OP ERDF Galicia 2014-2020

"A way to make Europe"

4th ANNUAL MEETING

BOOK OF ABSTRACTS

CINBIO

galicia

CINBIO Universidade de Vigo

Xacobeo 2021

UNIÓN EUROPEA

XUNTA DE GALICIA

Universidade Vigo

Congreso cofinanciado grazas á convocatoria de Axudas propias á investigación da universidade de Vigo para o ano 2020

galicia
CINBIO Universidade Vigo
Xacobeo 2021
UNIÓN EUROPEA
XUNTA DE GALICIA

Prologue

It is our pleasure to introduce the proceedings of the 4th CINBIO ANNUAL MEETING in Vigo, Spain. Our major goal is to bring academic scientists, students and industry researchers together to promote, encourage and recognized excellence in scientific research.

This annual meeting started in 2017 is fully promoted by the Postdoctoral Researchers of CINBIO providing a forum for interdisciplinary discussions with the aim of better exchanging and sharing experiences and research results about most aspects of science and social research, and discuss the practical challenges encountered and the solutions adopted.

Participants from 15 countries made this meeting truly international in scope. Of the total number of presented communications, 17 oral communications and 48 posters of these are included in this proceedings volume. These communications formed the heart of the conference and provided ample opportunity for discussion.

The program consisted of invited sessions, and technical workshops and discussions with eminent speakers covering a wide range of topics in science. This rich program provides all attendees with the opportunities to meet and interact with one another.

We hope your experience with 4th CINBIO Annual Meeting was and will be fruitful and a long lasting one. With your support and participation, the conference will continue its success for a long time.

We take also the opportunity to express our gratitude to the organization staff, the members of the scientific committee, speakers and poster´s participants.

We look forward to seeing all of you next year at the conference.

Miguel Ángel Correa Duarte
Director of CINBIO

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JULY 1ST

4th ANNUAL MEETING

1-2 July 2021
Online

Mónica Gómez Malmierca
Dpto. Biología Funcional,
Área Microbiología
Universidad de Oviedo
Asturias, Spain

Marina Romaní Pérez
Instituto de Agroquímica y Tecnología
de Alimentos (IATA-CSIC)
Valencia, Spain

Fernando Reyes
Fundación MEDINA
Granada, Spain

Belén Rodríguez Sánchez
Servicio de Microbiología Clínica
y Enfermedades Infecciosas
Hospital Gregorio Marañón
Madrid, Spain

09:00h OPENING

09:15h Discovery and combinatorial biosynthesis of sipanmycins: a new family of glycosylated macrolactams
Mónica Gómez Malmierca
Invited Speaker

09:45h The microbiota and the gut-brain axis in the control of energy homeostasis
Marina Romaní Pérez
Invited Speaker

10:15h Comparison of methods for the isolation and characterization of exosomes in plasma of Relapsing Remitting Multiple Sclerosis patients: interference of soluble protein, lipoproteins and another extracellular vesicles
Araceli Piñeiro Abuín, CINBIO (UVigo)
Oral

10:30h MUC16 is overexpressed in idiopathic pulmonary fibrosis and induces fibrotic responses mediated by transforming growth factor-β1 canonical pathway
Beatriz Ballester Llobell, Helmholtz Zentrum München
Oral

10:45h Functional and phenotypic characterization of human TBX4 sequence variation: gain of function mutations result in late-onset lung disease
Mauro Lago Docampo, CINBIO (UVigo)
Oral

11:00h POSTER SESSION

12:30h Structural elucidation of complex natural products assisted by biosynthetic gene cluster analysis
Fernando Reyes
Invited Speaker

13:00h MALDI-TOF Mass Spectrometry and MachineLearning. Applications in Clinical Microbiology
Belén Rodríguez Sánchez
Invited Speaker

13:30h Highly pleiotropic variants of human traits and diseases associated with strong background selection
Irene Novo, UVigo
Oral

13:45h Algae as a source of nutraceutics and functional foods
Franklin Chamorro, Nutrition and Bromatology Group (UVigo)
Oral

JULY 1ST

4th ANNUAL MEETING

1-2 July 2021
Online

Jeanine Houwing-Duistermaat
Department of Statistics, University of Leeds, UK
Department of Statistics, University of Bologna, Italy
Department of Biostatistics and Data Science,
University Medical Centre Utrecht, The Netherlands

Rebecca Betensky
Department of Biostatistics
School of Global Public Health
New York University

14:00h LUNCH TIME

15:30h Probabilistic partial least squares methods for data integration
Jeanine Houwing-Duistermaat
Invited Speaker

16:00h Reconsideration of the Kaplan-Meier Estimator: Censoring and Time-varying Covariates
Rebecca Betensky
Invited Speaker

16:30h Parametric Landmark estimation of the transition probabilities in survival data with multiple events
Gustavo Soutinho, ISPUP - Instituto de Saúde Pública da Universidade do Porto
Oral

16:45h Where is the mice? - Exploring the R package
Susana Rafaela Martins, Instituto Politécnico de Viana do Castelo
Oral

17:00h DTDA, the renewed R package to analyse doubly truncated data
Carla Moreira, Centre of Mathematics - University of Minho
Oral

17:15h Re-analysis of miRNA-Seq differential expression analyses in neuropsychiatric diseases
Daniel Pérez-Rodríguez, NeuroEpigenetics Lab, Translational Neuroscience Group-CIBERSAM, Galicia Sur Health Research Institute (IIS Galicia Sur)
Oral

17:30h END OF DAY 1

JULY 2ND

4th ANNUAL MEETING

1-2 July 2021
Online

María del Puerto Morales
Instituto de Ciencia de Materiales de Madrid del Consejo Superior de Investigaciones Científicas (ICMM/CSIC)
Madrid, Spain

Andreas Tittl
Chair in Hybrid Nanosystems,
Nanoinstitute Munich,
Ludwig-Maximilians-Universität München,
Germany

09:00h Controlling local temperature at the magnetic nanoparticle surface for improving bio and catalytic efficiency
María del Puerto Morales
Invited Speaker

09:30h Controlled plasmonic coupling for advanced photocatalysis
Yoel Negrín, CINBIO (Uvigo)
Oral

09:45h Selective and Ultransensitive SERS Sensor based on Plasmonic Supercrystals
Daniel García Lojo, CINBIO (UVigo)
Oral

10:00h Scaling on transport properties of nanofluids
José I. Prado, CINBIO (UVigo)
Oral

10:15h ALEHOOP Project: Biorefineries for the valorisation of macroalgal residual biomass and legume processing by-products to obtain new protein value chains for high-value food and feed applications
Camila Jiménez González, UVigo
Oral

10:30h POSTER SESSION

12:00h Spectrally selective metasurfaces for molecular biospectroscopy and sensing
Andreas Tittl
Invited Speaker

12:30h Chiral plasmonic photocatalysts
Lucas Vázquez Besteiro, CINBIO (UVigo)
Oral

12:45h Electroactive polypyrrole/linseed oil nanocapsules for drug delivery applications
Gabriela de Alvarenga, Federal University of Paraná (UFPR) / CINBIO
Oral

13:00h Manufacture of Magnetic Spiky Structures
Ecem Tiryaki, CINBIO (UVigo)
Oral

13:15h Bacterial surface display for the expression and engineering of functional glycan-binding proteins
Alba Vázquez Arias, CINBIO (UVigo)
Oral

13:30h CLOSING SESSION

ORGANIZING COMMITTEE

NOELIA FLÓREZ FERNÁNDEZ

Biomass and Sustainable development (EQ-2)

SERGIO GÓMEZ GRAÑA

Functional NanoBioMaterials (FunNanoBio)

HUGO LÓPEZ FERNÁNDEZ

Phenotypic Evolution Gorup, Instituto de Investigaçao e Inovaçao em Saúde (I3S), Universidade do Porto, Portugal

FÁTIMA OTERO DIOS

Management Unit

ANA SOUSA CASTILLO

Chair in Hybrid Nanosystems, Nanoinstitute Munich, Faculty of Physics, Ludwig-Maximilians Universität München, Germany

MARÍA DOLORES TORRES PÉREZ

Biomass and Sustainable development (EQ-2)

SCIENTIFIC COMMITTEE

NOELIA FLÓREZ FERNÁNDEZ

Biomass and Sustainable development (EQ-2)

SERGIO GÓMEZ GRAÑA

Functional NanoBioMaterials (FunNanoBio)

HUGO LÓPEZ FERNÁNDEZ

Phenotypic Evolution Gorup, Instituto de Investigaçao e Inovaçao em Saúde (I3S), Universidade do Porto, Portugal

ANA SOUSA CASTILLO

Chair in Hybrid Nanosystems, Nanoinstitute Munich, Faculty of Physics, Ludwig-Maximilians Universität München, Germany

JOSE ANTONIO SOUTO SALGADO

Organic Chemistry (ORCHID)

MARÍA DOLORES TORRES PÉREZ

Biomass and Sustainable development (EQ-2)

LORENA VÁZQUEZ IGLESIAS

Functional NanoBioMaterials (FunNanoBio)

SARAH DE MARCHI LOURENÇO

Functional NanoBioMaterials (FunNanoBio)

1st July 2021

Invited Speakers

Discovery and combinatorial biosynthesis of sipanmycins: a new family of glycosylated macrolactams

Mónica Gómez Malmierca, PhD

Dpto. Biología Funcional, Área Microbiología, Universidad de Oviedo, Asturias, Spain

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The rapid emergence of infectious diseases and multi-resistant pathogens and the need for more effective chemotherapeutics has risen the necessity for new bioactive compounds, thus novel strategies have to be developed to find them. Actinomycetes isolated in symbiosis with insects have attracted attention in recent years as producers of metabolites with important bioactivities. Many of these bioactive natural products are glycosylated compounds in which the sugar components usually participate in interaction and molecular recognition of the cellular target. Therefore, the presence of sugar moieties is important, in some cases essential, for bioactivity. The great majority of these sugar moieties belong to the 6-deoxyhexoses and share two common biosynthetic steps catalyzed by a NDP-D-glucose synthase and a NDP-D-glucose-4,6-dehydratase 4,6DH.

Here we described the genome mining strategy to search for novel glycosylated compounds in a collection of Streptomyces strains isolated from the surface of leaf-cutting ants of the Tribe Attini. As a result, we discovered the sipanmycin family, a group of macrolactam disaccharyl glycosides that exerts antibiotic and cytotoxic activities. Also, we characterized the gene cluster responsible for their biosynthesis (sip cluster) in Streptomyces sp. CS149 and the steps involved in the glycosylation of the final compounds were unraveled.

Finally, we reported the generation of several sipanmycin analogues with different deoxysugars by introducing plasmids leading to the biosynthesis of various nucleotide-activated sugars. In addition, modifications in the macrolactam ring were introduced by mutasynthesis approaches, substituting the 3-aminobutanoic acid (derived from β-glutamic) acid naturally used as starter unit in the native sipanmycin biosynthesis pathway by 3-aminopentanoic acid.

The microbiota and the gut-brain axis in the control of the energy homeostasis

Marina Romaní Pérez

Instituto de Agroquímica y Tecnología de Alimentos (IATA-CSIC), Valencia, Spain

E-mail: marina.romani@iata.csic.es

Obesity currently represents a major societal and health challenge worldwide. Its prevalence has reached epidemic proportions and trends continue to rise, reflecting the need for more effective preventive measures. Also its comorbidities such as type 2 diabetes (T2D) and cardiovascular diseases greatly impair the quality of life and involve considerable socioeconomic pressures on the individual and global health economies.

In recent years, obesity research has made an important progress on the identification of molecular signalling pathways involve in the meal-related communication between the gastrointestinal tract and the brain. This evidence helps to advance in the design of anti-obesity strategies targeting the gut-to-brain nutrient signalling, whose malfunction contributes to obesity.

Gut microbiota–diet interactions might interfere in nutrient sensing and signalling from the gut to the brain, where the information is processed to control energy homeostasis. This gut microbiota–brain crosstalk is mediated by metabolites, mainly short chain fatty acids, secondary bile acids or amino acids-derived metabolites and subcellular bacterial components. Accordingly, strategies based on the modification of the gut microbiota composition to beneficially modulate the gut-brain communication are now conceived as potential approaches for combat obesity and its comorbidities.

Our research is focused on the identification of human intestinal bacteria capable of improving the metabolic health through the modulation of the immune, endocrine and neural intestinal signalling pathways mediating the gut-brain axis functionality. With the final aim of developing efficient next generation of probiotics (NGP) for metabolic diseases, here, we illustrate the process for designing microbiome-based strategies and we show some examples with potential clinical use.

Structural elucidation of complex natural products assisted by biosynthetic gene cluster analysis

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Natural products frequently possess complex structures, including multiple chiral centers, that make their structural elucidation a challenging task requiring the use of many different spectroscopic approaches and computational techniques. Different structural classes may need the use of different strategies and in general a combination of several spectroscopic and chiroptical techniques, chemical derivatization, degradation reactions, and molecular modelling are used to solve the full structure and absolute configuration of complex molecules.

The latest developments in next generation sequencing technologies have facilitated an enormous breakthrough in whole genome sequencing of microbial strains in the last decade, transforming it nowadays in a fast and affordable process. Bioinformatics tools have also been developed in parallel to mine the biosynthetic potential of the sequenced genomes and, among others, they allow to predict the absolute configuration of most chiral centers in a wide range of natural products encoded by the different biosynthetic gene clusters (BGCs). The combined use of those predictions with NMR analysis has become a very efficient and powerful approach to establish the full absolute configuration of complex natural products, especially in the case of polyketides, non-ribosomal peptides, and RiPPs. In these structural classes, the configuration of chiral centers predicted by BGC analysis is usually confirmed and complemented with that determined by NMR for other chiral centers that might be present in the molecule, originated from post-translational tailoring steps.

This combination of NMR and BGC analysis has been successfully applied in our laboratory over the last years and my talk will present a few recent cases where BGC analysis has been succesfylly used in combination with traditional structural elucidation approaches to solve the structure of complex actinomycete natural products, including polyketides and RIPPs.

MALDI-TOF Mass Spectrometry and Machine Learning.

Applications in Clinical Microbiology

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MALDI-TOF Mass Spectrometry has become an essential identification tool in the clinical microbiology laboratories over the last decades. It has replaced many conventional identification methods based on biochemical tests, that are no longer in use due to the use-friendliness, reliability and cost-effectiveness of MALDI-TOF. Most bacterial species can be easily identified from the colonies grown overnight on agar plates. Yeasts and fungal species are becoming easier to identify by MALDI-TOF thanks to the improvement of the available databases and the sample preparation procedures. Besides, direct identification of pathogens from clinical samples (blood cultures, urine or cerebrospinal fluid) has been demonstrated to be a clinically impacting application of MALDI-TOF since the identification of microorganisms can be achieved in an early manner, allowing the streamlining of optimal antibiotic treatment. The detection of resistant mechanisms (beta-lactamases, carbapenemases) almost simultaneously to the identification of the host microorganisms has been another important milestone of MALDI-TOF, showing that the information contained in the protein spectra can provide much more information than the mere name of the microorganism analysed.

Our MALDI-TOF-based research group has initiated collaborations with bioinformaticians and biomedicine engineers in order to classify MALDI-TOF protein spectra according to the presence of antibiotic/antifungal resistance mechanisms, the sequence type (ST) or the serotype/ribotype they belong to or their connection with hospital outbreaks. The algorithms applied so far have allowed the correct classification of azole-resistant Aspergillus fumigatus, vancomycin-resistant Enteroccocus faecium or high-risk clones of Pseudomonas aeruginosa. Some of our studies are still in a preliminary stage, but the robustness of the results shows that MALDI-TOF is a promising tool for rapid screening of clinically important strains.

