



Universitat
de les Illes Balears



«Towards a *Redox Healthy Aging*»

WGs Meeting of the NutRedOx COST Action CA16112

15th – 16th February 2018

Palma, Mallorca





Scientific Committee

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 Prof. Josep A. Tur, University of the Balearic Islands & CIBEROBN, Palma, Mallorca, Spain

Sponsors: COST Action CA16112; Directorate General of Research, Development and Innovation of the Balearic Islands Government; University of the Balearic Islands; CIBEROBN.



Venue: Son Espanyol Auditori, c/ Laura Bassi, ParcBit, Palma-Valldemossa road.
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Foreword

The NutRedOx COST Action (CA16112) entitled “*Personalized Nutrition in aging society: redox control of major age-related diseases*” started in March 2017. Its scientific items are closely linked to the NutriOx network. The NutRedOx COST Action is gathering experts from 35 European, Asiatic and Mediterranean countries, and from different disciplines that are involved in the study of biological redox active food components that are relevant to the ageing organism, its health, function and vulnerability to disease.

In the tradition of the previous NutriOx and NutRedOx workshops, this WGs Meeting will provide an international forum for early stage scientists, graduate and PhD students, as well as Post-Docs. Early stage scientists are strongly encouraged to present their research (oral communications and posters) and to network with more senior scientists and research experts from the region, but also from across Europe. This WGs Meeting also facilitates contacts with potential host laboratories.

Timetable

- The scientific Meeting of the NutRedOx COST Action CA16112: 15th and 16th February (2 full days).
- The Core Group Meeting CA16112: 14th February (1/2 day).
- The Gala Dinner: 15th February 20:30 h.

Transportation to Meeting site: Line 19 bus EMT from city downtown to ParcBit (last stop): <http://www.emtpalma.cat/en/route/-/L/19/universitat>

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EDIFICIS

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|-----------------------------|------------------------------------|
| 1 Son Espanyol (AUDITORI) | 12 Edifici Europa |
| 2 Edifici Telecomunicacions | 13 Edifici Logitravel |
| 3 Escoleta Maria Serra | 14 Edifici ATB |
| 4 Edifici Adduno | 15 Edifici TUI |
| 5 Edifici Closell | 16 Blue Building |
| 6 Edifici Lleret | 17 Edifici Disset |
| 7 Edifici Mungra | 18 Edifici Naorte |
| 8 Complex d'R+D | 19 Edifici W |
| 9 Edifici NTIC | 20 Col·legis Alxa, Llaüt i Aladern |
| 10 Edifici Estel | 21 Edifici Manteniment |
| 11 Edifici U | 22 Planta d'Energia |

LLEGGENDA

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| Aparcament | Informació | Cafeteries/serveis |
| Aturada de bus | Auditori | Dubus |
| Edifici | Sense edificar | Vianants |



PROGRAM

Thursday 15 February 2018

08:30-09:00 Arrival and registration, coffee

09:00-09:30 Welcome and Opening remarks

Prof. Josep A. Tur (University of the Balearic Islands & CIBEROBN)

Prof. Mustapha Cherkaoui Malki (U. Burgundy) & Prof. Agnieszka Bartoszek (U. Gdansk) – Chair & Co-Chair NutRedOx COST Action CA16112

Mr. Josep Lluís Pons, Director General R+D+I, Government of the Balearic Islands

Prof. Llorenç Huguet, Rector, University of the Balearic Islands

Session 1: Nutrition & Health (1)

Chairperson: Prof. Mourad Elhabiri (University of Strasbourg)

09:30-09:45 *Communication 1:* J. Antosiewicz¹, A. Borkowska¹, M. Halon-Golabek¹, J. Koratsa², E. Ziemann² A. Herman-Antosiewicz³. (¹Medical University of Gdansk, Gdansk, Poland. ²University of Physical Education and Sport, Gdansk, Poland. ³University of Gdansk, Gdansk, Poland). **Insulin signalling and iron metabolism –role of exercise**

09:45-10:00 *Communication 2:* V. Mocanu, R.E. Haliga, C. Galesanu, C.G. Dascalu, E.M. Carausu. (Grigore T. Popa University of Medicine and Pharmacy, Iasi, Romania). **Changes of lipid peroxidation and 25-hydroxyvitamin D associated in nursing home residents with metabolic syndrome**

10:00-10:15 *Communication 3:* V. Mocanu, R.E. Haliga, C. Galesanu, C.G. Dascalu, E.M. Carausu. (Grigore T. Popa University of Medicine and Pharmacy, Iasi, Romania). **Changes of lipid peroxidation and 25-hydroxy vitamin D associated in nursing home seniors residents with metabolic syndrome**

10:15-10:30 *Communication 4:* D. Marko, G. Aichinger. (University of Vienna, Vienna, Austria). **Differential protective profile of anthocyanins and isoflavones against pro-oxidative food contaminants**

10:30-10:45 *Communication 5:* P. Martínez, K. Forman, M. Martorell. (University of Concepción, Chile). **Hypotensive effect of an infusion of olive leaves in humans: possible antioxidant and anti-inflammatory mechanism of oleuropein in blood pressure**

10:45-11:30 **Poster session and networking + Coffee Break**

Session 2: Nutrition & Health (2)

Chairperson: Prof. Patrick Chaimbault (University of Lorraine)

11:30-11:45 *Communication 6:* C. Busquets-Cortés; X. Capó; E. Argelich; M.D. Ferrer; D. Mateos; A. Arenas; J.A. Tur; A. Sureda; A. Pons. (University of the Balearic

- Islands & CIBEROBN). **Effects of chronic hydrogen peroxide exposition on inflammatory parameters in 'ex vivo' neutrophils**
- 11:45-12:00 *Communication 7: A.S. Yalçın¹, A.M. Yılmaz¹, E.M. Altundağ², S. Koçtürk^{1,3}. (1Marmara University, İstanbul, Turkey. 2Eastern Mediterranean University, Famagusta, North Cyprus. 3Dokuz Eylül University, İzmir, Turkey). **Mechanism of anti-cancer effects of curcumin, quercetin and tea catechins***
- 12:00-12:15 *Communication 8: T. Ozben¹, A. Cort². (1Akdeniz University, Antalya, Turkey. 2Sanko University, Gaziantep, Turkey). **Natural redox modulators of oxidative stress and chemoresistance in cancer therapy: beneficial versus deleterious effects***
- 12:15-12:30 *Communication 9: M. Baranowska, K. Suliborska, J. Namieśnik, A. Bartoszek. (Gdansk University of Technology, Gdansk, Poland). **Flavan-3-ols interaction with antioxidant defence system of cells***
- 12:30-14:00 **Poster session and networking + Lunch**

Session 3: Isolation, (Bio)characterization, Analysis and Effects of Antioxidants

Chairperson: Prof. Rosita Gabbianelli (University of Camerino)

- 14:00-14:15 *Communication 10 N. Sahakyan, M. Zareyan, S. Hambardzumyan, M. Petrosyan, A. Trchounian (Yerevan State University, Yerevan, Armenia). **In vitro antioxidant potential of ethanol extracts of different plants from Armenian flora***
- 14:15-14:30 *Communication 11: M. Đorđević, M. Mihailović, J. A. Jovanović, N. Grdović, A. Uskoković, M. Sinadinović, J. Rajić, A. Tolić, G. Poznanović, M. Vidaković, S. Dinić. (University of Belgrade, Belgrade, Serbia). **Protective effect of Centaurium erythraea methanol extract against oxidative challenge in red blood cells of diabetic rats***
- 14:30-14:45 *Communication 12: V. Todorovic¹, D. Jancic², N. Dabetic¹, B. Vidovic¹, I. Djuricic¹, D. Žnidarčić³, S. Sobajic¹. (1University of Belgrade, Belgrade, Serbia; 2LLC Center for Ecotoxicological Research, Podgorica, Montenegro; 3Biotechnical Faculty University of Ljubljana, Ljubljana, Slovenia). **Dietary antioxidants in green leafy vegetables***
- 14:45-15:00 *Communication 13: K. Suliborska¹, M. Baranowska², A. Bartoszek², J. Namieśnik³, W. Chrzanowski¹. (Gdansk University of Technology, Gdansk, Poland). **The relationship between standard reduction potential and thermodynamic constants of antioxidant compounds – creation of Antioxidant Power Series***
- 15:15-15:30 *Communication 14: E. Arranz¹, A. R. Corrochano^{1,2}, M. Villalva³, L. Jaime³, S. Santoyo³, E. Murphy¹, L. Giblin¹. (1Teagasc Food Research Centre, Fermoy, Ireland; 2University College Dublin, Dublin, Ireland; 3Institute of Food Science Research (CIAL, CEI UAM+CSIC). Madrid, Spain). **Antioxidant activity of whey-based beverages: Effect of shelf life and gastrointestinal transit on bioactivity***
- 15:00-15:30 *Communication 15: C. Gaucher. (Université de Lorraine, Nancy, France). **IMPACT Biomolecules***
- 15:30-16:00 **Poster session and networking + Coffee Break**

