L. monocytogenes* to grow. On the contrary, the surface of smear ripened cheeses (pH>6.5) created a suitable environment for the growth of persistent listerias. Inhibition by LAB in order to ensure the safety of smear ripened cheeses was not sufficient. The antilisterial effect of LAB in all used model systems was confirmed as highly-strain specific.

Keywords: Persistent listerias, lactic acid bacteria, bacteriocins, smear ripened cheeses

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D-16
COMPARISON OF NUTRITIONAL COMPOSITION OF INTEGRATED, ORGANIC AND HYDROPONIC GROWN GREENHOUSE LETTUCE

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High consumption of vegetables is associated with lower risk of many types of chronic diseases. Organic agriculture includes management practices such as no-till and soil mulching to minimize erosion, reduction of pests through rotation or biological methods of pest control, and the use of manure or natural fertilizers. Integrated production in agriculture consists of a set of practices such as minimum tillage, nutrient balancing and integrated pest management. On the other hand, hydroponics is a technique of growing plants in a soil-less medium and nowadays probably the most intensive and effective form of cultivation which characterized by heavy use of pesticides and chemical fertilizers. To establish how the certain crop management practice could influence nutritional composition of lettuce, Slovenian lettuce cultivar 'Marija' with high yield potential was used. For this purpose, lettuce plants were grown in the research greenhouse at the University of Ljubljana, Slovenia during spring 2009. Fertilization, disease and pest control were carried out in compliance with the main principles of hydroponic, integrated and organic crop management systems. As a conclusion, the results showed that organic growing lettuce had a higher percentage of palmitic (16:0), stearic (18:0), oleic (18:1), linoleic (18:2), linolenic (18:3) and arachidic (20:0) acid that lettuce from the hydroponic and the integrated growing system. Ash and soluble fibre content were significantly higher in organically grown lettuce. Comparative analysis also showed that organic lettuce has the greatest values of vitamin C in outer and inner leaves. According to other nutritional components of lettuce no differences was found between growing systems. Vitamin C was also analyzed up to 7 days of storage. Refrigerated storage resulted in the lowest retention of vitamin C in case of hydroponic system.

Keywords: Lettuce, integrated growing, organic growing, hydroponic growing, nutritional composition

D-17
QUALITY OF COW MILK FROM ORGANIC AND CONVENTIONAL FARMING

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Chemical and microbial analyses of raw milk samples from organic and conventional farming were statistically compared. Samples were analysed during the twelve-month period (6/2008 – 5/2009). There were altogether 2208 samples from which 530 were from organic farming. After the raw milk was processed, sensory hedonic quality of 170 pairs of organic and conventional non-standardized pasteurised whole milk samples was evaluated using pair comparison preference test. There were two components in raw milk, fat and somatic cells count, where no significant differences were found between both types of production. There were statistically significant higher contents of proteins (3.33>3.28 g/100 g, P<0.01), casein (2.66>2.61 g/100 g, P<0.01), lactose (4.83>4.78 g/100 g, P<0.01), non-fat solids (8.74>8.64 g/100 g, P<0.01) and urea (25.34>20.14 mg/100 g, P<0.01) in conventional milk, as well as lower freezing point (-0.5279<-0.5267°C, P<0.01). On the contrary, significantly higher contents were found in case of total mesophillic bacteria count (76 000 > 35 000 CFU/ml, P<0.01), coliform bacteria count (55>38 CFU/ml, P<01) and free fatty acids (0.94>0.59 mmol/100 g, P<0.05) in organic milk samples. Sensory analysis results of pasteurised milk did not shown any statistically significant hedonic difference between organic and conventional samples.

Keywords: Organic milk, composition, microbiological quality, hedonic evaluation

D-18

CHARACTERIZATION OF SLOVENIAN APPLE JUICE WITH RESPECT TO AGRICULTURAL PRODUCTION PRACTICE

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Apple is one of the most affordable and healthy fruit in the world, thus the production increased in recent years. Fruit production increased for 58% in the last 20 years also in Slovenia, along with apple production. In our county the most common is integrated apple production, however importance of organic production has been increased recently. Growth of the market of organically produced apples, which are also processed into juice, is mainly due to increasing demand for healthy food requirements, protection of environment and the promotion of biotic diversity. Organic foods are also more expensive, because their production has still slightly lower yields and higher risk of production. According to a EC Regulation (834/2007 and 889/2008) organic producers are subjected to strict inspections about the production, but a need to find authentication tools that can distinguish whether organically certified supermarket products are indeed organically grown, is still under investigation. With the experiment called OCAC (Organic ~ Conventional Apple Cultivation) 2010, we want to determine the influence of agricultural practice on selected parameters in apples and apple juice. It was implemented at the Faculty of Agriculture and Life Sciences, University of Maribor on the property under the castle HompoS in the experimental Gala apples orchard in 2010. Four different fertilizers were applied namely: Biosol (organic) and Nitrogen, KAN and UREA (mineral). The amounts of fertilizers added were calculated depending on the content of nitrogen in fertilizers (according to producers certificate data sheet) in a way that provided 60 and 120 kg of nitrogen per hectare. Various parameters were determined in apple juice made from apples in respect to different fertilizers that are legally allowed either in integrated/conventional or in organic agriculture production including: isotopic ratios of light elements ($^{13}C/^{12}C$, $^{15}N/^{14}N$); multi micro- and macro- element composition; major volatile compounds and selected chemical and physical parameters of apples and apple juice (fruit weight, pH, titratable acidity, total soluble solids, starch test of ripeness, flesh firmness, antioxidant activity, content of free amino acids, organic acids, sugars and ascorbic acid and total phenols). First evaluation of results indicated, that average fruit mass of apples manoured with fertilizers allowed in organic production was lower, but the difference was not statistically confirmed ($p>0.05$). Mass fractions of individual elements determined with TXRF showed that potassium was the most abundant in apple juice followed by phosphorus, calcium, sulphur and chlorine, regardless the treatment. Statistical evaluation using Kruskall-Wallis test indicated that $\delta^{15}N$ value of pulp was the most significant parameter to distinguish between organic and conventional fruits ($p<0.05$). Further statistical analyses are still in progress.

