XV Jornadas de Genética e Biotecnologia
V Jornadas Ibéricas de Genética y Biotecnología

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XV Genetics and Biotechnology Conference | V Genetics and Biotechnology Iberian Conference

The Genetics and Biotechnology Conference (JGB) of the University of Tras-os-Montes and Alto Douro (UTAD) is an annual scientific event organized jointly by the Nucleus of Students of Genetics and Biotechnology (ADNGB) of UTAD and the Direction of the Course of Genetics and Biotechnology in collaboration with the teaching staff of the Department of Genetics and Biotechnology (DGB). As a result of the scientific-pedagogical partnership established among professors of DGB (UTAD) and of Faculty of Biological and Environmental Sciences of the University of León (UL), Spain, it was considered important to repeat the shared organization of this event between professors and students of the UTAD and UL designating it as XV Genetics and Biotechnology Conference | V Genetics and Biotechnology Iberian Conference (XV JGB | V JIGB).

The main objective of the XV JGB | V JIGB is to update knowledge in the area of Genetics and Biotechnology. To this end, the focus of this event is the conferences given by renowned national and international scientists and the thematic workshops that will constitute more practical sessions. The XIV JGB | IV JIGB will also focus on interaction, exchange of experiences and scientific debates between Portuguese and Spanish students and professors.

The best oral and posters presentations will be awarded.

The target audience is Portuguese and Spanish students, researchers and university professors from the scientific areas of Biological Sciences and Biotechnology as well as High School teachers from the Biology area.

A wide variety of topics will be discussed, in the different areas of Genetics and Biotechnology, such as Plant, Animal, Human, Microbial, Evolutionary, Cancer, Forensic, Ethics, Entrepreneurship, among others.

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PROGRAM

Wednesday, 22nd of March (*morning session*) (Portugal – at UTAD)

UTAD (ECVA Auditorium): Zoom videoconference in streaming

PROGRAM

PT	ES	
09:00	10:00	**OPENING SESSION (*In Person*)**
09:30	10:30	Plenary Conference (*In Person*) **Dr. Luís Portela (Fundação Bial)** *Science and Spirituality*
10:30	11:30	BioPortugal – Company Presentation **BioPortugal Representative**
10:45	11:45	**Coffee break + Posters show (digital exposition)**
		FORENSIC GENETICS
11:05	12:05	Conference (*In Person*) **Prof. Maria João Prata (University of Porto)** *Forensic Genetics: From the imaginary to the real role in the justice system*
12:00	13:00	Lunch

Wednesday, 22nd of March (*afternoon session*) (Portugal – at UTAD)

WORKSHOPS (*In Person at UTAD*)

18:00 - 20:00	**Prof. Paula Martins-Lopes and Prof. Helena Gonçalves** *DNA and RNA Biosensing Platforms*
18:00 - 20:00	**Prof. Márcia Carvalho and Prof. Isaura Castro** *SSRs for genotyping plant genetic resources*
18:00 - 20:00	**Prof. Isabel Pires and Prof. Anabela Alves** *Collection and preservation of samples in forensic science from necropsy to lab*
18:00 - 19:30	**Prof. Daniela Ferreira, Prof. Sandra Louzada and Dr. Mariana Lopes (M.Sc.)** *The power os Microscopy in Research*
18:00 - 20:00	**Prof. Guilherme Sousa, Dr. Tiago Duarte (Ph.D.) and Prof. Verónica Bermudez** *Silk, a biopolymer as material for flexible sensors*
18:00 - 20:00	**Prof. Ana Escudeiro and Prof. Filomena Adega** *Breaking the code – Bioinformatic tools in cancer research, diagnosis and treatment*
18:00 - 20:00	**Prof. Estela Bastos, Prof. Nuno Osório, Prof. Jorge Pereira, Dr. Sofia Monteiro, Dr. Ana Vieira and Dr. José Pinheiral** *Making bridges – solving problems. A Hands-on practical* in silico *workshop*

Thursday, 23rd of March (*morning session*) (Portugal – at UTAD)

UTAD (ECVA Auditorium): Zoom videoconference in streaming

PROGRAM

MEDICAL GENETICS

PT	ES	
09:00	10:00	Conference (*In Person*) **Dr. Ricardo Ribeiro, Ph.D.** (Centro Hospitalar Universitário de Santo António) *Liquid Biopsy in Oncology*
09:50	10:50	Conference (*In Person*) **Dr. Joana Marques, Ph.D.** (Faculdade de Medicina da Univ. do Porto) *Epigenetic alterations in human infertility*
10:40	11:40	BioPortugal – Company Presentation **BioPortugal Representative**
10:55	11:55	**Coffee break + Posters show (digital exposition)**
		ORAL COMMUNICATIONS – SESSION 1 (In person and online)
11:15	12:15	**Ana M. Faria** - *Variants in CDKN1C gene and Beckwith-Wiedemann Syndrome: Bioinformatic review*
11:25	12:25	**Tiago Azevedo** - *Chemopreventive potential of a* Santolina chamaecyparissus *aqueous extract in a rat model of breast cancer*
11:35	12:35	**Vânia Santos** - *Characterization of multidrug-resistant* Escherichia coli *as a zoonotic pathogen of wild birds*
11:45	12:45	**Mónica Ramos** - *Identification of* Staphylococcus aureus *isolated from samples obtained from food and handlers and detection of virulence genes*
11:55	12:55	Discussion of all oral communications (Session 1)
12:05	13:05	**Lunch**

Thursday, 23rd of March (*afternoon session*) (Portugal – at UTAD)

UTAD (ECVA Auditorium): Zoom videoconference in streaming

PROGRAM

ENVIRONMENTAL GENETICS

PT	ES	
14:00	15:00	Conference (*In Person*) **Dr. Elena González-Toril, Ph.D.** (Centro de Astrobiologia de Madrid, CSIC-INTA) *Extreme Environments*
14:40	15:40	Conference (*In Person*) **Dr. Luis Sáenz de Miera, Ph.D.** (University of León) *Effect of forest on soil microbial communities. An example of the use of environmental DNA*

EVOLUTIONARY GENETICS

15:20	16:20	Conference (*In Person*) **Dr. Angelica Crottini, Ph.D.** (CIBIO/InBio) *Evolutionary genetics and genomics in a biodiversity hotspot*
16:00	17:00	**Coffee break + Posters show (digital exposition)**

ORAL COMMUNICATIONS – SESSION 2 (In person and online)

16:20	17:20	**Joana Paiva** - *Prevalence, antibiotic resistance and genotypic characterization of* Staphylococcus aureus *isolated from meat and meat products (Online)*
16:30	17:30	**Sara Araújo** - *High prevalence of ESBL producing* Klebsiella *spp. in surface waters (Online)*
16:40	17:40	**Beatriz Magalhães** - *Effect of potassium and magnesium on the expression of genes related to cell wall mechanisms in sweet cherry*
16:50	17:50	**Francisca Castro** - *Unveiling the role of transposable elements in bats' genome evolution*
17:00	18:00	**Sara Costa** - *Karyotypic analysis of Portuguese chiropterans: a step towards the genomic characterization of these model organisms*
17:10	18:10	Discussion of all oral communications (Session 2)
17:20	18:20	***ROUND TABLE WITH FORMER STUDENTS (In person and online)*** *Exclusive event for former and current students of the 1st, 2nd and 3rd cycles of study in the field of Genetics and Biotechnology*
18:30	19:30	***CLOSING SESSION WITH MUSICAL MOMENT (In person and online)***

Friday, 24th of March (Spain – at ULE)

University of León ("Aula Magna" - FCCBA)

PROGRAM

PT	ES	
11:00	12:00	Registration at the ULe (*UTAD participants*)
11:30	12:30	**Visits to the companies**: • Aquilon CyL S.L. [https://www.aquiloncyl.com/] • Bianor Biotech S.L. [https://bianorbiotech.es/] • Dentale Biomedica [https://dentalebiomedica.com/] • Neural Therapies [https://neuraltherapies.com/]
12:30	13:30	**Lunch**
14:30	15:30	Registration at the ULe (*ULe participants*) (*Main entrance of the FCCBA*)

University of León ("Aula Magna" - FCCBA): Zoom videoconference in streaming

15:15	16:15	Welcome by the Dean of Faculty
15:30	16:30	***ROUND TABLE: Y DESPUÉS ¿QUÉ?*** **Marta García Gil** **Patricia De la Madrid Salmerón** **Dr. Elsa González Cubero** **Elena Gallego Clemente**
16:30	17:30	**Coffee break + Posters show (digital exposition)**
17:00-18:00	18:00-19:00	Conference (*In Person*) **Dr. Francisco Aulestia, Ph.D, y Dr. Arsenio Fernández, Ph.D.** *Face to Face: business and entrepreneurship*

Saturday, 25th of March (*morning session*) (Spain – at ULE)

University of León ("Aula Magna" - FCCBA): Zoom videoconference in streaming

PROGRAM

PT	ES	
8:30	9:30	Conference (*On line*) **Dr. Aitor Balmaseda Rubina, M.Sc.** *Omic approaches for understanding the microbial physiological state:* Oenoccocus oeni *and the adaptation to wine*
		ORAL COMMUNICATIONS – SESSION 3 (In person and online)
09:15	10:15	**Garrido-Chamorro S** - *Optimization and validation of Corynebacterium glutamicum scale-down processes*
09:25	10:25	**Guío J** - *Unravelling the FurC regulon in the cyanobacterium* Anabaena *sp. PCC 7120 and its role in the control of processes dependent on carbon/nitrogen balance*
09:35	10:35	**Carnicero-Mayo Y** - *Addition of non-digested gluten to a digested-gluten containing culture medium does not affect bacterial density of intestinal communities derived from gluten metabolism*
09:45	10:45	**Cerezo C** - *How could nettle infusions protect beans from halo blight disease?*
09:55	10:55	Discussion of all oral communications (Session 3)
10:15	11:15	**Coffee break + Posters show (digital exposition)**
11:00	12:00	Conference (*In Person*) **Dr. Javier Villoch Fernández, Ph.D.** *Exploring p73 inhibition as a therapeutical target against Glioblastoma Stem Cells*
		ORAL COMMUNICATIONS – SESSION 4 (In person and online)
12:00	13:00	**Guío J** - *Redox-sensitive control by thioredoxin of the ferric uptake regulator Fur from the strict anaerobe pathogen* Clostridioides difficile
12:10	13:10	**Montero-Villacorta L** - *Understanding the genetic control of heat-induced seed coat browning in cowpea (*Vigna unguiculata*)*
12:20	13:20	**Nhhala N** - *Effect of liquid extract from Ulva Lactuca algae, in the alleviation of salt stress on crops of the common bean, Phaseolus vulgaris L.*
12:30	13:30	**Llano J** - *Gene deletion in Rhodococcus fascians and phenotypic evaluation of a mycoredoxin-deficient mutant under oxidative stress*
12:40	13:40	Discussion of all oral communications (Session 4)
13:00	14:00	**Lunch (*Catering at the FCCBA*)**

Saturday, 25th of March (*afternoon session*) Spain – at ULE)

University of León ("Aula Magna" - FCCBA): Zoom videoconference in streaming

PROGRAM

PT	ES	
14:30	15:30	***ROUND TABLE:*** ***STUDENTS FROM ABLE (ULe) AND FROM ADNGB (UTAD)***
15:30	16:30	Conference (*In Person*) **Dr. Michal Letek Polberg, Ph.D.** *Superbug and a host hunt*
16:30-18:00	17:30-19:00	***AWARDS CEREMONY AND CLOSING SESSION***

SPEAKERS

Dr. Luís Portela

Nascido em 1951 no Porto, é licenciado em Medicina pela Universidade do Porto, tendo feito algumas ações de formação em gestão. Exerceu atividade clínica no Hospital de S. João apenas durante três anos e foi docente da Universidade do Porto durante seis anos, onde lecionou a cadeira de Psicofisiologia.

Desligou-se da carreira médica e universitária para se dedicar à gestão da empresa de sua família - Bial. Iniciou a atividade empresarial com vinte e um anos e aos vinte e sete assumiu a presidência executiva da empresa (1979-2011), tendo depois passado a presidente não executivo (2011-2021). Foi também presidente do Health Cluster Portugal (2008-2017) e do Conselho Geral da Universidade do Porto (2009-2013), vice-presidente da Fundação de Serralves (2001-2008) e membro da Direção da Cotec (2006-2012). Em 2021 retirou-se da vida profissional, para se dedicar à Fundação Bial, a que preside, à leitura, à escrita e à família.