Session 4: Chairpersons: Prof. Elke Richling (Univ. Kaiserslautern) & Prof. Josep A. Tur (Univ. Balearic Islands & CIBEROBN)

16:00-18:00 **WG 2 Meeting:** Discussion of future WG2 actions. Final remarks

WG 2 Goals:

- WG 2 - GP goal A1: To assess the impact of redox active food and individual food microcomponents on healthy, ageing organisms, as well as those suffering from age-linked diseases.
- WG 2/4 - GP goal A2: To define redox biomarkers considered as indicators of a changing health.
- WG 2 - GP goal B: To assess relationships between dietary intake, lifestyle and socio-demographic determinants on one-side and plasma levels of specific biomarkers.
- WG 2/4 - GP goal C: To assess relationships between redox biomarkers levels and ageing organisms, as well as those suffering from age-linked diseases
- WG 2/4 - GP goal D: To assess reversion of age-linked diseases after intervention by redox biomarkers.

20:30 Conference Dinner

Friday 16 February 2018

Session 5: Biological Activity and Intracellular Effects

Chairpersons: Prof. Antoni Sureda (University of the Balearic Islands & CIBEROBN)

- 09:00-09:15 *Communication 16:* R. Leontiev^{1,2}, J. Nasim², Y. Ney², P. Denezhkin², A. Uebachs¹, M.C.H. Gruhlke¹, A.J. Slusarenko¹, C. Jacob². (¹Aachen University; ²Saarland University). **Deciphering modes of action: from chemogenetic phenotype profiling to intricate intracellular diagnostics**
- 09:15-09:30 *Communication 17:* S. Sasson¹, G. Maulucci², C. Ferreri³. (¹The Hebrew University, Jerusalem, Israel; ² The Catholic University of the Sacred Heart, Rome, Italy; ³Consiglio Nazionale delle Ricerche, Bologna, Italy). **Role of lipid overload on the fluidity of plasma and organelle membranes and subcellular distribution of lipids in cells**
- 09:30-09:45 *Communication 18:* K.E. Kypreos. (University of Patras, Panepistimioupolis, Greece). **Site-specific effects of apolipoprotein E expression on white adipose tissue mitochondrial substrate oxidation for energy production**
- 09:45-10:00 *Communication 19:* K. Parchem, A. Bartoszek. (Gdansk University of Technology, Gdansk, Poland): **Qualitative assessment of oxidized phospholipids generated as a result of enzymatic oxidation and their digestion by phospholipase A₂**
- 10:00-10:15 *Communication 20:* I. Bazukyan, T. Aleksanyan, A. Trchounian. (Yerevan State University, Yerevan, Armenia). **GUT microbiota and probiotics: In vitro and in vivo hydrophobicity and adhesion of lactic acid bacteria**
- 10:15-10:30 *Communication 21:* R. Gabbianelli, L. Bordoni, D. Fedeli, C. Nasuti. (University of Camerino, Italy). **Preventive strategies to counterbalance food pesticide effect on epigenome and gut microbiota**



10:30-11:15 **Poster session and networking + Coffee Break**

Session 6: Chairpersons: Prof. Nina Hermans (Univ. Antwerpen) & Prof. Mustapha Cherkaoui Malki (U. Burgundy)

11:15-13:15 **WG 3 Meeting:** Discussion of future WG3 actions. Final remarks

WG 3 Goals:

- WG 3 - GP goal A1: To list implemented models and methods in different WG3 member labs.
- WG 3 - GP goal A2: To list known natural product (NP) molecules metabolized by the microbiota.
- WG 3 - GP goal B: Catalogue the known NP derivatives issued from microbiota metabolism.
- WG 3/4 - GP goal C: To address known microbiota composition changes or signalling pathways impacted by NP molecules.
- WG 3/4 - GP goal D: To review health effects of NP microbiome-derived metabolites.

13:15-14:30 **Poster session and networking + Lunch**

Session 7: Chairperson: Prof. Ana Fernandes (Universidade Lusofona)

14:30-15:00 Closing Lecture: A. Bartoszek¹, Klaudia Suliborsk², Monika Baranowska¹, Barbara Kusznierevicz¹, Agata Kot-Wasik³, Wojciech Chrzanowski², Jacek Namieśnik³ (Gdansk University of Technology, Gdansk, Poland). **Antioxidant Power Series project; state of art after three years.**

15:00-15:30 **Poster session and networking + Coffee Break**

Session 8: Chairpersons: Prof. Marc Diederich (Laboratoire de Biologie Moléculaire et Cellulaire du Cancer & Univ. Seoul) & Dr. Linda Giblin (Teagasc Food Research Centre)

15:30-17:30 **WG 4 Meeting:** Discussion of future WG4 actions. Final remarks

WG 4 Goals:

- WG4 - Year 1: To list known natural product molecules investigated by NutRedOx related to ageing; To list cellular models of NutRedOx labs that are indicators of ageing or age-linked diseases; To list food/nutrients investigated by NutRedOx, that are bioavailable (cross the intestinal barriers) and have known bioactivity relevant to ageing.
- WG 4 - Year 2: To describe the effects of individual (or combinations of) food bioactives on cellular models; To catalogue known natural products derivatives with these pathways.
- WG 4 - Year 3 & 4: To investigate epigenetic regulators including HDAC modulators; To investigate anti-inflammatory/antioxidant effects of natural products; To link in vitro biomarker outputs with in vivo biomarkers and thereby validate in vitro models.

Session 9: Chairpersons: Prof. Mustapha Cherkaoui Malki (U. Burgundy) & Prof. Agnieszka Bartoszek (U. Gdansk)

17:30-18:30 WGs Meeting: Final remarks



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CLOSING LECTURE



IL1 - Antioxidant Power Series project; state of art after three years

Agnieszka Bartoszek^{1*}, Klaudia Suliborska², Monika Baranowska¹, Barbara Kuzniewicz¹, Agata Kot-Wasik³, Wojciech Chrzanowski², Jacek Namieśnik³

¹Dept. of Food Chemistry, Technology and Biotechnology, ²Dept. of Physical Chemistry, ³Dept. of Analytical Chemistry, Gdansk University of Technology, 11/12 Narutowicza St., 80-233 Gdansk, Poland, *agnieszka.bartoszek@pg.gda.pl

Over past two decades, plantborne antioxidant substances have been a hot topic in the area of food and nutrition, chemoprevention of civilization diseases, as well as cosmetology. Plant antioxidants are generally recognized as synonyms of nutraceuticals or nutricosmetics, at least among consumers and producers. Popularity and associated with it commercial success of antioxidants has been based on mechanistic studies that suggested the involvement of reactive oxygen species (ROS) in the aetiology of chronic diseases. However, it has not been overlooked by research community, that the results of extensive epidemiological studies conducted among different populations, including various risk groups, have not provided unequivocal confirmation of the protective activity of plantborne antioxidants in people exposed on oxidative stress. Such an ambiguity caused certain disappointment and decreased the interest in antioxidants as chemopreventive agents.

Meanwhile, the importance of ROS in the regulation of several vital processes responsible for the proper functioning of eukaryotic organisms at all stages of the development has become evident. In this new context, ROS are no longer perceived solely as cell damaging factors (as in oxidative stress), but also regulators of pivotal cell functions, where their depletion impairs cellular processes (as in reductive stress). In what way and by what mechanisms, exogenous antioxidants can support proper cellular redox homeostasis remains to be established.