Keywords: Apples, agricultural practice, tracers, stable isotopes, Slovenia

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D-19

NUTRITIONAL AND MORPHOLOGICAL CHARACTERISTICS OF CARROT (*DAUCUS CAROTA* L.) FROM ORGANIC FARMING

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Recently, the demand for organic food products is steadily increasing, mainly due to the expected health advantages of organic food consumption. Improved taste and freshness of foods, as well as environment protection are important reasons for organic production, although the possible health benifits of organic food consumption are not considered scientifically well-documented. The aim of our study was to compare nutritional (the contentrations of sugars, organic acids, ascorbic acid, carotenoids and single phenolic compounds) and some morphological characteristics of three carrot cultivars (Fanal, Rodelika, Rolanka) grown under organic and integrated conditions. Carrot from the organic farming had higher concentrations of organic acids, α-carotene and β-carotene and ascorbic acid (not always significantly) compared to the carrot from integrated growing system and from the control. No significant differences of the sugars content in carrots regarding the cultivation system were recorded. In carrots, four different phenolic compounds belonging to the group of hydroxycinnamic acids were quantified: feruoylquinic acid respresented the major, 5p-cumaroylquinic acid the second prevailing and 3-caffeoylquinic and ferulic acid the minor hydroxicinnamic acids. The levels of feruoylquinic acid and 5p-cumaroylquinic acid were higher in carrots from the organic farming than in the carrot from integrated production. Cultivation type had an important infuence on some morphological and nutritional characteristics. Carrot from organic production showed higher content of karotenoids, ascorbic acid, hydroxycinnamic acids and organic acids than carrots from integrated cultivation.

Keywords: Organic farming, carrot, primary metabolites, carotenoids, phenolic compounds

D-20

CHARACTERIZATION OF CONVENTIONALLY AND ECOLOGICALLY GROWN PEPPERS WITH STABLE ISOTOPES OF NITROGEN AND CARBON

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Previously known methods can not distinguish between biologically and conventionally grown fruits and vegetables. The purpose of the study was to find physical and chemical parameters that may enable to distinguish between conventionally grown (artificial fertilisers) from ecologically grown (organic fertilisers) sweet pepper. Following parameters were determined: composition of carbon and nitrogen stable isotopes ($\delta^{15}N$ and $\delta^{13}C$), content of ascorbic acid, total polyphenols and antioxidative potential. Mineral fertilisers significantly enhance fruit mass, height, wideness and thickness of mesocarp. Sweet pepper fuits grown by means of organic fertilisers contained more total phenols, more ascorbic acid and have higher antioxidative potential. Fertilisation influenced fruit colour parameters but the differences are not important from the technological point of view. Organic fertilisers contribute to statistically significantly higher $\delta^{15}N$ values and to lesser extent but still higher $\delta^{13}C$ values.

Keywords: Pepper, fertilisation, biological, stable isotopes, $\delta^{15}N$, $\delta^{13}C$

D-21
DIFFERENCES IN EWE'S MILK COMPOSITION DUE TO THE PRODUCTION SYSTEM CONVENTIONAL *vs* ORGANIC

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Among the distinctive features of organic livestock production organic farming regulations restrict the use of concentrates, and with a lower intake of energy from concentrates some changes in milk characteristics can be expected. However, the scarce available data show that fat, protein and lactose percentages in ewe's milk are unaffected by the organic production system (Revilla et al., 2009). That study included only few flocks from the same geographical area, and hence the aim of the present work was to include a higher number of flocks, geographical areas and managing techniques within conventional system and also to consider parameters related to the health of the flocks such as somatic cell counts. Bulk tank ewe's milks were obtained from 20 flocks raised in six different provinces of the Region of Castile & Leon. Samples were analysed for fat, protein and total solids (Milko Scan) and somatic cell counts (SCC) (Fossomatic Foss) five times a month from January until December at the Analysis Service of the Interprofessional Dairy Laboratory of the Junta of of Castile & Leon (Spain) (LILCYL; Palencia, Spain). Two types of production systems - conventional (CS, 18 flocks) and organic (OS, 2 flocks) and two breeds- the local breed Churra (6 flocks under CS and 2 flocks under OS) and the foreign breed Assaf (12 flocks under CS)- were studied. The sheep under the conventional system remained on a feedlot and were allowed ad libitum access to the commercial concentrate, although in some flocks they also went to pasture for a few hours a day after the lamb weaning. Sheep under the organic system went to pasture, where they were allowed to graze ad libitum and their diet was supplemented (maximum 30% of the ration) with a mixture certified by the Organic Agriculture Council of Castile & Leon (CAECYL). The results show there were no statistically significant differences due to the production system (CS vs OS) as regards fat, protein or dry matter. However, the SCC were significantly lower ($p<0.001$) in the organic (573.636 cell mL^{-1}) than in the conventional flocks (1.208.665 cells mL^{-1}). A significant effect of month was observed on fat, protein ($p<0.001$) and dry matter ($p<0.01$) maximum values being observed in December and January and minimum values in June and July. Nevertheless, a statistically significant production system-month interaction was observed for fat. A statistically significant effect of breed was also observed for fat ($p<0.05$), protein ($p<0.001$) and SCC ($p<0.001$), the Assaf breed showing the lowest values for all this parameters. Since all the Assaf breed flocks were under the conventional production system, the effects of the production system were analysed only for Churra breed flocks. The results show there were no statistically significant effect of the production system for fat, protein or dry matter although, as previously observed, the SCC were significantly lower ($p<0.001$) in the organic than in conventional milk samples.