Sob a sua presidência, Bial tornou-se a primeira empresa farmacêutica internacional de inovação de origem portuguesa, operando atualmente em cerca de 60 países. No Grupo Bial criou e desenvolveu um Centro de Investigação, especializado na investigação de fármacos. Nesse centro foram criados os dois primeiros medicamentos de investigação portuguesa a serem comercializados no mercado global: a partir de 2009 um antiepilético e de 2016 um medicamento para a Doença de Parkinson.

Em 1994 criou, conjuntamente com a Bial e o Conselho de Reitores das Universidades Portuguesas, a Fundação Bial, tendo como objetivo incentivar a investigação sobre o ser humano, tanto sob o ponto de vista físico como espiritual. A Fundação teve desde então mais de 1.600 bolseiros em investigação científica, de 29 países. Também atribui três prémios: o Prémio Bial de Medicina Clínica, o Bial Award in Biomedicine e o Prémio Maria de Sousa, este último em parceria com a Ordem dos Médicos.

O seu prazer pela leitura e pela reflexão levou-o à escrita, tendo-o feito com caráter permanente em alguns órgãos de comunicação social. Publicou dez livros, dos quais se mantêm nas livrarias Serenamente, O Prazer de Ser, o best-seller Ser Espiritual, em 32ª edição, Da Ciência ao Amor, em 14ª edição, e o mais recente The Science of Spirit - Parapsychology, Enlightenment and Evolution, publicado nos EUA pela Toplight/McFarland.

Foi agraciado com três condecorações do Estado Português: Comendador da Ordem do Mérito, Grã-Cruz da Ordem do Mérito e Grã-Cruz da Ordem da Instrução Pública. Foi distinguido com quatro doutoramentos Honoris Causa, pelas Universidades de Cádis, Porto, Coimbra e Lisboa. Em 1998 foi distinguido com o Prémio de Neurociências da Louisiana State University, nos E.U.A. Em 2007 com a Medalha Municipal de Mérito - Grau Ouro – pelo Porto, em 2014 com a Medalha de Honra - Grau Ouro - pela Trofa, e ainda, desta última, em 2021, com a primeira chave da cidade. Em 2008 foi distinguido como “Empresário do Ano” pelo Rotary International. Em 2009 foi eleito Académico Correspondente pela Academia Portuguesa de Medicina. Em 2010 foi eleito Membro Honorário da Parapsychological Association of the American Association for the Advancement of Science. Em 2016 foi distinguido com a Medalha de Ouro de Serviços Distintos do Ministério da Saúde. Em 2017 com o Prémio Carreira pela Associação Nacional de Jovens Empresários - ANJE. Em 2019 com a Medalha de Mérito da Ordem dos Médicos. Em 2021 com a Medalha de Mérito Científico do Ministério da Ciência. Em 2023 com o Prémio Excelência na Liderança pela revista Exame.

Professor Maria João Prata

Professora associada com agregação do Departamento de Biologia da FCUP; Investigadora sénior do Grupo de Genética Populacional e Evolução do i3S.

Tem desenvolvido atividades de investigação na área da Genética Humana, abrangendo tópicos diversos com base na análise dos padrões de diversidade genética em populações humanas contemporâneas.

Foi uma das promotoras da criação do Mestrado Genética Forense da FCUP, curso em que tem tido grande envolvimento, quer em cargos diretivos quer na docência.

Dr. Ricardo Ribeiro, Ph.D.

Ricardo Jorge Teixeira Ribeiro (M.D., Ph.D.) é Médico do Departamento de Patologia no Centro Hospitalar Universitário do Porto, Clinical Affiliate Researcher do Tumor & Microenvironment Interactions Group do Instituto de Investigação e Inovação em Saúde (i3S). Desempenhou e mantém atividade como orientador de alunos de mestrado e doutoramento na área da investigação em cancro (> 20).

Foi Research Fellow no Laboratório de Investigação Metabólica/Laboratório de Metabolómica Funcional da Universidade de Navarra em Espanha, no Departamento de Oncologia da McGill University no Canada e no NEW Therapies Group do INEB/IPATIMUP da UP.

É autor ou co-autor de mais de 50 artigos full-text publicados em revistas indexadas. É co-autor de uma patente que visa a utilização de microfluídica para isolamento selectivo de células em líquidos corporais baseado em padrões de glicosilação. Para além do contributo com investigação original, é revisor ativo de mais de 10 revistas científicas indexadas internacionais. É Associate Editor do Frontiers in Oncology – Cancer Genetics e Editorial Board Member do Nutrients e da Revista Portuguesa de Oncologia. O seu trabalho foi distinguido com mais de 25 prémios científicos e distinções.

Dr. Joana Marques, Ph.D.

Licenciada em Biologia pela FCUP, Doutorada em Biologia Humana pela FMUP, realizou projetos de pós-doutoramento em Cambridge, no Reino Unido e no ICVS, Universidade do Minho, até se tornar investigadora principal na FMUP desde 2016.

É autora/co-autora de artigos científicos em revistas científicas como The Lancet, Nature, Nature Communications, Molecular Psychiatry e New England Journal of Medicine.

Realizou estágios em laboratórios internacionais como University of Pittsburgh (Pensylvannia, USA), Institute of Molecular Genetics of Montpellier (France), University of Saarlandes (Germany), Centre René Huguenin (INSERM), Wellcome Trust Genome Campus (UK), and Blizard Institute (UK).

Foi galardoada com vários prémios entre os quais a Medalha de Honra L'Oreal/FCT para as Mulheres na Ciência em 2010.

Realiza investigação em regulação epigenética em vários modelos celulares (células germinais humanas, células estaminais embrionárias, células precursoras neuronais e neurónios glutamatérgicos) e modelos clínicos (infertilidade masculina e feminina, perda de gravidez e restrição de crescimento intra-uterino) assim como modelos animais (KO condicional da enzima Tet3).

Outras áreas de interesse incluem utilização de células pluripotentes induzidas no desenvolvimento de terapêuticas inovadoras na área das doenças neurogenéticas e farmacogenética.

Dr. Elena González-Toril, Ph.D.

Elena González Toril is researcher at Centro de Astrobiología (INTA-CSIC) in Madrid, Spain. She has degree in Molecular Biology from the Universidad Autónoma de Madrid, 1996. She carried out her PhD. at Centro de Biología Molecular Severo Ochoa (CSIC-UAM), and MaxPlanck Institute for Marine Microbiology en Bremen, Alemania (2002). 2002-2003 Post-Doc at Centro de Biología Molecular Severo Ochoa (CSIC-UAM). Since 2003, she is researcher in the Centro de Astrobiología, Madrid, Spain. She has more than 20 years of experience in biodiversity and microbial ecology studies, mainly related to extreme ecosystems, to understand the physiology and adaptation mechanisms of the organisms that inhabit them.

Research topic: She has a solid knowledge of the main methodologies of molecular ecology. She has worked in extreme acid environments, cold environments such as Antarctica, high mountains and hydrothermal environments. Her experience in microbial ecology focuses mainly on understanding the physiological, ecological and genetic mechanisms of adaptation to extreme environmental conditions, belonging to one of the pioneering groups in the application of metagenomic, metaproteomic and metatrascriptomic methodologies to the study of these ecosystems. She reported the first molecular description of the microbial diversity of Rio Tinto, a Martian analog. This gives rise to numerous documents describing the different niches of the river (water column, sediments, biofilms, etc.). The first of these works has been cited more than 240 times. She has set up different techniques of molecular ecology in environments of astrological interest. These techniques have been the basis of many of the works that have been carried out. Some examples are FISH, CARD-FISH, DGGE, cloning of 16/18S RNA genes or NGS.

Dr. Luis E. Sáenz de Miera, Ph.D.

After a PhD in Genetics at the University of León (1995), I completed my research training with a stay at the University of California in Irvine and other postgraduate studies such as “Use of Networks and Databases in Molecular Biology”, by the Universidad de Valencia (1992), “Advanced Methods in Applied Statistics” by UNED (2011) and “Data Analysis for Genomics” through online courses at Harvard University (2015). My research began with studies of Plant Population Genetics, mainly grasses and legumes, using Molecular Biology techniques, initially isozymes, to pass quickly to DNA markers or directly to the amplification, sequencing and analysis of DNA fragments. My field of research was extended with evolutionary studies, focusing on the phylogenetic analysis of genes included in gene families.

Later, I began working with two research groups of the University of León, one dedicated to the study of the relationship of human pathologies of the digestive system with the microbiome and another dedicated to the ecological study of soils that have suffered anthropic or natural disturbances, with a special interest in wastewater treatment through low cost systems and in soils from ecosystems affected by wildfires. With these groups my research was focused on the investigation of bacterial communities from the sequences that encode the 16S ribosomal RNA gene. Initially from clone libraries and currently using high-performance mass sequencing for a subsequent metagenomic analysis using bioinformatics tools. That is, environmental genomics using environmental DNA.

Dr. Angelica Crottini, Ph.D.

Angelica Crottini graduated in Biology at the University of Milano (Italy) in 2004 and she completed a PhD in Animal Biology at the same institution in 2008. During her PhD she started a parallel research activity on the evolution of the herpetofauna of Madagascar, and she spent 1 year at the University of Braunschweig (Germany), where in 2008 she obtained a postdoctoral grant to work with one of the top leading experts on Malagasy biota. She obtained 2 FCT post-doctoral grants (2010, 2013) to work at CIBIO and in 2014 she secured an Investigador FCT starting grant that enable her to develop her independent research career at InBIO where, since 2018, she leads the Biogeography and Evolution group (BIOEVOL). Since graduation she published 103 ISI-indexed and 17 non ISI-indexed papers, 4 books, 23 book chapters, and 13 popular science contributions.

Research topic: Broadly, I am interested in characterizing and describing the herpetological diversity of Madagascar, understand how this has evolved, and use this knowledge to suggest conservation measures aimed at safeguarding Madagascar ecosystems. Depending on the different questions (and together with many international collaborators), we target single species, specific groups of organisms, or investigate the host-microbiome relationships. To do so we are combining traditional collection-based approaches (specimen collection and comparative specimen inspection), molecular phylogenetics and phylogenomics, population genetic and genomics, molecular taxonomic identification, biogeography, bioinformatics, modelling and ecology.

Dr. Francisco Aulestia, Ph.D.

Dr. Francisco Aulestia is a professional with experience in technical and business projects. He started as a research project assistant, then became a project manager, and is currently the technological director of a portfolio of Cellus Group projects for the pharmaceutical development of advanced medicinal products. He has acquired expertise in applied PMI and agile methodologies for project control. Additionally, he has experience in laboratory, manufacturing, and clinical good practices for the development and validation of therapies in humans.

Francisco Aulestia has held the positions of Vice President at BIOTECYL, and is currently the Chief Scientific Officer and Director Científico at Cellus Medicina Regenerativa S.A., where he is responsible for developing advanced cell therapies. He has a Doctorate in Biomedicine from the Universidad de Valladolid, and has completed postdoctoral studies at the New York University and the Université Paul Sabatier Toulouse III. He is also competent in Spanish, English, and French.

Francisco Aulestia has experience leading the implementation of advanced therapies and has contributed to the entry of Cellus Biomedica EU España into the market with a high-value-added offer. He is currently enrolled in a high-level management program at IE Business School.

Dr. Arsenio Fernández, Ph.D.

Graduated in Biology and Ph.D. in Biology, Universidad de Oviedo. Professor titular in Cell Biology in 1983 and Catedrático in Cell Biology in 2000 at the Universidad de León. Teaching in different matters in Cell Biology and Biotechnology (Cell Biology, Neurobiology, Comparative Microscopic Anatomy, Techniques in Cell Biology, etc.). Expertise in central nervous system (CNS) research and focused on stroke from 2008. Supervisor of more than 25 theses (PhD, MD and Pharmacy). Mention international in all the theses in the last ten years. More than 70 publications in international journals and more than 25 articles in Q1 journals. Associate editor in Oxid Med Cell Longev, reviewer in many journals (Neuroscience, Neurobiol Dis, Neural Reg Res, Brain res, Redox Biol, etc.). Communications mainly in the Society for Neuroscience (USA) and Forum for Neuroscience (UE). Expertise in HPLC, anatomical, histological, electrophysiological, molecular, and antibody-based techniques applied to studying the CNS.

Founder and CEO of Neural Therapies in 2015. Attendance to Biotechnical meetings in Spain (BioSpain), Europe (BioEurope), and the USA (BIO International Conventions). Development of models for contract research organization (CRO) services in Neural Therapies (models of pathologies in "in vivo", "in vitro" and cell culture research for SNC including hippocampal organotypic slice culture and behavior test (stroke models, Parkinson disease models) and other developments addressed to study inflammation (osteoarthritis or LPS models), also antiviral and antibacterial models of screening.