Probabilistic partial least squares methods for data integration

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Many studies collect multiple omics datasets to gather novel insights about different stages of biological processes. For joint modelling of these datasets, several data integration methods have been developed. These methods address high dimensionality, within and across datasets correlation, and the presence of heterogeneity among datasets due to representing different biological processes and using different measurement technologies. Most methods, neither provide statistical evidence for a relationship between the datasets nor identify relevant variables that contribute to this relationship.

We propose a probabilistic latent variable modelling framework for inferring the relationship between two omics datasets. These methods reduce dimensionality and capture correlations by forming components that are linear combinations of the variables. The correlation structure is modelled by joint and data specific components. We propose maximum likelihood estimation of the parameters and formulate a test statistic for the null hypothesis of no relationship.

We evaluate our methods via simulations. Under the null hypothesis, the test statistic appears to approximately follow the normal distribution for sample size larger than 500. Our method outperforms existing methods for small and heterogeneous datasets in terms of selecting relevant variables and prediction accuracy. We illustrate the methods by analysing omics datasets from various studies.

Reconsideration of the Kaplan-Meier Estimator: Censoring and Time-varying Covariates

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In this talk I will present two extensions of the Kaplan-Meier estimator that address a nuance of censoring and incorporation of time-varying covariates.

Clinical studies with time to event endpoints typically report the median follow-up (i.e., censoring) time for the subjects in the trial, alongside the median time to event.

The reason for this is to provide information about the opportunity for subjects in the study to experience the event of interest. In most clinical studies, the censoring time is a composite measure, defined as the minimum of the time to drop-out from the study and time to the administrative end of study. The time to drop-out component may or may not be observed, while the time to the end of study is observed for each subject. I consider whether this decomposition of the censoring time into a time that is itself potentially censored and a time that is fully observed offers any improvement of the estimation of the censoring distribution.

Extensions of the Kaplan-Meier estimator have been developed to illustrate the relationship between a time-varying covariate of interest and survival, however they are limited to displaying survival for patients who always have a certain value of a time-varying covariate. I present extensions of these estimators that provide crude and covariate-adjusted estimates of the survival function for patients defined by covariate paths.

Oral communications

Oral communication (O1)

Comparison of methods for the isolation and characterization of exosomes in plasma of Relapsing Remitting Multiple Sclerosis patients: interference of soluble protein, lipoproteins and another extracellular vesicles

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Exosomes are membranous vesicles from 40-150 nm released from a multitude of cell types, including nerve and immune cells. These nanovesicles together with microvesicles (100-150nm) are correlatively referred to as small extracellular vesicles (sEVs) and they are physiologically secreted for cellular communication. Exosomes and their nucleic acid and protein cargo have gained significant interest as potential diagnostic biomarkers because their number, markers and composition change in pathological processes. Besides, they can be found in all biological fluids and can cross the blood-brain barrier. Human plasma is a rich source of readily accessible sEVs; however, the identification of exosome from other extracellular vesicles and separation from plasma proteins and non-EV lipid particles represents a considerable challenge. In our study we identified the best exosomal marker in plasma of relapsing remitting multiple sclerosis (RRMS) patients, combining cytoplasmic and membrane markers. Moreover, we compared commonly and new used isolation techniques, such as ultracentrifugation and SmartSEC columns, respectively. Concentration and size of particles isolated by each method was evaluated by nanoparticle tracking analysis, using the variation in particle size distribution to check the relative impact of lipoproteins and protein aggregates on the isolated sEV population. Isolation of sEVs was confirmed by transmission electron microscopy and the expression of CD9, CD81 and CD63 tetraspanin markers by flow cytometry. Purification of sEVs from soluble protein was determined by the ratio sEV particle count/protein concentration. Finally, lipoprotein particles and microvesicles co-isolated with exosomes was analysed by flow cytometry using APOB and ARF6 markers, respectively. Overall, this study reveals that the choice of exosome markers and isolation procedure significantly impacts sEV yield from human plasma, specifically in RRMS patients, together with the presence of lipoprotein and protein contaminants in the sample.

Oral communication (O2)

MUC16 is overexpressed in idiopathic pulmonary fibrosis and induces fibrotic responses mediated by transforming growth factor-β1 canonical pathway

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Background: Several transmembrane mucins have demonstrated to contribute intracellularly to induce fibrotic processes. The extracellular domain of mucin 16 (MUC16) is considered as a biomarker for disease progression and death in Idiopathich pulmonary fibrosis (IPF) patients. However, there is no evidence regarding the signalling capabilities of MUC16 that contribute to IPF development.

Objective: To analyze the implication of MUC16 in IPF.

Methods and results: MUC16 is overexpressed in lung tissue of IPF patients (n=20) compared with healthy subjects (n=17) and localized in fibroblasts and hyperplastic alveolar type II cells. Repression of MUC16 expression by small interferent RNA (siRNA)-MUC16 transfection inhibits the transforming growth factor (TGF)-β1-induced fibrotic processes such as mesenchymal/ myofibroblast transformations of alveolar type II A549 cells and lung fibroblasts, as well as fibroblast proliferation. SiRNA-MUC16 transfection also decreases the TGF-β1-induced SMAD 3 phosphorylation, thus inhibiting the smad binding element activation. Immunoprecipitation assays and confocal immunofluorescence show the formation of a protein complex between MUC16/p-SMAD3 in the cell membrane after TGF-β1 stimulation.

Conclusions: MUC16 is overexpressed in IPF and collaborates with TGF-β1 canonical pathway to induce fibrotic processes. Therefore, direct or indirect targeting of MUC16 could be a potential drug target for human IPF.

Oral communication (O3)

Functional and phenotypic characterization of human TBX4 sequence variation: gain of function mutations result in late-onset lung disease

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TBX4 is one of the main causal genes in Pulmonary Arterial Hypertension (PAH), for a long time it was mostly related to paediatric forms of the disease, but in the past few years, mutations have been increasingly found in adult patients.

Most of the studies describing TBX4 mutations have a low number of patients with missing clinical data that has made it extremely difficult to study the hallmarks of TBX4-mediated PAH. In an international effort to study the effect of these mutations inTBX4, we have assembled a cohort of 100+ patients of different types of PAH, developmental lung disease, and Small Patella Syndrome (all with mutations in TBX4).

First, we developed a method to assess TBX4 activity using over-expression vectors and luciferase reporter plasmids. Then, we selected 51 TBX4 variants (42 missense, 3 splice-site, and 6 in-frame insertions/deletions) with different in silico predictions, carried out site-directed mutagenesis, and tested the missense and indels with this method. For the splice variants, we opted for a minigene assay.

Our results showed that TBX4 pathogenesis can be caused both by Loss of Function (LoF) and Gain of Function (GoF), our method allowed for the classification of 32 variants as Pathogenic, 12 as benign, and 4 as Variants of Uncertain Significance. We have reclassified up to 70 % of the variants analyzed, highlighting the importance of using functional data to improve genetic diagnosis. Also, coupling functional and phenotypic data, we have analyzed how the effect of the mutation affects patients' outcomes.

In all, we have performed functional analyses for the largest TBX4 mutation set to date and used the results to reclassify the mutations and analyze patients' outcomes.

Oral communication (O4)

Highly pleiotropic variants of human traits and diseases associated with strong background selection

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Pleiotropy is a phenomenon in which a single locus has an effect in more than one trait. It has been shown to be common in the human genome, with between 50 and 60 % of SNPs found to be pleiotropic. Recent studies have found that rare variants are less pleiotropic than common ones, which suggests that highly pleiotropic variants (i.e. variants associated with a large number of traits) might be under strong purifying selection. However, it has also been found that highly pleiotropic variants have greater average effect sizes. In order to investigate this apparent contradiction, we analysed data from the NHGRI-EBI GWAS Catalog for 41 traits and diseases. We studied if the average effect size, minor allele frequency and heritability contribution of the variants in the catalog vary with their degree of pleiotropy (the number of traits and diseases a variant is associated with). To investigate whether the highly pleiotropic variants are enriched in genomic regions under strong purifying selection we also analysed data from other three pleiotropy studies. As a measure of purifying selection we used the B statistic, which indicates the reduction in genetic diversity in a given genomic region due to the action of background selection (selection on deleterious mutations, particularly in low recombination regions). Our results indicate that the average effect size, minor allele frequency and heritability contribution of the variants increase with their pleiotropy degree, and that highly pleiotropic variants are enriched in regions of strong background selection.

Oral communication (O5)

Algae as a source of nutraceutics and functional foods

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The global demands for functional and nutraceutical products is growing rapidly. In this sense, there is important evidence that both nutraceuticals and functional foods derived from algae are increasingly being consumed, due to their therapeutic role in the prevention of certain diseases. Bioactive compounds from algae, including pigments such as fucoxanthin (Fx), have various biological activities, including antioxidant, antibacterial, anticancer, antidiabetic, antitumor, antiviral, anti-inflammatory, and anticoagulant activities, which play a health-enhancing role on the consumer. In this sense, the extraction of Fx from Undaria pinnatifida was carried out in the laboratory, by means of heat-assisted extraction. The optimal extraction parameters were determined, combining 100% acetone as solvent, at a temperature of 50 °C for 90 minutes, resulting in the extraction of, approximately, 2.8 mg of fucoxanthin per gram of dry weight (dw) of algae sample (mg Fx /g dw), with an extract yield of ≈17 mg extract/g dw, achieving a purity of ≈150 mg Fx/g of extract. Once the extraction of Fx was optimized at a laboratory scale, we proceeded to carry out the implementation of this procedure at an industrial level, by the establishment of a pilot plant with a dual aim: i) the large-scale production of Fx, and ii) the design of 5 Fx-enriched nutraceuticals, derived from four different algal species, namely U. pinnatifida, Himanthalia elongata , Porphyra purpurea, and Ulva lactuca, and the mixture of two microalgae:Spirulina and Chlorella. Overall, our results provided evidence about the successful transference of bioactive natural product research results to their application at an industrial level. Consequently, such strategy must be regarded as a reliable approach for the production of new nutraceuticals in the food industry.

Keywords: Fucoxanthin; extraction, nutraceuticals, functional products

Acknowledgements: The research leading to these results was supported by MICINN supporting the Ramón y Cajal grant for M.A. Prieto (RYC-2017-22891) and the FPU grant for A. Carreira-Casais (FPU2016/06135), the program BENEFICIOS DO CONSUMO DAS ESPECIES TINTORERA-(CO-0019-2021) that supports the work of F. Chamorro and the program Grupos de Referencia Competitiva (GRUPO AA1-GRC 2018) that supports the work of J. Echave; by University of Vigo for supporting the predoctoral grant of M. Carpena (Uvigo-00VI 131H 6410211). Authors are grateful to Ibero-American Program on Science and Technology (CYTED—AQUA-CIBUS, P317RT0003), to the Bio Based Industries Joint Undertaking (JU) under grant agreement No 888003 UP4HEALTH Project (H2020-BBI-JTI-2019) that supports the work of P. Garcia-Perez and C. Lourenço-Lopes and to AlgaMar enterprise (www.algamar.com) for the collaboration and algae material provision. The JU receives support from the European Union's Horizon 2020 research and innovation program and the Bio Based Industries Consortium. The project SYSTEMIC Knowledge hub on Nutrition and Food Security, has received funding from national research funding parties in Belgium (FWO), France (INRA), Germany (BLE), Italy (MIPAAF), Latvia (IZM), Norway (RCN), Portugal (FCT), and Spain (AEI) in a joint action of JPI HDHL, JPI-OCEANS and FACCE-JPI launched in 2019 under the ERA-NET ERA-HDHL (n° 696295).

Oral communication (O6)

Parametric Landmark estimation of the transition probabilities in survival data with multiple events

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The estimation of transition probabilities is of major importance in the analysis of survival data with multiple events. These quantities play an important role in the inference in multi-state modeling providing in a simple and summarized manner long-term predictions of the process.

Recently, de Uña-Álvarez and Meira-Machado (2015) proposed nonparametric estimators based on subsampling, also known as ladmarking, which have already proved to be more efficient than other nonparametric estimators in case of strong violation of the Markov condition. However, as the idea behind the landmarking is to use specific portions of data when the subsample sizes are reduced or in the presence of heavily censored data this may lead to higher variability of the estimates.

To avoid the high variability of the nonparametric landmark estimator proposed by the de Uña-Álvarez and Meira-Machado (2015), we introduce parametric estimators for the transition probabilities that are also based on subsampling. We have considered several flexible distributions to handle this issue appropriately. One of the proposed approaches, which provides good results, with high flexibility, is based on the generalized gamma distribution.

Results of simulation studies confirm the good behavior of the proposed methods. We also illustrate and compare the new methods to the nonparametric landmark estimator through a real data set on colon cancer.

Oral communication (O7)

Where is the mice? - Exploring the R package

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The R mice package is a package that appeared in 2001 and imputes multivariate data using chained equations. We are currently using this package to map missing data in a multivariate database with multiple missing data. The application of this package required a more detailed analysis. In this presentation we will explore this package a bit. First, let's summarize the features of the package. Second, we are going to show you some simple applications that we will try to illustrate with some examples. At the end we will summarize some practical problems of the package.

Oral communication (O8)

DTDA, the renewed R package to analyse doubly truncated data

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The analysis of doubly truncated data is relevant in epidemiological applications, when the observation of the lifetime of interest is limited to events between two specific calendar dates. This implies that small or large times are less probably observed and thus properly corrections to the estimators must be done in order to avoid biased estimations that may lead to wrong conclusions. Given the aforementioned motivation, the interest of the scientific community in the phenomenon of random double truncation has significantly grown, particularly in fields like Epidemiology and Survival Analysis and this has motivated the development of software routines that could facilitate a proper analysis of this kind of data. DTDA, built in 2010, was the first R library spreading the methods for the analysis of doubly truncated data. Since the release of the original DTDA, new statistical methods for analyzing doubly truncated data have been proposed. Apart from the classical algorithms for estimating the cumulative distribution function, the renewed DTDA package outfits smoothing methods to estimate the kernel density function and hazard function, including the bandwidth selection procedures for the density function. Additionally, these new functionalities are accompanied by some real datasets from different areas.

Oral communication (O9)

Re-analysis of miRNA-Seq differential expression analyses in neuropsychiatric diseases

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MiRNAs are attracting considerable interest as potential biomarkers on neuropsychiatric diseases due to their expression plasticity. Consequently, many studies have been published in this regard in the last decade, suggesting a large number of potential biomarkers. However, there is widespread concern about the reproducibility and validity of these results. This study compares the differentially expressed miRNAs reported by 5 recent studies of neuropsychiatric diseases, with those obtained using the miARma-Seq pipeline. In general, we found a low reproducibility (0-74%), and some variations related to the software used for the differential expression analysis. Our results match the idea that miRNAs reported as potential biomarkers in neuropsychiatric diseases are strongly correlated with the analytical methodology and the biological references used; nonetheless, more research is needed to unravel the magnitude of this problem and determine its main causes.

Poster session

Poster

Red seaweed mediated synthesis of gold nanoparticles with antitumoral and antioxidant potential

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Porphyra linearis (PL) is a winter red macroalga in the order Bangiales, widely distributed in the northern Atlantic and Mediterranean coasts. The genus Porphyra contains important algal species that are cultivated and/or harvested for human consumption since they are rich in protein (25-50%), vitamins, minerals and fibres. Regardless the interest of PL for human nutrition, studies dealing with its biological properties are still scarce.