Keywords: Organic dairy farming, ewe, milk composition, somatic cell count

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D-22

FOODCAP: THE SEARCH FOR BIOMARKERS OF HETEROCYCLIC AMINE EXPOSURE IN HUMAN BLOOD

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Heterocyclic amines (HCA) are food-derived carcinogenic mutagens produced when meat or fish are cooked. They are formed mainly in muscle foods providing creatinine and other precursors including amino acids and sugars or other aldehydes. Over 25 HCAs have been identified in various foods, the most abundant being PhIP, DiMeIQx and MeIQx. HCA dietary intake is influenced by type of meat, frequency of consumption, portion size, thickness of meat, cooking method/temperature/duration, ingestion of pan residues/gravy, use of marinades and frequency of turning food during cooking. Thus, assessment of HCA dietary exposure has proven difficult. The US National Cancer Institute has compiled the only database of measured HCA content of foods (CHARRED). One strategy to overcome shortcomings of epidemiological and HCA intake studies is to use biomarkers of status. Biomarkers reflect true net exposure and are a more objective measurement of status/exposure. Protein adducts as biomarkers of medium term HCA exposure have been proposed and measured in human serum (e.g. PhIP adducted to serum albumin and haemoglobin). The FoodCAP project, funded by the World Cancer Research Fund, aims to develop cost effective immunochemical assays to measure HCA-protein adducts in blood and test their validity in human dietary studies. Blood samples and 7-day food diaries were collected from 150 volunteers in a cross-sectional study to estimate habitual HCA intake based on the CHARRED database. Forty participants from the highest tertile of HCA intake took part in a 6-week controlled dietary intervention study, eliminating all meat and fish with the aim of reducing nominal HCA intake to zero. Quantitative analytical methods, currently being developed, will be used to measure HCA-protein adducts in blood samples from each study, correlating measured adduct levels with estimated HCA intakes, and demonstrating the efficacy of dietary intervention to reduce levels of circulating adducts. Analyses will also be conducted on stored blood from 2000 adolescents in the Young Hearts 2000 cohort of which Queen's University is the custodian. This will elucidate further the relationship between biomarkers of dietary carcinogens and adolescents' dietary patterns. Adducts of PhIP and MeIQx bound to human serum albumin and haemoglobin were prepared in vitro using a liver microsome system to mimic metabolic adduction in vivo. HCA incorporation was confirmed by mass spectrometry. Initial attempts to raise monoclonal antibodies specific to HCA-protein adducts proved unsuccessful, presumably due to poor immunogenicity of the HCA haptens. In a parallel approach, screening of phage display libraries for peptide binders to adducted HCAs is currently ongoing. The aim is to incorporate binders (peptides or antibodies) into rapid quantitative assays based on ELISA or SPR biosensor technologies to measure HCA-protein adducts in blood samples generated by FoodCAP: Food, Cyclic Amines and Protein.

Keywords: Heterocyclic amines, biomarkers, protein adducts, meat

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D-23

EVALUATION OF BENZO(a)ANTHRACENE, BENZO(a)PYRENE, BENZO(b)FLUORANTHENE AND CHRYSENE CONTENT IN SMOKED FISH

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Polycyclic aromatic hydrocarbons (PAH´s) can significantly influence smoked meat and fish quality and safety. Toxicological studies on individual PAHs in animals, mainly on the PAH benzo(a)pyrene, have shown various toxicological effects, such as haematological effects, reproductive and developmental toxicity and immunotoxicity. It has been concluded that benzo(a)pyrene is a probable human carcinogen. One significant source of BaP in the human food chain is smoking of products. Smoke not only gives special taste, colour and aroma to food, but also enhances preservation due to the dehydrating, bactericidal and antioxidant properties of smoke. Therefore the aim of our investigation was to determine the conetents of BaP in industrially smoked different fish products. Results were summarized and compared with maximum acceptable levels set by European Commision regulation (EC) No 1881/2006 Forteen different smoked fish products samples were taken and packing according to the sampling procedure. Benzo(a)anthracene, benzo(a)pyrene, benzo(b)fluoranthene and chrysene in (PAH4) content in the studied samples were determined The results obtained in this application were all performed on Waters HPLC system consisting of the Waters 2695 separations module, Waters 2998 Photodiode Array detector, Column: ZORBAX Eclipse PAH, 4.6 mm × 150 mm, 3.5 µm. The sum PAH4 was the highest 2.09 µg.kg^{-1} in hot-smoked fish samples and the lowest 0.34 µg.kg^{-1} in cold smoked saples.

Keywords: Polycyclic aromatic hydrocarbons, smoked fish, benzo(a)pyrene

D-24

EVALUATION OF POLYCYCLIC AROMATIC HYDROCARBONS COMPOUNDS COMPOSITION OF FRIED AND GRILLED DEER MEAT

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A game animals every autumn and winter period provide an excellent investment, diversification of many consumer meals. In last years consumption and assortiment of game meat products significantly increase. The meat of wild animals is more favourable for human health because it has lower SFA content but higher content of protein. When preparing meat as edible products to enjoy the aroma and taste, we heat treat a variety of ways, such as cooking and frying. Polycyclic aromatic hydrocarbons (PAH's) can influence meat quality. Investigations about PAH`s composition of game meat are not very much. Therefore, the aim of the investigation was evaluate PAH`s composition of fresh and processed deer (*Cervus elaphus*) meat in Latvia. In order to make PAH`s analyses meat samples after hunting in Latvia were collected. After processing and meat ripening, samples were fried and grilled, as the consumers do it at home. Later according to the sampling procedure PAH's – benzo(a)anthracene, benzo(a)pyrene, benzo(b)fluoranthene and chrysenein content in the studied samples were determined. The results obtained in this application were all performed on Waters HPLC system consisting of the Waters 2695 separations module, Waters 2998 Photodiode Array detector, Column: ZORBAX Eclipse PAH, 4.6 mm × 150 mm, 3.5 µm.The sum of benzo(a)pyrene, 1,2 benzanthracene, benzo(b)fluoranthene and chrysen was 0.42 µg.kg^{-1} in fresh meat samples, 0.56 µg.kg^{-1} in cooked saples and 0.81 µg.kg^{-1} in grilled meat samples. Results were compared with acceptable levels set by European Commision regulation (EC) No 1881/2006.

Keywords: Deer meat, cooked meat, polycyclic aromatic hydrocarbons

D-25
VACUUM FRYING: UNIQUE TECHNOLOGY FOR MINIMIZING ACRYLAMIDE IN POTATO CRISPS

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Acrylamide, processing contaminant formed during heating of foodstuff such as frying, baking, roasting or smoking is classified according to the International Agency for Research on Cancer (IARC) as “probably carcinogenic to humans” (Group 2A). Its formation starts at 120°C and is described via Maillard reaction with amino acid asparagine and reducing sugars as crucial precursors. Much attention has been drawn especially to potato products because of their high consumption and high contribution of acrylamide, which content has not been regulated in food, yet. Many experiments focusing on optimizing conventional frying conditions have been implemented and described in recommendations intended especially to manufacturers. According to Food Drink Europe Acrylamide Toolbox 2011, food should not be fried at temperatures higher than 175°C and their color should be golden yellow. A unique technology, vacuum frying, offers an alternative thermal system which enables to process food at lower temperatures and has significant benefits such as the improvement of fried product safety and quality. This research was focusing on optimizing vacuum and conventional frying conditions and to compare the quality of both different fried products. Special potato variety (Saturna) with low reducing sugars and asparagine content was used for the preparation of potato crisps. Potato crisps were fried at different temperatures and frying times. To find optimal frying condition, not only acrylamide content was observed but also the moisture content which is important to avoid microbiological spoilage during storage. To evaluate the food nutrition value, fat content was determined. Finally, sensory evaluation of both vacuum and conventional fried potato crisps was also done.