Dr. Aitor Balmaseda Rubina, M.Sc.

Dr. Aitor Balmaseda is a biotechnology graduate with a BSc from UPV/EHU and an MSc in Fermented Beverages from URV. He has extensive research experience in the areas of *Aspergillus fumigatus*, non-*Saccharomyces* yeasts, and *Oenococcus oeni*.

Aitor has published eight indexed scientific articles and collaborated in the supervision of several students.

He has participated in different teaching and scientific activities, organized various scientific conferences, and held positions in scientific associations.

He is also actively involved in university life and has reviewed publications for high impact indexed scientific journals.

Dr. Javier Villoch Fernández, Ph.D.

Dr. Javier Villoch studied Biotechnology at the University of León and obtained his Ph.D. in Molecular Biology and Biotechnology from the same University.

His research has focused on the role of transcription factors belonging to the p53 family, especially TP73, in the development of the central nervous system, and on understanding how the control of their activities can be used as a target therapy against certain types of glioblastoma. In relation to this field, he has published several papers in prestigious international journals and is co-inventor of a European patent.

His research has been carried out mainly at the University of León, although he has also moved to other centers such as the Max Planck Institute in Munich, thanks to obtaining a grant funded by the European Molecular Biology Organization (EMBO), the Catholic University of Leuven or the Francisco de Vitoria University. Additionally, he has also shown great interest in teaching activities, co-supervising several Master's and Final Degree Projects related to the fields of Cell Biotechnology and Biomedicine.

Dr. Michal Letek Polberg, Ph.D.

My research work has always been focused on identifying novel ways to control bacterial pathogens.

During my PhD, I studied bacterial cytokinesis and cell wall synthesis to understand how Actinobacteria grow and divide. In particular, I identified and functionally characterized proteins involved in cell division and elongation present only in Actinobacteria. This group of bacteria includes important human pathogens that are becoming increasingly resistant to currently available antibiotics, such as *Mycobacterium tuberculosis*. The proteins involved in bacterial cytokinesis and cell wall synthesis could be used as targets for novel antibiotherapies. However, I have also an interest in studying actinobacterial virulence to identify novel ways to block the pathogenesis of these bacteria. During my first postdoctoral years, I functionally analysed the genome of an intracellular pathogen, *Rhodococcus equi*. The reconstruction of the regulatory network of this pathogen led to the discovery of novel virulence factors used by *R. equi* to survive inside macrophages. I am now studying the redox biology of this pathogen during host cell infection.

My research work is also focused on the identification and characterization of the host molecular factors that are involved in the intracellular survival and proliferation of *Staphylococcus aureus*, another major human pathogen. This could be crucial to design novel therapeutic strategies to control MRSA and to identify the major determinants of susceptibility to *S. aureus*.

CONFERENCES

Science and Spirituality

Luís Portela [1]

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Throughout the 20th Century, humanity made enormous progress in science and technology, which has made possible a great knowledge of matter in general and of the human body. However, the existence of one or more non-physical entities within human beings, life beyond physical death, and the so-called parapsychological phenomenology, all have gone relatively unstudied, hampering the spiritual enlightenment.

Apparently, the fascination for the material discoveries put a greater focus on materialism, causing some imbalance in a considerable number of humans who are highly focussed on the physical world, in what they have and how they appear, giving less importance to universal values, to being, to spiritual life. However, given the significant results of the scientific research in parapsychology over the last few decades, science has the obligation to continue to research in this area, seeking to contribute to the spiritual enlightenment of humanity.

It would be reasonable to admit that some of the traditional phenomena we hear about will be shown as pure fantasy. But it would be also reasonable to admit that some traditional psychic phenomena we hear about will be confirmed as true, permitting a more comprehensive perspective of reality, with a greater capacity for individual and collective fulfilment and better understanding of ourselves as one particle of "universal energy" that is interconnected with all other particles.

Thus, it seems that science may contribute to humans living in a higher level of consciousness, developing their ability for respecting and loving themselves, but also the other and the Universal Whole.

Forensic Genetics: From the imaginary to the real role in the justice system

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Genética Forense: do imaginário ao real papel no sistema de justiça

O imaginário que se tem gerado sobre as Ciências Forenses, tem tido repercussões complexas em diversos sistemas de justiça.

A par da mediatização, tem aumentado o escrutínio sobre todas as ciências auxiliares da justiça. Depois de abordar o que distingue a Genética Forense de outras Ciências Forenses, convocar-se-á a importância de estreitar a colaboração entre todos os intervenientes no sistema de justiça, mas garantindo uma criteriosa compartimentação de papéis, de forma a minimizar conflitos entre Ciência e Lei.

Liquid Biopsy in Oncology

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Precision medicine emerged as a model for personalizing healthcare based on genomic analyses. The introduction of new technologies allowed the identification of significant genetic variants in the human genome, with the results progressively being used to help determine the best therapeutic option for cancer patients. Liquid biopsy, particularly analysis of cell-free DNA (cfDNA) from peripheral blood, is used in clinical practice for the molecular characterization of tumors without the need for invasive biopsy, having shown additional interest for detection of minimal residual disease, monitoring of response to treatment and early detection of cancer.

Its performance is concordant with solid tumor sequencing, and useful for identifying actionable DNA changes with targeted therapies. Liquid biopsy represents the preferred modality to successively determine the mutational profile of the tumor genome after each line of treatment.

Liquid biopsy may include beyond detection of tumor-derived nucleic acids in the form of ctDNA, analysis of circulating tumor RNA, circulating microRNA, RNA or DNA from extracelular vesicles and circulating tumor cells, all identified from a sample of peripheral blood or body fluid, obtained in a minimally invasive way.

Liquid biopsy is increasingly recognized to provide important contribution for clinical oncology reasoning, representing a fundamental means of implementing personalized medicine.

Epigenetic alterations in human infertility

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Infertility represents a growing emergence worldwide affecting about one out of seven couples who attempt to generate a child. In the last 20 years, our group has been working on the occurrence of epigenetic defects, namely DNA methylation errors at imprinted genes, in spermatogenic cells from infertile patients. We described, in a seminal paper in 2004, the occurrence of imprinting errors in human spermatozoa from patients with oligozoospermia, i.e., decreased sperm count. Here, we observed hypomethylation of the Differentially Methylated Region (DMR) of H19 imprinted gene. Later, in 2008, we described that hypermethylation also occurs at the MEST gene in sperm from
oligozoospermic patients. Regarding azoospermia, i.e. absence of sperm in the semen, we have also observed imprinting errors in testicular sperm and other more immature spermatogenic cells. In normal spermatogenesis, we have described that imprinting marks are already established in human adult spermatogonia and are kept in subsequent stages, across meiotic divisions and in terminal differentiation phases. Our studies have contributed to novel insights into the molecular basis of human infertility.

Extreme Environments

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Extreme ecosystems, such as terrestrial hot springs, deep sea hydrothermal vents, glaciers and permafrost, hypersaline habitats, extreme pH habitats and subsurface, are widely distributed throughout the world. These environments and the microorganisms that inhabit them, called extremophiles, have always been of great scientific interest. Their study has provided pioneering discoveries that challenge some of the paradigms of modern biology.

Extremophiles live in habitats that are very hostile or even lethal to other forms of life, and have evolved different molecular strategies to cope with such extreme environmental conditions. They define the physical and geochemical limits of life, and they have attracted a great deal of interest from a fundamental and astrobiological perspective.

Moreover, although extremophiles were initially regarded as a “scientific curiosity”, the biotechnological potential of these microorganisms and their cellular products has boosted their research in recent years. The fields of biotechnology that could benefit from extremophiles are numerous and include the search for new bioactive compounds for industrial, agricultural, environmental and pharmaceutical uses. Thus, biotechnology developed from extremophiles has reached such a level of commercial development that it has led numerous research initiatives, biotechnology companies and the continuous search for new extremophile microorganisms and new genes across the globe.

Effect of forest fires on soil microbial communities. An example of the use of environmental DNA

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Environmental DNA or eDNA analysis is a useful tool for determining the biodiversity of different samples, whether they come from soil, water, sediments or air. The DNA is extracted directly from the samples and not necessarily from particular individuals. Specific gene sequences are normally amplified by PCR from the eDNA, the amplicons obtained are sequenced using high-throughput sequencing techniques. Although on occasions and when it is intended to analyse viruses or prokaryotes, all the extracted DNA can be sequenced. The extracted eDNA can come from communities of whole organisms, usually living or not living microorganisms. And they can also come from remains of skin, mucosa, saliva, sperm, eggs, blood, roots, leaves, pollen or fruits. The bioinformatic analysis of the sequences obtained makes it possible to identify species or enzymatic functions of the sample by comparing them with databases.

As an example of the usefulness of eDNA, the results obtained in the analysis of communities of bacteria and fungi from forest soils that have suffered a large fire will be presented in this paper. The analysis immediately after the fire allows us to know how the structure and composition of the communities' changes depending on the severity of the fire and what is the degree of resistance of each one of the components of the communities, it even allows the identification of species or taxonomic groups which behave as indicators. A new sampling after some time and under different conditions, allows us to understand how these communities recover and estimate their degree of resilience.

Evolutionary genetics and genomics in a biodiversity hotspot

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Madagascar hosts an almost unparalleled concentration of endemic, diverse and endangered flora and fauna which has evolved in isolation over millions of years. Despite having four centuries of biological explorations, undescribed diversity is still widespread and is occurring at both poorly explored and in better-studied areas, and field exploration is still playing a crucial role in new species discoveries. The product of our field-based research resulted in the identification and description of several new species, in proposing new classifications, uncovering the distribution of genetic diversity and demographic history, and is currently contributing to uncover the patterns of species diversification of multiple radiations of amphibians and reptiles. Its native amphibian fauna is constituted by five independent radiations with 100% species-level endemism. Among these, the mantellid frogs is the most diversified family of anuran that here underwent a wide species radiation that resulted in the evolution of a plethora of morphological, ecological and reproductive traits. We combined the use of evolutionary genomics with the collection of life history traits and the use of comparative phylogenetic methods to investigate the evolutionary association between life-history and morphological traits and test for their contribution to the diversification of this group.

Face to Face: Business and Entrepreneurship

Francisco Aulestia y Arsenio Fernández

Cell therapies to resolve medical needs. CELLUSPHERES® innovative product as spearhead

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Cellular therapies are a medical treatment approach that uses living cells or their components to treat disease or injury. Throughout the world, there is increasing interest in the use of cell therapies to treat a wide range of diseases, including musculoskeletal, gastric, cardiovascular, neurological disorders and autoimmune diseases.

One of the most common forms of cell therapy is stem cell or mesenchymal cell (MSC) therapy. These are obtained from different sources through bone marrow tissue, umbilical cord blood or adipose tissue.

In some countries, such as the United States and Europe, cell therapies are regulated by government agencies (FDA or EMA) and must meet stringent prerequisites to be approved for therapeutic use. These requirements involve animal and human studies that allow a characterization of the therapy as well as its toxic effects and efficacy.

Despite the challenges and controversies, cell therapies continue to be an exciting area of research and development around the world, with the potential to revolutionize the way many diseases are treated.

Cellus Biomedica has promoted for 4 years the development of an innovative advanced therapy based on mesenchymal cells called Cellusphere®. The therapy aims to be a platform for the treatment of diseases with an inflammatory component such as ostearthritis, ulcerative colitis or macular degeneration.

Creating a biotechnology spin-off company

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Keywords: Biotech enterprises, spin-off, entrepreneurs, enterprise requirements

There are many types of biotech companies. Small companies are usually based on a molecule's property or an experimental procedure that can use for clinics, agriculture, veterinary, nutrition, etc. Biotech enterprises can also be created from technical skills (proper biotechnical skills, but also skills in statistics, computational, regulatory, legal, etc., applied to biotech) and even from market knowledge (databases, client portfolio, etc.). Creating a spin-off from the University represents translating academic expertise, both conceptual knowledge and technical skills, into a usually specialized enterprise that must be competitive enough to survive in a crowded market of companies with plenty of new ideas, procedures, or promising molecules. Therefore you need a clear and realistic notion of what you want to do and look at the possible competitors. Once convinced that it is worth translating your basic idea or skills into a business idea, you must obtain the funding to create the enterprise and consider whether employees or partners are required and the legal type of company (individual, limited society, foundation, etc.). It is also necessary to look for a place and the equipment needed, a name, a logo, a convincing web page, and to think of the possible clients and how to contact them. Usually, there are many administrative and economic difficulties to sort out, especially when employees are required in the company. Contracting an agency to deal with the Administration is practically mandatory (countability, taxes, social security, VAT dealing, etc.). All these aspects must be considered before taking the plunge and creating the company, whose costs require immediate funding (paying taxes, rents, social security, attending biotechnology meetings to look for clients, etc.). Another aspect, basic for the company's survival, is to have enough plasticity to adapt to the market's requirements and optimize resources.