In this study, we proposed the application of PL in the green synthesis of gold nanoparticles. An aqueous extract of PL was prepared and used to perform the synthesis of nanoparticles by the reduction of HAuCl4. Obtained nanoparticles were fully characterized, confirming the formation of spherical units with mean diameter of 15.0±3.0 nm. To study the possible biomolecules involved in the reducing and stabilizing process, Fourier transform infrared spectra of the PL extract and the Au@PL were recorded. The carbohydrates in the extract were analyzed by high performance anionic exchange chromatography coupled to pulsed amperometric detection before and after nanoparticles formation.

The antioxidant activity of the extract before and after the synthesis were analyzed by determining the reducing activity, total phenolic compounds and DPPH scavenging effect. Likewise, the capability of the PL aqueous extract and Au@PL to reduced ROS release under oxidative stress conditions was also measured in promyelocytic cells. Finally, we tested the potential antitumor activity of the PL aqueous extract and Au@PL in a monocytic cell line and a lung epithelial adenocarcinoma. We found a selective antitumor effect of the samples against the monocytic cells, indicating its potential use in treating monocytic leukemia. Au@PL showed a superior antioxidant and antitumor activity compared to the PL extract, hence confirming the potentiality of gold nanoparticles to function as carriers of biological active molecules into the human cells.

Poster

Photosensitizer-synthesized AuNP for application in photodynamic therapy

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Cancer is the second leading cause of mortality in the world. Traditional therapeutic approaches still present severe limitations such as low efficacy and unspecific accumulation derived side effects. Therefore, the development of new alternative therapies is of uppermost importance. Photodynamic therapy (PDT) has recently arisen considerable attention as a new more efficient and less side-effect causing therapeutic approach. PDT consists in the administration of a non-toxic organic photosensitizer agent (PS) followed by irradiation at a specific wavelength to produce cytotoxic singlet oxygen and/or reactive oxygen species (ROS) that kill cancer cells. A major drawback for the clinical application of PDT is the low specificity of the organic PS and their low bioavailability due to their high hydrophobicity. The combination of organic PS with metallic nanoparticles can overcome these limitations and enhance their cytotoxicity by synergistic photothermal effect. In this work, the natural photosensitizer riboflavin (RF), otherwise known as vitamin B2, was selected to achieve the green synthesis of gold nanoparticles with potential application in PDT.

The synthesis of gold nanoparticles was achieved by adding 0.02 mM of HAuCl4 to 0.05 mM of RF and 10 mM of SBS at 100 ºC. All the reactions were followed by UV-visible spectroscopy and the nanoparticles obtained were characterize by means of transmission electron microscopy (TEM), High Resolution TEM (HRTEM), X-ray diffraction analysis (XRD), energy dispersive X-ray analysis (EDX), Z-potential analysis, Fourier transformed infrared spectroscopy (FTIR) and magnetic nuclear resonance spectroscopy (NMR).

In summary, an efficient method for the synthesis of AuNP has been described, by direct reduction with photosensitizer RF. The nanoparticles obtained could be further used for the development of improved phototherapy against a wide variety of diseases, by combining the photodynamic cytotoxic effect of the natural photosensitizer and the photothermal effect of gold nanoparticles.

Poster

Specific substitution models for the HIV-1 protease and integrase

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The HIV-1 protease (PR) and integrase (IN) are common molecular targets of HIV-1 antiretroviral therapies. However, HIV-1 often evolves rapidly, leading to resistance against therapies, and thus its evolution should be considered to develop effective therapies. By contrast, and despite the HIV-1 is the most sequenced organism so far, only two empirical substitution models (HIVb and HIVw), based on the entire viral proteome, have been developed. Here we inferred new empirical substitution models to mimic the HIV-1 PR and IN evolution considering thousands of protein sequences and we found that these models more accurately describe (in terms of likelihood) the evolutionary process of HIV-1 PR and IN than currently available substitution models. The results also suggest that efforts should be made in developing protein-specific substitution models in order to improve the accuracy of modeling protein evolution.

Poster

Development of biocompatible liposomal nanocarriers for the efficient transfection of siRNA into pancreatic tumor cells

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KRAS is a protein frequently mutated in pancreatic ductal adenocarcinomas (PDAC), inducing the upregulation of some transcription factors and nuclear effectors, such as FOSL-1 and YAP, both implicated in tumor progression and poor prognosis (1-3). For that reason, we developed a new therapy based on the use of specific small interfering RNAs (siRNAs) encapsulated in liposomes to downregulate these nuclear effectors.

Liposomes composed of the cationic lipid dioctadecyldimethylammonium bromide (DODAB) and the neutral lipid 1-monooleoyl- rac-glycerol (MO) at two different ratios (2:1 and 1:2) were synthetized to obtain different nanostructures (4). Then, different siRNAs against FOSL-1 and YAP were encapsulated into the liposomes to form lipoplexes that are able to transport the siRNA inside the tumoral cells. The transfection efficiency was evaluated in vitro using different PDAC tumor cell lines by measuring FOSL-1 and YAP protein silencing. Biocompatibility studies were also performed (cell viability, hemotoxicity and complement activation).

A decrease of about 40-60% in the expression of the proteins was detected 48-72 hours after the incubation of PDAC cells with the lipoplexes in comparison to the untreated cells. Both, liposomes and lipoplexes were cytocompatible at relevant therapeutic concentrations and showed a good transfection efficiency in vitro, showing better performance the DODAB:MO (1:2) liposomes. Moreover, they were hemocompatible and did not induce complement activation or hemolysis in plasma and whole blood samples, respectively.

The DODAB:MO liposomes are efficient transfection agents to introduce the siRNAs into PDAC tumor cells and show excellent biocompatibility in vitro. The silencing of FOSL-1 and YAP factors could be a promising adjuvant therapy for the treatment of PDAC.

We thank Dr. Barbacid's laboratory for providing us the mouse pancreatic tumor cells. This work was funded by the MEC (ref. BIO2017-84974-R) and Xunta de Galicia (ref. GRCED431C2020/02). L.D-G acknowledges a fellowship from Xunta de Galicia (ED481A-2018/294) and a grant from IACOBUs program (2019/2020-190).

Poster

Voltaje-gated potassium channel expression in schizophrenia and multiple sclerosis lymphocytes

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Schizophrenia is a psychiatric disorder with a wide spectrum of clinical and biological manifestations. The usual treatment is the administration of different antipsychotics by the psychiatrist. A new drug, unique in its mode of action, is NW-3509 (in phase III of the clinical trial), a selective blocker of voltage-gated Na + channels ($Na_v1.3$, $Na_v1.7$ and $Na_v1.8$). Currently unknown mechanisms that seem to be also present in the neuronal membrane involve voltage-gated K + channels (Kv) (both types of channels modulating NMDA receptors). Therefore, an experimental work was proposed that tries to find new mechanisms that can be used in schizophrenia as a predictor biomarker of response to the new antipsychotic drug in the immune system. In this way, the presence of Kv in lymphocytes of patients with schizophrenia and controls has been studied, as well as the changes in the expression of the different subtypes of Kv (Kv1.1, Kv1.2, Kv1.3, Kv4.2 , Kv4.3 and Kv7.2). Furthermore, multiple sclerosis is a neuroinflammatory disease for which one of the therapeutic targets is Kv1.3, which is why it was decided to add this disease to the study. The study was carried out with blood samples from patients diagnosed with schizophrenia (n = 42) according to the DSM5 criterion of EOXI-Vigo, patients diagnosed with multiple sclerosis (n = 40) and controls (n = 42). Lymphocyte extraction was performed using a Ficoll gradient. The study of the presence of Kv in lymphocytes was carried out by immunofluorescence and the study of expression levels by western blot. All selected Kv subtypes are present in the lymphocytes of patients with schizophrenia and controls. In addition, statistically significant differences have been found in the level of expression of these channels between both groups, this being higher in both schizophrenia and multiple sclerosis. Due to the novelty of the study, the lack of bibliography has not allowed to justify the data obtained for the quantification of protein. Consequently, and given that this study shows a clear relationship between Kv and these pathologies, it would be of great interest to carry out a broader study that relates them and delves into the structure, tissue expression, cell location and functioning of Kv, as well as an electrophysiology study to find out if these channels function differently between controls and patients

Poster

Vitamin D analogue Gemini UVB1 against colorectal cancer

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UVB1 is a Gemini type vitamin D analogue recently synthesized in our research group. We showed that this analogue inhibits colorectal carcinoma progression and lacks hypercalcemic activity and toxicity effects in in vivo assays. The first synthesis of this compound was based on a serendipitous result. However, we could not reproduce the same transformation, thus we have developed a new pathway to obtain it from a building block previously synthetized in our laboratory.

Poster

Synthesis of a non-Gemini vitamin D analogue of UVB1

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1α, 25-(OH)2-vitamin D3 (calcitriol) has an important antiproliferative activity with possible use as a therapeutic agent. However, the necessary therapeutic doses carry a risk of hypercalcemia.

The search for new more selective analogues has been promoted, thus giving rise to a second generation of analogues due to modifications in the side chain. Second-generation vitamin D3 analogues may provide interesting improvements over the pharmacological and pharmacokinetic properties of the parent analog by simple C20 epimerization.

In search of new analogues, we serendipitously synthesized a new type Gemini vitamin D3 analogue which was named as UVB1. The biological evaluation showed that it has potent antitumoral effects over a wide range of tumor cell lines and lacks hypercalcemic activity as well as it inhibits colorectal carcinoma progression.

This work focuses on the design of a synthetic route to obtain a novel non-Gemini UVB1 analogue which corresponds with a 20-epi vitamin D3 analogue.

Poster

Remote monitoring of cancer patients at high risk of relapse through an injectable nanobiosensor

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Remote patient monitoring (RPM) with cancer can be potentially used to detect the disease in early or relapse stages to provide greater probabilities to survival. The research of novel tools to monitor cancer in patients should be used to detect it since onset or during progression.[1] Herein, we propose the development of a remote nanobiosensor for high-risk profile cancer patients, to be injected in the dermis layer of the skin. To develop this nanobiosensor, plasmonic nanoparticles such as gold nanostars (GNSs) labelled with a Raman reporter (RaR), were selected to be embedded in a biocompatible matrix, to support these nanoparticles.[2] This hybrid nanobiosensor (hydrogel + GNSs with RaR) will be injected subcutaneously, and the biomolecules in the interstitial fluid (ISF) will diffuse through the sensor and will be subsequently detected by means of surface-enhanced Raman scattering (SERS) spectroscopy.[3] The main function of SERS will be to obtain the fingerprint of the cancer biomarkers in close vicinity to the GNSs. In previous results, it was shown that hydrogels with more concentration of GNSs have a higher signal than those with a lower concentration of gold, as expected. Moreover, we performed the SERS analysis of individual ISF compounds and of artificial ISF to first have a library of the expected Raman signals. The next steps are to train a machine learning algorithm for the classification of patients based on their ISF/serum SERS fingerprinting to later apply to the analysis of real samples from prostate cancer patients in a retrospective preclinical study.

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Poster

Metastasis-on-a-chip System to Study the Invasive Capacity of Patient-Derived Circulating Tumour Cells

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Metastasis is responsible for the majority of cancer-related fatalities, however it is still poorly understood [1]. With such impact in morbidity and mortality of cancer patients, it is crucial to better understand this process, to be able to develop better diagnostic tools and therapies. Circulating Tumour Cells (CTCs) are malignant cells that escape the tumour and travel through the blood stream, potentially invading other organs and causing metastasis. Being the key factor of this hallmark, understanding the mechanisms for CTC intra- and extra-vasation can be crucial to comprehend metastasis. To this end, it is necessary to build 3D biomimetic models of metastasis that replicate truthfully the in vivo tumour microenvironment. Microfluidics was selected as the ideal technology to accomplish this task, since it overcomes the limitations of traditional 2D static models and provides interesting features such as low cost, high throughput, and miniaturization.

We have developed a multichannel microfluidic organ-on-a-chip system to mimic cancer vasculature, fabricated by standard photolithography and replica-moulding techniques. The system was provided with the appropriate channel surface functionalization to favour cell attachment and growth. Our preliminary results demonstrate the growth of an endothelial monolayer onto the channel walls, and a 3D culture of tumour cells embedded in a hydrogel matrix in an adjacent channel. Tests are ongoing to demonstrate that our system correctly mimics the tumour environment. Future work will be dedicated to study the influence of cell phenotype in cancer cell migration and invasion.

Acknowledgments: This work was developed under the METASTARG project that received funding from the “European innovative research & technological development projects in nanomedicine” EuroNanoMed III program.

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Poster

Protein folding stability of HIV-1 protease resistance mutations

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The emergence of variants resistant to host immune systems and antiretroviral therapies is fundamental for viruses. However, resistance mutations can also result in some costs to the viral protein, such as decreasing the normal protein activity and folding stability. Concerning the latter, here we explored the influence of relevant resistance mutations on the folding stability of the HIV-1 protease (PR) and the overall evolution of folding stability of this protein. In particular, we analyzed the protein folding stability in a variety of HIV-1 PR variants with and without common resistance mutations to therapies and we found that these mutations decrease (in average) the folding stability, although some of them can also maintain or increase it. We also found that the folding stability of the HIV-1 PR fluctuated widely over time, without showing a clear trend. We conclude that resistance mutations can decrease the HIV-1 PR folding stability but this pattern cannot be extended to all resistance mutations and, that the evolution of the HIV-1 PR folding stability is complex with multiple fluctuations over time.

Poster

Peloids from different parts of the world for therapeutic uses: A review

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The use of clays for therapeutic and / or cosmetic purposes is ancestral and dates back to the existence of human beings. The term peloid or thermal mud was defined for the first time in 1949 and subsequently evolved until 2013, the year in which Gomes and collaborators [1] published a review of the different definitions of the term over time, their classification according to different authors and their historical evolution. General way, peloids are defined as heterogeneous mixtures of a solid phase (organic, inorganic or mixed) mixed with mineral-medicinal or sea water, which is used for therapeutic or cosmetic purposes in thermal centres.

In the last decades, many publications [2-5] collect different studies on the peloids, being able to group them into those that have a mainly therapeutic and / or cosmetic indications, others that report their chemical and thermophysical properties; and we also find studies on the production and maturation process or those that focus on its components, both organic and inorganic.

This work presents a review of the use of peloids in different parts of the world carried out by geographical areas. Thus, the Mediterranean area is studied, in which there is a great tradition of using thermal peloids, the central area of Europe and those from countries of the former USSR. The situation of the peloids in the American continent, as well as Asia, Oceania and Africa, is also reviewed.

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Poster

Design and development of an automatic microcalorimetric control system for the study of bacterial growth

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The use of a Calvet microcalorimeter allows the monitoring of numerous physical, chemical and biological processes of long duration. The remote use of a microcalorimetric experience allows continuous monitoring of the data from any point, without the need to be "in situ" where the experience is carried out.

In this work we present an automatic data acquisition device through a remote system, through a web interface, connected to a microcalorimeter, for the study of bacterial growth that occurs in joint prosthesis infections.

The system used to collect data from the Calvet microcalorimeter is a microvoltmeter, which registers the potential differences, connected through an RS232 interface to a data acquisition and control system consisting of a Raspberry PI minicomputer. This board is a free hardware platform, which allows you to adapt your configuration. Thus, using various modules, the design can be endowed with various functionalities. The operating system that runs on the minicomputer is LINUX, which provides the system with great robustness and reliability against failures.

The application consists of the creation of an API Rest server, which will be in charge of providing an interface for read and write operations (CRUD) on a Postgree database, which are carried out from the frontend.

For the backend programming of the application, we use the Python FastAPI framework, which allows us to interact with the database through REST requests through HTTP.

For the development of the frontend, React is used, a JavaScript framework developed by Facebook, which provides dynamic web pages through the use of reusable components.