Keywords: Acrylamide, vacuum frying, conventional frying, potato crisps, sensory evaluation

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D-26

EVALUATION OF THE ACID TRANSESTERIFICATION AND SALTING-OUT CONDITIONS DURING THE INDIRECT DETERMINATION OF 3-MCPD ESTERS IN VEGETABLE OILS

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Fatty acid esters of 3-monochloropropane-1,2-diol (3-MCPD esters) are processing contaminants that can occur in refined vegetable oils at significantly high concentrations. These compounds can be formed during the deodorization step of the oil refining process, in which a higher temperature is needed to provide acceptable color, odor and low fatty acid content. The suggested mechanism involves the formation of a cyclic acyloxonium ion from triacylglycerol, followed by reaction with chloride ions. Several indirect quantitative methods have been proposed for the determination of 3-MCPD esters. They are based on the conversion of individual 3-MCPD esters into a single compound (free 3-MCPD) that is subsequently quantified. In general, these methods present high sensitivity and do not require a large series of standards compounds. On the other hand, the sample preparation is complex and comprises a number of chemical transformations that may affect the results, especially due to the presence of glycidyl esters that could lead to an additional formation of 3-MCPD during the analysis. The objective of this work was to conduct a preliminary evaluation of some critical steps involved in the indirect determination of 3-MCPD esters, such as the transesterification and salting-out. Experiments were conducted with standard solutions of 1,2-dipalmitoyl-3-MCPD (PP-3-MCPD), a refined soybean oil (that should contain 3-MCPD esters) and an extra virgin olive oil (that should not contain significant levels of 3-MCPD esters), using as internal standard deuterated 1,2-dipalmitoyl-3-MCPD (PP-3-MCPD-d5). The analytical procedure included the addition of a PP-3-MCPD-d_5 solution to the sample, the transesterification employing a mixture of sulfuric acid and methanol, the neutralization by adding sodium hydrogencarbonate solution, the salting-out of lipophilic compounds, the derivatization of the released 3-MCPD with phenylboronic acid and the analysis by gas chromatography coupled to mass spectrometry. Incubation times of 4, 16 and 24 h were evaluated for the transesterification step while two salts (sodium chloride and ammonium sulfate) were tested for salting-out. When sodium chloride was used, unrealistic high levels of 3-MCPD esters were verified in the samples, although data reported in the literature indicates that the results obtained by using the acid transesterification approach are not dependent on the type of salt. Moreover, the levels of 3-MCPD esters increased by up to 245% with the increase of incubation time from 4 to 24 h. On the other hand, expected concentrations of 3-MCPD esters were achieved when ammonium sulfate was used for salting-out and no significant differences were observed within different transesterification times. It can be concluded that the type of salt was the most critical parameter in these experiments. The influence of the transesterification time was also affected by the type of salt.

Keywords: Vegetable oils, 3-MCPD esters, acid transesterification

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D-27
OCCURRENCE OF 3-MONOCHLOROPROPANE-1,2-DIOL (3-MCPD) IN BRAZILIAN SMOKED FOODS

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3-Monochloropropane-1,2-diol (3-MCPD) is a chemical contaminant belonging to the group of chloropropanols that demonstrated to affect male fertility and kidney functioning as well as to induce cancer when administrated to experimental animals at high doses over a long period of time. 3-MCPD can be formed during the processing of several foods and food ingredients, including smoked foods. In these products, the proposed mechanism involves the reaction of chloride ions with acetol (3-hydroxyacetone), a thermal degradation product of cellulose and wood smoke. Mean concentrations between 11 and 43 µg/kg were already reported for smoked foods such as bacon, sausage and ham. The objective of this work was to evaluate the levels of 3-MCPD in smoked foods commercialized in Brazil, since no data is available so far. A total of 30 samples were collected in the city of Campinas-SP, in September 2011. The sampling included smoked cheese, sausage, bacon, turkey breast paté, bologna, tuna, and turkey and chicken breast. For the determination of 3-MCPD, NaCl 5M solution and Extrelut NT20® were added to the homogenized samples, mixed and transferred to a glass column. Elution was performed with a mixture of hexane/diethyl ether and then with diethyl ether. Dried and concentrated extracts were derivatized with HFBI prior to analysis by gas chromatography coupled to mass spectrometry (GC–MS). Quantification was carried out by employing deuterated 3-MCPD-d5 as internal standard. Levels of 3-MCPD above the limit of quantification were verified in 14 samples (47%). The concentrations ranged as follow: not detected (nd)–14.9 µg/kg (sausage); 6–48.5 µg/kg (cheese); nd–11.7 µg/kg (bacon); nd–6 µg/kg (turkey breast); nd (turkey breast paté); below 5.9 µg/kg (chicken breast); nd–6.8 µg/kg (bologna); nd (tuna). The highest levels were found in provolone cheese. These results can be compared to those previously published. Besides the supposed formation from acetol and chloride ions, the contamination of smoked foods with 3-MCPD may also be due to the high concentrations of triacylglycerols and sodium chloride, to the presence of epichloridrine in sausage casings and to the hydrolysis of previously existing 3-MCPD esters.