Omic approaches for understanding the microbial physiological state: *Oenoccocus oeni* and the adaptation to wine

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Omics are a set of technologies for comprehensive study of biological components such as genes, proteins, and metabolites. Omics techniques include genomics, transcriptomics, proteomics, metabolomics, epigenomics, and microbiomics. The application of omics has revolutionized our understanding of biological systems, providing unprecedented insights into molecular mechanisms underlying health and disease. These techniques allow researchers to study large sets of molecules in a single experiment, identify disease biomarkers, drug targets, and develop personalized medicine. The integration of multiple omics datasets allows a comprehensive understanding of biological systems. Omics has opened up new avenues for biomedical research and clinical practice, with the potential for significant advances in understanding the complexity of biological system.

Transcriptomics and proteomics have been powerful tools to study the response of Oenococcus oeni to different stresses and how it varies depending on the inoculation of different non-Saccharomyces yeast strains. In order to process and analyze the data obtained from these experiments, bioinformatics tools and pipelines were used to filter, normalize, and statistically analyze the transcriptomics and proteomics data. This process allowed the identification of differentially expressed genes and proteins, which can provide insights into the molecular mechanisms underlying the stress response. This is a valuable model for beginners in the world of transcriptomics, as it demonstrates the importance of appropriate data processing and analysis to obtain meaningful results.

Exploring p73 inhibition as a therapeutical target against Glioblastoma Stem Cells

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Glioblastoma multiforme (GB) is the most prevalent and severe central nervous system malignant tumor, with a median of survival of 15 months, and almost no long-term survivors. This high mortality rate is caused by a combination of its highly invasive behavior, high rate of tumor relapse and the low effectiveness of currently available treatments. Multiple studies have demonstrated the presence of a tumor cell population in GB, known as Glioblastoma Stem Cells (GSC) with self-renewal and multipotency. These cells show high resistance to actually available GB treatments, and are highly tumorigenic, characteristics that make these cells, at least in part, responsible for the high mortality of GB. The TP73 gene is a transcription factor that belongs to the p53 family. P73, and most specifically, the TAp73 isoform, was initially described as a tumor suppressor, similar to p53. However, contrary to p53, TAp73 is not usually found mutated in cancer. Indeed, many human tumors, including GB, display high p73 expression, suggesting a pro-oncogenic function. P73 is involved in several developmental and homeostatic processes, and some of these functions could be responsible for the pro-oncogenic role of TAp73. One of the most studied p73 function is its role in the central nervous system development. TAp73 is an essential regulator for the maintenance of stemness of neural stem cells, which share many common features with GSC. Considering this, our working hypothesis was that p73 could have a relevant function in GSC biology, and thus, be a potential therapeutic target against GB. Inactivation of the TP73 gene in GSC using the CRISPR/Cas9 gene editing system demonstrated that this gene is an important regulator of GSC cell growth and stemness. Next, we optimized a screening system that allow the identification of a novel natural compound from the collection of natural compounds of *Biomar Microbial Technologies* with TAp73 inhibitory capacity in GB cells. This compound presented a higher cytotoxic effect on GB cells than the drugs commonly used for GB treatment, and showed specificity against GSC. Moreover, we demonstrated that this treatment was able to inhibit invasion capacity and stemness potential in GSC. Transcriptomic analysis suggested that these effects could be, at least in part, due to the capacity of this novel natural compound of inhibit TAp73 expression in GSC. Thus, considering all the above, we conclude that we have identified a TAp73 novel natural compound that is a good candidate treatment against GB.

Superbug and a host hunt

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Keywords: *Staphylococcus aureus*, intracellular, bacteria, resistance, host-pathogen interactions

Staphylococcus aureus is considered a major human pathogen worldwide and the emergency of multidrug-resistant strains, such as methicillin-resistant *S. aureus* (MRSA), represents a severe issue for healthcare providers (Ippolito *et al.* 2010). As an intracellular pathogen, MRSA is able to invade and survive within different types of mammalian cells (Fraunholz & Sinha, 2012). Intracellular survival may allow *S. aureus* to elude host immune responses and antibiotic treatments (Lehar *et al*, 2015). In contrast to the virulence factors involved in MRSA pathogenesis, which have been widely studied, host factors and metabolic pathways employed by MRSA, remain mostly unknown. Since invasion and intracellular proliferation are closely connected to the host cell metabolism for most intracellular pathogens, it is likely that MRSA interferes with some pathways or regulatory networks of the host cell metabolism to trigger primary carbon and nitrogen sources. This study aims to investigate interactions between MRSA and mammalian cells during intracellular survival to identify novel host-targeted therapies against MRSA infections.

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ORAL COMMUNICATIONS

O1

Variants in *CDKN1C* gene and Beckwith-Wiedemann Syndrome: Bioinformatic review

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Keywords: *CDKN1C* gene, Genetic Variants, Beckwith-Wiedemann Syndrome

Introduction: The *CDKN1C* gene is a negative regulator of cell proliferation involved in growth regulation. Underexpression of this gene underlies the overgrowth phenomena often observed in cases of Beckwith-Wiedemann Syndrome (BWS). Pathogenic variants in the maternal allele of *CDKN1C* gene are a BWS-associated cause when there are no changes in the genomic imprinting. The sequencing of the *CDKN1C* gene is frequently neglected in the diagnostic workup. Objective: Genotype-phenotype correlations regarding the existence of pathogenic variants and clinical traits suggestive of BWS, and the development of a descriptive compilation of all pathogenic/likely pathogenic variants in *CDKN1C* gene associated with BWS. Results and Discussion: Presently, more than 100 pathogenic/likely pathogenic variants in the *CDKN1C* gene are associated with BWS. According to ACMG guidelines, there are some discrepancies in the information published in the databases consulted, highlighting the requirement of continuous database updates as well as the implementation of functional and segregation studies to eliminate ambiguities regarding variants of uncertain significance.

Acknowledgements: *Grant References: UIDB/00215/2020; UIDP/00215/2020; LA/P/0064/2020.*

O2

Chemopreventive potential of a *Santolina chamaecyparissus* aqueous extract in a rat model of breast cancer

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Keywords: mammary cancer, MNU, natural compounds, Santolina, Wistar rats

Breast cancer is the most often diagnosed cancer worldwide, being a leading cause of death in women. *Santolina chamaecyparissus* L. has been shown to inhibit cancer cells' proliferation, especially the human breast adenocarcinoma cell line. This study assessed the ability of a *S. chamaecyparissus* aqueous extract (SCE) to prevent mammary cancer induced by *N*-methyl-*N*-nitrosourea (MNU) in female rats. UTAD's ORBEA approved this study (834-e-CITAB-2020), which included 28 female Wistar rats divided into four groups (*n*=7/group): Control, IND, SCE and SCE+IND. SCE was supplemented in drinking water (120µg/mL) and replaced every 3 days due to the compounds' stability. A total of nineteen compounds were identified in the extract, being myricetin-*O*-glucuronide and 1,3-*O*-dicaffeoylquinic acid the most abundant. At 50 days of age, MNU (50mg/kg) was administered by intraperitoneal route. Tumour development was monitored weekly. After twenty-one weeks, animals were sacrificed by ketamine/xylazine overdose and tumours were collected for histopathological and gene expression analysis. The transcript levels of four genes (*PCNA*, *VEGF*, *ER-α* and *ER-β*) were assessed by qRT-PCR, with relative expression levels calculated by the ΔCT method and *GAPDH* used as a housekeeping gene. Results showed that the first tumour appeared in SCE+IND six weeks after the first one appeared in the IND group. Tumour incidence in SCE+IND (29%) was lower than in IND (57%). Gene expression analysis revealed that only *VEGF* transcript levels were significantly altered, being lower in SCE+IND (*p*=0.016). Histopathological analysis revealed that the tumours from IND group were all classified as being malignant lesions, while only two lesions (in a total of six) from the SCE+IND tumours were malignant. In conclusion, SCE appears to be a promising chemopreventive agent for breast cancer, as it reduced tumour incidence and latency and altered tumour gene expression and histopathology in a rat model of breast cancer induced by MNU.

O3

Characterization of multidrug-resistant *Escherichia coli* as a zoonotic pathogen of wild birds

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Keywords: *Escherichia coli*, multidrug resistance, wild birds, aminoglycosides, zoonoses

Multidrug-resistant strains of *Escherichia coli* are commonly found in diverse species of wild birds. It's of extreme importance the continuous study of resistant bacteria in different animals and ecological niches due to the increase of zoonoses. In this study, 32 *E. coli* strains were isolated from fecal samples of 8 wild bird species using selective and differential media. The susceptibility test was performed following the Kirby and Bauer method testing 16 different antibiotics according to EUCAST guidelines. Extended-spectrum β-lactamases (ESBL) production was tested by double-disc synergy test and resistance genes were detected by Polymerase Chain Reaction (PCR). All strains were resistant to ampicillin, amoxicillin-clavulanate, gentamicin, amikacin, and tobramycin, and susceptible to cefoxitin, cefepime, ceftazidime, aztreonam, and meropenem. No ESBL were detected. The most frequent genes detected were *aadA1*, *aac(3)-II*, *sul1* and *sul3*. Thus, the results highlight a high percentage of resistance to penicillins and aminoglycosides antibiotic classes which is expected since these two classes are frequently used in human and veterinary medicine. A better use and control of antibiotics is urgent to prevent the emergence of resistant bacteria in wild animals.

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O4

Identification of *Staphylococcus aureus* isolated from samples obtained from food and handlers and detection of virulence genes

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Keywords: *Staphylococcus aureus*, food poisoning, enterotoxins

Staphylococcus aureus is a Gram positive, human commensal bacterium. However, it is considered an opportunistic microorganism, associated with several invasive infections such as staphylococcal food poisoning. Its efficiency in causing infections depends of the amount of virulence factors, namely toxins. In the present work, samples obtained both from food and handlers were analysed to verify the presence of *S. aureus* and then, some virulence genes were studied: *hla* (hemolysin α), *tsst1*, *sed*, *seg* and *sei* (enterotoxins). By amplification of *nuc* gene (thermonuclease), the presence of *S.aureus* was confirmed in all analysed samples. The virulence profile of the 12 samples obtained from foods allowed to identify 4 of the 5 analysed genes in 10 samples while 3 virulence genes were presented in 2 samples. Concerning to the 12 samples collected from handlers, 11 of them presented 4 virulence genes and 1 sample presented 3 genes. Since these samples were collected in bars and canteens that serve food directly to the population, the presence of these genes in the samples is worrying. Raising awareness on the subject, for proper hygiene by those who make these foods, is extremely important to avoid contamination that can result in serious food poisoning.

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O5

Prevalence, antibiotic resistance and genotypic characterization of *Staphylococcus aureus* isolated from meat and meat products

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Keywords: *Staphylococcus aureus*, meat, antimicrobial resistance, genotypic resistance, genes

Staphylococcus aureus is a major bacterial species that can be a potential cause of foodborne diseases, having an important impact on public health. This study aims to provide an overview of *S. aureus* isolated from fresh meat and meat products from the North of Portugal, as well as its antibiotic resistance profiles. A collection of 75 samples of meat preparations was undertaken, from retail shops, including fresh meats (minced meat, meatballs, hamburgers, fresh sausage, meat breading and skewers) and meat-based products ("*alheira*" and "*moura*"). *S. aureus* strains were identified using morphological and molecular (*nuc* gene) methods. Antibiotic resistance of *S. aureus* was determined using the Kirby-Bauer disk diffusion method. The presence of antimicrobial resistance genes was investigated by PCR. The overall prevalence of *S. aureus* among screened samples was 12%. A higher number of the isolates were found resistant to benzylpenicillin and tetracycline (50% and 42.9% respectively). Chloramphenicol was resistant in 37.7% of the samples, followed by tobramycin (28.6%) and kanamycin (7.1%). There was no resistance registered to cefoxitin, linezolid, erythromycin, clindamycin, fusidic acid, trimethropim-sulfamethoxazole nor mupirocin. The *bla*Z gene, conferring resistance to penicillin, and the *tet*K gene, conferring resistance to tetracycline, were identified, recording the highest percentage (both with 28.57%), followed by *ant*(4')-Ia and *aph*(3')-IIIa (aminoglycoside resistance) (with 14.28% and 7.14%, respectively), while no isolates harboured the *aac*(6')-*aph*(2''), *tet*M, *tet*O, *tet*L, *cat*pC194, *cat*pC221 or the *cat*pC223 genes. Collected data reinforce the need to consider meat products as potential vehicles of *S. aureus* transmission through the food chain, highlighting the importance to establish food safety practices for food handlers.