From the control panel of the app, all the necessary operations can be carried out to create a new experience and monitor its evolution by viewing the generated graphs. In addition, there will be tools to handle the data of the experiences, allowing to filter and analyse the registered signals.

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Poster

Altered expression of cytoskeletal proteins in lymphocytes in Alzheimer's disease.

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Introduction and objectives: The immune system is involved in the functioning of the central nervous system, and with this, one of the main cell types, lymphocytes, somehow regulate brain function. In addition, from studies with proteomic techniques we know that cytoskeleton proteins from lymphocytes and other cells such as Vinculin, PIP5K1C, RIAM, β3 Integrin, β2 Integrin are involved in the pathophysiological mechanisms of various mental disorders such as major depression, bipolar disorder, as well as neurological disorders such as Alzheimer's disease. All of them form a network in the plasma membrane in order to establish cellular communication. Since lymphocytes serve as a comparative study model to investigate what happens in neuronal pathways, we set out to investigate whether there are alterations in these proteins that allow us to find out how the cytoskeleton is altered in this disease.

Methods: A comparative study was developed with 10 patients with Alzheimer's disease and 10 healthy controls, where the levels of Vinculin, PIP5K1C, RIAM, β3 Integrin, β2 Integrin in lymphocytes were measured. Patients with Alzheimer's disease were evaluated by Unit of Dementia at Hospital Alvaro Cunqueiro.

Results: We compared the levels of the theses proteins between the total number of patients (N=10) and controls (N=10) by unpaired two-tailed t-test. Levels of Vinculin, β3 Integrin, β2 Integrin, RIAM, PIP5K1C in lymphocytes were lower in Alzheimer's disease patients compared to control, however, only with PIP5K1C we detected significantly lower levels in patients group compared with controls ($P<0.005$).

Conclusions: The levels of cytoskeleton proteins (Vinculin, β3 Integrin, β2 Integrin, RIAM, PIP5K1C) are involved in cellular communication and are altered in Alzheimer's pathology. This would indicate that there are molecular changes in lymphocytes that may somehow modulate the pathophysiology of Alzheimer's disease.

Poster

Experimental determination of heat loses on silica-coated iron oxide nanocrystals in magnetic hyperthermia

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Iron oxide (Fe_3O_4/gamma-Fe_2O_3) nanocrystals when exposed to an alternating magnetic field can transform power losses to thermal energy and therefore dissipate heat. This ability renders them highly interest for different applications including those in the biomedical field, such as localized cancer therapy. Depending on the shape and anisotropy, size, concentration and magnetic properties (ferrimagnetic nature, superparamagnetic regime, etc.) the amount of generated thermal energy can differ. [1] Since the surface of Fe3O4 nanoparticles has to be functionalized to remain non-harmless to the human body, the biocompatible coating, while inevitable, can also cause heat losses in the system. [2] Accordingly, with the aim of investigating on the heat transfer through the interface of an amorphous silica coating over crystalline magnetic core to the media, different shapes and sizes of Fe3O4 naked and coated with a SiO2 layer with various thicknesses were investigated. For the experimental determination of the heating capability (SAR) of these magnetic nanocrystals in water, a calorimetric method was employed and the data fitted by using corrected slope method. [3]

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Poster

Extraction and proteomic analysis of human breast milk exosomes

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Besides being source of nutrients, breast milk (BM) contains a wide variety of biologically active components that guide the maturation of the new-born's systems. Exosomes are small membrane vesicles of endosomal origin, secreted by a wide variety of tissues to extracellular space, blood, cerebrospinal fluid, urine and also BM. Initially described as organelles for removing debris and unwanted molecules, the discovery that they contained proteins, mRNAs, and miRNAs suggests a very interesting role as mediators in cell-cell and tissue-to-tissue communication.

During lactation, babies are fed exclusively on BM. Thus, we postulate that in exosomes there are genetic regulators that cause proper brain development. Under this hypothesis, we intend to extract exosomes from BM to identify new regulators that modulate the expression of genes involved in neurodevelopment. For this, we will analyse milk samples from term mothers and from mothers of preterm infants (infants born between 28 and 37 weeks of gestation). In addition, within each milk fraction we will analyse two states, colostrum and mature milk.

Five groups of samples according to the time of delivering were recruited: 4 mature milk (group 1) and 4 colostrum (group 2) at term; 4 premature milk (group 3) and 4 premature colostrum (group 4); and 4 preterm mature milk (group 5). Exosome extraction were done with EX04 Exo-spinTM midi column, optimized by our laboratory for human BM. WB control was done by using exosome markers CD9, CD63, CD81. Electron microscopy was realised to measure the size of the micro vesicles. Proteomic analysis of exosomes was performed by LC-MS / MS. Data was analysed with two software: Proteome DiscovererTM and PEAKS.

With the Proteome Discover software we identified 2009 proteins, of which 74 are possible co-pollutants, leaving 1935 proteins identified in the milk exosomes. 45 of them had quantitative data in all samples. If we analyse the data with the Peaks software and the identification is performed with the Spider with an FDR of 0.5%2 we identified 1312 proteins. Quantification with Label free software with 20ppm and 20min mass error and retention time shift tolerances, and with the filter's significant protein greater than or equal to 20, fold change greater than or equal to 2 and at least 1 unique peptide, we obtained that 51 proteins are significantly modulated in the 5 groups.

Proteins related to the mammary glands and milk are beta, kappa, and alpha S1 caseins, Lactose synthase B protein, Bile salt-activated lipase, Prolactin induced protein, and Zinc-alpha-2-glycoprotein. These last two related to each other and more in groups 2 and 4.

This is the first molecular analysis of the milk samples, the next step will be to identify potential regulators of the central nervous system, which will be tested in cell cultures of neurons (SH-SY5Y) and glia (U87). This will allow us to investigate the changes that occur in different groups and correlate this with the different stages of maturation of the child.

Poster

Antiproliferative features of ultrasound treated hybrid carrageenans from *Mastocarpus stellatus*

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Carrageenophyte red algae as *Mastocarpus stellatus* (Gigartinales, Rhodophyta) are used mainly industrially to obtain biopolymers with gelling or thickening properties. However, these algae are also rich sources of compounds with potential as antitumoral, antiviral or anticoagulant agents. The valorisation of algae components with potential biological activities, in a sustainable way, with interest for food and non-food applications should be further studied. The main objective of this study is to recover extracts from ultrasounds treated *Mastocarpus stellatus* red seaweed and analyse their antitumoral potential.

Mastocarpus stellatus employed as raw material to the hybrid carrageenan extraction was supplied by Portomuíños company (A Coruña, Spain). All alga samples were sonicated employing an ultrasound bath at fixed temperature (80 °C) and frequency (80 kHz), monitoring some other relevant processing conditions such as time, alga/water ratio, or ultrasounds amplitude. Hybrid carrageenans extracted at the optimal extraction conditions were evaluated by different human carcinoma cell lines such as lung (A549), ovarian (A2780), cervix (HeLa 229) and colon (HT-29) cells.

Outcomes indicated that the achievement of hybrid carrageenan with promising biological activities was highly dependent on the ultrasound processing conditions. Biopolymers recovered at the optimum extraction conditions (i.e. sonication time ~35 min, solid liquid ratio ~2 g/100 g, and ultrasound amplitude ~79%) were cytotoxic against four human carcinoma cell lines (i.e. A549; A2780; HT-29; HeLa 229). In all cases, extracted hybrid carrageenan exhibiting IC_{50} values lower than 51.9 µg/L. Overall, the suggested green ultrasound treatment is a simple and flexible method that offers an integral valorisation of Mastocarpus stellatus that could be extensible to other carrageenophyte red alga.

Poster

Analysis of endothelin-1 (EDN-1) UTR regions

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Pulmonary Arterial Hypertension (PAH) is a disease characterized by an increase of secretion and deregulation of Endothelin-1 (ET-1). This peptide is secreted by the endothelium of blood vessels and promotes vasoconstriction. We carried out the characterization of the UTR regions of endothelin-1gene (EDN-1), in order to determine common variations that may modulate disease outcome.

The analysis was carried out in 60 patients with different classes of PAH, testing a fragment of 2 kb for both UTR region. An in silico analysis was performed to evaluate binding transcription factors. Luciferase assay was done to evaluate in vitro the SNP influence in gene expression. Data revealed the presence of a deletion in the promoter region (rs397751713), while a transversion in the 3' UTR region was found (rs2859338). The distribution of the genotype frequencies in our PAH patients were: for rs397751713: A/A: 0.08; A/-: 0.27; -/-: 0.66; for rs2859338: A/A: 0.15; A/G: 0.60; G/G: 0.25. Variations are located in a KLF4and PPARg binding sequence and a vitamin D receptor binding sequence respectively. Both transcription factors are related to PAH development.

In conclusion, these SNPs in the UTR regions of EDN1 are related with gene expression levels, as we measured higher expression rates for patients with A/A and G/G genotype. Moreover, we hypothesized that this overexpression is due to the inability of KLF4, PPARg and vitamin D receptor to attach the target sequence and to regulate the expression of EDN1, as KLF4 is probe to avoid PAH when present and vitamin D is an anti-hypertrophic factor.

Poster

Neurochemical effects of microplastics in experimental rats

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Plastic pollution is considered one of the main environmental problems. Multiple studies have documented that microplastics are ingested by various marine organisms, accumulate in specific tissues, and are transported through the food chain to mammals, posing a potential risk to the health of human populations. Experimental data shows that, in fish, these derivatives can cross the blood-brain barrier, reach the brain and cause positive regulation of genes in the CNS, affect the activity of acetylcholinesterase and the dopaminergic system. The main objective of the present work is to evaluate the effects of the administration of microplastics on the biochemical parameters in the levels of acetylcholinesterase and the antioxidant capacity determined by colorimetric methods in different areas of the nervous system (cerebellum, trunk, cortex, striatum and hippocampus). Sprague-Dawley female rats (250-300 g, n = 5 / experimental group) were used for the experiments, performing two experimental groups: control and treated with 5 mg / kg of 1 micrometer microplastics for 4 days of intraperitoneal administration. The data were evaluated with an ANOVA / Dunnet statistical analysis. No significant differences were found in acetylcholinesterase levels in different brain areas. There was a significant increase in the concentration of antioxidants in the hippocampus of the treated animals (1.12 ± 0.07 and 1.90 ± 0.55 nmol / microliter of antioxidant, $p < 0.05$). In the rest of the areas, no significant differences were obtained. With the results obtained, we see that microplastics produce an effect on oxidative stress, but more experiments are needed with other doses of administration and other treatment times.

Poster

Optimization of microwave-assisted extraction, from an edible marine alga of the Galician coastline, using a response surface methodology

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Macroalgae are a suitable source for the obtention of new functional ingredients. Besides their high content in carbohydrates, proteins, and minerals they are also rich in secondary metabolites like phenolic compounds, carotenes, and carotenoids with many different properties and applications. One of these compounds is fucoxanthin (Fx), a pigment majorly found in brown algae, that has gathered the attention of many researchers in the last few years, because of its antioxidant, anti-inflammatory, anticancer, neuroprotective, and antiobesity activities. Undaria pinnatifida (UP) is one of the most common edible brown algae species, generally found in the Mediterranean sea as well as the Atlantic ocean, and it is known for its high Fx concentrations. In this study, we applied a response surface methodology (RSM) with 3 independent variables: ethanol concentration (0-100%), time (3-25 min), and pressure (2-20 bar) in 28 experimental points. This study aimed to simultaneously optimize the total phenolics content (TPC), total flavonoids content (TFC), extraction yield (EY), 2,2-diphenyl-1-picrylhydrazyl radical scavenging activity (DPPH), and Trolox equivalent antioxidant capacity (TEAC) from UP extracts using microwave-assisted extraction. The optimal extraction conditions were determined at 10.25 ± 0.34 min, 20.00 ± 4.00 bar, and 0% ethanol. These results suggest that water was a more receptive polar solvent for the microwave energy absorption than ethanol, enabling the enhanced extraction of bioactive compounds with antioxidant activity from UP, by avoiding the use of organic solvents. A final validation step was also performed to verify the correlation between the experimental and predicted values, indicating the suitability of the model employed and the success of RSM as a predictive tool optimizing the extraction conditions under evaluation.

Acknowledgments: The research leading to these results was supported by MICINN supporting the Ramón y Cajal grant for M.A. Prieto (RYC-2017-22891) and the FPU grant for A. Carreira-Casais (FPU2016/06135); by Xunta de Galicia for supporting the pre-doctoral grant of P. García-Oliveira (ED481A-2019/295), the program Grupos de Referencia Competitiva that supports the work of J. Echave and M. Barral-Martinez (GRUPO AA1-GRC 2018) and the program EXCELENCIA-ED431F 2020/12; by the Bio Based Industries Joint Undertaking (JU) under grant agreement No 888003 UP4HEALTH Project (H2020-BBI-JTI-2019) that supports the work of P. Otero, P. Garcia-Perez and C. Lourenço-Lopes and by the Ibero-American Program on Science and Technology (CYTED—AQUA-CIBUS, P317RT0003). The JU receives support from the European Union's Horizon 2020 research and innovation program and the Bio Based Industries Consortium. The project SYSTEMIC Knowledge hub on Nutrition and Food Security, has received funding from national research funding parties in Belgium (FWO), France (INRA), Germany (BLE), Italy (MIPAAF), Latvia (IZM), Norway (RCN), Portugal (FCT), and Spain (AEI) in a joint action of JPI HDHL, JPI-OCEANS and FACCE-JPI launched in 2019 under the ERA-NET ERA-HDHL (n° 696295). The authors are grateful to AlgaMar enterprise (www.algamar.com) for the collaboration and algae material provision.

Poster

CRISPR-Cas9-mediated ALMS1 knockout inhibits TGF-β signalling and epithelial-mesenchyme transition

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ALMS1 is an ubiquitous gene associated with Alström syndrome (ALMS). The main symptoms of ALMS affects multiple organs and tissues, generating at last, multi-organic fibrosis in the lungs, kidneys and liver. TGF-β is one of the main pathways implicated in fibrosis, controlling the cell cycle, apoptosis and epithelial-mesenchymal transition (EMT). Nevertheless, the role of ALMS1 gene in fibrosis generation and epithelial-mesenchymal transition via the TGF- β pathway has not been elucidated yet.

Initially, we evaluated how depletion of ALMS1 affects different processes like apoptosis, cell cycle arrest and mitochondrial activity. Then we performed proteomic profiling with TGF-β stimuli and validated the results examining different EMT biomarkers by qPCR.

Depletion of ALMS1 generated apoptosis resistance to thapsigargin (THAP) and C2-Ceramide (C2-C) and cell cycle arrest in HeLa cells. For mitochondrial activity, results did not show significant differences between ALMS1 +/+ and ALMS1 +/+. Proteomic results showed an inhibition of downstream pathways regulated by the TGF-β. The protein coding genes (PCG) were associated with processes like focal adhesion or cell-substrate adherens junction. Finally, EMT biomarkers like VIM, DSP, EDIL3 and SNAI1 shown an opposite pattern to what would be expected when activating the EMT.

ALMS1 has a role controlling the cell cycle and the apoptosis processes. Moreover, the depletion of ALMS1 inhibits the signals transduction through the TGF -β and others routes regulated downstream by it, such as the EMT.

Poster

Temperature modulates the behaviour of intracardiac neurons through TREK channels

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It is widely known that the activity of the heart is mainly determined by the intrinsic activity of cardiac pacemakers and is also strongly modulated by the sympathetic and parasympathetic branches of the autonomic nervous system. In contrast to the extensive knowledge of the general mechanism controlling cardiac activity, very little is known about the electrical properties and functioning of the parasympathetic neurons of the intracardiac ganglion (ICG).