Keywords: Smoked foods, chloropropanols, GC–MS

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D-28
DETERMINATION OF FREE AND BOUND VOLATILE AND NON-VOLATILE NITROSAMINES IN NITRITE CURED MEAT PRODUCTS

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Nitrite and nitrate has for many decades been used for the preservation of meat, because these compounds effectively inhibit the growth of Clostridum botulinum and thereby also the production of the botulism toxin by this organism. Preserving of meat with nitrite is often referred to as ”curing”. The presently authorised uses of nitrite results in a risk of exceeding the ADI, especially for Danish children, which have a relatively high consumption of processed meat products, e.g. luncheon meat. This is of concern to the Danish Authorities together with the formation of *N*-nitrosamines in nitrite preserved meat products. It is well established that *N*-nitrosamines (NA), of which many are carcinogenic, can be formed in cured meat products and numerous publications report a link between meat intake and colorectal cancer risk (Corpet 2011). For a renewed evaluation of the risk associated with the consumption of nitrite preserved meat products it is relevante to determine free and bound NAs. The bound NAs are to be determined to prevent an underestimation of the risk. Earlier studies have shown that the amount of bound N-Nitrosoproline exceeds the amount of free N-Nitrosoproline (Dunn and Stich, 1984; SEN et al., 1989). This may also be the case for other NAs. An analytical method has therefore been developed and validated for the determination of free and protein/peptide bound volatile and non-volatile NAs in meat products. The method includes 11 volatile and 5 non-volatile NAs. The free NAs were extracted from the meat samples with acetonitrile/water acidified with formic acid. The extracts were frozen in order to precipitate any low soluble fractions. Following centrifugation and transfer of the supernatant to a clean tube, the acetonitrile was allowed to evaporate from the extract, facilitated by a stream of nitrogen, leaving an aqueous extract. Whereby co-extracted interferences of low water solubility precipitate. For determination of the NAs the samples were incubated with Protease Type XIV for 2 hours prior to the extraction. These extracts were analysed by LC–MS/MS using either APCI or ESI ionization. To allow for adequate retention and separation of the generally highly polar non-volatile NAs and generally non-polar volatile NAs, the compounds were separated on a two column system with a Kinetex C18 and an Acclaim PAII column in series. A gradient elution was employed using a mobile phase of water/methanol with 0.1% formic acid. The two column systems provides enough retention of the polar NAs and co-extracted interferences co-eluting with the solvent front (e.g. sodium chloride, nitrite) was sent to waste and thereby avoiding contamination of the MS. Two MRM transitions were determined for most of the NAs included in the method. The method has been used for the analyses of real samples and the results will be presented.

Keywords: *N*-Nitrosamines, nitrite, curing, processed meat, LC–MS/MS

D-29
FORMATION OF FRYING MUTAGENS DURING HOME-GRILLING AND DURING GRILLING UNDER CONTROLLED CONDITIONS

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The use of barbecues or outdoor grills for cooking has increased considerably in Denmark over the last decades. It is well known that during heat-treatment of meat and especially barbecuing or smoking, harmful components can be formed with increased cancer risks as a result. Both heterocyclic aromatic amines (HCA) and polycyclic aromatic hydrocarbons (PAH) are such compounds. During barbecuing both types of compunds are formed. HCA's are generated as a reaction between precursors in the meat during temperature treatment and PAH's are formed either from the charcoal itself or from lipids dripping on the hot charcoal. The levels of HCA and PAH were studied in barbecued beef, pork and chicken using two types of set-up. One set-up included home-grilling by Danish consumers, grilling an extra piece of meat for research purpose, while preparing their own meal. The other set-up included controlled barbecuing conditions at the Danish Meat Research Institute using either open or closed charcoal barbecues, or gas heaters. Product parameters included two end points measured as meat temperatures (65 or 72°C) and wrapping or non-wrapping in foil of roast pork. HCA's were extracted from the meat crust using hollow-fiber extraction followed by LC–MS/MS analysis. PAH's were extracted from the homogenized piece of meat using pressurized liquid extraction followed by two clean-up steps and a final GC–MS analysis. Home-grilling was found to result in highest concentrations of PAH 4 (sum of benzo[a]pyrene, chrysene, benz[a]anthracene and benzo[b]fluoranthene]) in beef (up to 65 µg/kg), with lowest concentrations in barbecued chicken (up to 5 µg/kg). For HCA highest concentrations of 2-amino-1-methyl-6-phenylimidazo[4,5-b]pyridine (PhiP) were found in barbecued pork (up to 1.8 µg/kg), whereas for 9H-pyrido[3,4-b]indole (Norharman) highest concentrations (up to 8.2 µg/kg) were found in barbecued chicken. For controlled barbecuing, the formation of PAH was low no matter the type of product. For HCA formation, PhiP was detected for roasted pork at 72°C with charcoal (2.4 µg/kg) and gas heaters (1.8 µg/kg), also pork chops and beef prepared with gas heaters were found to contain PhiP. Calculation of human exposure included margin of exposure (MOE) for PAH 4 well above the critical value of 10,000 set by the European Food Safety Authority (EFSA) for carcinogenic compounds. Only a worst case scenario with consumers eating all their meat from home-grilled barbecues all at maximum levels of PAH 4 resulted in a MOE below 10,000 and therefore consumers at health risk.

Keywords: PAH, PAH 4, HCA, barbecuing, MOE

D-30
IMPACT OF VACUUM BAKING ON FURAN AND ITS PRECURSORS IN BISCUITS

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Furan, which can be formed in a variety of heat-treated commercial foods, has been shown to be carcinogenic in animal laboratory studies. The International Agency for Research on Cancer (IARC) has classified furan as possibly carcinogenic to humans (Group 2B). The latest results on the monitoring of furan levels in food realized by The Food Safety Authority (EFSA) between years 2004-2010 show that the major contributors to its exposure were brewed coffee for adults and fruit juice, milk-based and cereal-based products for toddlers and other children. The innovative technologies including ohmic heating, vacuum baking and treatment with high hydrostatic pressure enable to reduce the temperature and time during food processing. The aim of this study was to evaluate the impact of vacuum applied during baking on furan formation in comparison with conventional oven systems. Moreover, the impact on its precursors originating via lipid degradation and/or Maillard reaction was also investigated. For the analysis of furan, a routine solid-phase micro-extraction based method coupled to gas chromatography and ion trap mass spectrometry (SPME/GC–ITMS) was used. This method is adequate to detect low furan levels formed in biscuit samples. The analysis of precursors (e.g. furfural and 5-hydroxymethylfurfural) was performed by a newly developed solid-phase micro-extraction followed by gas chromatography and time-of-flight mass spectrometry (SPME/GC–TOFMS), which enable to monitor flavor released during baking.