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O6

High prevalence of ESBL producing *Klebsiella* spp. in surface waters

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Keywords: ESBL; *Klebsiella* spp.; surface waters; antimicrobial resistance; public health

In recent years, *Klebsiella* spp. has emerged as a major public health threat mainly due to its increasing prevalence in healthcare-associated infections caused by multidrug-resistant strains producing extended-spectrum β-lactamases (ESBL). This pathogen can survive as a commensal and can be transferred to humans and animals. The efficacy reduction of antimicrobial therapy, or failed treatment, caused by the resistance conferred by these enzymes, is one of the main concerns. Therefore, we aimed to investigate the prevalence of ESBL producing *Klebsiella* spp. in surface waters. Fifty samples were collected from different surface waters in the North of Portugal, of which twelve (n=12; 24%) were positive for *Klebsiella* spp. The antimicrobial susceptibility was performed by the Kirby-Bauer disc diffusion method against 11 antibiotics. The screening of phenotypic ESBL production was carried out by the double-disk synergy. ESBL production was detected in 6 (50%) out of 12 *Klebsiella* spp. isolates. Isolates showing resistance to three or more antibiotic classes were considered as multidrug-resistant (MDR), thus in this work 3 (25%) of 12 isolates were categorized as MDR. Lastly, high rates of antibiotic resistance were observed among these isolates for cefotaxime (n=5; 41,7%). Monitoring the evolution of the ESBL situation and applying a One Health approach is essential to keep this problem under control. In addition, the development of novel antimicrobials is required for successful treatment along with infectious disease control.

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O7

Effect of potassium and magnesium on the expression of genes related to cell wall mechanisms in sweet cherry

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Keywords: Cracking Mitigation, Potassium, Magnesium, qRt-PCR

Sweet cherry's (*Prunus avium* L.) surface integrity has a significant influence on its quality, making cracking a critical economic issue since a crop becomes unprofitable with a cracking rate of just 20–25%. Many strategies have been developed to mitigate fruit cracking, one of them the pre-harvest application of foliar sprays, like potassium (P) and magnesium (Mg), which have been reported as essential nutrients in maintaining the structural integrity and stability of cell walls, making the cherries more resistant to cracking. The objective of this study was to investigate how the pre-harvest foliar application of potassium (50 g/hL and 100 g/hL) and magnesium (125 g/hL and 250 g/hL) as well as a combination of both nutrients (100 g/hL of K and 250 g/hL of Mg) in sweet cherry trees affects the expression of genes related to cell wall mechanisms, such as *PaExp1*, *PaExp2*, *PaXTH*, *PaB-Gal*, *PaEG*, and *PaCYP78A9*, during two different phenological growth stages: green/red and ripe phases. Thus, using fruits collected from different treatments, total RNA was extracted from the fruit's exocarp, followed by cDNA synthesis. Posteriorly, gene expression was studied by qRT-PCR. The results showed differential levels of expression between the two growth phases. In general, for all genes, during the green/red stage, a higher expression was observed in cherries treated with the combination of both nutrients, while during the ripe stage, a higher expression was found in cherries treated with 250 g/hL of Mg.

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O8

Unveiling the role of transposable elements in bats' genome evolution

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Keywords: Transposable elements, bats, genome evolution

Transposable elements (TEs) consist of genetic sequences capable of mobilising in genomes, thus being considered one of the major drivers of genomic diversity. Constituting one-third to one-half of mammalian genomes, TEs significantly impact genome size, structure, and organisation, as well as their host's gene expression. Bats constitute the most diverse group of extant mammals, being *Myotis* one of the most diverse genera among the Chiroptera order. The 103 species that make part of this genus present a global distribution, showing a great adaptability to different environments and habitats. The genomic diversity found in this group of species has been associated with recently found TEs' activity, suggesting that the mechanisms underlying these elements transposition could strongly contribute to unveil the events behind genomes' remodelling and speciation process. Through the isolation, molecular analysis, and physical mapping of different classes of TEs (*LINE-1*, *Ves4*, and *Helitron-1*), we sought to establish a relationship between the distribution and transposition of these sequences in bat genomes and their karyotypes diversity. So far, our study has shown a dispersed distribution pattern of these elements along the genomes of two *Myotis* species, apart from *LINE-1*, which showed block accumulations in some chromosomal regions. On the other hand, *Ves4* appears to be the TE that shows a greater sequence conservation, being *Helitron-1* the one that presents greater interspecific variability. In that sense, we intended to propose some hypothesis concerning these sequences and their association with the genome diversity of the species under analysis.

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O9

Karyotypic analysis of Portuguese chiropterans: a step towards the genomic characterization of these model organisms

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Keywords: Bats, Evolution, Karyotype, Zoo-FISH, Ribosomal DNA

Bats belong to a unique and diverse order, Chiroptera, among the mammals with a worldwide distribution. Their genome and its evolution are, for various reasons, an important matter of study. Besides the increase of the scientific knowledge at its most fundamental basis, the understanding of their immune resistance to a range of pathogens that can cross the species barrier and the potential of these animals as model organisms to evolutionary studies are strong reasons for a closer “look” at the genomic and cytogenomic levels.

Here we present some preliminary results on the genomes of two bat species from the Iberian Peninsula: *Myotis blythii* and *Myotis daubentonii* of the *Vespertilionidae* family. With this work we intend to contribute to the knowledge of the evolutionary dynamics of some genomes of this group, using classical and molecular cytogenetic techniques such as G-banding, physical map of ribosomal genes and comparative chromosomics using the human genome as reference, in order to establish phylogenetic relationships between the species in analysis, understand their genomic evolutionary pathways and ultimately, hypothesize their ancestral karyotype.

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O10

Optimization and validation of *Corynebacterium glutamicum* scale-down processes

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Keywords: Scale-down, *Corynebacterium glutamicum*, fermentation, aminoacids

Scaling up is a challenging task in the microbial fermentation processes. Several inhomogeneities and factors (e.g., nutrients or oxygen transfer) result in performance losses and development delays. Thus, scale-down approaches have been proposed as suitable solution to mimic these issues in a smaller scale to tackle the optimization in a highthroughput way. *Corynebacterium glutamicum* is an industrial non-pathogenic and fast-growing microorganism recognized as an aminoacids producer, which is being used in the present research as a workhorse for update the scale-down *versus* traditional flask fermentation.

Aimed to define the scale-down *C. glutamicum* culture conditions, different parameters, such as: media, aeration, inoculum, etc. have been tested to implement the process.

Acknowledgements: *Horizon Europe project, ESTELLA (Project no.: 101058371).*

O11

Unravelling the FurC regulon in the cyanobacterium *Anabaena* sp. PCC 7120 and its role in the control of processes dependent on carbon/nitrogen balance

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Keywords: cyanobacteria, ferric uptake regulator, carbon metabolism, nitrogen metabolism, C/N balance

Cyanobacteria are photosynthetic prokaryotic microorganisms able to fix atmospheric nitrogen and CO_2. In the cyanobacterium *Anabaena* sp. PCC7120, FurC is a global regulator that controls processes such as nitrogen metabolism, photosynthesis and oxidative stress. In this work a putative FurC DNA-binding sequence was searched in the whole genome of *Anabaena*, allowing the identification of nearly 20 novel FurC targets. Some of these targets play relevant roles in nitrogen fixation and carbon assimilation processes suggesting a key regulatory role of FurC in the control of C and N metabolisms. For this reason, some signaling molecules which inform about the C/N balance were tested as ligands of FurC protein, revealing that 2-oxoglutarate (2-OG) was able to bind to FurC. Taken together, these data suggest that apart from redox signals and metal availability, FurC could also respond to C/N ratio, being an additional player for the harmonization of carbon and nitrogen metabolisms.

O12

Addition of non-digested gluten to a digested-gluten containing culture medium does not affect bacterial density of intestinal communities derived from gluten metabolism

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Keywords: communities, gluten, intestinal microbiota, RT-PCR, bacteria quantification

Gluten can cause Celiac Disease in predisposed individuals. Celiac Disease has been linked to intestinal microbiota dysbiosis. Two media for studying intestinal communities derived from gluten metabolism were designed. Medium G1 contained 0.5 % gluten peptone. Medium G2 contained 0.5 % gluten peptone and 0.2 % undigested gluten. The objective of this study was to assess if different gluten concentration in G1 and G2 affected microbial density of communities. Fecal communities of 3 healthy individuals were maintained for 15 passages by daily subculturing in G1 and G2. Genomic DNA was extracted from cultures of passages 0, 4 ,6 , 8 and 15. Bacterial concentration was studied via RT-PCR of 16S rDNA. Marginal means analysis for 4 time points based on a mixed effect generalized model showed bacterial density differences between G1 and G2 at only one time point. In conclusion, G1 and G2 can harbor communities with similar bacterial concentration.

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O13

How could nettle infusions protect beans from halo bligth disease?

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Keywords: *Phaseolus vulgaris, Pseudomonas syringae* pv. *phaseolicola*, *Urtica dioica,* oxidative damage, biotic stress

The treatment of common bean (*Phaseolus vulgaris* L.) *in vitro* plants with nettle (*Urtica dioica* L.) aqueous preparations (U-PBP) reduced the symptoms of halo blight disease caused by *Pseudomonas syringae pv. phaseolicola* (De la Rubia *et al*., 2022). Also U-PBP negatively affected the Pph growth evaluated by bioassay. The objective of the present work was to probe if nettle infusions (Ui) –obtained by autoclaving and filtering U-PBP- have similar effects and, if so, what it could be their way of action. The results suggest that Ui reduced *in vitro* Pph growth, at least at the highest concentrations, and that Ui pretreatment of bean plants prevented the foliar oxidative damage caused by Pph infection. Moreover, it seems that Ui effects could be independent of the triggering of plant defense responses, because tissue pretreatment with Ui reduced the H2O2 production in bean leaf discs, even in the presence of defense elicitors as flg22. These preliminary results point out that the infusions have a high antioxidant capacity.

O14

Redox-sensitive control by thioredoxin of the ferric uptake regulator Fur from the strict anaerobe pathogen *Clostridioides difficile*

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Keywords: *Clostridioides difficile*, ferric uptake regulator, transcriptional regulation, redox sensing, thioredoxin

Clostridioides difficile is a strictly anaerobic pathogen that produces gastrointestinal diseases. As iron is an essential element for this microorganism but high levels of iron can be lethal, iron uptake is tightly controlled at transcriptional level by the ferric uptake regulator Fur. Apart from iron homeostasis, Fur from *C. difficile* (*Cd*Fur) is thought to play key roles in virulence and antibiotic resistance but little is known about its mechanism of action. In this work we have found that *Cd*Fur DNA binding activity is specific under reducing conditions and that it is hindered by the presence of metal ions. We have also determined that this regulator binds to the promoter regions of genes involved in iron homeostasis and that its activity is modulated by thioredoxin through a reversible redox mechanism that controls its oligomerization state. Taken together, these results suggest that the activity of *Cd*Fur might undergo redox-dependent apo-regulation under iron deficiency.

O15

Understanding the genetic control of heat-induced seed coat browning in cowpea (*Vigna unguiculata*)

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Keywords: cowpea, genome-wide association studies (GWAS), seed quality, single nucleotide polymorphisms (SNPs), *Vigna unguiculata*

Cowpea (*Vigna unguiculata*) is a climate-resilient legume crop that is very important for food security, especially in sub-Saharan Africa. Consumer acceptance of cowpea seeds is essential; heat-induced browning of seed coats (Hbs) occurs in certain cowpea varieties and it is associated with reduced seed quality and commercial value. The objective of the study is to identify loci and genetic markers associated with this trait that could be used in breeding programs.

A diverse germplasm collection including 368 accessions has been genotyped and used for genetic mapping of *Hbs* via genome-wide association studies (GWAS). A significant SNP associated with heat-induced browning has been identified on chromosome 8 (Vu08), in a region that overlaps with a *Hbs* QTL (quantitative trait locus) identified previously. The high mapping resolution achieved in this study has allowed the identification of a strong candidate gene encoding an (R)-specific enoyl-CoA hydratase. Next steps include sequencing of the candidate gene and the development of a PCR-based marker for marker-assisted selection.