We have recently discovered that mouse ICG neurons express two-pore domain potassium channels (K2P) of the TREK subfamily, channels characterized by their great sensitivity to changes in temperature. In this work we investigated the effect of temperature on the behaviour of ICG neurons and TREK channels by applying patch-clamp electrophysiological techniques (perforated-patch) on cultured ICG neurons.

In “current-clamp” experiments, the excitability of ICG neurons was clearly reduced when the temperature was increased from 24 to 37 °C, neurons resulted hyperpolarized, action potential firing rate decreased and some action potential characteristics were also affected. Consistently, in “voltage-clamp” the increase in temperature induced an outward current and an increase in potassium conductance. This current shows properties similar to the current through TREK-type channels. Our results show for the first time that temperature could modulate cardiac activity through TREK-like potassium channels.

Poster

Spontaneous A-Site interchange: From double to triple cation Perovskite Nanocrystals with tunable Optical properties

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Post-synthetic halide exchange in colloidal perovskite nanocrystals (NCs) and bulk thin films have been greatly exploited to fine-tuning their optical and electronic properties[1]. However, cation exchange in these systems has been relatively less investigated, and few existing studies showed that the cation exchange is much slower compared to halide exchange[2]. Herein, we report fast and controllable interparticle A-site cation exchange in perovskite NCs made of different cations (Cs, FA, and MA) at room temperature. Surprisingly, we find that this cation exchange is even slightly faster than the halide exchange. It is also found that the cation exchange rate increases with increasing the concentration of colloidal NCs and also depends on the solvent type. More importantly, we demonstrate the preparation of bi (MACs, MAFA, CsFA)- and triple (MACsFA)-cation perovskite NCs with an optical bandgap precisely tunable by their composition. In fact, this is the first demonstration of triple-cation perovskite NCs. Furthermore, we demonstrate the preparation of triple-cation mixed halide perovskite NCs with fine-tuning their emission across the visible spectrum of light. We strongly believe that these findings will have implications not only in the exploration of fundamental properties but also device application of mixed cation perovskite NCs in the future.

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2nd July

Invited Speakers

Controlling local temperature at the magnetic nanoparticle surface for improving bio and catalytic efficiency

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Magnetic hyperthermia emerged as a valuable alternative for some kind of cancers difficult to remove surgically, such as some brain tumours, but the number of approved products and their clinical performance are still modest [1]. Some of the main drawbacks that this therapy faces are the lack of effective accumulation of nanoparticles at the tumour site after intravenous injection, the alteration of the heating due to nanoparticle agglomeration in lysosomes after cell uptake and the uncertainty on the temperature at the nanoscale.

Several strategies will be proposed in this talk about the development of more efficient magnetic nanoagents able to overcome these difficulties by maximizing the heating at low dose (Fig. 1), and/or on the design of chemical modification of the nanoagent surface to increase its retention within the tumour extracellular matrix, binding them to the cell membrane or increasing the cell internalization [2]. Finally, the progress on controlling the temperature at the nanoscale on the nanoparticle surface allows to expand the use of these nanoparticles in other applications such as nanoheaters and enzyme supporters to achieve simultaneous or sequential control of multienzymatic bioprocesses of industrial interest [3].

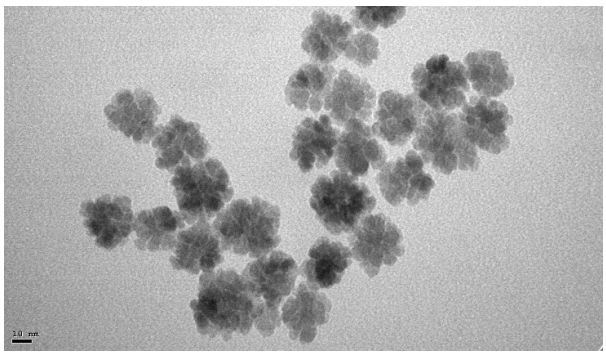

Fig. 1. Flower-like magnetic iron oxide nanoparticles

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Spectrally selective metasurfaces for molecular biospectroscopy and sensing

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All-dielectric resonators have emerged as a powerful toolkit for controlling light on the nanoscale, providing significant advantages over their plasmonic counterparts. In an effort to push all-dielectric nanophotonics even further, new physical concepts based bound states in the continuum (BICs) have recently been introduced to realize optical metasurfaces with unprecedented spectral control over the nanophotonic enhancement.

In my talk, I will introduce several examples of such spectrally selective metasurfaces and highlight how they can provide new insights for bio-nanophotonics, covering a wide range of applications from lipid membrane and vesicular processes to the detection of antibodies and biomarkers for disease.

Oral communications

Oral communication (O10)

Controlled plasmonic coupling for advanced photocatalysis

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Plasmonic photosensitization of large bandgap semiconductors such as TiO2 can be substantially enhanced in those geometries in which high electromagnetic fields are created at the metal-semiconductor interface. In the particular case of Au nanorods (NRs) such enhancement can be obtained thanks to the specific interparticle coupling between neighbor NRs in a tip-to-tip configuration.

Herein, we introduce the use of titanate nanowires (TiNWs) and poly(N-isopropylacrylamide) (pNIPAM) as substrates for the assembly of Au NRs and TiO2 nanoparticles. The results obtained with the proposed hybrid materials illustrate that the rational assembly of different components at the nanoscale is a characteristic of paramount importance towards the development of broadband photocatalysts with enhanced activities.

Oral communication (O11)

Selective and Ultransensitive SERS Sensor based on Plasmonic Supercrystals

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Surface-enhanced Raman spectroscopy, SERS, is an advanced analytical technique that can be used for the ultrasensible detection of analytes. It is based in the enhancement of the Raman signal of a molecule at the surface of a plasmonic nanostructure mainly due to the presence of strong electromagnetic fields generated after the plasmon excitation. Moreover, this effect could be more intense in the case of plasmonic supercrystals due to antenna effects as demonstrated by recent simulations.

As we previously reported, microfluidic platforms allow generating a highly-ordered assembly of uniform gold nanoparticles inside their microchannels through the slow pervaporation of the solvent. While plasmonic supercrystals made by drop-casting show poor uniformity that limits their potential plasmonic applications. Microfluidic approach enables the fabrication of supercrystals of any dimension or morphology. Furthermore, the integration of a plasmonic supercrystal inside microfluidic platform guarantees the infiltration of the desired analyte, even without affinity for gold surface, within the plasmonic supercrystals and therefore its ultrasensitive detection.

Herein, we show the fabrication and characterization of plasmonic supercrystals using gold octahedra nanoparticles synthesized through a wet chemical method. Besides, the study of the sensing capabilities of these platforms showed a highly uniform and intense SERS activity being both key parameters to achieve quantitative and ultrasensitive analysis (LOD). In fact, LOD as low as 10-19 M was achieved for the detection of Crystal Violet (CV), which is several orders of magnitude lower than those found in the literature. Also, we investigate the possibility of combining the capabilities of our SERS sensor with the chromatographic properties of silica nanoparticles to develop a sensor device with the ability of separate analytes by charge.

Oral communication (O12)

Scaling on transport properties of nanofluids

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The increasing worldwide demand of thermal energy storage (TES) applications at low temperature, the so-called cold TES, fosters the development of new technological solutions based on the most recent scientific advances in the field. In this context, the use of nanofluids can enhance the efficiency of TES systems leading to eco-friendlier and less cost-effective thermal devices. MgO/n-tetradecane nanofluids are designed in this work to be used in cold TES, a thorough characterization being performed in terms of stability analysis and experimental determination of their thermophysical properties (thermal conductivity, viscosity, density, isobaric heat capacity and thermal diffusivity). Furthermore, a useful relationship on thermal conductivity and viscosity of nanofluids is proposed based on previous works by Andrade, Osida and Mohanty. The reliability of this proposed relationship is checked using the new results reported in this work and also experimental data for TiO2/H2O, Ag/PEG400, and Al2O3/([C2mim][CH3SO3]:H2O) nanofluids sets available in the literature.

ALEHOOP Project: Biorefineries for the valorisation of macroalgal residual biomass and legume processing by-products to obtain new protein value chains for high-value food and feed applications

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Population growth and socio-economic changes such as increased incomes, urbanisation and aging population is leading to changes in consumption patterns, in particular showing an increasing demand for proteins. Almost 60% of protein eaten worldwide comes from plant sources such as soy, cereal and potatoes, while the rest come from eggs, meat, fish and dairy. Current sources of proteins are becoming unsustainable from a growing health concern, economic and environmental perspective for Europe resulting in concerns for sustainability and food security leading to search for new alternative proteins. The EU ALEHOOP project focuses on the remains resulting from the processing of vegetal origin materials (legumes) and residual macroalgae as a source to obtain demanded proteins in an eco-friendlier way. This project intends to demonstrate at pilot scale the feasibility of macroalgae and legume-based biorefineries for the recovery of low-cost biofunctional and technological proteins. ALEHOOP will last 48 months, receiving funding from the European Union's Horizon H2020 research and innovation programme (call H2020-BBI-JTI-2019) and involves sixteen partners out of six different countries from academia, industries and research & technology centres. The raw materials will be sourced locally to reduce imports of currently used raw materials. Green and brown macroalgae will be collected in Rias Baixas (Spain) and Norway, respectively. By products generated after the processing of legumes will also be harvested in Spain. The University of Vigo will be involved in the physical-chemical characterization of both extracted proteins from legumes and seaweeds (purity, amino acid composition, presence of antinutritional factors, etc). Furthermore, technological properties such as solubility, emulsification, foamability and gelation will be tested. Biological indices of the protein extract will be determined following in vivo metabolic and behavior studies in rats. For that purpose, variables such as body composition, basic haematological parameters, protein anabolism, hormone profile along with others will be assessed.

Oral communication (O14)

Chiral plasmonic photocatalysts

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Metal nanoparticles support the excitation of plasmonic modes, collective oscillations of the electrons in their conduction band. These resonant modes couple strongly with incoming light, at wavelengths that we can control with the shape and size of the nanoparticles. This can be generalized to the creation of chiral resonators that respond preferentially to a given polarization of circularly polarized light (CPL), be it by creating single nanocrystals with chiral geometries or by arranging achiral nanocrystals in chiral configurations. This talk will present theoretical work exploring the usage of chiral plasmonic resonators in the context of photocatalysis. Plasmonic nanocrystals are being extensively studied as photocatalysts, often as photosensitizers in heterogeneous photocatalysis with wide bandgap semiconductors, but also as direct promoters of chemical reactions. They are useful in this context because their resonant modes localize effectively light's energy and provide several physical channels to share it with neighboring materials. Notably, they can excite electrons and holes up to the total energy of the incoming photons, so that they are able abandon the metal and contribute to chemical reactions on their environment. We will explore several chiral configurations exploiting this mechanism in a manner that is sensitive to the circular polarization of impinging light, as well as scenarios where we can use the photocatalytic mechanism to preferentially modify the geometry of left-handed or right-handed chiral complexes through the photocatalytic crystal growth of the metal.

Oral communication (O15)

Electroactive polypyrrole/linseed oil nanocapsules for drug delivery applications

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Stimuli-responsive capsules are effective signal-triggered carriers, useful in different areas, from self-healing smart materials (1) to controlled and targeted drug delivery (2) systems. To form such a capsule, the shell must be composed of a stimuli-responsive material, as a conducting polymer. Polypyrrole, for instance, can change its conductivity and volume depending on its oxidation state (3), property that can be explored in a capsule to control the shell porosity and allow controlled release of an active from the capsule core.

Among the many available techniques to form capsules, the interfacial polymerization is an interesting approach, since emulsion droplets act as templates for shell formation, and the surfactant and emulsification technique play a key role on shell size and morphology.

In the present work, pyrrole was chemically synthesized at the interface of an oil-water emulsion droplet, using ammonium persulfate and iron chloride as oxidizing agents, and the influence of their concentration on the shell thickness was evaluated. TEM and DLS data show that by increasing the concentration of oxidizing agent the capsule size increases, mainly due to the formation of thicker shells, with a greater amount of polymer formed at the interface. The next steps include the evaluation of these capsules as a controlled drug delivery system.

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Oral communication (O16)

Manufacture of Magnetic Spiky Structures

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Three-dimensional (3D) hierarchical nanostructured materials exhibit highly advanced properties compared to their bulk counterparts thanks to their controllable morphology, orientation, and dimensionality. Among them, rough and spiky magnetic structures attract a lot of attention and they are preferred in many applications (since their effective interaction with bacteria and cells in biological systems, imaging ability and environmental applications) thanks to their magnetic properties, surface properties and morphology. In this study, we have synthesized magnetic spiky nanostructures by growing akaganeite (β-FeOOH) nanospindles on polystyrene (PS) beads. These akaganeite elongated nanospindles were formed taking advantage of an easy hydrothermal process in very moderate conditions, resulting in highly spiky and stable nanocomposites. Finally, the oxohydroxy material was reduced using hydrazine (N_2H_4) to obtain the magnetite phase. The investigations were performed to understand the effect of synthesis conditions on the final structure of the particles.

Oral communication (O17)

Bacterial surface display for the expression and engineering of functional glycan-binding proteins

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Glycans are cell-surface carbohydrates present on glycoproteins that are involved in a wide range of key biological processes including cell−cell adhesion, cell communication, development, inflammation, pathogen recognition and malignant transformation. Recombinant lectins, defined as glycan-binding proteins, can be used as reagents to evaluate the expression of glycans for a variety of basic research and clinical applications including cancer diagnosis and glycome profiling.

However, most readily available recombinant lectins present inherent problems including inconsistent activity, broad specificity, or poor affinity for their target carbohydrates. The development of alternative platforms for the expression and engineering of lectins is important to overcome current limitations. Bacterial surface display has become an increasingly useful tool for protein engineering and selection, which has never been applied to non-bacterial lectins.

Here we report on the use of the intimin bacterial surface display system as an efficient and robust method for the expression of functional glycan-binding proteins on the surface of Escherichia coli. We show that the intimin system efficiently translocates functional lectins belonging to the human galectin-3 and mannose-binding lectin subfamilies enabling bacterial adhesion to surfaces coated with different carbohydrates and glycoproteins.

Our study, establishes for the first time the application of bacterial display as a suitable platform for lectin expression, thereby increasing the current toolbox for glycomics and glycobiology studies.

Poster session

Poster

Functional development of the lamprey system, a reflect of its evolution

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Even though lampreys belong to the oldest group of extant vertebrates, it has been shown that many of their neural mechanisms have been preserved throughout the evolution, being present also in humans, as in the case of the visual system. The visual system of the lamprey develops stepwise during its larval period, in which the extraocular muscles are formed, the retina expands and the connections that form the visual maps are established in the optic tectum, a structure present in all vertebrates that controls where to direct gaze. This development is believed to reflect the evolution of the vertebrate visual system, allowing the isolation of mechanisms common to all vertebrates. Besides, the in vitro survivability of the lamprey nervous system is astonishing, and it is relatively easy to expose its central nervous system and keep it alive together with sensory organs for in-depth study of sensorimotor processes. For all these reasons, this animal has been chosen to carry out the present work, focused on the study of the functional development of the visual system.