Keywords: Furan, vakuum baking, biscuits, flavor, SPME/GC–MS

Acknowledgement: *The financial support by the European Commission (project PROMETHEUS, PROcess contaminants: Mitigation and Elimination Techniques for High food quality and their Evaluation Using Sensors & Simulation; FP7-KBBE-2010-4-265558) and the Ministry of Education, Youth and Sports of the Czech Republic (Specific University Research MSMT No. 21/2012) is gratefully acknowledged.*

D-31

STUDY OF PAHs ELIMINATION FROM MODEL MATRIX BY MIGRATION TO THE PLASTIC PACKAGING MATERIALS

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It was found that the polymer packaging could play an important role in eliminatig the harmful organic contaminants from food because of the high affinity of some organic compounds to certain plastic materials. The ability of PET (polyethylene terephthalate) to lower polycyclic aromatic hydrocarbons (PAHs) concentrations in polar and non-polar liquid media has already been unambiguously proven [1]. The aim of this study was testing the capability of three commercially available plastic package materials (LDPE – low density polyethylene, PET - polyethylene terephthalate, PA/PE – polyamide/polyethylene) to eliminate PAHs from a model food matrix. PAHs may be formed on the surface of smoked meat products during the technological processing. Therefore the polyamide/artificial sausage casing was chosen as a food matrix and benzo[a]pyrene (BaP) as a representative of PAHs. The changes of BaP concentration on the surface of casing covered by plastic package were followed for 10 h under controlled conditions in climate chamber and analyzed by high-performance liquid chromatography with fluorescence detection. Obtained results show that the BaP concentration decreased as a reason of the interaction between BaP on the surface of casing and plastic packaging. It was due to the sorption of BaP on packaging material, in which BaP was found during the experiment and also at the end of the experiment. These findings could bring new philosophy in safety and quality of packaged food.

[1] Šimko, P. - Šimon, P. - Belajová, E.: Lowering of concentration of polycyclic aromatic hydrocarbons in liquid media by sorption into polyethylene terephtalate – a model study. European Food Research and Technology, 2004, 219, 273–276

Keywords: PAH, plastic packaging materials, elimination

Acknowledgement: *This work was supported by Science and Technology Assistance Agency of Slovak Republic under the contract No. APVV-0168-10.*

D-32
PRODUCTION OF BENZOIC ACID IN FERMENTED GOAT AND EWE MILK

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The production of goat and ewe milk in the Czech Republic has a growing tendency in the last years, primarily on private farms. Here most of the milk is processed into fermented milk products. A natural component of fermented milk products is also benzoic acid. The level of benzoic acid depends primarily on the level of hippuric acid in raw milk and may also depend on the process of dairy product manufacture, especially on the used microorganisms. Although since the year 2004 there has not been established hygienic limit for the content of benzoic acid in fermented milk products in the Czech Republic due to the inhibitory effects of benzoic acid on some enzymes and the possible irritant effects on sensitive persons the lowest content of benzoic acid in fermented dairy products is desirable. The content of hippuric acid in the raw goat and ewe milk and thereafter the content of benzoic acid in model fermented milk drinks prepared using six different cultures of bacteria of milk fermentation were determined using HPLC-DAD method. Milk samples from two farms were collected monthly during the whole lactation period. Data were statistically analyzed using ANOVA. The contents of hippuric acid in raw milk as well as the contents of benzoic acid in fermented milk products during the lactation period were comparatively variable. A statistically significantly lower quantity of hippuric acid 15.5 with limit 8.3 mg/kg was established in milk from Farm A (goat milk) than in milk from Farm B 43.3 with limit 12.3 mg/kg (ewe milk). All fermented milk products contained benzoic acid. Higher quantity of benzoic acid in fermented milk products made from ewe milk corresponded also to statistically significantly higher quantity of hippuric acid in raw ewe milk. The contents of benzoic acid in model fermented milk drinks ranged from 2.0 (made from goat milk) to 78 mg/kg (made from ewe milk). No statistically significant differences among used cultures of bacteria of milk fermentation and the quantity of benzoic acid in fermented milk drinks has been demonstrated. In addition, the quantity of benzoic acid in various commercial goat and ewe cheeses produced in five farms from milk of different animal breeds was analysed. No statistically significant difference in the quantity of benzoic acid from goat and ewe cheeses has been found, whereas statistically significant differences in the quantity of benzoic acid in cheeses produced in individual farms were demonstrated. The contents of benzoic acid in cheeses ranged from 2.2 to 90 mg/kg. The lowest and the highest content of benzoic acid were determined identically in goat cheeses. The results led to the conclusion, that the content of hippuric acid in raw milk and after that the level of benzoic acid in fermented goat and ewe milk products were not influenced by the differences between goat and ewe milk, but mainly by specific conditions of breeding in individual farms.

Keywords: Benzoic acid, goat milk, ewe milk

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D-33
ACRYLAMIDE FORMATION DURING HEAT PROCESSING OF TRADITIONAL CZECH POTATO PANCAKES

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Potato products represent dietary items containing the highest amounts of food carcinogen acrylamide (IARC 1994, group 2A–probably carcinogenic to humans). As the main way of acrylamide formation in food, Maillard reaction was identified. Free amino acids, mainly asparagine, and reducing sugars have been known as its key precursors in sugar rich heat processed food. Processing conditions, such as time, temperature, water activity, and, as well matrix composition largely influence the kinetics of acrylamide formation and degradation. Intensive research is still ongoing in this research area. In our study we focused on the production of potato pancakes, a popular and widely consumed traditional Czech meal prepared either by baking, frying or deep-frying. We used commercial available powder and frozen semi-finished products. Our aim was to identify an optimal time and temperature for preparation of a final product with low acrylamide level and, at the same time, with good sensory properties. According to our results, the use of higher temperature and longer heat-processing time resulted in higher content of acrylamide in deep-fried and oven baked potato pancakes. Typically, the acrylamide levels in deep-fried and baked potato pancakes with acceptable sensory quality ranged from 59 to 673 µg/kg and from 111 to 813 µg/kg, respectively. In case of frying, the pancakes made of powder mixture contained lower levels of acrylamide compared to those made of semi-finished frozen product. The content of acrylamide in powder and frozen pancakes ranged from 15 to 31 µg/kg and from 253 to 702 µg/kg, respectively. When comparing the sensory acceptable products, lowest levels (15–31 µg/kg) of acrylamide were found in fried powder pancakes. At the same time a slight difference in taste/texture exists between the powder and frozen pancakes which can have an impact on consumers' preferences.