Acknowledgements: *The development and genotyping of the germplasm collection used in this work was funded by the Feed the Future Innovation Lab for Climate Resilient Cowpea (AID-OAA-A-13-00070).*

O16

Effect of liquid extract from *Ulva Lactuca* algae, in the alleviation of salt stress on crops of the common bean, *Phaseolus vulgaris L.*

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Keywords: Biostimulant, *UC* algae, common bean, enzymatic activities, salt stress

The use of seaweed extracts has been proven to provide positive effects on enhancing crop quality and resistance to abiotic and biotic stresses. Salinity is one of the major abiotic stressors which limits the yield of major crops. The macroalgae Ulva lactuca (UL) produced naturally creates a huge problem due to its stranding on the local coast, congestion, decomposition, and release of bad smell. To contribute to mitigating this environmental problem UL algae were chosen as a source of bio-stimulant for experiment purposes on common bean plants (Phaseolus vulgaris L.) with the absence and presence of salt stress with two levels (34,2 mM NaCl and 68,4 mM NaCl) also with and without UL extract (ULE) treatment using three levels (1%, 3% and 6%). Results showed that ULE has a great benefit on common bean growth both in the absence and under salt stress conditions especially at 3%. These results proved that UC extract significantly improved the activities of enzymes involved in the activation of the carbon-nitrogen, the antioxidant enzymatic system, and osmolytes (PEPc, GPX, SOD, GST, GS, GDH, GR activities, IAA content,…etc.) and also improved the chlorophyll and the morphological parameters, thus, we can say that ULE could be successfully used to overcome the negative effects of salt stressors conditions for common bean plants for example in areas where water for irrigation contains some salt content.

O17

Gene deletion in *Rhodococcus fascians* and phenotypic evaluation of a mycoredoxin-deficient mutant under oxidative stress

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Keywords: ROS, Gene Deletion, Hydrogen peroxide, Phytopathogen, Mycoredoxin

Oxidative stress, an imbalance between the production and detoxification of Reactive Oxygen Species (ROS), plays a crucial role in many physiological processes, including fighting bacterial infections. To protect against oxidative stress, bacteria use a wide variety of strategies such as the glutathione metabolism and the thioredoxin system. Mycoredoxins are critical for redox homeostasis in *Actinobacteria*. This study presents a methodology for generating gene mutations in the plant pathogen *Rhodococcus fascians*. Using this method, a mutant lacking three mycoredoxin-coding genes was generated. The mutant strain's response to oxidative stress was evaluated, and unexpectedly, it showed higher resistance facing hydrogen peroxide-induced oxidative stress.

POSTERS

P1

Egg-straordinary Nutrition: A Biuret Method Comparison of Protein Content in Hen Eggs

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Keywords: Eggs, protein, Biuret, spectrophotometer

Proteins are macronutrients responsible for body functions such as tissue building and repair. Excessive protein intake can lead to renal overload and increase the risk of long-term kidney damage, and can cause dehydration and disrupt the balance of important nutrients in the body, leading to potential health problems.

In this study, we aimed to determine if there were significant differences in the protein content of hen eggs from different sources. We compared eight different sources of hen eggs, including various commercial brands as well as eggs obtained from local farmers.

After analyzing the data collected from our study, we discovered significant differences in the protein content of eggs. This finding is important as it may impact the nutritional value of eggs consumed by individuals and could have implications for those with specific dietary requirements or health concerns. Further research is needed to investigate the underlying factors contributing to these differences.

P2

Karyotypic evolution analysis of two bat species (*Chiroptera*) through ZOO-FISH

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Keywords: Chromosome painting, Chiroptera, ZOO-FISH, evolution

Bats are a group of mammals belonging to the order *Chiroptera,* being the second largest order of species with a worldwide distribution, especially abundant in tropical and subtropical regions. A total of 1456 bat species belonging to 21 families have been recorded. In Portugal, there are 26 bat species, belonging to four families (*Vespertilionidae, Rhinolophidae, Miniopteridade and Molossidae*). The widespread distribution of bats habitats indicates that they had a highly successful radiation in mammalian evolution, and it is necessary to understand at the karyotype level how this evolution occurred, so it would be possible to obtain an ancestral karyotype of the species.

With this work we intend to contribute to the knowledge of the evolutionary dynamics of the genomes of two species from the Iberian Peninsula, which are, *Myotis blythii* e *Myotis daubentonii* of the *Vespertilionidae* family, analyzing their karyotypes through the construction of a partial comparative chromosomal map for each specie using the ZOO-FISH technique with 4 human chromosome-specific DNA probes, these being, HSA 14, HSA 15, HSA 18 and HSA 20.

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P3

An overview of dermatophytosis in shelters and clinics of Northeast Portugal

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Keywords: Dermatophytosis; Shelters, Clinics, Pets, Portugal

Dermatophytosis, which can cause superficial mycoses in both humans and animals by invading keratinised tissue, is considered an important zoonotic skin disease. Dermatophyte carriage data are crucial for assessing their epidemiology and designing potential control strategies. Dermatophyte species are categorised into three ecological groups: anthropophilic, zoophilic, and geophilic. Based on the new classification system, dermatophytes are classified into seven genera: *Arthroderma Epidermophyton Lophophyton, Microsporum*, *Nannizzia*, *Paraphyton*, and *Trichophyton*. Infections caused by dermatophytes are common among humans and animals, making them one of the most frequent dermatologic infections. Fur samples were obtained from 341 animals from shelters and veterinary clinics. Dermatophyte culture was performed using Dermatophyte test medium® in Petri dishes. The Petri dishes were handled under sterile conditions and incubated at 28°C for up to 21 days. The dermatophytes isolated was *Microsporum canis* (3.2%; 95% CI: 1.8–5.7%) and *Microsporum audouinii* (0.29%; 95% CI: 0–1.6%). The occurrence of dermatophytosis in this study was low. Nevertheless, this study increased the knowledge about dermatophytosis in pets in the Northeastern region of Portugal.

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P4

Unveiling *Alternaria alternata*: the hidden allergen lurking in our pets' fur

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Keywords: *Alternaria alternata*, One Health, pets

Alternaria alternata, a common species of the genus, is a ubiquitous filamentous Ascomycete fungus that can be found in a variety of environments, including soil, air, plants, and indoor spaces. *A. alternata* is clinically related to various manifestations, such as asthma, allergic rhinosinusitis, hypersensitivity pneumonitis, invasive rhinosinusitis, oculomycosis, onychomycosis and skin infections, and also allergic bronchopulmonary mycosis.

Animals are the third leading cause of allergic asthma, after mites and pollens. Young people are often allergic to animal fur, and sometimes it is *Alternaria* that is found in the animal's fur or body since it has long been identified as part of the normal fungal flora of dogs and cats. Samples were sent to the Medical Microbiology Laboratory of the University of Trás-os-Montes and Alto Douro, where cultures were performed in Potato Dextrose Agar and Sabouraud Dextrose agar and incubated at 25 ºC and 37 ºC for 3 to 7 days. *Alternaria* spp. were identified in culture in 85 animals. The occurrence in pets was 24.9% (CI 95%: 20.6-29.8%). More studies are required to understand better the relevance of the isolation of this important allergen in the pets' fur and their importance for humans in a One Health approach.

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P5

Modulatory effect of melatonin and *Akkermansia muciniphila* on the gut microbiota in an *in vivo* model of liver fibrosis

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Keywords: *Akkermansia muciniphila*, gut microbiota, liver fibrosis, melatonin

Background: Early liver fibrosis is a global health concern that has been associated with gut dysbiosis. Antioxidant, anti-inflammatory and anti-fibrogenic effects have been related to melatonin. *Akkermansia muciniphila* has been named as a new generation probiotic. Aim: To evaluate the effect of melatonin and *A. muciniphila* on gut microbiota in an *in vivo* model of early liver fibrosis. Methods: C57BL/6J mice were fed with control diet (C) or Western Diet, supplemented with fructose in drinking water and injected with CCl_4 (WD) for 8 weeks. Then, they were subdivided according to diet and treatment: melatonin and/or *A. muciniphila* for 4 weeks. Gut microbiota composition was analysed. Results: WD induced gut dysbiosis showing changes at phylum, class and genus levels. Treatment with melatonin and *A. muciniphila* changed gut microbiota composition in this group. Conclusion: Combination of melatonin and *A. muciniphila* could be linked with a modulatory effect on gut microbiota composition.

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P6

Molecular characterization of thyroid tumors of dogs – a multicentric Portuguese series

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Keywords: Comparative pathology; Canine tumours; Sequencing; Oncobiology; Thyroid carcinoma

Thyroid carcinoma (TC) is the most common endocrine malignancy in both humans and dogs. In human medicine, mutations in *BRAF*, *NRAS*, *HRAS*, *KRAS*, and in the promoter of *TERT* are key predictors of diagnosis, prognosis, and response to treatment of TC. However, the molecular dynamics of veterinary thyroid tumours is still unclear, and the potential of dogs as spontaneous models of TC remains to be clarified. Therefore, our aim was to assess if the same molecular players are present in canine thyroid tumours and if they have the same biological consequences as in humans. We collected 57 samples of formalin-fixed paraffin-embedded tissues canine thyroid tumours (5 adenomas (9%), and 52 carcinomas (91%)) and performed DNA extraction, PCR and Sanger sequencing of exon 16 of *BRAF* (n = 53), exon 2 of *NRAS* (n = 42), exon 3 of *HRAS* (n = 43), and exon 3 (n = 31) and 4 (n = 20) of *KRAS*. We detected silent variants on *HRAS* (p.(asp47=) / c.307T>C) (n = 16/43, 37%) and *NRAS* (p.(glu106=) / c.693G>A) (n = 1/42, 2.4%), but no mutations were found. Our preliminary analysis suggests that the tumorigenesis of canine TC may be triggered by different players or by different mechanisms.

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P7

Exploring natural compounds as potential alternatives to antibiotics for intracellular treatment of *Staphylococcus aureus* infections

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Keywords: nosocomial, toxins, antimicrobials, MRSA, multiresistance

Staphylococci are pathogenic bacteria that can cause nosocomial infections, and some species are known to cause severe illnesses due to their ability to produce various toxins and their resistance to multiple antimicrobials. One such species is *Staphylococcus aureus*, which is not only an extracellular bacterium but also has the ability to survive inside host cells, making it a facultative intracellular pathogen. Therefore, our focus is on combating methicillin-resistant *S. aureus* (MRSA), which has become a major concern due to its spread from hospitals to the community (community-acquired MRSA) and livestock (livestock-associated MRSA). To address the problem of antibiotic resistance, we are screening for natural compounds that can function as an alternative to conventional antibiotics and effectively combat these pathogens. Our results indicate that some natural compounds seem to increase the cell viability percentage on experiments, such as antivirals, fungicides or antibacterials.

P8

Cytomolecular characterisation of maritime pine post-fire regenerated stands

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Keywords: Fire recurrence, ISSR, mitotic cell cycle, natural regeneration, *Pinus pinaster*

Fire recurrence can impact the canopy seed bank of maritime pine (*Pinus pinaster* Ait.), influencing the post-fire natural regeneration. This work intends to evaluate how fire recurrence can affect the: (i) germination of *P. pinaster* seeds isolated from fully closed pinecones collected in stands (NE of Portugal) that burned once (A), twice (B), and three times (D); (ii) root cell division in germinated seeds; and (iii) genomic template stability (GTS) and genetic variability of the regenerated plants using inter-microsatellite (ISSR) markers; using an unburned stand (C) as a control. The lowest germination rate was found in the control stand (22.86%) and the highest (87.14%) in stand A. The mitotic index differed significantly (p<0.001) among stands, except between the control and A stands. The lowest and highest percentage values of dividing cells with anomalies (%DCA) were detected in stands A and D, respectively. The ISSRs revealed the lowest GTS (44.90%) and the highest polymorphism percentage (89.41%) in stand D (burnt three times). Overall, a high percentage of ISSR polymorphism (>72%) was assessed in each stand. Although, the control seeds showed the lowest germination rate in this work, probably due to other stress factors, the species developed strategies to protect its canopy seed bank to ensure natural regeneration. Also, the evidenced maintenance of genetic variability in the post-fire regenerated stands confers high adaptive potential helpful to cope with actual and future climate changes.