Lamprey larvae present immature eyes covered with skin. However, some of the neural circuits involved in stabilizing the eyes - mediated by the vestibulo-ocular (VOR) and the optokinetic reflexes (OKR) - have been described. Stabilizing eye movements compensate head movements to clamp the visual scene and avoid image blurring. In the present study, we investigate when in development these circuits appear, and when they become functional. In addition, we analyse these circuits in adult lampreys to understand their functioning when the visual system is completely formed. To achieve this, we combine anatomical techniques (tracer injections, immunohistochemistry) to study the connectivity of neuronal circuits, with electrophysiological recordings and eye tracking to analyse their functionality. Besides, we have established an innovative experimental setup, with a platform specifically designed to apply visuovestibular stimuli to an in vitro preparation isolating the brain with the eyes and vestibular organs, while allowing coordinated electrophysiological recordings. Studying the development of eye movements provides relevant information to understand their evolution underlying neural mechanisms, with implications for all vertebrates.

Poster

Liquid biopsy for the early detection of the serrated pathway of colorectal carcinogenesis

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Colorectal cancer (CRC) is a very heterogeneous disease arising from multiple pathways. Epigenetic modifications define the CpG island methylator phenotype (CIMP), highly associated with the serrated neoplasia pathway, accounting for 15-30% of CRC. Advanced serrated lesions (ASL), such as traditional serrated adenomas (TSA), sessile serrated lesions (SSL), and large hyperplastic polyps (HP) seem to be precursor lesions of CRC developed via the serrated pathway. The subtle morphology of SSL makes them likely to be missed during colonoscopy and more prone to incomplete resection. The most common non-invasive test for CRC screening, the fecal immunochemical test, has no sensitivity for the detection of ASL. In this study, we performed an epigenome-wide analysis of serum cfDNA pools to explore the methylation signatures in patients with serrated lesions, with the aim to propose non-invasive biomarkers suitable for the detection of advanced serrated lesions that could lead to CRC.

We extracted cfDNA from serum samples from 110 individuals (30 with no colorectal findings, 30 non-advanced serrated lesions (HP and serrated polyps <10mm without dysplasia), 20 HP ≥10mm, and 30 ASL (serrated polyps with dysplasia or ≥10mm, and TSA). Pooled samples were prepared for each group using equal amounts of cfDNA from 10 individuals, sex- and age-matched. DNA methylation levels of 866,091 epigenome-wide CpG positions were measured with MethylationEPIC array.

We found a differentially methylated signature that can discriminate advanced stages of the serrated pathway (HP >10mm, TSA and serrated polyps >10mm) from individuals without colorectal findings or with non-advanced serrated polyps. We found 27 differentially methylated regions and 13,578 differentially methylated positions, of which 330 have at least 10% difference in the methylation levels. Some of the differentially hypermethylated positions are located within CpG islands of genes proposed as CIMP panel, such as MLH1, MINT1, CACNA1G, NEUROG1 and IGF2. We selected 5 differentially methylated regions as candidate methylation biomarkers.

Each candidate region was individually validated using MS-qPCR in individual cfDNA serum and tumor/mucosa tissue samples, to determine their utility for diagnosis.

Such differentially methylation regions are a source of potential non-invasive biomarkers that could be implemented in screening programs to improve the rate of detection of serrated lesions and reduce the incidence of serrated CRC.

Funding: AES-ISCIII FEDER (PI15/02007), Centro Singular de Investigación de Galicia (ED431G2019/06, Xunta de Galicia-FEDER), predoctoral fellowship (FPU15/02350).

Poster

Alterations in Event-Related Potentials in patients with Substance Use Disorder

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Substance use disorder is a pathology that affects the brain and alters its functioning, making the individual unable to control the consumption of the substance and disturbing their cognitive functions (attention, memory, etc.). In Spain, cocaine is the second most widely used illegal drug (2.8%), and its long-term use is associated with memory and learning problems. The aim of our study was to investigate auditory attentional processing through N2 and P3 ERPs in substance use disorder, since these components are considered a neural reflection of attentional processing.

Evoked potentials (EPs) allow us to study the electrical brain activity associated with these cognitive processes, analyzing possible deficits in mental function.

We recorded N2 and P3 by means of an auditory task, in which the subjects had to discriminate and mentally count the different beeps within a series of identical stimuli.

We found alterations in both components, for both latency and amplitude, in patients when compared to the control group.

The increase in N2 latency and the decrease in P3 amplitude point to a strong alteration in the discrimination and processing of auditory information in substance use disorders, which point to the potential of ERPs as neurophysiological markers of attentional decline in this pathology. ERPs could help as reliable markers to predict the evolution of impairment attentional in psychiatry.

Poster

Effects of 5-methylisatin, an inhibitor of MAO-B, and of harmine, an inhibitor of MAO-A, on the in vivo release of dopamine in the striatum of rats

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The use of monoamine oxidase (MAO) inhibitors, responsible for the degradation of dopamine, is one of the strategies for the treatment of Parkinson's disease. Here we evaluate the effects of administration of different concentrations of an isatin derivate, 5-methylisatin, a potential MAO-B inhibitor, and compare them with the effects of harmine, another indole that inhibits MAO-A. The substances were administered locally into the striatal nucleus for 60 minutes via a microdyalisis probe, and the in vivo release of dopamine and its main metabolites (DOPAC and HVA) was analyzed by HPLC-EC, with the animals awake and freely moving. Adult female Sprague-Dawley rats (250-350 g, 4-6/group) were used in all experiments, maintained under standard animal house conditions. Statistical analysis of the data was performed by ANOVA/Student-Newman-Keuls test. Intrastriatal administration of 1 mM 5-methylisatin produced no significant effects on dopamine, DOPAC or HVA levels. The infusion of 5 or 10 mM increased dopamine release up to 348.3±81% and 783.4±110%, relative to baseline levels, respectively. The infusion of 5 mM 5-methyllysatin lowered DOPAC levels to 80.4±10.5%. The infusion of 1, 5, or 10 mM of harmine also increased striatal dopamine levels to 2421±434.6%, 1782±512.7%, and 1148±114%, compared to baseline, respectively. All three harmine concentrations produced significant decreases in DOPAC or HVA levels. These data indicate that both 5-methylisatin and harmine significantly increased the in vivo release of dopamine in a concentration-dependent manner. Harmine exerted a significantly greater effect than 5-methylisatin, whose effect did not appear to be concentration-dependent, which is probably related to its inhibitory action on the MAO-A isoform.

Poster

Environmental-friendly extraction of hops antioxidant compounds

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Nowadays, the use of hops is not limited to its traditional role within the brewery industry. The growing community interest in natural bioactive compounds makes the scientific community look for new attractive sources as hops. The presence of phenolic compounds contributes to the antimicrobial, antifungal or antioxidant properties of the hops, which leads to an increased interest in expanding their knowledge and research on new applications in non-traditional industries and environments. In this context, the aim of this work was to study the content of antioxidant features of industrial hops.

Two industrial hops variety were kindly provided by Hijos de Rivera S.A. (A Coruña, Spain). Fundamental physicochemical and phytochemical analysis were carefully performed. An environmentally friendly extraction technology was selected to extract high value compounds considering their advantages. In this context, ultrasound assisted extraction was the extraction process to recover bioactive components using water as extraction solvent with a solid:liquid ratio 1:15 (w/w). According to antioxidant compounds, the content in total phenolics and antioxidant capacity were determined at least in triplicate by spectrophotometric techniques.

The results indicated that the total phenolic content for the industrial hops were 2,27±0,06 and 1,33±0,01 g GAE/100 g extract, respectively. The values obtained are consistent with other authors using this raw material. Overall, this information could be relevant from the point of view of future studies on innovative applications to an integral valorisation of industrial disposals.

Poster

Revalorization of industrial fruit waste by recovery of bioactive extracts with Green Technology extraction

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In industrial fruits processing, approximately 30% of the raw material is discarded. The need to revalue these waste and/or by-products arises from the interest of the scientific community for the phytochemical compounds present in the fruits and their potential applicability so much in the food industry like cosmetic or pharmaceutical field. These wastes (seeds, peel, and pulp leftovers) without commercial value, hold a significant amount of antioxidant substances, such as phenolic compounds, whose antioxidant capacity is related with the prevention of cardiovascular illnesses. Therefore, the aim of this study is the physicochemical and phytochemical analysis of industrial fruits waste by means of an environmentally friendly extraction.

The extraction technique was selected based on resource optimization criteria, using a non-polluting solvent (distilled water). For this reason, an ultrasound probe was used, operating at a fixed frequency (0.5 Hz) and a solid: liquid ratio (1:15 w/w). The effect of the amplitude (20-50-80%), time (4-8-12 min) and temperature (30-50-70ºC), was studied using an Experiments Design to establish the optimal conditions. The liquid extracts were analysed at least in triplicate by spectrophotometric techniques to determinate the content in total phenols and antioxidant capacity.

The obtained results indicated that the total phenolic content and the antioxidant capacity, varied between 88.93 –138.72 mg GAE/g extract and between 40.79 -54.14 mg Trolox/g extract, respectively. These values show that the industrial waste fruits would be an attractive source of bioactive compounds and that they are valuable raw materials for the generation of high-value products, suitable for food and non-food fields.

Poster

Evaluation of the effects of individual and combined administration of psychoactive substances on in vivo dopamine release from nucleus accumbens and striatum of rats.

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Addiction is defined by the World Health Organization (WHO) as a physical and psycho-emotional illness that creates a dependency or need for a substance, activity, or relationship. Addictive behavior is mediated by the brain reward system which mediates conditioning responses to stimuli and comprises several structures of the CNS, including the nucleus accumbens (NAc) and striatum. In this system, the most important neurotransmitter is the dopamine. Currently, one of the serious problems associated with the phenomenon of addiction is the issue of polyaddictions, which greatly complicates its monitoring and, above all, its treatment. It is common to see associations of addictions to substances, such as nicotine, caffeine, and alcohol. The objective of this study is to evaluate the neurochemical effects of the individual and combined administration of these three substances on the in vivo release of dopamine in the NAc and striatum of rats. Nicotine (5 mM), caffeine (5 mM), and alcohol (20%) were administered locally into NAc and the striatum individually or in combination. Its effects on the release of dopamine were evaluated using brain microdialysis technique associated with HPLC-EC. In all experiments, female Sprague-Dawley rats (250-350 g, n = 4-8/group) housed under standard conditions were used. The data were statistically analyzed using the ANOVA/Student-Newman-Keuls test. The results obtained indicate that the intrastriatal administration of nicotine, caffeine, and ethanol, administered individually or in combination, produced significant increases in the extracellular levels of dopamine both in the striatum and NAc. We also observed that the co-administration of the three substances produced significantly greater increases than the individual administration, which characterizes an additive effect produced with the combined infusion of the three substances in the NAc and striatum of rats.

Poster

“Rational design of high efficiency SERS TAGs for detection by immunoassay and bioimaging”

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SERS nanotags have been widely used in different bio-related applications such as disease diagnosis or bioimaging. The ideal SERS tag must offer a number of requirements such as high sensitivity, uniform and reproducible SERS response, targeting ability, biocompatibility andstability in a long-term period. Particularly, SERS tags have been demonstrated as a promisingtool for the development of ultrasensitive SERS-based lateral flow immunoassays (LFIA).

Herein, we propose a SERS-based LFIA for the detection of a tumoral biomarker, HER 2, related with breast cancer. SERS-based LFIA will allow us to overcome some drawbacks of traditional colorimetric LFIA such as low sensivity and/or limit of detection/quantification. A sandwich format will be performed employing nanobodies as conjugate antibodies. SERS nanotags will consist on plasmonics nanoparticles based on gold silver core-shell nanorods (Au@Ag NRs), that will be encapsulated within zeolitic imidazolate frameworks (ZIFs) and codified with different dyes acting as Raman reporter molecules. The ZIF coating is topologically isomorphic with zeolites and present high chemical stability, being constructed from tetrahedral metal cations (Zn2+,Co2+) bridged by imidazolate ligands. ZIF-8 based on Zn2+ cations has been chosen since it will favour the bioconjugation step through its coordination with histidine-tagged nanobodies. After the synthesis and characterization of the Au@Ag@ZIF nanotags, their bioconjugation with his-tagged antibodies will be tested. Finally, the LFIA test will be developed in order to obtain a calibration curve and the limit of detection and quantification for the biomarker.

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Poster

Role of microRNAs in Pulmonary Arterial Hypertension, functional and biomarker analysis

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Pulmonary arterial hypertension (PAH) is a vascular disease characterized by proliferative remodeling of the precapillary pulmonary arteries, leading to an increase in pulmonary arterial pressure, right heart failure, and ultimately death. The pathogenesis of PAH is multifactorial and includes the dysregulation of micro-RNAs (miRNAs). Since the diagnosis of this disease currently uses invasive and expensive methods, we propose the use of circulating miRNAs as biomarkers. The expression of 13 candidate miRNAs (based on previous transcriptomic results) was analyzed in a cohort of 50 patients and 50 controls for prevalidation. RNA was extracted from plasma samples and miRNA levels were quantified by RT-qPCR using TaqMan probes. Only 7 of the candidate miRNAs amplified by this technique: let-7a-5p, let-7b-5p, let-7c-5p, and let-7f-5p showed lower levels in patients than in controls; while miR-9-5p, miR-31-5p, and miR-3168 were overexpressed in patients. Finally, using logistic regression, we generated a simplified classification model using 3 of the candidate miRNAs (let-7a, miR-9, and miR-31) with an AUC of 0.878. The results suggest that a panel based on these three miRNAs could be useful for the diagnosis of PAH.

Poster

Orthologous genes mining and evaluation of cell migration capacity in Alström syndrome

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Alström syndrome is a rare genetic disorder caused by mutations in the ALMS1 gene and whose symptoms mainly include visual problems, obesity, diabetes and multiple organ fibrosis. This disease is included in the ciliopathies group, since in it there is a dysfunction of primary cilia.

Initially, a comparative analysis was carried out between fibroblasts from patients and a zefrafish model for this syndrome, in order to find common genes that could play a relevant role in the disease development. The expression of various epithelium-mesenchymal transition markers was also determined by qPCR in an ALMS1 KO cell line and in a control line, after 24 and 48 hours TGF-β stimulation. Finally, to compare the migration capacity in the two previous cell lines, a wound healing assay was made.

The comparative analysis results showed 4 possible genes related to the disease (MFAP4, IL7R, LGALS3BP and SERPINH1) and statistically significant differences in expression between the control and KO lines for the epithelium-mesenchymal transition markers. EDIL3, ACTA2 and POSTN markers were the most relevant, as they showed differences at 24 and 48 hours.

Regarding the migration capacity, no statistically significant differences were found between the two cell lines.

Poster

Antiproliferative effect of the extracts obtained from Himanthalia elongata and their ability to produce polymeric nanoparticles

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Brown seaweeds have great potential for biomedical applications due to the exhibited variety of components. The most important polymer present in brown seaweeds is fucoidan, which is mainly comprised of fucose units and sulphate groups. Its structure and the position of the sulphate groups have been associated to the inherent biological activities. Antioxidant, antiproliferative, anticoagulant, anti-inflammatory and antiangiogenic have been the most relevant activities reported in the literature. Extraction technologies and conditions influence fucoidan properties, but other abiotic factors may also affect its composition. Apart from the intrinsic biological properties, fucoidan structure has also been supporting its application in the formulation of nanoparticles by polyelectrolyte complexation. This technique relies on the complexation of two oppositely charged polymers, mediated by electrostatic interactions. In this context, the aim of this work was to evaluate the antiproliferative effect of the extracts obtained from the edible brown seaweed Himanthalia elongata by subcritical water extraction (SWE), and to study their ability to produce polymeric nanoparticles, assessing their size and surface charge.