The aim of the project „Antioxidant Power Series as a tool rational design and assessment of health promoting properties of functional food based on antioxidant phytochemicals” is to fill in this gap by elaboration of Antioxidant Power Series (APS) for antioxidants that may be produced endogenously or are found as natural food components or components of plants (and other organisms) of medical importance. This series is supposed to resemble the electrochemical series, which enables chemists to predict behaviour of redox pairs in different systems. Similarly as hydrogen constitutes the point "zero" in electrochemical series, in the case of APS as such a reference point glutathione (GSH), the main physiological antioxidant, is proposed. All other substances to be assessed will be compared to GSH based on their reduction potential. They will possess negative or positive values of antioxidant power depending on the value of their reduction potential in comparison with the reference compound.

I will share with the COST Action CA16112 members our observations and more general presumptions which research on the development of APS enabled us to come up with.

Project „Antioxidant Power Series as a tool rational design and assessment of health promoting properties of functional food based on antioxidant phytochemicals” (number of the application 2014/14/A/ST4/00640) financed by National Science Centre, Poland in a programme „MAESTRO 6”



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ORAL COMMUNICATIONS



OC1 - Insulin signalling and iron metabolism –role of exercise

Jedrzej Antosiewicz¹, Andzelika Borkowska¹, Malagorzata Halon-Golabek¹, Jakub Koratsa², Ewa Ziemann², Anna Herman-Antosiewicz³.

¹Medical University of Gdansk, Department of Bioenergetics and Physiology of Exercise, 80-211 Gdansk, Debinki 1, Poland. ²Department of Physiology and Pharmacology, University of Physical Education and Sport, Kazimierza Gorskiego 1, 80-336 Gdansk, Poland. ³Department of Molecular Biology, University of Gdansk, Gdansk, Poland.

An increase in a body iron stores and tissue iron accumulation is a significant risk factor for age related morbidities like cancer, heart attack, diabetes and many others. Regular exercise has been shown to reduce oxidative stress and increase insulin sensitivity. Interestingly, evidence exists for a positive correlation between the levels of a body iron stores and insulin resistance. Thus, the main goal of our study was to investigate the relationship between tissue iron accumulation and insulin signaling.

In this communication, we provide evidence that impairment of insulin signaling leads to iron accumulation and skeletal muscle atrophy. In transgenic animals expressing SOD1 G93A, which is a good model for impaired insulin signaling, a decrease in Akt activity was accompanied by an increase in ferritin L and H proteins and rise in muscle iron content. This was accompanied by an increase in the active form of FOXO3a, upregulation of atrogen-1, catalase, and muscle atrophy. In SH-SY5Y cells stably expressing SOD1 or SOD1 G93A we observed elevated levels of ferritin L, H and non-hem iron. Insulin treatment significantly downregulated ferritin L and H proteins in the cell which confirms a role of insulin signaling in iron metabolism. Conversely, cells transfected with siRNA against Akt1, 2 or 3 showed a significant increase in the ferritin and FOXO3a levels. In order to assess the role of FOXO3a in the ferritin expression, we constructed a SH-SY5Y cell line expressing FOXO3a fused at the C-terminus with the ligand-binding domain of the estrogen receptor (TM-ER) being activated by 4-hydroxytamoxifen (4OHT). Treatment of cells with 4OHT significantly upregulated ferritin L and H proteins level.

Experiments performed on elderly women who regularly exercise confirmed the results obtained on animals and cell lines. We observed that 12 weeks of training reduced body iron stores and this was accompanied by lower oxidative stress. Interestingly, fasting blood glucose concentration negatively correlated with blood ferritin level (body iron stores). Concluding, our data suggest that impairment of insulin signaling leads to alteration in iron metabolism and vice versa.



OC2 - Changes of lipid peroxidation and 25-hydroxyvitamin D associated in nursing home residents with metabolic syndrome

Veronica Mocanu, Raluca Ecaterina Haliga, Corina Galesanu, Cristina Gena Dascalu, Elena Mihaela Carausu.

Grigore T. Popa University of Medicine and Pharmacy, 16, Universitatii street, Iasi, 700115, Romania.

Objective. To examine the association of serum oxidative stress and 25hydroxyvitamin D with metabolic syndrome (MetS) in nursing home residents.

Method. We enrolled 50 institutionalized seniors (15 men and 35 women), age 72.6+ 6.5 years living in a nursing home in Iasi, Romania. We investigated the oxidative stress by measuring serum malondialdehyde (thio barbituric acid reactive substances, TBARS), the erythrocyte reduced glutathione, GSH (using di thio bis nitro benzoic acid, DTNB), the superoxide dysmutase activity, SOD (SOD's inhibition of the reaction of superoxide anion, from xanthine by xanthine oxidase and the reduction of nitroblue tetrazolium, NBT), and the catalase activity, CAT (residual hydrogen peroxide after incubation with the enzyme). We evaluated the metabolic syndrome parameters using the reference values of the WHO Definition of the Metabolic Syndrome criteria.

Results. The prevalence of MetS in participants of this study was 54% (27 of 50 patients). The lipid peroxidation (TBARS) was significantly increased ($p<0.05$) in nursing home residents with MetS as compared with those without MetS. GSH, SOD and CAT did not significantly change between the groups. The serum concentration of 25-hydroxyvitamin D was significantly decreased ($p<0.05$) in patients with MetS. MNA score, bone mineral density, and BMD did not change significantly between old aged patients with and without MetS.

Conclusion. Our data revealed that oxidative stress status and 25hydroxyvitamin D could be used as markers of pathophysiological changes associated with MetS in seniors.

Keywords: metabolic syndrome, oxidative stress, 25-hydroxyvitamin D, aging, nursing home.



OC3 - Changes of lipid peroxidation and 25-hydroxy vitamin D associated in nursing home seniors residents with metabolic syndrome

Veronica Mocanu, Raluca Ecaterina Haliga, Corina Galesanu, Cristina Gena Dascalu, Elena Mihaela Carausu.

Grigore T. Popa University of Medicine and Pharmacy, 16, Universitatii street, Iasi, 700115, Romania.

Objective. To examine the association of serum oxidative stress and 25hydroxyvitamin D with metabolic syndrome (MetS) in nursing home residents.

Method. We enrolled 50 institutionalized seniors (15 men and 35 women), age 72.6+ 6.5 years living in a nursing home in Iasi, Romania. We investigated the oxidative stress by measuring serum malondialdehyde (thio barbituric acid reactive substances, TBARS), the erythrocyte reduced glutathione, GSH (using di thio bis nitro benzoic acid, DTNB), the superoxide dysmutase activity, SOD (SOD's inhibition of the reaction of superoxide anion, from xanthine by xanthine oxidase and the reduction of nitroblue tetrazolium, NBT), and the catalase activity, CAT (residual hydrogen peroxide after incubation with the enzyme). We evaluated the metabolic syndrome parameters using the reference values of the WHO Definition of the Metabolic Syndrome criteria.

Results. The prevalence of MetS in participants of this study was 54% (27 of 50 patients). The lipid peroxidation (TBARS) was significantly increased ($p<0.05$) in nursing home residents with MetS as compared with those without MetS. GSH, SOD and CAT did not significantly change between the groups. The serum concentration of 25-hydroxyvitamin D was significantly decreased ($p<0.05$) in patients with MetS. MNA score, bone mineral density, and BMD did not change significantly between old aged patients with and without MetS.

Conclusion. Our data revealed that oxidative stress status and 25hydroxyvitamin D could be used as markers of pathophysiological changes associated with MetS in seniors.

Keywords: metabolic syndrome, oxidative stress, 25-hydroxyvitamin D, aging, nursing home

OC4 - Differential protective profile of anthocyanins and isoflavones against pro-oxidative food contaminants

Doris Marko, Georg Aichinger.

University of Vienna, Dept. of Food Chemistry and Toxicology, Währingerstr. 38, 1090 Vienna, Austria.

Many dietary flavonoids are described as antioxidants, acting as scavengers of reactive oxygen species and/or inducing the expression of anti-oxidant enzymes, in particular via the Nrf2 pathway. Those effects are usually shown in test systems involving synthetic model compounds to induce oxidative stress like e.g. hydrogen peroxide. We addressed the questions a) if comparable positive effects might be observed against naturally occurring pro-oxidants, e.g. microbial food contaminants and b) whether the induction of protective enzyme systems or direct interaction might be crucial for the biological activity.