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P9

Comet assay optimization in *Drosophila melanogaster* sperm cells

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Keywords: infertility, *Drosophila*, DNA damage, sperm cells

Exposure to toxic substances in humans has increased and is a current problem nowadays. As a consequence, infertility in mammals is growing due the genome instability in germinal cells. The aim of this study is the impact of two environmental agents (temperature and tobacco smoke) on male fertility *in vivo* using *Drosophila melanogaster* as a model. The comet assay allows the detection of DNA chain breaks in eukaryotic cells and is widely used. This method allows to identify DNA damage in the sperm, therefore there is no established protocol for this study yet, and it's still being optimized. Two strains were used; one wild-type (*Oregon K, OK*), and one deficient Mus308 protein, homologue of the human POLQ DNA polymerase (*mus 308*) to increase sensibility. Virgin males (9 days old) were used, the sperm isolation and lysis were optimized and adapted. To isolate the sperm, male seminal vesicles placed in microtube, Ringer's solution is added and centrifuged. Initially, it was carried out in three different countries with different constituents, adapted from various protocols (from human sperm and human lymphocytes). In the literature it is described that the thermal stress in the male organs caused by the exposure to high temperature results from the modification of the integrity of the DNA of the spermatozoa. In the case of tobacco, it causes malformations and a decrease in sperm, leading to an increase in DNA damage. The results obtained were 338 arbitrary units for *mus 308*. For future studies, the *mus 308* assay will be repeated the assay will be performed on the *OK* strain and the application of these two agents.

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P10

Bioinformatic analysis of a familial balanced translocation

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Keywords: Cytogenetics, translocation, clinical report, diagnosis

Bioinformatics is a branch of science that involves utilizing computer technology to gather and analyze biological data, such as DNA sequences. Clinicians and scientists use databases that organize and index this biological information. In genetics, bioinformatics using databases, can be used to identify what type of DNA sequences are present in chromosomal breakpoints, allowing to understand the clinical implications that a chromosomal rearrangement, such as a translocation, may have. Generally, reciprocal translocations don't have any phenotypic implications. However, gene disruption or altered gene expression can happen, and in these situations, bioinformatics can be an important tool to understand the phenotype. A cytogenetic study was requested on the grandparents and an aunt due to the presence of a familial translocation (detected in the grandchild and also present in the father). There was no history of miscarriage, but two grandmother relatives had intellectual disability of uncertain etiology. All cytogenetic cultures and methods were performed according to the protocols established in the Genetics Laboratory of the Trás-os-Montes e Alto Douro Hospital Centre. Cytogenetic analysis was accomplished according to the guidelines present in ISCN (2020). Bioinformatic tools such as the UCSC Genome Browser database and Ensembl were used to study the translocation's breakpoints and to identify which genes or repetitive elements may be present. Cytogenetic analysis revealed the reciprocal translocation t(4;16)(q21.1;q22) in the grandmother and aunt. The bioinformatic analysis showed that the breakpoints have a lot of repetitive DNA sequences. The 16q22 region is even described as being susceptible to chromosomal breakage. This may explain the absence of clinical implications in carriers of this balanced translocation. According to the literature review, no reference was found involving the chromosome breakpoints of this translocation. This report highlights the importance of bioinformatic databases as a very useful resource that helps to understand the translocation's stability, allowing an efficient genetic counselling.

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P11

Estimating the variability and genetic structure of roe deer Iberian population using mtDNA

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Keywords: roe deer, populations, genetic structure, mitochondrial DNA, haplotype

Capreolus capreolus, commonly known as roe deer, presents a widespread geographical distribution. Over the last four centuries, there has been a decrease in size as well as in distribution of roe deer on account of several fluctuations in populations of this animal, mainly due to anthropogenic activities, like hunting. These alterations aggregated with their translocations may influence the genetic structure, diversity, and fitness of populations. For this reason, the study of the diversity and genetic structure of this deer is of great importance. Mitochondrial DNA (*mtDNA*) has been used in several studies in cervids, as a molecular marker, with a high percentage of success, since it is easy to amplify, is cheap to work, and has lower recombination. This work aims to analyze the genetic diversity and structure of a roe deer population from the Iberian Peninsula using a specific *mtDNA* conserved region. The sequencing of 28 samples from roe deer demonstrated the presence of six haplotypes already identified in other populations of roe deer in Iberian Peninsula using *mtDNA*. In conclusion, this type of analysis is quite relevant for the management, conservation, and restocking programs of this species.

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P12

Forensic DNA Phenotyping: Advancements, Application, and Ethical Considerations

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Keywords: Forensic DNA Phenotyping, Epigenetics, environment, SNPs, STRs

Forensic DNA Phenotyping (FDP) is a technique used to predict the appearance of suspects or victims from biological material found at crime scenes.

Epigenetics can be described as changes in gene expressions without altering the DNA base sequences and resulting in phenotype changes. Therefore, by combining conventional techniques of Short tandem repeats (STRs) and Single Nucleotide Polymorphisms (SNPs) variation analysis with epigenetic modifications, FDP has become useful in forensic analysis. It is often used to determine an individual's sex, identify tissue samples, and distinguish between monozygotic twins. It is expected that, with the advancements in science and in order to understand how epigenetic modifications affect gene expression regulation, more genetic markers revealing physical characteristics and diseases will be discovered.

Epigenomics emerged from the necessity of analyzing the whole Human Epigenome. Epigenomic studies made possible to determine that lifestyle choices have an impact on the Human epigenome and may vary from person to person, hence making it necessary that companies that specialize in FDP have epigenetic variations in regard. It was also disclosed that traces that are highly influenced by the environment have lower accuracy rates than traces that are highly heritable. In addition to being useful for investigation, FDP raises several ethical issues, in particular, concerns about privacy protection and discrimination prevention arose when using FDP, as it utilizes markers located within coding regions which can disclose information for genetic predisposition to certain diseases and possibly reveal unique physical characteristics. As this technique is not 100% accurate (apart from sex determination), it already led to critical investigation mistakes, for example, geographical markers only estimate the continent a person originates from and not the exact country. FDP uses DNA found at the crime scene which can lead to the identification of individuals who are not involved in the crime but for some reason their DNA is present at the location. Thus, it is required a careful analysis based on other evidence in addition to FDP. Concluding, although ethical issues such as privacy and accuracy remain a concern, FDP is a powerful tool for narrowing down the suspect pool.

P13

Relation between *Fusobacterium nucleatum* and colorectal cancer

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Keywords: Colorectal Cáncer, microbiota, Fusobacterium, miRNA

Colorectal cancer (CRC) shows the third highest incidence amongst all cancer cases. Microbial flora happens to play a big role in CRC development and promotion.

The consensus used to be that Microbial flora played a role as either a shield or an indicator of cancer: if the bacterial population decayed, cancer is more prone to occur and develop a worse prognosis. Fact is that some bacteria thrive in cancer microenvironment, some of them even promote it.

This is the case of *Fusobacterium nucleatum*. *F.nucleatum* is a commensal opportunistic bacteria that inhabits the human oral cavity, where it can cause diseases in a wide range of severity. This bacteria can also be found infecting colon and rectum cells, where is thought to promote cancer by activating a signal chain leading to high expression levels of a particular type of miRNA. High presence of both *F.nucleatum* and this miRNAwhich are related with bad prognosis.

The goal of the study is to dig deeper into the relation between *F. nucleatum* and CRC in order to broaden its understanding.

P14

Molecular evaluation of wheat and triticale plants exposed to polyethylene glycol-induced water stress

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Keywords: Genomic template stability (GTS), inter-microsatellites (ISSRs), osmotic stress, ×*Triticosecale* Wittmack; *Triticum aestivum* L.

Concerning climate change projections, a higher frequency and severity of heat waves and drought episodes will occur. The selection of crop varieties more tolerant to drought is required to overcome the grain yield and quality decrease. In this work, we aimed to analyse the genomic template stability (GTS) in young plants of two varieties of hexaploid wheat ('Jordão' and 'Nogal') and triticale ('Fronteira' and 'Gavião') whose seeds were exposed to 0% (control treatment, distilled water), 10% (-0,4 MPa) and 20% (-0,8 MPa) of polyethylene glycol (PEG) during three weeks (encompassing the germination stage and seedling emergence), using inter-microsatellites (ISSRs).

Based on the molecular data, the GTS per species, variety and treatment was determined. The bread wheat variety 'Nogal' demonstrated the highest GTS value. The triticale variety 'Gavião' presented the lowest GTS, indicating higher molecular instability in response to the induced water stress. However, this was the only variety whose seeds germinated upon exposure to 20% PEG, which might suggest a higher tolerance of triticale to water stress than bread wheat.

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P15

Genotoxicity studies in mice genetically modified for HPV16 and exposed to an *Aloysia citrodora* extract

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Keywords: *Aloysia citrodora*, HPV16, mice, comet assay, genotoxic

Aloysia citrodora, also known as lemon verbena, is used in traditional medicine to prepare infusions due to its antispasmodic, digestive, sedative, and antipyretic properties. In addition to these characteristics, it also has antioxidant and antimicrobial properties. This study aims was to evaluate *in vivo* genotoxic effects through the comet assay in *wild-type* mice and mice genetically modified for HPV16 during ingestion of a lemon verbena extract. The experimental protocol lasted 28 days authorized by the ORBEA (UTAD) and approved by DGAV (014139).The lemon verbena infusion was prepared fresh every 48 hours and made available as drinking water. Thirty female mice were divided into six groups (n=6): GI (WT, control), GII (HPV, control), GIII (WT, 0.013g/ml), GIV (HPV, 0.006 g/ml), GV (HPV, 0.008 g/ml), and GVII (HPV, 0.013 g/ml). Body mass, food and water consumption and *humane endpoints* were recorded weekly. At the end of the experimental assay, all animals were sacrificed by anaesthetic overdose (FELASA recommendations) and blood was collected for the comet test. Mean food and food intake was higher in the HPV groups (G2, G4, G5 and G6). The mean body mass of the animals increased throughout the trial, with no statistically significant differences between groups. Regarding the humane endpoints, we obtained higher values in the HPV groups, with statistically significant differences between groups G2 and G4, and groups G4 and G5 ($p<0.05$). Regarding the comet assay, baseline damage and total damage were higher in groups G3, G4, and G6. About oxidative damage, they were higher in groups G2 and G4, but no statistically significant differences were observed. According to our results, the *Aloysia citrodora* extract interfered in the variables studied, seems to be safe in terms of animal welfare but other studies should be conducted to understand its effects in animals' healthy, such as liver oxidative stress and histopathology studies. Regarding DNA damage the concentrations 0.006 and 0.013 seemed to induce basal DNA damage, whereas concentrations 0.008 and 0.013 seemed to provide a reduction in oxidative damage to DNA, indicating an antioxidant effect.

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P16

Toxicological activity of *Quercus ilex* beverage in mice genetically modified for HPV16

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Keywords: *Quercus ilex*, HPV16, mice, toxicological, extract

Quercus ilex is a very common holm oak in Portugal, namely in the Montado Alentejano. It should be noted that the acorns from this tree are widely used in animal feed and can also be used in the production of flour. Its potential should be highlighted due to its antioxidant, antibacterial, antitumor and antidiabetic activity. Our work aimed to evaluate the effects of *Quercus ilex* beverage in a transgenic model for HPV16, to understand its influence on liver and kidney oxidative stress parameters. The study was authorized by the ORBEA (UTAD) and DGAV (014139). Thirty-six female mice were used. The beverage was freshly prepared every two days and made available as drinking water. Thus, mice were divided into six groups (G1 to G6, n=6): G1 (WT, control), G2 (HPV16, control), G3 (WT, 0.09g/ml), G4 (HPV16, 0.03g/ml), G5 (HPV16, 0.06g/ml), G6 (HPV16, 0.09g/ml). For 28 days, body mass, food, and water consumption, and humane endpoints were recorded. At the end of the test, the animals were sacrificed, and samples were collected, such as liver and kidneys, to perform oxidative stress determinations. Animals' body weight gain was statistically significant different between groups 2 and 5 ($p<0.05$). The water consumption and beverage were higher in transgenic animals from G2, G4, G5 and G6. Food consumption was higher in transgenic animals from G2, G4, G5 and G6. There are statistically significant differences in ROS in the kidneys for G2 compared to G4 and G5. Regarding, superoxyde dismutase activity there aren´t statistically significant differences. In the liver, catalase activity G2 was statistically higher than G1, G5 and G6. And in the remaining parameters (GSH, GPx, GR, GST, GSH, GSSG, LPO and carbonyls) there are no statistically significant differences between groups. In conclusion, according to our results, the administered beverage does seem to interfere with the presented oxidative stress indicators. However, more studies are being processed to better understand the relationship between the extract under study and HPV16.