Dried Himanthalia elongata alga was purchased to Algamar (Pontevedra, Spain). The extraction was performed in a stainless-steel reactor with stirring at 120, 160 and 200 °C using a solid:liquid ratio of 1:30 (w/w). The extracted liquid phases were tested regarding the antiproliferative effect in alveolar carcinogenic cells (A549) by the MTT assay, evaluating exposure times of 24 and 48 h. Additionally, the extracts were assessed regarding their ability to formulate polymeric nanoparticles using chitosan (CS) as counterion. Different mass ratios of chitosan/Himanthalia elongata extract (4/1, 1/1 and 1/4) were tested.

Himanthalia elongata extracts generally showed an antiproliferative effect dependent on time of exposure and concentration, which was more pronounced for the extract prepared at 220 °C. The smaller nanoparticles (246 nm) were obtained for CS/extract = 1/4 (w/w), using the extract obtained at the aforementioned temperature. The zeta potential reached +35.5 mV. Zeta potential was positive in all tested formulations/conditions, indicating higher charge density of CS comparing with the interacting polymer. Further studies are required to deepen the evaluation of the potential of algae extracts to produce marine-based polymeric nanoparticles with biomedical applications.

Poster

Gold nanoparticles as suppress migration and proliferation agent on radiotherapy treatments applied to DU145 cell line

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Gold nanoparticles (AuNPs) are being extensively used in biomedical fields due to their unique chemical, optical and physical properties. Besides that, AuNPs are ease of synthesis in a variety of size and shapes, are relatively inert, biocompatible and can be functionalized with different (bio)molecule including ligands, diagnostics, or therapeutic agents.1, 2 Over the past two decades, AuNPs have been explored in therapeutic field, mainly in radiotherapy (RT) treatments. AuNPs can be used as radiosensitizer because of high X-ray photon capture cross-section of gold.3

In this study, we evaluated the effect of spherical gold nanoparticles (AuNPsp) with 50nm on DU145 prostate cancer cell line for biocompatible studies. Cells were incubated with different concentrations of AuNPsp (0-1.0 mM) during 24h, then subjected to irradiation of 2,5 Gy per fraction for 3 days using a PRIMUS linear accelerator with 6 MV photon beam. After RT treatments, their viability was analyzed using PrestoBlue assay (Invitrogen™) and migration was explored doing the in vitro wound healing assay.

The results showed that AuNPsp decreased the viability of DU145 cells and consequently improve the radiotherapeutic effect. Also, AuNPsp contributed to suppressed migration of DU145 cells when submitted to RT treatments.

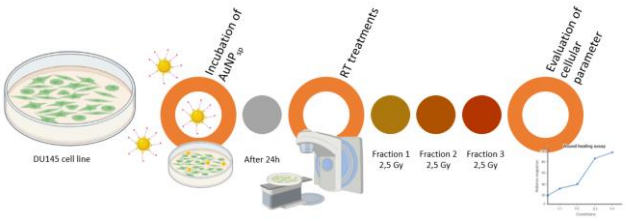

Figure 1: Graphical abstract scheme

Acknowledgements: SS is grateful for the financial support of Fundação para a Ciência e Tecnologia (grant reference: SFRH/BD/138271/2018) and thanks the disposal to all those involved in the radiotherapy service at Centro Hospitalar Universitário de São João for carrying out radiotherapy treatments.

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Poster

Development of an animal model for fish allergy using BALB/c mice

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Food allergy is considered a major concern in food safety. Around 6-8% of children, as well as 2-4% of the adult population suffer from this condition, and prevalence is increasing all over the world, being more prominent in industrialized countries due to a combination of environmental and lifestyle changes. Fish is one of the eight major foods causing type I food allergy, an immunoglobulin E (IgE)-mediated hypersensitivity disease resulting from the breakdown of immune tolerance. The major allergen of fish has been identified as β-parvalbumin (β-PRVB), a Ca2+-binding acidic protein that is abundant in muscle. The allergenic properties of β-PRVBs seem to be related to their resistance to heat and to certain gastrointestinal proteases.

To date, the only proven and effective treatment for this type of hypersensitivity is to consume a diet free of the allergenic food and its derivatives. Significant progress has been made with oral, sublingual, and epicutaneous immunotherapy in the treatment of food allergy; however, these emerging treatment options have important limitations in efficacy, durability of effect, and safety. Further studies are needed to get successful treatments for this complex disease.

We have developed an animal model for fish allergy using BALB/c mice as a previous step for developing a novel oral immunotherapeutic strategy. Mice were sensitized to fish β-PRVBs after intraperitoneal administration of protein in presence of aluminium based adjuvant. A Th2-like immune response was induced with production of PRVB specific IgE and IgG1. This mouse model will be further used in basic biomedical, preclinical and applied food/nutrition research on fish food allergy.

Poster

Biosynthesis of gold nanoparticles by Pseudomonas aeruginosa with therapeutical potential in prostate carcinoma.

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Gold nanoparticles (AuNPs) applications to anticancer research have been gaining great interest due to its excellent biocompatibility, low toxicity and stability. However, some limitations have arisen regarding its synthesis by conventional chemical methods. These limitations are related to the presence of chemical contaminants or its biocompatibility.

Biosynthetic AuNPs (bAuNPs) have been arising as a disruptive alternative method for its production. This study focuses on optimizing different biosynthesis conditions of AuNPs, such as temperature, pH and incubation period using Pseudomonas aeruginosa and how it affects their antitumor effect on prostate cancer cells PC3. bAuNPs were characterized by TEM (Transmission Electron Microscopy) and spectroscopy (spectrophotometry). Cell metabolic activity was determined by MTT.

So far, we conclude that bAuNPs produce a significant effect on cancer cells. Promise results were achieved with the preparation of bAuNPs at 50 ºC for 48h under pH values of 9, presenting the most homogeneous structures with a greater effect on PC3 cell lines.

Poster

Evaluation of bioactive compounds of the plant *Chamaemulum nobile* from the family Asteraceae

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Traditional plants have been used since ancient times for therapeutic purposes. They have bioactivities, which make them usefully candidates to be employed in different formulas such as decoctions, infusions, creams, among others. In the latest years, medicinal plants are also evaluated for the recovery of bioactive compounds with applications in the food, cosmetic and pharmaceutical industries. Specially, the plant Chamaemelum nobile which belong to Asteraceae family, has been traditionally used as a remedy to treat fever, insomnia, back pain, and skin conditions, among others. In this work, an in vitro study of the antioxidant, antimicrobial, cytotoxic and neuroprotective activities of C. nobile was carried out. This plant showed significant antioxidant effects in DPPH and β-carotene assay. In the antimicrobial test, extracts of the plant inhibited the growth of both bacterial and fungi species. Cytotoxic effect against different cancer cells lines were also observed. Finally, the enzymatic assays reported the inhibition of acetylcholinesterase (AChE) and butyrylcholinesterase (BuChE), two enzymes involved in neurodegenerative disorders. These results indicate that this plant may be a source of interesting compounds to the development of new applications in industrial fields.

Acknowledgements

The research leading to these results was supported by MICINN supporting the Ramón y Cajal grant for M.A. Prieto (RYC-2017-22891), by Xunta de Galicia for supporting the program EXCELENCIA-ED431F 2020/12, the pre-doctoral grant of P. Garcia-Oliveira (ED481A-2019/295), the program Grupos de Referencia Competitiva (GRUPO AA1-GRC 2018) that supports the work of M. Barral-Martinez and by EcoChestnut Project (Erasmus+ KA202) that supports the work of B. Nuñez-Estevez. Authors are grateful to Ibero-American Program on Science and Technology (CYTED—AQUA-CIBUS, P317RT0003), to the Bio Based Industries Joint Undertaking (JU) under grant agreement No 888003 UP4HEALTH Project (H2020-BBI-JTI-2019) that supports the work of P. Otero. The JU receives support from the European Union's Horizon 2020 research and innovation program and the Bio Based Industries Consortium. The project SYSTEMIC Knowledge hub on Nutrition and Food Security, has received funding from national research funding parties in Belgium (FWO), France (INRA), Germany (BLE), Italy (MIPAAF), Latvia (IZM), Norway (RCN), Portugal (FCT), and Spain (AEI) in a joint action of JPI HDHL, JPI-OCEANS and FACCE-JPI launched in 2019 under the ERA-NET ERA-HDHL (n° 696295). Authors are also grateful to Foundation for Science and Technology (FCT, Portugal) for financial support through national funds FCT/MCTES to the CIMO (UIDB/00690/2020). L. Barros and R. Calhelha thank the national funding by FCT, P.I., through the institutional and individual scientific employment program-contract for their contracts, respectively. The authors would like to thank the EU and FCT for funding through the project PTDC/OCE-ETA/30240/2017- SilverBrain - From sea to brain: Green neuroprotective extracts for nanoencapsulation and functional food production (POCI-01-0145-FEDER-030240).

Poster

Chemical approach to biomolecules from *Calendula offinalis*

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Medicinal plants have been traditionally used throughout time as therapeutic treatment. These plants possess different compounds with antioxidant, anti-inflammatory and antimicrobial properties of great interest at an industrial level. In particular, the plant Calendula officinalis L, belonging to Asteraceae family, have shown relevant applications including the preparation of foods, dyes, cosmetics, and traditional remedies, although its consumption is currently decreasing. [1]. The aim of this study was to evaluate the development of new products derived from C. officinalis, which may be of interest to the pharmaceutical, nutraceutical, and cosmetic industries. For this reason, after an initial review and a study of its main bioactivities of interest such as antioxidant, antimicrobial and neuroprotective capacity, it has been possible to detect a relevance in the results obtained in the enzymatic assays. For this purpose, after an initial review and a study of its main bioactivities of interest such as antioxidant capacity with ABTS, β-carotene-white and TBARS assays, antimicrobial against bacteria and fungi, cytotoxic in four tumor cell lines (MCF-7, CaCo, AGS, and NCI-H460) and neuroprotective. The results for antioxidant activity indicated that C. officinalis extracts showed a high rate of antimicrobial and antifungal activities with active concentrations ranging from 0.25 to 0.5 mg/mL of extract. The results for anti-inflammatory and cytotoxic activities showed growth inhibitory concentration 50 (GI50) values of 72.6 µg/mL for anti-inflammatory activity, and GI50 values ranging from 310 to >400 µg/mL for cytotoxic activity. The results obtained in the enzyme assays were also significant, showing greater inhibitory effects on two enzymes related to Alzheimer's disease, acetylcholinesterase (AChE) and butyrylcholinesterase (BuChE). Overall, this study provides scientific evidence for the evaluation of the potential of medicinal plant extracts for the development of new products.References

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Acknowledgements: The research leading to these results was supported by MICINN supporting the Ramón y Cajal grant for M.A. Prieto (RYC-2017-22891), by Xunta de Galicia for supporting the program EXCELENCIA-ED431F 2020/12, the pre-doctoral grant of P. Garcia-Oliveira (ED481A-2019/295) and the program Grupos de Referencia Competitiva (GRUPO AA1-GRC 2018) that supports the work of M. Barral-Martínez and by EcoChestnut Project (Erasmus+ KA202) that supports the work of B. Nuñez-Estevez. Authors are grateful to Ibero-American Program on Science and Technology (CYTED—AQUA-CIBUS, P317RT0003), to the Bio Based Industries Joint Undertaking (JU) under grant agreement No 888003 UP4HEALTH Project (H2020-BBI-JTI-2019) that supports the work of P. Otero. The JU receives support from the European Union's Horizon 2020 research and innovation program and the Bio Based Industries Consortium. The project SYSTEMIC Knowledge hub on Nutrition and Food Security, has received funding from national research funding parties in Belgium (FWO), France (INRA), Germany (BLE), Italy (MIPAAF), Latvia (IZM), Norway (RCN), Portugal (FCT), and Spain (AEI) in a joint action of JPI HDHL, JPI-OCEANS and FACCE-JPI launched in 2019 under the ERA-NET ERA-HDHL (n° 696295). The authors would like to thank the EU and FCT for funding through the project PTDC/OCE-ETA/30240/2017- SilverBrain - From sea to brain: Green neuroprotective extracts for nanoencapsulation and functional food production (POCI-01-0145-FEDER-030240). The authors are grateful to the Foundation for Science and Technology (FCT, Portugal) for financial support through national funds FCT/MCTES to CIMO (UIDB/00690/2020); and to the national funding by FCT, P.I., through the institutional scientific employment program-contract for L. Barros contract.

Poster

Single-index mixture cure models

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Survival analysis is a branch of Statistics that studies time until an event of interest occurs. Conventional survival models suppose that if individuals could be followed up for an infinite time, they will all experience the event of interest. However, in many situations it is realistic to suppose that a fraction of the individuals will never experience the event (e.g. cancer studies). In such cases, there will be a cure fraction. In order to take this fraction into account, classical survival analysis has been extended to cure models. Specifically, mixture cure models are used to estimate the probability of cure and the survival function for the uncured subjects (latency). A completely nonparametric method for the estimation of both functions, in the presence of a univariate continuous covariate, was proposed by López-Cheda et al. (2017a, 2017b). In this work, this methodology is extended to the presence of a vector covariate. A single-index model is proposed for dimension reduction in this context. This approach has been studied in the presence of censoring in Strzalkowska & Cao (2013), but not in the presence of cure. In the future, functional covariates will be considered. The methods will be applied to a database of cardiotoxicity from the University Hospital of A Coruña (CHUAC).

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Poster

Development of a Computer-Aided Diagnosis System Based on Deep Learning for Colorectal Polyp Detection and Classification

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In the last years, Deep Learning (DL) has gained a lot of attention in the field of medical image analysis due to its higher performance in image classification when compared to previous state-of-the-art techniques. Moreover, recent meta-analyses found that the diagnostic performance of DL models is equivalent to that of health-care professionals and that systems based on AI can increase both adenoma and polyp detection ratios. In this context, we are working on the PolyDeep project (polydeep.org), where we are developing of a computer-aided diagnosis (CAD) system to assist the endoscopist during the colonoscopy. Such system consists of two DL models, one for polyp detection and another one for polyp classification, developed using a new polyp video and image dataset created in collaboration with the CHUO Hospital (Ourense, Spain). The detection model, based on a pre-trained YOLOv3 architecture and fine-tuned with our own dataset of 13 873 images, achieves an F1 of 0.88 (sensitivity=87%, predictive positive value=89%) on a frame-based evaluation with a test set comprising 8 658 images. To perform a polyp-based evaluation on polyp and non-polyp videos, we classify a video as polyp-positive if it contains at least one segment of 50 frames where at 75% or more of them have been predicted as containing a polyp. In such evaluation, using a test set comprising 628 polyp-videos and 171 non-polyp videos, the model achieved a sensitivity of 71.18% [95% CI: 67.52 – 74.58] and a specificity of 86.55% [95% CI: 80.63 – 90.87]. To develop the classification model, a dataset with 2 755 NBI images from 250 polyps (214 adenomatous and 36 hyperplastic) was created. A preliminary 5-fold cross-validation evaluation of the classification model, based on a pre-trained InceptionV3 architecture fine-tuned with our own dataset, shows an adenoma sensitivity of 93,07% [95% CI: 88.85 – 95.76] and a specificity of 44.28% [95% CI: 29.39 – 60.26].