Thus, we investigated the impact of the berry anthocyanidin delphinidin and the soy isoflavone genistein, on the toxicity of selected mycotoxins. Despite all technical development, the contamination of food with mycotoxins is still an unsolved problem. Advances in the production chain, monitoring and regulation brought great efforts towards food safety. Nevertheless, although within the regulatory limits, we are continuously exposed to, at least, low levels of mycotoxins by consumption of plant-derived food. In this low level exposure levels, several mycotoxins are known to induce oxidative stress. Recently, we demonstrated that alternariol (AOH) and altertoxin-II (ATX-II) secondary metabolites formed by the black molds *Alternaria alternata*, possess potent pro-oxidative properties [1]. The main sources of *Alternaria* toxins in the human diet are cereals and cereal-based products, tomato and sauces, sunflower seeds and oil, fruits, beer and wine.

In HT29 colon carcinoma cells, delphinidin was found to suppress the enhanced oxidative stress induced by AOH and ATX-II. However, this effect was not mediated by an activation of Nrf2, as a pre-incubation with delphinidin did not result in a reduction of contaminant-induced ROS levels in the DCF assay. However, in contrast to the anthocyanidin, genistein did not show any protective effect against the pro-oxidative properties of the mycotoxins [2].

Despite the different effectiveness of delphinidin and genistein against the oxidative properties of the mycotoxins, both flavonoids decreased the DNA damaging impact of the *Alternaria* toxins in the comet assay [2]. In the case of AOH, both flavonoids interfered with the topoisomerase-targeting properties of the mycotoxin, thus suppressing genotoxicity. Furthermore, delphinidin was found to diminish the level of reactive ATX-II, which might prevent the formation of genotoxic DNA-adducts [3].

Taken together, our studies demonstrate the high potential of flavonoids to counteract pro-oxidative and genotoxic effects of food contaminating mycotoxins, with anthocyanidins showing stronger protective effects as compared to isoflavones.

References:

1. Tiessen, C., et al., Modulation of the cellular redox status by the *Alternaria* toxins alternariol and alternariol monomethyl ether. *Toxicol Lett*, 2013. 216(1): p. 23-30.
2. Aichinger, G., J. Beisl, and D. Marko, Genistein and delphinidin antagonize the genotoxic effects of the mycotoxin alternariol in human colon carcinoma cells. *Mol Nutr Food Res*, 2017. 61(2).

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Keywords: anthocyanins, delphinidin, genistein, mycotoxins, oxidative stress, topoisomerase.

OC5 - Hypotensive effect of an infusion of olive leaves in humans: possible antioxidant and anti-inflammatory mechanism of oleuropein in blood pressure

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Background: The beneficial effects of olive leaf extract are known by the Mediterranean population since antiquity for the alleviation of inflammatory diseases, gout and blood pressure. The most abundant component in dry leaf extracts is oleuropein which has exhibited a variety of biological actions such as antioxidant, antimicrobial, anti-inflammatory, vasodilator, antihypertensive and hypoglycemic. Arterial hypertension is a chronic disease worldwide distributed and its treatment is based on drug and diet therapy which can be complemented with natural therapies such olive leaf infusions. The aim of this study was to evaluate the effect of an olive leaf infusion on blood pressure levels in humans.

Methodology: N-of-1 trial, randomized single blind, was carried out with 2 parallel study groups alternating 3 treatments: the daily consumption of 100 mL of infusion (2 or 1 g/L olive leaf or water) for 4 consecutive days in a week. The number of participants was 24 and the duration of the study was 3 weeks. Systolic and diastolic blood pressure was measured before and 5-10 min after infusion intake.

Results: The intake of 1 and 2 g/L of olive leaf infusion decreased systolic and diastolic blood pressure and a chronic effect was observed only on systolic blood pressure. Men presented higher blood pressure than women and participants aged over 40 years also presented higher values than participants under 40 years. Woman and participants under 40 years not presented significantly differences between the consumption of 1 or 2 g/L olive leaf infusion, but men and the participants aged over 40 years presented differences between doses, showing a major effect of 2 g/L olive leaf infusion.

Conclusion: Olive leaf infusion intake decreases systolic and diastolic blood pressure. Men and the participants aged over 40 years usually present elevated blood pressure values and may be more benefited with the effects of the higher dose of olive leaf tested. Further studies will be needed for determine the effects of olive leaf infusion and the role of their components on hypertension disease.



OC6 - Effects of chronic hydrogen peroxide exposition on inflammatory parameters in 'ex vivo' neutrophils.

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Reactive oxygen species (ROS) are considered to be unavoidable by-products of aerobic metabolism that can act as signal molecules produced during functional activities but also play a key role in various pathological conditions when antioxidant capabilities are overwhelmed by oxidant production. Exercise is associated with an increase in the generation of free radicals in a bell-shaped curve: low concentrations show stimulating effects (signalling, receptor or enzymatic stimulation), while a massive level of ROS inhibits enzyme activity and causes apoptosis or necrosis.

We have developed a methodology to validate 'ex vivo' the effects of oxygen peroxide (H₂O₂) on gene expression of differentiated human neutrophils. Neutrophils from 35 patients with metabolic syndrome were exposed for 2 hours to a continuous H₂O₂ production by glucose oxidase (15 and 0,5 µg glucose oxidase/ml culture medium) catalysing the glucose oxidation to gluconate in order to emulate an in vitro a physical activity situation.

We observed an exposure–response relationship in inflammatory gene expression. Some of the genes studied responded independently of the H₂O₂ concentration (eg. COX2, IL6, IL8, TLR4, and CuZnSOD mRNAs expression augmented significantly under both treatments), while other genes showed a dose-related response (TNFα mRNA expression only increased under low H₂O₂ exposure and catalase only augmented under high H₂O₂ exposure) or did not experiment a significant change in their mRNA expression (MnSOD, TLR2, IL1β, IL1α, NFκB).

We have established a methodology that allows tracking 'ex vivo' differential cellular responses to external stimuli. The developed system is fully working and is available to determine dissimilar sensibilities of various interesting pro-inflammatory, anti-inflammatory and antioxidant genes to continuous H₂O₂ exposure or to the action of key biomolecules.

Keywords: neutrophils, oxidative stress, oxygen peroxide, ROS.

This study was supported by Instituto de Salud Carlos III (PI11/01791, PI14/00636, PI17/01827, RETIC RD06/0045/1004, & CIBEROBN CB12/03/30038), EU-COST Action CA16112, Grant of support to research groups no. 35/2011 (Balearic Islands Gov.) and EU-FEDER funds.



OC7 - Mechanism of anti-cancer effects of curcumin, quercetin and tea catechins

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Polyphenols are present in high amounts in all parts of plants including roots, seeds, flowers, leaves, branches and trunk as well as plant derived products such as tea, coffee and wine. Extensive amount of information is available on biological effects of polyphenols including antioxidant, anticancer, anti-inflammatory, anticoagulant and antimicrobial activities. In recent years, researchers have turned their interest towards identification of molecular mechanisms underlying the anticancer effects of these compounds. However, the limited bioavailability of polyphenols and the existence of differences in cancer cells in terms of intracellular mechanisms resulted in the use of specific approaches to individual cancer cell types as well as methods of increasing bioavailability. In this presentation molecular mechanisms of anticancer effects of curcumin, quercetin and tea catechins in different cancer cells will be discussed.

Keywords: Curcumin, quercetin, tea catechins, anti-cancer effects, apoptosis.

OC8 - Natural redox modulators of oxidative stress and chemoresistance in cancer therapy: beneficial versus deleterious effects

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The roles of oxidative stress in physiology and pathology have been intensively studied over the last decades, but the problem is still far beyond our full comprehension. The roles of free radicals and antioxidants have been entirely redefined recently. Free radicals widely recognized as absolute evils causing damage to biologically important molecules and structures, have been recently transformed into positive actors, in the appreciation of their essential impact in the intracellular signaling and regulation of apoptosis. In contrast, the great hope that antioxidants could be the panacea resolving practically many health problems has vanished, due to the growing number of inconclusive or negative data from studies. Multiple drug resistance (MDR) may develop against chemotherapeutic agents with unrelated chemical structure and mechanism of action used for the treatment of cancer, reduces the efficacy of drugs, and remains as a major challenge in the treatment of cancer. A complex redox pattern underlies MDR problem. Natural product modulators of MDR are used as low toxicity chemosensitizers to enhance the efficacy of anticancer protocols and to overcome MDR. Redox active drugs could provide a valid and promising way to overcome MDR in cancer therapies via targeting an axis consisting of drug transporters, aryl hydrocarbon receptor, phase I/II metabolic enzymes, and the inducible Nrf2-linked pathway. The mechanism underlying the MDR inhibition by natural products obtained from plants and fungi lies in the blockade of the drug binding site, interference with the ATP hydrolysis process, alteration in integrity of cell membrane lipids, and decrease in Pgp or/and MRP1 expression. During coadministration, natural modulators compete with cytotoxic agents for binding to the active site of the transporters and reduce drug efflux. However, beneficial versus deleterious effects of these substances must be well evaluated in chemoresistance and cancer therapy.