Keywords: Acrylamide, potato pancakes, heat-processing

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D-34

UV LEDS – A NEW TECHNOLOGY FOR IMPROVING FOOD QUALITY, SAFETY AND SHELF-LIFE

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Food waste is becoming a major problem around the world, with up to 50% of edible and healthy food wasted in households, supermarkets, restaurants and along the food supply chain. According to the US Natural Resource Defense Council (NRDC) "about 40% of all food produced in the US goes to waste". The European Parliament earlier this year stated that "up to 50% of food is wasted" and called for "urgent measures to halve food waste by 2025" claiming that if nothing is done, "wastage will grow 40% by 2020". Sensor Electronic Technology, Inc. (SETi) offers a new platform for diverse solutions to this growing problem: deep ultraviolet light emitting diodes (DUV LEDs). Today, visible LEDs are found in almost every electrical product and have recently found their way in to general lighting applications, replacing incandescent and compact fluorescent bulbs. SETi's technology has enabled extension of the wavelength range of LEDs deep into ultraviolet down to 210 nm. However, until recently, DUV LEDs have only been found in niche applications. This is about to change: DUV LEDs have many uses in production, transportation, monitoring and storage of food. SETi's DUV LEDs operating in the germicidal light range (250–280 nm) have the ability to destroy pathogens on surfaces, in air and in water. A DUV LED water disinfection chamber has been demonstrated by SETi and applications will be laid out where these devices can be used to reduce pathogens on the surface of meat and fish and in the air flow of a recirculating air storage system. A joint program of SETi and the US Department of Agriculture (USDA) has recently demonstrated that SETi's DUV LEDs operating in the range of 280–305 nm increase the nutritional quality of crops grown in green houses with a several fold increase in flavonoids generated in leaf lettuce. This technology is now being applied to a long-term post-harvest treatment of various fruits and vegetables to substantially increase shelf-life and retain nutritional value of produce. The applications for DUV LEDs in the food industry area are not limited to just treatment. These novel devices can also be used for real-time monitoring and control of the storage environment for pathogens, gases emitted by aging vegetables, such as ethylene, and for potential disinfection gases, including ozone. Unlike other source of ultraviolet light, DUV LEDs are uniquely suited to be used with foods: their cool operation temperature means that they do not cook or spoil food; unlike mercury lamps, they perform better in chilled environments; their compact physical size (size of grain of salt) and flexible design rules mean that they can be placed virtually anywhere; and their low power requirements enable them to be powered by a battery or solar cell, making this technique even suitable for transportation. The demonstrated results point to the potential of DUV LEDs for revolutionary changes in food quality, food safety and shelf-life.

Keywords: Ultraviolet, LED, disinfection, flavonoid, nutrition

Acknowledgement: *US Department of Agriculture*

D-35
NEW STANDARDS FOR PAPS – EMERGING FLUORINATED SURFACTANTS IN FOOD CONTACT MATERIALS

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Polyfluoroalkyl phosphate surfactants (PAPS) are a type of fluorinated chemicals that are used to grease and water proof food wrappers. The substances are applied to food contact paper as coating because they prevent oil soaking into paper from fatty food. A study in 2005 showed that PAPS and similar compounds used in these applications can leach from microwave-popcorn packaging into the food. Another study by theUniversity of Toronto chemists in 2007 showed that once ingested, PAPS are bioavailable and can be metabolized to form PFOA and other perfluorinated chemicals (PFC). PAPS and other fluorinated surfactants in food contact materials therefore seem to be a significant source of exposure of PFC to both humans and the environment. The European Commission recommend the monitoring of perfluoroalkylated compounds in food of plant and animal origin to enable an accurate estimation of exposure. Especially perfluorooctane sulfonate (PFOS), perfluorooctanoic acid (PFOA), their precursors and PAPS in particular need to be monitored in order to estimate the relevance of their presence in food. However, the lack of commercially available standards of PAPS is making it difficult to determine the exposure of humans to these compounds. A project for synthesis of standards for PAPS has been established at CHIRON and a series of fluorotelomer alcohol substituted phosphate surfactants, both native and duterated such as 8:2 monoPAPS, di-PAPS and tri-PAPS, has been synthesized and analyzed. Structure of deuterated 8:2 di-PAPS

Keywords: PAPS, PFAS, food wrapping, internal standards, surfactants

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D-36
IDENTIFICATION AND QUANTIFICATION OF „NON-PHTHALATE“ PLASTICIZERS IN PACKAGING MATERIALS USING DART TECHNIQUE

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The ambient ionization technique direct analysis in real time (DART) was used for the screening of food packaging materials for the presence of non-phthalate plasticizers (NPPs) using a mass spectrometer TOFMS. Optimization of DART-TOFMS response was characterized across various configurations (temperature, helium flow rate, voltage, sample speed) for a model solution of bis(2-ethylhexyl)adipate (DEHA). Molecular ion (M^+H^+) and ammonium adducts were obtained as the typical ionization products of NPPs (for example DEHA, DINA, ATBC, DBS). The possibility of quantification of NPPs was evaluated using internal standardization and isotopically labeled standards. Determination of NPPs in the commercially produced packaging materials was based on their direct extraction into the solvents (hexane, toluene, and methanol). It was also studied determination of NPPs directly without prior sample preparation or after corrupting of surface of sample by selected solvents.

Keywords: DART, packaging materials, non-phthalate, plasticizers

D-37

FOOD SAFETY, GMO QUANITATIVE AND QUALITATIVE ANALYSIS AND REGULATIONS IN GEORGIA

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Due to the market introduction of genetically modified organisms (GMOs) in crops, foods, and ingredients, legislation worldwide came face to face with the question of the use and labeling requirements on GMO crops and their derivatives. In this review, protein- and DNA-based methods, such as enzyme linked immunosorbent assay, western blots, and qualitative and quantitative polymerase chain reaction PCR (Q-PCR) are reviewed. Qualitative detection methods for genetically modified (GM) sequences in foods have evolved rapidly during the past years. The sensitivity of these systems is extremely high, even for processed foodstuffs. However, the availability of quantitative detection methods for GMO analysis is an important prerequisite for the introduction of threshold limits for GMOs in food. In Georgia Consumers continuously lack effective mechanisms to force government entities to react on their complaints. When a consumer's disagreement with the entrepreneur is about non-food products, the only available dispute resolution mechanism is to go to court. When the issue concerns a food product, then a consumer is able to address the National Food Agency (NFA). However, for the NFA to undertake an unplanned inspection of this enterprise, the consumer has (1) to detail the manner in which he or she obtained the said product sample (no sample form is available to provide guidance on as to how it should be done), (2) provide the results of a laboratory analysis of the product (lab testing is fairly expensive), and (3) to present a doctor's note confirming that the consumer was indeed poisoned after ingesting the product. With these requirements, the law puts undue burden on Georgian consumers to protect their rights, as the public authorities' obligation to ensure proper food safety and consumer protection is premised on the ordinary consumer's capacity to follow this complicated and expensive route of action. In addition, it is questionable to what extent the ! consumer is expressly authorized by this law to collect official food product samples.