Poster

Antimicrobial and cytotoxic activity of Amaranthaceae famyly extract: source of new antibiotics and anticancer treatments

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Two of the most prevalent diseases around the globe are currently cancer and resistant bacteria-mediated infections. Despite the numerous advances that have been made in recent years against tumoral diseases and in the discovery of antibiotics, these diseases are still leading causes of worldwide deaths. Therefore, the search for compounds with cytotoxic and antimicrobial activity is increasing in the last years. In this sense, secondary plant metabolites, namely phenolic compounds constitute a promising source of natural bioactive compounds. In this study, three Amaranthaceae species largely used in the traditional Asiatic medicine (Alternanthera sessilis, Dicliptera chinensis and Dysphania ambrosioides), were selected to test the in vitro cytotoxic and antibacterial activities of their ethanolic extracts. The results for antibacterial activity showed low minimal inhibitory concentrations (MIC) in the three species, ranging from 5 to 20 mg/mL in gram-positive (E. faecalis, L. monocytogenes, etc) and gram-negative species (E. coli, K. pneumoniae, etc). In parallel, D. ambrosoides showed cytotoxic activity against three tumoral cell lines (CaCo, MCF-7, NCI-H460), with growth inhibitory concentration 50% (GI50), ranging from 188 ± 14 to 263 ± 12 µg/mL. In a previous study a characterization of the phenolic compounds present in the studied species was performed. In order to find possible correlations between the detected phenolic compounds and the obtained bioactivities, in silico molecular docking studies were performed using AutoDock Vina software. A total of 7 proteins were selected as known protein targets for anti-cancer (COX-1; COX-2 and PPARgamma2) and anti-microbial agents (B-cetoacyl-ACP sintase; Type IIA topoisomerase; Dihydrofolate reductase and DNA gyrase). The discovered phenolic compounds were docked against all 7 proteins. The most potent predicted inhibition activity was obtained for Quercetin-3-O-glucosyl-pentoside-7-O-glucuronide (Alternanthera sessilis) and Luteolin-6-C-hexosyl-8-C-pentosyl (Dysphania ambrosioides), against Type IIA topoisomerase, with predicted Ki values of 0.58 nM and 0.81 nM, respectively. These two compounds may be, at least partially, responsible for the observed antimicrobial activity.

Acknowledgements: The research leading to these results was funded by Xunta de Galicia supporting the Axudas Conecta Peme, the IN852A 2018/58 NeuroFood Project and the program EXCELENCIA-ED431F 2020/12; to Ibero-American Program on Science and Technology (CYTED—AQUA-CIBUS, P317RT0003) and to the Bio Based Industries Joint Undertaking (JU) under grant agreement No 888003 UP4HEALTH Project (H2020-BBI-JTI-2019); by MICINN supporting the Ramón y Cajal grant for M.A. Prieto (RYC-2017-22891); by EcoChestnut Project (Erasmus+ KA202) that supports the work of B. Nuñez-Estevez,. The JU receives support from the European Union's Horizon 2020 research and innovation program and the Bio Based Industries Consortium. The project SYSTEMIC Knowledge hub on Nutrition and Food Security, has received funding from national research funding parties in Belgium (FWO), France (INRA), Germany (BLE), Italy (MIPAAF), Latvia (IZM), Norway (RCN), Portugal (FCT), and Spain (AEI) in a joint action of JPI HDHL, JPI-OCEANS and FACCE-JPI launched in 2019 under the ERA-NET ERA-HDHL (n° 696295). Foundation for Science and Technology (FCT, Portugal) for financial support through national funds FCT/MCTES to the CIMO (UIDB/00690/2020). L. Barros and R. Calhelha thank the national funding by FCT, P.I., through the institutional scientific employment program-contract for their contracts.

Poster

Optimization of phenolic compounds production from Fucus spiralis by microwave-assisted and ultrasound-assisted extraction using Response Surface Methodology (RSM)

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Fucus spiralis (FS) is a brown alga, widely distributed in the Galician coast, which biosynthesizes different secondary metabolites of great interest at an industrial level, such as phenolic compounds and fucoxanthin [1]. Specifically, phenolic compounds present important biological activities, acting mostly as antioxidant agents [2]. This study is aimed at optimizing the conditions of microwave-assisted extraction (MAE) and ultrasound-assisted extraction (UAE) to obtain the maximum yield of phenolic compounds-enriched FS extracts, using response surface methodology (RSM). RSM was performed to decipher the influence of the independent variables involved in MAE (ethanol concentration (0 - 100% v/v E), pressure (2 - 20 bar) and time (3 - 25 min)) and UAE (power (100 - 500 W), time (5 - 55 min) and temperature (35 - 100 ºC), in order to maximize the extraction yield. Our results indicated that MAE was more efficient than UAE in terms of extraction yield. The optimal MAE conditions were reported to be the combination of 36.94% E, 9.11 bar, and 25.00 min, obtaining an extraction yield of 467.62 mg dry extract per g of dry weight. Moreover, for the optimized FS extracts, the determination of total phenolic and flavonoid contents and antioxidant capacity, by means of 2,2-diphenyl-1-picryl-hydrazyl (DPPH), and 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) radical scavenging capacity were carried out. The results showed that FS extracts present a total polyphenol content of 61.2 mg phloroglucinol equivalents per g of dry weight, and a total flavonoid content of 14.2 mg quercetin equivalents per g of dry weight. Additionally, the results for antioxidant capacity showed inhibitory concentration 50 (IC50) values of 58.37 nM for DPPH and 102.92 nM of Trolox Equivalents per g of dry mass for ABTS. Overall, this work provides evidence about the efficiency of sustainable extraction procedures, such as MAE and UAE, for the production of bioactive compounds from natural sources, as it is the case of algae.

Poster

A possible mathematical model to solve the quantification procedures used in Single Electron Transfer antioxidant assays

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The development of a convenient mathematical application for testing the antioxidant potential of standard and novel therapeutic agents is essential for the research community to perform evaluations in a more precise way. Despite the abundance of procedures to describe the antioxidant effects in Single Electron Transfer (SET) assays, based in theoretical standpoints, they are somehow inadequate. Some of the causes hindering the advances in a field which is per se very complex are due to the lack of: (1) unambiguous experimental design, as a result of the extended use of simplistic procedures to quantify the effects of joint responses, based in single-dose and single-time values; and (2) detailed mathematical hypotheses to quantify the kinetic dose-response values, which in addition causes difficulties for assessing the statistical consistence of the results. This communication aims to illustrate a mathematical procedure for SET antioxidant assays to adequately quantify the capacity of antioxidants.

The model was developed by studying the kinetic dose-response behaviour of butyl-hydroxytoluene (BHT) (0.0-600.0 μM) as the reference antioxidant and evaluating the results of the DPPH bleaching reaction by two approaches: (A): kinetic dose-response of BHT; (B): non-linear dose-response of BHT at the fixed points selected in the previous study (t = 5, 30 min, 90 min). Then, the inhibitory concentration, IC50 was computed for each respective fixed time and compared with the computed kinetic IC50 value for all asymptotic values found by the proposed model. Although the approach was experimentally demonstrated just in one classical SET assay (DPPH), the results can be directly expanded to other types of classical SET assays. The methodology proposed is more complex than some relatively common approaches, nevertheless its main purpose is to leave behind some of the most controversial aspects of quantifying antioxidant activity. The model helps to accurately describe the antioxidant response as a function of time and dose. Furthermore, it allows to easily perform comparisons between different compounds.

Poster

Quantification of Small Pollutants in Natural Waters by Direct SERS using Microporous Plasmonic Capsules.

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Fast and versatile optical SERS methods represent a major advance in chemical analysis of environmental samples such as water. To date, however, these ultrasensitive methods are hindered by two key drawbacks: (i) colloidal stability and (ii) chemical diversity, both arising from the compositional complexity of natural samples. Here, we present an engineered material that, due to its unique microporous structure, imparts colloidal stability and provides selectivity while confining a densely populated film of gold nanoparticles optimized for the generation of large electromagnetic fields. The material is tested against natural water for the ultraquantification of dichlorodiphenyl-trichloroethane (DDT), a ubiquitous environmental pollutant.

Poster

CTAB-based nanostructures for photocatalysis

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Artificial organic compounds present in the water environments can be harmful to human health. One of the most preferred promising strategies to overcome these damages consist in a photodegradation strategy than can be implemented in wastewater treatment plants. For this purpose, different catalytic systems containing wide bandgap semiconductors, such as TiO2, are often used. In this work, a hybrid nanostructures composed of TiO2 nanocatalyst deposited onto mesoporous SiO2 colloidal templates was designed. A high improvement of the photodegradation activity of different dyes was demonstrated.

Poster

Ascophyllum nodosum as a source of antioxidant compounds: optimization of microwave-assisted extraction

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Algae constitute a group of organisms with promising applications as a source of antioxidant compounds, with a strong potential in both the prevention and treatment of different diseases and in food preservation. In practical terms, the optimization of the extraction conditions is necessary to obtainthe bioactive compounds present in algal matrices. Among the different extraction procedures currently available, microwave-asssited extraction (MAE) has emerged as a green technique with high efficiency rates as largely reported in plant matrices. In this work, the extraction of the antioxidant compounds from the alga Ascophyllum nodosum was carried out by MAE, using the multiwave-3000 microwave extraction system (Anton-Paar, Germany) . The critical extraction parameters that were studied in depth were ethanol concentration (% Et), pressure (P) and time (t). Such parameters ranged from 0–100% v / v for ethanol concentration, 3–25 min for extraction time, and 2–20 bar for pressure. These intervals were selected based on practical considerations and the previously available literature, while microwave power was set at its maximum value of 1400 W. In order to optimize the extraction procedure, the experimental results were subjected to the modeling by response surface methodology (RSM), with a circumscribed compound central design (DCCC). The parameters involved were total phenolic content, total flavonoid content, antioxidant capacity (determined by different antioxidant assays: sequestering capacity of the diphenyl-2-picryl-hydrazyl radical (DPPH), Trolox equivalent antioxidant capacity (TEAC) and β-carotene discoloration method (BC) and extraction yield. The predicted values given RSM modeling showed that all variables presented an adequate correlation coefficient with the experimental values, confirming the potential of these models to predict and optimize extraction performance, regarding the extraction of polyphenols and the determination of antioxidant capacity of A. nodosum extracts. As a result, the optimal conditions that maximize the simultaneous extraction of total polyphenols and flavonoids were obtained by the combination of 5.05 ± 1.68 min, 10.53 ± 1.58 bar, and 37.26 ± 2.01 % Et (v / v). According to our results, MAE constitutes an efficient solution to respond to global environmental concerns, suggesting that the application of green extraction techniques must be considered as a reliable methodology in terms of performance, time and cost, as well as being environmentally friendly.

Acknowledgments The research leading to these results was supported by MICINN supporting the Ramón y Cajal grant for M.A. Prieto (RYC-2017-22891) and the FPU grant for Anxo Carreira Casais (FPU2016/06135); by Xunta de Galicia for supporting the post-doctoral grant of M. Fraga-Corral (ED481B-2019/096), the pre-doctoral grants of P. García-Oliveira (ED481A-2019/295) and A. González Pereira (ED481A-2019/0228) and the program EXCELENCIA-ED431F 2020/12 that supports the work of F. Chamorro; by University of Vigo for supporting the predoctoral grant of M. Carpena (Uvigo-00VI 131H 6410211); by EcoChestnut Project (Erasmus+ KA202) that supports the work of B. Nuñez-Estevez, by UP4HEALTH Project that supports the work of P. Otero, P. Garcia-Perez and C. Lourenço-Lopes.

Poster

Green biopolymers from by-products as carrier materials for spray drying microencapsulation of bioactive green tea extracts. Applicability and future outlook

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Green tea is sourced from Camelia sinensis (L.). Nowadays, tea cultivation spread globally, being tea the second most common beverage worldwide. Its industry generates many waste materials due to regular pruning, required to improve quality and harvest yields. "Clean label" trend, popularity and well-documented health benefits of green tea (e.g., antioxidant capacity, anticarcinogenic properties, and neuroprotectiveness, among others) offer a unique opportunity to incorporate tea extracts obtained from by-products into nutraceuticals and clinical applications. However, sensibility to external factors such as light and temperature, are major disadvantages hindering their effective utilization in the pharmaceutical/food industry and limiting its clinical applicability. Therefore, the aim of this study is to implement and characterize suitable delivery systems, as microencapsulation, to stabilize and protect bioactive compounds of tea. A parallel goal is to explore potential applications of these systems as 3D printing materials or food additives.

Based on previous studies of our research group, ultrasound bath was used as an optimized clean extraction technique using exclusively water as extracting agent. The resulting liquid extracts were spray-dried and microencapsulated in green alternative-sourced carrier materials, such as alginate, carrageenan and starch. To characterize and compare the resulting microparticles, microencapsulation efficiency, particle-size distribution, SEM analysis, antioxidant capacity, total phenolic content and shelf-life properties were tested. Rheological features of resuspended microparticles were also studied to explore their potential as 3D printing materials.

At a time when sustainability and circular economics are a must in the industry, the achievements obtained here show a promising future for the revalorization and recycling of waste materials like those used in this study.

Poster

Valorization of *Bifurcaria bifurcata* as a marine source of bioactive compounds through the optimization of fucoxanthin extraction

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In the past, seaweed has been traditionally consumed in Asian countries [1]. However, current food trends have pointed to the re-evaluation of seaweed for its consumption in Europe, motivating its use as a food product. In this sense, seaweed is considered as natural source of proteins, minerals, oligosaccharides, and pigments with associated bioactive properties. The seaweed Bifurcaria bifurcata Ros. is a brown alga, native to the North Atlantic coast, and it is commonly found in the Galician coasts, anchored to the seabed forming a false rhizome. B. bifurcata extracts were reported to exhibit different health-promoting effects in humans, including antioxidant and neuroprotective effects, as determined on different in vitro models [2]. Fucoxanthin, a carotenoid pigment found in high concentrations on B. bifurcata, has been considered as a major responsible of its beneficial properties [3]. As a promising bioactive compound, the extraction of fucoxanthin from B. bifurcata by Ultrasound Assisted Extraction (UAE) was optimized by Response Surface Methodology (RSM), following an experimental design of three variables with 5 levels each: extraction time (t, 5 – 55 min), ethanol proportion of solvent (S, 35 – 100%), and ultrasound power (P, 100 – 500 W). The application of RSM promoted the optimization of fucoxanthin extraction, showing that the highest extraction yields were achieved at medium-high times, using solvents with high ethanol proportions and high extraction power values. Overall, these results will contribute to the valorization of brown algae, especially B. bifurcata, as promising sources of bioactive compounds, regarding its introduction as a functional food.

Acknowledgments: The research leading to these results was supported by MICINN supporting the Ramón y Cajal grant for M.A. Prieto (RYC-2017-22891) and the FPU grant for Anxo Carreira Casais (FPU2016/06135); by Xunta de Galicia for supporting the program EXCELENCIA-ED431F 2020/12, the predoctoral grant of A.G. Pereira (ED481A-2019/0228) and the program BENEFICIOS DO CONSUMO DAS ESPECIES TINTORERA- CO-0019-2021 that supports the work of F. Chamorro; by University of Vigo for supporting the predoctoral grant of M. Carpena (Uvigo-00VI 131H 6410211); and by the Bio Based Industries Joint Undertaking (JU) under grant agreement No 888003 UP4HEALTH Project (H2020-BBI-JTI-2019) that supports the work of P. Otero and P. Garcia-Perez. The authors are grateful to Ibero-American Program on Science and Technology (CYTED—AQUA-CIBUS, P317RT0003). The JU receives support from the European Union's Horizon 2020 research and innovation program and the Bio Based Industries Consortium. The project SYSTEMIC Knowledge hub on Nutrition and Food Security, has received funding from national research funding parties in Belgium (FWO), France (INRA), Germany (BLE), Italy (MIPAAF), Latvia (IZM), Norway (RCN), Portugal (FCT), and Spain (AEI) in a joint action of JPI HDHL, JPI-OCEANS and FACCE-JPI launched in 2019 under the ERA-NET ERA-HDHL (n° 696295).

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ISBN: 978-84-09-31756-1

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Xacobeo 2021

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