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OC9 – Flavan-3-ols interaction with antioxidant defence system of cells

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To pinpoint actual role of dietary antioxidants in supporting endogenous antioxidant defence system in cells, the relation between their physicochemical, electrochemical and biological properties must be established. Unfortunately, despite the large number of publications on antioxidants, attempts to find such connections have been only rarely taken. It is still not fully recognized which properties of dietary antioxidants make real difference to organism redox homeostasis.

The aim of the study was to clarify the relationship between electrochemical properties of 5 flavan-3-ols, their biological behaviour as well as molecular implications for oxidative stress response and antioxidant defence system in colon adenocarcinoma HT29 cell line. Our findings showed, that standard reduction potential of antioxidants can be helpful to predict their behaviour in cellular model. We observed that catechol moiety in catechins structure had great influence on their reducing power, which was seen both in chemical and biological tests. Protection against oxidative DNA damage in HT29 cells strongly depended on standard reduction potential of antioxidants. Furthermore, low concentrations of catechins, which are potentially achievable in blood stream, played a key role in antioxidant defence system of cells, especially at genomic level, while higher concentrations relevant only for intestinal epithelium did not show such a strong impact.

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Keywords: antioxidants, reduction potential, antioxidant defence system of cells.

OC10 - In vitro antioxidant potential of ethanol extracts of different plants from Armenian flora

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This work aims to reveal some mechanisms of antioxidant action, using ethanol extracts of different plants (*Lamiaceae* family) from Armenian flora. *Thymus vulgaris* Lam, *Ajuga genevensis* L. and *Origanum vulgare* L. plant aerial part ethanol extracts were prepared. Scavenging free radical potential was studied using 1,1-diphenyl-2-picrylhydrazyl. Chelation power on ferrous (Fe^{2+}) ions was calculated, using $1 \text{ mg}\cdot\text{mL}^{-1}$ extract. Tyrosinase inhibitory activity was determined using mushroom tyrosinase. The antioxidant activity (AOA) was determined with thiobarbituric acid reactive species (TBARS) assay. The AOA was estimated also from its capacity to support aerobic growth of *Escherichia coli* QC 772 (GC 4468 sodA49) gene-engineered strain (derivative from K12) with fusion *sodA::lacZ*, received from D. Touati and kindly provided by prof. Oktyabrskii O.N. (Institute of Ecology and Genetics of Microorganisms, Perm, Russia) in the presence of H_2O_2 (4mM). Specific growth rate was monitored by measuring the optical density at 600 nm. Some mechanisms of extract antioxidant activity was revealed by the ability of *E. coli* GC 4468 sodA49 to utilize lactose, while growing in the lactose broth in the presence of H_2O_2 and extract. Antiradical activity of ethanol extracts of *T. vulgaris*, *A. genevensis*, *O. vulgare* and catechin (positive control), expressed with half-maximal inhibitory concentrations were $32.1 \pm 1.9 \text{ }\mu\text{g}\cdot\text{mL}^{-1}$; $175.1 \pm 5.1 \text{ }\mu\text{g}\cdot\text{mL}^{-1}$; $18.2 \text{ }\mu\text{g}\cdot\text{mL}^{-1}$ and $13.08 \pm 0.9 \text{ }\mu\text{g}\cdot\text{mL}^{-1}$, respectively. The further investigations were carried out with oregano extract. The chelating activity of extract was $74.5 \pm 0.2 \%$, the tyrosinase inhibitory activity was $6.5 \pm 0.2 \%$. Percentage antioxidant index (TBARS-assay) in case of using extract ($1 \text{ mg}\cdot\text{mL}^{-1}$) was $77.3 \pm 1.5 \%$. Treatment of bacteria with oregano extract without the oxidant had little effect on their growth. Pretreatment of the bacteria with extract before adding H_2O_2 (20 min) increased (approx. two fold) the resistance of *E. coli* to oxidant action. These bacteria did not lose the ability to use lactose. The results obtained indicate that oregano ethanol extract supports the growth of *E. coli* QC 772 bacteria by the activation of *sodA* gene as well as due to high antiradical activity.

Keywords: antioxidant activity, plant extract, *sodA::lacZ* gen fusion.

OC11 - Protective effect of *Centaurium erythraea* methanol extract against oxidative challenge in red blood cells of diabetic rats

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In light of increasing prevalence of diabetes over the past few decades and increasing lifespan of human population, there is a need to better monitor the quality of life of diabetic patients. Epidemiological evidence shows protective effect of regular consumption of diets rich in plant polyphenols against development of diabetes. *Centaurium erythraea* Rafn (CE) is traditionally used in Serbia for diabetes treatment. Previous phytochemical studies with aerial parts of CE showed it can substantially alleviate oxidative stress, one of the major pathogenic factors that lead to diabetes and its complications. Chronic hyperglycemia, the hallmark of diabetes, leads to increased production of free radicals which affect red blood cells (RBCs) structure and function. Considering RBCs role as oxygen transporters and consequences of impaired oxygen delivery in diabetes, the main goal of this research was to evaluate the protective effect of the methanol extract of aerial parts of CE against oxidative challenge in RBCs of rats with streptozotocin (STZ)-induced diabetes. The CE extract (100 mg/kg) was given daily and orally two weeks before, during diabetes induction (i.p. injection of STZ (40 mg/kg) for five consecutive days), and for four weeks after the STZ injections (animals designated as pre-treated group), or for four weeks after diabetes induction (post-treated group). Daily application of CE extract to STZ-induced diabetic rats improved the redox status of RBCs, observed as reduced lipid peroxidation and alleviated oxidative damage due to improved glutathione system and antioxidant enzyme activity, such as catalase (CAT), superoxide dismutase (SOD) and glutathione reductase (GR). Ameliorated RBCs' redox status was accompanied with improvement of major biochemical indicators of diabetes. CE extract increased serum insulin level, reduced blood glucose and glycated hemoglobin concentrations and improved lipid profile of diabetic rats. Furthermore, the CE extract reduced non-enzymatic glycation and enzymatic glycosylation and improved parameters which correlate with RBC aggregation and deformability. The protective effect of CE extract was more pronounced in pre-treated diabetic group. According to these results, *Centaurium erythraea* methanol extract has a great potential for use as dietary supplement in diabetes management. Since plenty of bioactive compounds, including polyphenols were determined in CE extract, identification of active metabolites from CE and their tissue distribution after intake could provide a useful source of potential novel antidiabetic pharmaceutical entities.

Keywords: *Centaurium erythraea*, diabetes, oxidative stress, red blood cells, antioxidant

OC12 - Dietary antioxidants in green leafy vegetables

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Green leafy vegetables are excellent dietary sources of different micronutrients and phytochemicals that have certain antioxidant properties (1). The aim of this study was to estimate the profile of antioxidant ingredients in the samples of cultivated green leafy vegetables such as chicory (*Cichorium intybus* L.), lettuce (*Lactuca sativa*), spinach (*Spinacia oleracea*) and Swiss chard (*Beta vulgaris* L. var. cicla L.). In this regard the amount of carotenoids, total polyphenols, flavonoids and some phenolic acids, as well as the content of α , γ , δ – tocopherols was determined in lyophilised vegetable samples. In addition, the antioxidant activity of green leafy methanol extracts was determined using three different antioxidant tests (DPPH, FRAP, ABTS) (2). All results were calculated on 100 g fresh weight basis. The obtained results indicated that spinach was the best source of α -carotene, while the highest concentration of β -carotene was determined in chicory samples ($p < 0.05$). Total polyphenol and flavonoid content of chicory was significantly higher in comparison with other vegetables. Other polyphenol compounds like chlorogenic and caffeic acids were detected only in chicory and lettuce samples. Tocopherols' content varied the most among all antioxidants with α – isomer as the most prominent one. Significant correlation was noted between polyphenol, flavonoid and γ -tocopherol and all performed antioxidant tests ($p < 0.05$). Amount of α -carotene was in good correlation with results obtained in DPPH and FRAP tests, while β -carotene and α -tocopherol concentrations had good correlations with ABTS test (0.891, 0.906, 0.966, 0.788, respectively). From mentioned results it can be concluded that various biologically active compounds of green leafy vegetables are potent contributors to different levels and mechanisms of antioxidant protection. Considering an increasing number of studies suggesting that consumption of vegetables can reduce the risk of cancer and cardiovascular diseases (3,4), our study confirmed that green leafy vegetables could be very good way to protect and improve human health.