Keywords: GMO, guanitative, qualitative, analysis, food, safety, Georgia

D-38
DECOMPOSITION OF CHLOROPROPANOL ESTERS IN THE MODEL SYSTEMS

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3-chloropropane-1,2-diol (3-MCPD) and its esters are classified as process contaminants of foods. The aim of this work was to design a model system that would allow to monitor the kinetics and reactivity of chloropropanol esters and related compounds (esters of glycidol with fatty acids). With regard to sporadic knowledge of the mechanism of these substances, these models should also help to confirm the expected mechanisms that lead to the formation of esters chloropropanols in foods, especially in vegetable oils. Kinetics of decomposition were observed from the starting materials 3-MCPD dipalmitate and 3-MCPD monopalmitate. Palmitic acid was chosen not only because of its frequent occurrence in the oils and hence food, but also with respect to its oxidative stability. It was established GC/MS method, which allows analysis of all expected compounds simultaneously. Deuterated analogues of above mentioned analytes were used as the internal standards together with deuterated palmitic acid for quantification. As an inert material to simulate the food matrix was chosen silica. In kinetic study are examined the effect of the chloride ion content, water content, and influence of temperature and heating time on the formation and decomposition of all compounds.

Keywords: 3-MCPD esters, model system, decomposition

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D-39
SELECTIVE POLYMERS FOR EXTRACTION OF DEMANDING COMPOUNDS FROM FOOD STUFFS

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'Designer polymers' are a class of selective polymer resins with built-in affinities for particular target molecules or 'classes' of molecules. Molecularly imprinted polymers are just one example of this type of polymer. Designer polymers are obtained by careful design of the required interaction chemistry. The choice of polymer building block is also important to ensure that the required surface chemistry and material morphology, properties of an adsorbent that can impact selective binding, are compatible with the application desired for a particular food matrix. An area where selective, designed polymers has significant practical applicability is in the preparative clean-up of liquid food stuffs; one important example is in the clean-up of flavour oils. Flavour oils are important ingredients in the food and beverage industry. Where such flavour oils may be contaminated with high levels of different agricultural residues (AR's), such as pesticides, removal of such AR's is critically important. Traditional unit operations such as distillation are not effective in the removal of many of these pesticides without detrimental impact on taste and aroma through simultaneous loss of critical taste components. To meet this challenge we have explored other clean-up routes, in particular the development and application of tailor-made, designed resins. A selective polymeric adsorbent is now available (from laboratory to process scale) that can be used to selectively remove a large number of different pesticides from flavour oils. After only a single pass through the resin, the level of pesticides in the flavour oils is drastically reduced. For many pesticides, the reduction is quantitative and no residues can be detected in the purified flavour oil. Very importantly, the aroma and taste profile of the purified flavour oils remain virtually unaffected and passes professional sensory panels. In this lecture, we would like to describe the characteristics and the performance of these designed resins for efficient preparative removal of undesired substances from food stuffs. In addition, we would like to point out further examples and other applications such as trace analysis of undesired compounds in food samples utilizing designed polymers.

Keywords: Selective, polymers, purification, extraction, clean-up

D-40

EFFECT OF HOUSEHOLD AND INDUSTRIAL PROCESSING ON LEVELS OF SOME PESTICIDE RESIDUES IN CARROTS

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Pesticides are widely used in conventional agriculture to obtain a better yield for crops. They can cause toxic effects (headaches, cancer, reproductive harm and endocrine disruption). The main exposure to pesticides for humans is via food (especially by fruit and vegetables). Processing food can affect the level of pesticide residues and in some special case, more toxic by-products can be formed during processing. Different studies have been related to measure concentrations of pesticide residues after home or industrial processing. However, many processing factors (residues level in processed commodity/residue level in raw commodity) remain unknown and are necessary to estimate the level of pesticide exposure at the point of consumption after processing. To reach this objective, the combination carrots/pesticides (boscalid, chlorpyrifos, difenoconazole, dimethoate, linuron and tebuconazole) was chosen in collaboration with the Belgian Federal Agency for Safety of the Food Chain based on the number of non compliant samples, frequency and concentration level observed and on the toxicity of the pesticides. Treated carrots were grown in Belgium by Redebel sa. The amount of the pesticide residues observed in the raw material varied due to the different treatments, application times, weather conditions, the physico-chemical properties of each pesticide. Food processing (washing, peeling, blanching, microwave cooking, pasteurization and sterilization) was carried out on industrial pilot scale. Pesticides and the degradation products formed during processing were analyzed with GC–MS/MS and UHPLC–MS/MS. The washing step allowed decreasing the concentration of residues for all pesticides up to ~90%. The second process, peeling, results in a reduction comparable to washing. The blanching step, combining heat with a large quantity of water, enhanced the elimination of residues. Even residual concentrations were below 5 ppb, it was observed that microwave cooking did not reduce the level of residues while in-pack sterilization decreased most of the pesticide residues still present except difenoconazole. To conclude, most of the processing factors could be explained in terms of water solubility, the log-octanol-water-partitioning coefficients, the systemic properties of the pesticides studied and the agricultural practices.

Keywords: Pesticide residues, home processing, processing factor, carrots

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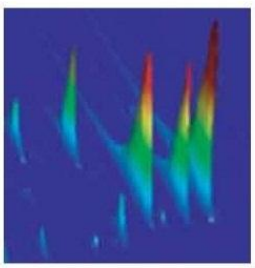

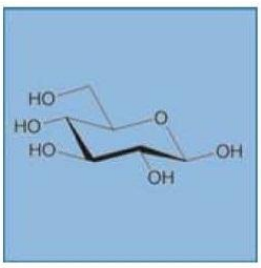

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