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Keywords: green leafy vegetables, polyphenols, tocopherols, flavonoid, antioxidant activity

OC13 - The relationship between standard reduction potential and thermodynamic constants of antioxidant compounds – creation of Antioxidant Power Series

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The Antioxidant Power Series (APS) is proposed as a way to organize antioxidants important for human health based on determinations of their reduction potentials, in relation to glutathione – the main physiological antioxidant.

In our study, the values of reduction potentials of antioxidants were determined by potentiometric titration. All measurements were carried out using an Ag/AgCl reference electrode and a Pt working electrode at 295.15 K, 298.15 K, 310.15 K, 314.15 K, 318.15 K in phosphate buffered saline (pH=7.4). On the basis of titration curves received, the inflection points were read by the non-linear regression method using the proposed sigmoidal, 5-parameter model (with determination coefficient r^2 almost equal to 0.999). The results obtained allowed to calculate the values of standard reduction potentials (E^0) and thermodynamic data for oxidation reactions: standard Gibbs free energy (ΔG^0), enthalpy (ΔH^0) and entropy (ΔS^0).

Investigated antioxidants could be divided into two groups, based on the obtained thermodynamic data: straight line and parabola-shaped of dependence $E^0=f(T)$ with minimum for the temperature near 310.15 K (37°C). Parabolic shape means that the reaction of oxidation at minimum point is the least spontaneous (the highest ΔG^0), which may have great importance in biological processes.

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Keywords: potentiometric titration, reduction potential, antioxidant, thermodynamic constants.

OC14 - Antioxidant activity of whey-based beverages: Effect of shelf life and gastrointestinal transit on bioactivity

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Consumer demand for foods with benefits beyond nutrition continues to grow and has led to significant efforts in food formulation to design food products with improved efficacy. For example, food products with dietary antioxidants can reduce oxidative stress which plays a significant role in cellular damage and ageing. Dairy proteins such as whey can be added to beverages at relatively high concentrations and evidence suggest they exhibit some antioxidant activity (Brandelli et al., 2015). The objectives of the present study were to formulate whey-based beverages and powder products for the ageing consumer and investigate (1) the antioxidant shelf life of these products and (2) the antioxidant activity after gastrointestinal transit. Pilot plant scale processing was used to prepare model whey beverages with a combination of whey protein, and the well-established antioxidants, plant polyphenols and astaxanthin. Formulated beverages were submitted to standard pasteurization, ultra-high temperature or spray drying processing technologies. Obtained products were analysed by means of rheological characterization, particle size distribution, colorimetric characterization and instability index. The shelf life study was conducted over a 24 week period, collecting samples every week during the first four weeks and once every four weeks after that. In vitro gastrointestinal digestion of test samples were performed using the standardised COST INFOGEST method (Minekus et al., 2014). Antioxidant activity of samples was determined using ORAC and ABTS. Results demonstrated that combination of ingredients and ultra-high temperature processing boosted bioactivity of formulated products. Antioxidant shelf life study provided evidence that beverages and powder products functionality was preserved using standard storage conditions. Moreover gastrointestinal transit increased antioxidant activity of tested products.

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Keywords: Antioxidant, whey, beverage, shelf life, digestion



OC15 - IMPACT Biomolecules

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The Impact Biomolecules project of Lorraine University of Excellence (LUE) financed by the great loan and the industry for 5 M€, develops a federative and finalized research in close collaboration between 16 research laboratories and industries. The final goal of this opened consortium is to mobilize expertises to become a major actor of the bio-economic development. This project aimed at the development of a BioEngineering Valley in the Région Grand-Est, which will produce new functionalized and targeted biomolecules adapted to the food, pharmaceutical and medical markets.

The project is focused on major biological activities: anti-inflammatory, anti-proliferative and antioxidant. This choice was motivated by: i) the positioning of the private partners associated to the project, and ii) the internationally recognized skills from all the laboratories included in the consortium. This private/public consortium allows for the validation of the biological applications of proposed molecules from the development to the preclinical evaluation and premarketing (food, agrochemicals).

Scientific issues for the Biomolecules Impact project are the study of the interaction between living organisms (microorganisms and plant), which synthesize those new compounds, the conception of bio-inspired molecules, the improvement of their bioactivity by chemical functionalization or encapsulation/targeting and the validation of their biological activity to introduce these bio-inspired molecules in industrial or medical products.

The main strength of this project is the complementary of the skills proposed by each laboratory or industry, which will cover all the steps of biomolecules development for their marketing, from the initial discovery to the validation of their action mechanism.

The general goals of the Impact Biomolecules project are to:

- Intensify public/private research collaborations in the bioengineering field at the local, regional and international level. The research activity starting from the identification and the production of new functionalized and targeted biomolecules until their integration in complex matrices and the characterisation of their biological activities.
- Develop interdisciplinary projects between biologists, biotechnologists, chemists and physicians to arise new research areas
- Generate marketable products and technologies in close collaboration with private partner.
- Develop a new master program in bioengineering and bioeconomy to educate future leaders in this field, by joining this formation on to advanced research and public/private partner skills.

Keywords: Biomolecules, antioxidant, anti-inflammation, private/public partners.



OC16 - Deciphering modes of action: from chemogenetic phenotype profiling to intricate intracellular diagnostics

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Redox-modulating substances derived from natural as well as synthetic resources, such as redox active secondary metabolites and organo sulfur and selenium compounds, have attracted the attention of scientist in recent years. These substances have potential applications in the field of chemoprevention, drug and phytoprotectant development. Biological assays based on Yeast offer an interesting and robust platform to explore the mechanism of action of compounds and also to probe the “redox link” which may exist between its activity and chemistry. Chemogenetic profiling based on yeast could be employed to explore possible intracellular mechanisms which may also be confirmed using intracellular diagnostics. These methods, however, provide only a first glimpse on the probable mode of action novel substances and set a direction for more complicated and in depth future investigations.



OC17 - Role of lipid overload on the fluidity of plasma and organelle membranes and subcellular distribution of lipids in cells

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Metabolic homeostasis of fatty acids is complex and well-regulated in all organisms. The biosynthesis of saturated fatty acids (SFA) in mammals provides substrates for β -oxidation and ATP production. Monounsaturated fatty acids (MUFA) are products of desaturases that introduce a methylene group in cis geometry in SFA. Polyunsaturated fatty acids (n-6 and n-3 PUFA) are products of elongation and desaturation of the essential linoleic acid and α -linolenic acid, respectively. The liver processes dietary fatty acids and exports them in lipoproteins for distribution and storage in peripheral tissues. The three classes of fatty acids are integrated in membrane phospholipids and determine their biophysical properties and functions. This study was aimed at investigating effects of fatty acid remodeling on membrane biophysical properties under varying nutritional and pathological conditions, by integrating lipidomic analysis of membrane phospholipids with functional two-photon microscopy (fTPM) of Laurdan staining of cellular membranes. In addition, Phasor analysis of Nile red imaging of the cells allows for determination of the distribution and subcellular localization of highly polar (e.g., free fatty acids), polar (e.g., phospholipids and sphingolipids) and non-polar (e.g., triglycerides and cholesterol esters) lipids. Our findings in INS-1E pancreatic beta cells exposed to high levels of glucose and palmitic acid indicate that these methodologies provide powerful means to investigate cellular and subcellular lipid structures in a variety of pathologies associated with the metabolic syndrome.

Keywords: Fatty acids, Lipidomics, Membrane fluidity, Phasor analysis, Phospholipids, Triglycerides.

OC18 - Site-specific effects of apolipoprotein E expression on white adipose tissue mitochondrial substrate oxidation for energy production

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Apolipoprotein E (APOE) has been strongly implicated in the development of diet induced obesity. In the present study, we investigated the contribution of brain and peripherally expressed human apolipoprotein E3 (APOE3), the most common human isoform, to diet induced obesity.

In our studies APOE3 knock-in ($ApoE3^{knock-in}$), $ApoE$ -deficient ($apoE^{-/-}$) and brain-specific expressing APOE3 ($ApoE^{3brain}$) mice were fed western-type diet for 12 week and biochemical analyses were performed. Moreover, AAV-mediated gene transfer of APOE3 to $apoE^{-/-}$ mice was employed, as a means to achieve APOE3 expression selectively in periphery, since peripherally expressed APOE does not cross barrier (BBB) or blood-cerebrospinal fluid barrier (BCSFB).

Our data suggest a bimodal role of APOE3 in visceral white adipose tissue (WAT) mitochondrial metabolic activation that is highly dependent on its site of expression and independent of postprandial dietary lipid deposition.

Our findings indicate that brain APOE3 expression is associated with a potent inhibition of visceral WAT mitochondrial oxidative phosphorylation, leading to significantly reduced substrate oxidation, increased fat accumulation and obesity. In contrast, peripherally expressed APOE3 is associated with a notable shift of substrate oxidation towards non-shivering thermogenesis in visceral WAT mitochondria, leading to resistance to obesity.

Keywords: Apolipoprotein e; Obesity; White Adipose tissue; Brown adipose tissue; Mitochondrial metabolic activity; Substrate oxidation; Energy production.

OC19 - Qualitative assessment of oxidized phospholipids generated as a result of enzymatic oxidation and their digestion by phospholipase A₂

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The results of numerous epidemiological studies indicate that the type and quality of food-delivered lipid compounds contribute to the prevention or promotion of diet related and metabolic diseases such as: type 2 diabetes, obesity, atherosclerosis, hypertension or coronary artery disease. Among the food-delivered lipids, phospholipids (PLs) attract increasing attention due to their high nutritional value and functional properties. PLs, especially those containing essential fatty acids (FAs) exhibit

a number of key biological activities. On the other hand, polyunsaturated FAs incorporated in the structure of natural occurring PLs are particularly susceptible to oxidation. Food-delivered oxidized phospholipids (OxPLs) can be generated during manufacturing, storage and final preparation of food, including frying, baking or grilling, as a result of non-enzymatic (autooxidation and photooxidation) and enzymatic mechanisms.

OxPLs delivered with food as well as products of their digestion can be potentially toxic molecules for to the epithelial cells of digestive tract, which are directly exposed on the chyme containing lipid emulsion. Accumulation of OxPLs can lead to gut pathologies, as a result of cell membrane modifications, as well as DNA and protein damage. In addition, previous research suggested that lipid hydroperoxides, which were not hydrolyzed during intestinal digestion, may be released to the blood stream after absorption by enterocytes and thereby contribute to the pathogenesis of atherosclerosis.

However, there are several mechanisms aimed at reducing the negative effect of OxPLs on the gastrointestinal tract cells. The key enzyme involved in these mechanisms is phospholipase A₂, which is characterized by a higher activity for OxPLs compared to their native counterparts.

In the our work, we characterised products of enzymatic oxidation of PLs isolated from hen egg yolk using offline two-dimensional liquid chromatography coupled with DAD, CAD and MS detection. The next step of our research was to determine the pancreatic phospholipase A₂ activity for enzymatically oxidized PLs isolated from hen egg yolk in comparison with non-oxidized PL fraction.

Keywords: dietary lipids; hen egg yolk; oxidized phospholipids; enzymatic oxidation; lipid digestion; pancreatic phospholipase A₂; offline 2D (HILIC-RP)-LC.



OC20 - GUT microbiota and probiotics: In vitro and in vivo hydrophobicity and adhesion of lactic acid bacteria

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Lactic acid bacteria (LAB) play an important role in the gastrointestinal tract (GUT) of human and animals. Some of them got the GRAS status of probiotic organisms. For being called the probiotic, they should survive in the intestine, be genetically stable, have aggregation specificity and antagonistic activity. They must not be toxic and pathogenic or change their properties in GUT stress conditions. The adhesion on the surface of epithelial cells is among the main criteria for the selection of probiotic strains. This is important because in adhesive status bacterial cells can have positive effects on human health by extruding pathogens and immunomodulating the host organism for a long period of time.

Approximately 35 strains of lactobacilli, enterococci and lactococci, isolated from different dairy products, fermented vegetables and GUT of honeybees, were screened according to their possibility to show high hydrophobicity. The strains *Lactobacillus helveticus* INRA-2010-H11, *Lactobacillus* spp. WT-10.2 and *L. acidophilus* JM-2012 were determined to have high hydrophobicity in toluene (approximately 66%, 83%, 89,5% correspondingly), xylene (approximately 70%, 88%, 90% correspondingly) and hexane systems (approximately 50%, 78%, 92.2% correspondingly). The adhesion degree of INRA-2010-H11 and *L. acidophilus* JM-2012 on the surface of epithelial cells of cow small intestine was 35% and 19.3%, large intestine was 18% and 50% and caecum was 48.67% and 63% respectively. CFU of adhered cells for these investigated strains was approximately 9×10^9 compared to the initial quantity of LAB cells 10^{10} CFU. In vivo experiments of LAB adhesion in GUT of rats revealed the presence of 2×10^8 , 2×10^8 and 8.3×10^7 CFU of LAB in control, fed by INRA-2010-H11 and mixture groups of rats, respectively. The reduction of microbiota biodiversity in rats' GUT was detected in case of feeding by mixture of LAB compared with control group. INRA-2010-H11 demonstrated high aggregation and adhesion activity, thus, it has the potential as a good probiotic strain. The amount of immunoglobulins was measured during the each week of feeding. The amount of IgA did not change at all during the feeding, while the amount of IgM decreased in all groups, including the control. Interestingly, the amount of IgG increased crucially on the 3rd week of feeding and decreased again on the last week compared to the control.

All these results suggested that there were typical probiotic effects on the organism; therefore, the investigated strains are important for the creation of functional food and correction of human GUT microbiota.

OC21 - Preventive strategies to counterbalance food pesticide effect on epigenome and gut microbiota

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Food pesticide residues have been identified in food as well as in people's urine [1]. Studies on early life exposure to the food pesticide permethrin demonstrate its ability to modulate gene expression, epigenome and gut microbiota leading to long term effects later in life [2-4]. In particular, permethrin pesticide, binding to sodium channels, induces neuron depolarization which modulate DNMTs activities responsible for DNA methylation [5].

Previously, we demonstrated changes in the DNMTs in striatum of animals exposed to permethrin during brain development [2]. Besides, we observed that permethrin is able to decrease global DNA methylation and dopamine level in mothers exposed to food pesticide during early life as well as in their untreated offspring, underlining the intergenerational effect of the food pesticide [6]. Of particular interest is the evidence that permethrin exposure can also promote significantly changes in gut microbiota.

With the aim to propose strategies for prevention, bioactive supplements have been studied and positive nutrigenomic effects able to counterbalance the brain and gut microbiota alterations will be discussed.

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Keywords: nutrigenomics, food pesticide, gene expression, epigenetic modulation, gut microbiota.

POSTERS



PO1 - Effects of Revamil® honey on autophagy in macrophages

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For thousands of years, honey has been recognized for its beneficial properties in traditional medicine, and has been commonly used as a first remedy to heal skin wounds and diseases of the gut. Many studies reported the antibacterial properties of honey, besides its anti-inflammatory and immunomodulatory effects, especially on macrophages. However, nothing is known in the literature about the putative effects of honey on subcellular structures and organelles such as mitochondria and lysosomes. Our project intends to test the modulatory effects of Revamil® (a medical-grade honey) on the abundance of lysosomes and mitochondria, on the functions of these organelles, and on the signaling pathways regulating their population in macrophages. As this project opens a new research topic in our labs, we first needed to optimize experimental conditions and characterize the models that will be used. We showed that a concentration of 8 mg/ml of Revamil® does not modify the pH and osmolarity of culture medium, and does not impair cellular viability and proliferation. In order to study the effects of honey and to discriminate the contribution of the sugars present in the Revamil® from the effect of other specific compounds, we used a control syrup made up of a similar sugar composition than the one found in Revamil®.

In this work, we particularly showed that Revamil® honey triggers a transient activation of the mTOR signaling, known to regulate many metabolic functions, and inhibit lysosomal biogenesis and autophagy. Moreover, we highlight that Revamil® is responsible for an inhibition of the autophagic flux through an increase in the abundance of acidic compartments as well as protein markers of the autophagic flux such as LC3-II and p62. Further studies will be needed to determine how Revamil® honey inhibits autophagic responses in macrophages, and which components of this honey are potentially involved in these mechanisms.

Keywords: Revamil® honey, lysosome, mitochondria, autophagy, mTOR.

PO2 - Association of trace elements with cardiovascular risk factors in an adult Serbian population: a pilot study

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In recent years there has been an increased investigation of the potential link between trace elements status and cardio-metabolic risk factors in the general populations, however with inconsistent results.

A total of 60 apparently healthy participants of both sexes, aged 20 to 65 years, were enrolled in the study. Anthropometric measurements, blood pressure and biochemical analysis of fasting blood were performed. The plasma selenium (Se), copper (Cu) and zinc (Zn) determination were measured by inductively coupled plasma–mass spectrometry.

The mean selenium, copper and zinc concentration was $81.7 \pm 15.7 \mu\text{g/L}$, $0.77 \pm 0.32 \text{ mg/L}$ and $0.98 \pm 0.24 \text{ mg/L}$ respectively. A higher plasma Se and Zn concentrations were found in older subjects (age range, 40 to 65 years) than in younger subjects (age range, 20 to 39 years). No associations were found among trace elements concentrations and gender, systolic and diastolic blood pressure, fasting glucose, smoking, and body mass index. Concerning lipid profile, plasma Zn correlated positively with low-density lipoprotein cholesterol ($r=0.270$, $p<0.05$), and the triglycerides/high-density lipoprotein ratio ($r=0.278$, $p<0.05$), while that of Cu correlated with triglycerides ($r=0.310$, $p<0.05$). The plasma Se concentration showed a positive correlation with total cholesterol ($r=0.324$, $p<0.05$) and low-density lipoprotein cholesterol ($r=0.320$, $p<0.05$), even after adjustment for age. A significant positive correlation was found between selenium and zinc concentrations ($r=0.481$, $p<0.001$).

This study demonstrates the association between Se, Cu, and Zn with lipid parameters. However, prospective studies including a large number of subjects are needed to confirm these preliminary results.

Keywords: selenium, zinc, copper, cardiovascular risk, dyslipidaemia.



PO3 – Inflammatory markers and Mediterranean diet

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The aim was to assess inflammatory markers among adults and adolescents in relation to the adherence to the Mediterranean diet. A random sample (219 males and 379 females) of the Balearic Islands population (12-65 years) was anthropometrically measured and provided a blood sample to determine biomarkers of inflammation. Dietary habits were assessed and the adherence to the Mediterranean dietary pattern calculated. The prevalence of metabolic syndrome increased with age in both sexes. The adherence to the Mediterranean diet in adolescent males was 51.3% and 45.7% in adults, whereas in females 53.1% and 44.3%, respectively. In males, higher adherence to the Mediterranean diet was associated with higher levels of adiponectin and lower levels of leptin, tumor necrosis factor alpha (TNF- α), plasminogen activator inhibitor 1 (PAI-1) and high-sensitivity C-reactive protein (hs-CRP) in adults, but not in young subjects. In females, higher adherence was associated with lower levels of leptin in the young group, PAI-1 in adults and hs-CRP in both groups. With increasing age in both sexes, metabolic syndrome increases, but the adherence to the Mediterranean diet decreases. Low adherence to the Mediterranean dietary pattern (MDP) is directly associated with a worse profile of plasmatic inflammation markers.

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PO4 - Western and Mediterranean dietary patterns are related to physical activity and fitness among Spanish older adults.

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The aim was to assess prevailing food patterns, and its association with physical activity and fitness among Spanish older adults. Cross-sectional study in Spain, collecting data from a sample ($n = 380$; 54% female) aged 55-80 years (men) and 60-80 years (women) with no previously documented cardiovascular disease. Body weight, body fat and waist circumference were assessed. Physical activity performed was measured using the Minnesota Leisure-time Physical Activity Questionnaire (LTPA). Physical fitness was assessed using a validated physical fitness test battery. Food consumption was assessed by a validated semi-quantitative food-frequency questionnaire. Factor analysis identified two major dietary food patterns: "Western" (WDP) and "Mediterranean" (MDP) dietary patterns. Participants in MDP's fourth quartile were classified in the second (men) and third (men and women) tertile of LTPA. After adjusting for age, body fat, waist-to-height ratio, and METs, in both sexes, a negative significant association was found between 30-s Chair stand and 6-min walking test, a positive significant association was found between 30-m Gait speed and 8-foot Time Up-and-Go (except in men) tests with WDP. The 30-m Gait speed test was negatively associated with MDP in men. MDP is associated with more time spent on LTPA, and this association was independent of body composition and a fast gait speed in men. WDP is associated with slower gait speed and lower body strength, agility and aerobic endurance. MDP has protective effect on healthy physical fitness, and WDP may be a contributor to frailty.

This study was supported by Instituto de Salud Carlos III (PI11/01791, PI14/00636, PI17/01827, RETIC RD06/0045/1004, & CIBEROBN CB12/03/30038), EU-COST Action CA16112, Grant of support to research groups no. 35/2011 (Balearic Islands Gov.) and EU-FEDER funds.



PO5 – Hesperidin from genus Citrus has anti-inflammatory effects

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The benefits of the Mediterranean diet for protecting against many diseases are usually attributed to high consumption of certain foods, characterized by the presence of bioactive substances such as polyphenols. Citrus fruits, which are cultivated and consumed worldwide, are typical products of the Mediterranean diet. Inflammation plays an important role in the pathogenesis of numerous diseases such as arthritis, allergies and neurodegenerative disorders, among others. Dietary polyphenols constitute a large family of bioactive substances with potential beneficial effects against a broad group of diseases. Citrus fruits and juices are a rich source of vitamin C and flavonoids, with a potential effect on the inflammatory response. Hesperidin is a flavonoid present in high concentration in citrus species and has numerous biological properties, principally antioxidant and anti-inflammatory. Several studies have been performed in order to evaluate the effects of hesperidin as anti-inflammatory agent using cellular and animal models and few clinical trials. In the present review, available literature about the anti-inflammatory effects of hesperidin is reported and discussed. Moreover, we also discuss the chemistry, bioavailability and proposed mechanisms of action of hesperidin.

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PO6 – Regular physical activity are related to trace element contents in older adults' toenails.

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The aim was to assess the trace element contents in toenails of older adults and its association with regular physical activity. Cross-sectional multicentre study in Spain, collecting data from a random sample of 380 participants (54% female) aged 55-80 years (men) and 60-80 years (women) with no previously documented cardiovascular disease. Physical activity performed was measured using the Minnesota Leisure-time Physical Activity Questionnaire. The 25 most inactive and 25 most active individuals for each sex were selected for this study (final sample n = 100). Anthropometric measurements were performed and toenail samples collected for calcium (Ca), chromium (Cr), iron (Fe), cobalt (Co), nickel (Ni), zinc (Zn), selenium (Se) and mercury (Hg) analysis. Significant differences between sexes were reported in Ca concentrations, women having lower concentrations than men. No differences were reported in trace element contents between active and inactive men. Active women showed higher Ca, Cr, Fe, Co, and Zn and lower Hg contents than their inactive peers (all $p < 0.05$). Inactive women showed lower Ca and Co levels (735.0 mg/kg and 4.5 $\mu\text{g}/\text{kg}$, respectively) than inactive men (1170.0 mg/kg and 7.9 $\mu\text{g}/\text{kg}$, respectively). Active women had lower Ca and higher levels of Cr (936.0 mg/kg and 1230.0 $\mu\text{g}/\text{kg}$, respectively) than active men (1070.0 mg/kg and 522.0 $\mu\text{g}/\text{kg}$, respectively). The present data added new information on the element contents in toenails of healthy Spanish older adults. The concentration of trace elements was similar in both sexes except for Ca which were lower in women. The trace element contents in women's toenails, but not in men, were markedly influenced by physical activity, with higher levels of Ca and Fe and lower Hg among active females.

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