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P17

Antimicrobial resistance and biofilm formation of *Klebsiella* spp. in healthy rabbits for human consumption

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Keywords: *Klebsiella,* antibiotic resistance, biofilms

Klebsiella spp. strains have been found to exhibit resistance to multiple classes of antibiotics. This fact together with their ability to form biofilms are making the infections very difficult to treat. The investigation of pathogens in animals for consumption, such as rabbits, may allow the implementation of effective strategies to mitigate the impact of these microorganisms on public health. Therefore, the main goal of the work was to characterize the antibiotic resistance and biofilm formation of *Klebsiella spp.* isolated from rabbits for consumption. The antimicrobial susceptibility testing was performed by the agar disk diffusion method. The Extended-Spectrum Beta-Lactamase (ESLB) phenotype of each isolate was also investigated. Biofilm formation of all strains was performed by the microtiter assay. The antimicrobial susceptibility testing showed that most of the strains were resistant to cefoxitin and tobramycin, but susceptible to meropenem and ciprofloxacin. Two isolates were positive for the ESBL test. In addition, all strains isolated from rabbits for healthy consumption produced biofilm, with a percentage mean of biomass production of 66.95%. These findings highlight the potential risks associated with consuming contaminated rabbits' meat and the importance of implementing effective strategies to reduce contamination and mitigate the impact of these microorganisms on public health.

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P18

Portuguese founder mutation c.156_157insAlu in *BRCA2* gene: a historical and practical perspective

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Keywords: c.156_157insAlu *BRCA2* variant, Hereditary breast/ovary cancer, NGS

Alu elements are retrotransposable sequences that are the most abundant mobile elements in the human genome. Association between Alu elements and genetic instability has been described in the literature for some cancer genes. Hereditary breast/ovary cancer (HBOC) is an autosomal dominant syndrome that can be caused by pathogenic/probably pathogenic variants in *BRCA1/2* genes. In Portugal, breast cancer is the most frequent type of cancer, accounted for 11.6% of new cancer cases in 2020, while ovary cancer represented 0.93% of them. In 2009, a team from Oporto demonstrated that c.156_157insAlu in *BRCA2* gene contributed to inherited predisposition to breast/ovary cancer in families from northern/central Portugal, having a founder effect in this region. This variant is an in-frame insertion that causes exon 3 skipping, and is considered pathogenic by leading to protein size change with loss of function. At Genetics/Andrology laboratory of CHTMAD, next generation sequencing (NGS) is performed to search for germline variants when there is a suspect of HBOC. Targeted sequencing is done using a specific panel that covers 25 genes associated with hereditary cancer. Firstly, DNA is extracted from peripheral blood, followed by library construction and target enrichment. Then, sequencing is performed in MiSeq platform from Illumina, and the results analysed and interpreted using a bioinformatic software. In the implementation of the process, a positive control for *BRCA2* c.156_157insAlu was used to assess the capacity of the platform to detect this variant. Although its capacity proven, the platform is only validated for the identification of small deletions/insertions and substitutions, so the presence of this variant in the samples has always to be confirmed by polymerase chain reaction (PCR), followed by capillary electrophoresis. The evidence regarding *BRCA2* c.156_157insAlu responsibility in HBOC was very important to establish a diagnosis in affected portuguese families which phenotypes could not be explained otherwise. Moreover, genetic testing including this variant in Portugal is essential not only to identify carriers, allowing their surveillance and counselling about reproductive rights, but also to help in the decision of the therapeutic to be used in an affected individual.

P19

Antioxidant enzymatic machinery is altered in the union tissues during tomato graft healing

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Keywords: antioxidant activities; graft healing; grafting; oxidative stress; tomato

Plant grafts have been exploited since ancient times, to combine a scion (upper part) on a rootstock (lower part) that provide a significant advantage in terms of crop yield, as is the case with tomato plants. Grafting causes a period of generalized stress and therefore the aim of this study was to know how the antioxidant enzymatic machinery functions in the union tissues during graft healing using tomato autografts. Total antioxidant capacity (TAC) and ascorbate peroxidase (A-POX); catalase (CAT); class III peroxidase (CIII-POX) and superoxide dismutase (SOD) activities were evaluated during 0–8 days after grafting in functional and non-functional grafts. Main results indicated that TAC, CAT and SOD activities showed consistent increases, especially in scion, thorough grafting; whereas A-POX was found to be irregular. Non-functional grafts revealed high levels of CIII-POX activity. Therefore, it is possible to conclude that the antioxidant enzymatic machinery is enhanced during tomato graft healing.

P20

Cracking the Egg Myth: Do Blue or Homegrown Eggs Really Have Lower Cholesterol?
A Comparative Analysis using the Lieberman-Buchard Method

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Keywords: Cholesterol, Hen eggs, Steroids, Lieberman-Burchard

Cholesterol is a vital compound in the human body. However, its excessive consumption is linked to cardiovascular issues, which cause 17.9 million deaths annually. As cholesterol is present in many foods, it is crucial to measure its levels in various products, like eggs.

The Lieberman-Burchard method is a reliable way of measuring cholesterol content. It involves several chemical reactions that turn cholesterol into a blue-green colour, which can be measured using a spectrophotometer. This study compared the cholesterol content of hen eggs from different brands and types, aiming to settle if there were differences between them.

Results showed no significantly higher cholesterol content between eggs regardless of their colour or production method. Unexpectedly, blue eggs did not have lower cholesterol content, which contradicts marketing campaigns that aim to sell them. The study highlights the importance of proper methods for quantifying cholesterol in foods to inform of healthy dietary choices.

P21

Cowpea roots: screening the most suitable drought stress tolerance genes

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Keywords: *Vigna unguiculata* L. Walp., roots, gene expression; water stress

Climate change is one of the most critical challenges for the near future, being drought a severe stress with major impact on plant development and productivity. Root system has an important role on plant physiology and its architecture may contribute to improvements of desirable traits such as drought tolerance and plants' yield. Cowpea (*Vigna unguiculata* L. Walp) is a member of Leguminosae family and native from Africa being the primary source of protein for millions of people of the developing world. This legume has a high tolerance to drought and high temperature, being important to understand the mechanisms and pathways responsible for these tolerances. The main objective of this study was to determine the most suitable genes related to drought tolerance using cowpea roots as biological material.

Two Portuguese cowpea genotypes were submitted to two drought stress conditions using polyethylene glycol (PEG, -0.75 and -1.5 bars) and a control (distilled water), for 10 days. Roots were collected from all the samples and RNA was extracted. Five genes related to drought tolerance and three housekeeping genes were evaluated by semi-quantitative PCR.

A differential gene expression was observed between drought stress treatments and control, being the up-regulation of the drought tolerance genes correlated with the severity of the drought stress. The genes *VuCPRD65* and *VuNced1* were selected as candidate genes underlying cowpea's drought tolerance. This information will be useful in future studies for selecting cowpea tolerant genotypes and increase the efficiency of plant breeding strategies.

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P22

Reversible Biosensor for Reactive Nitrogen Species for Photodynamic Therapy Application

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Keywords: Biosensor, Reactive Nitrogen Species, Photodynamic Therapy Application, reversibility, hypoxia environments

The therapeutic battle against cancer never stops. The traditional clinical available options in cancer treatment are limited due to high recurrence rate, systemic side effects, and cumulative radiation dose. Photodynamic therapy presents itself as an alternative and versatile technique for cancer treatment with minimal invasion, low poison, precise operation, and nondrug resistance. One of its major limitations is the need to have an oxygen rich environment on the therapeutic area. As such, the development of new nanoparticles that can be effectively used in low oxygen environments, e.g., solid tumours, is a new and exciting hot research topic in the nanomedicine field. Carbon Dots (Cdots) are nanoparticles that have been proven non-toxic, photostable and effective photosensitizers agents that can induce both ROS and RNS using a FRET process. In this particular case the Cdots were tailored to induce the formation of RNS when exposed to a given wavelength. The Cdots ability to be used not only as photosensitizer but also as *in situ* nanosensor is one of its most interesting characteristics.

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P23

A preliminary genome-wide association study for resistance to Tropical Theileriosis in Portuguese Mertolenga cattle breed

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Keywords: Tropical Theileriosis, SNP, QTL, Mertolenga

Tropical Theileriosis in cattle is a tick-borne disease with considerable production losses. The reduction of its incidence may be based on the genetic selection of resistant animals to be used as future breeders. In this preliminary study, we identified SNPs in cattle of the Mertolenga breed, using an array of 100 000 SNPs, to determine the putative association with resistance or tolerance to infection. Thus, we analysed 48 non-infected animals and 48 infected. For a suggestive significance of $p \leq 1x10^{-4}$, we identified twenty SNPs, of which four are already described as QTLs. This QTLs are associated with carcass and meat characteristics, creatinine content of muscle, milk glycosylated kappa-casein percentage and with urate oxidase. This is an enzyme responsible for the degradation of uric acid, a haematological parameter that may be increased in animals infected with *T. annulata*. Further in vivo studies should include these parameters to determine their association with the disease.

P24

Metagenomic assessment of fungal microbiota on *Vitis vinifera* leaves treated with plant extracts

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Keywords: Natural fungicides, grapevine microbiome, ITS, fungus

Viticulture is one of the most challenging cropping systems regarding crop protection. Moreover, climate change will have several impacts on grapevine such as on pests and diseases. Studies on sustainable vine protection strategies are necessary, and an example is the use of plant extracts as sources of natural fungicide effect, to reduce chemical fungicides. Nonetheless, the influence of foliar application of these products on the plant microbiome is disregarded.

Aiming to gain a better knowledge of the effect of sprayings based on plant extracts, searching for crop protection sustainable alternatives, in the present work was studied the fungal community on leaves, following a metagenomic approach. One field trial was installed at UTAD, with the cultivar 'Touriga Franca' and foliar applications were carried out with four different treatments: nettle extract, Japanese knotweed extract, conventional fungicide, and water (control), throughout the 2020 and 2021 growing seasons. Leaves were sampled at harvest and deep sequencing of fungal-directed ITS1 amplicon led to the detection of the main genera in leaves treated with conventional fungicide *Mycosphaerella, Cladosporium*, *Alternaria*, *Aureobasidium* (only in 2020) and *Sporobolomyces* and in leaves treated with plant extracts and control the four main genera, *Mycosphaerella*, *Cladosporium*, *Erysiphe* and *Sporobolomyces*. It should be noted that regarding the genus *Erysiphe*, which is the genus of the powdery mildew causal agent, *Erysiphe necator*, it was recorded a decrease of its abundancy in leaves treated with plant extracts compared to control, in averaged of 25.1% for nettle extract and 61.2% for Japanese knotweed extract. Crop protection urges for alternatives to *synthetic* fungicides and new formulations including plant extracts seem very promising.

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P25

FA-SAT ncRNA influence in anti-mitotic drug response in human cancer cell lines

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Keywords: *FA-SAT* ncRNA, taxol, resistance, sensitivity, anti-mitotic drug

FA-SAT is the major satellite DNA (satDNA) of the cat genome, being highly conserved in Bilateria species, including human. This satDNA sequence is transcribed yielding satellite non-coding RNAs with important cellular functions in proliferation and apoptosis. Preliminary data showed a potential role of *FA-SAT* transcripts in the cellular response to anti-mitotic drugs in cat mammary cells.

In order to understand the influence of *FA-SAT* expression in anti-mitotic drug (Taxol) response in human cell lines, a study has been developed with two human breast cancer cell lines, MCF7 and MDA-MB-231.These cell lines were treated with 500 nM of Taxol and the cell viability was accessed by MTT assay and phenotypic analysis was performed. Also, the *FA-SAT* ncRNA levels in these cells was analyzed, by RT-PCR, in Taxol treated cells.

Our results of cell viability analysis showed that MCF7 seems more sensitive to taxol than MDA-MB-231, which revealed higher resistance to this drug. Also,

FA-SAT ncRNA levels quantification showed that its expression decreases in taxol-treated MCF7 cells, suggesting the influence of *FA-SAT* levels in the cell fate under taxol treatment.

In the future, in order to better understand this interaction, taxol resistant cells, as MDA-MB-231, will be *FA-SAT* silenced in order to verify if they become more sensitive to taxol.

P26

Serological evidence of West Nile Virus infection in horses from the municipality of Castelo Branco

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Keywords: horse, ELISA, equine, seroprevalence, West Nile Virus

West Nile Virus (WNV) is an RNA virus from the family Flaviviridae. It is transmitted by several genera of mosquitoes, with *Culex* spp. as the main vectors identified in Europe and North America. Birds are amplifying agents and important to the transmission cycle, during the viremia period. Equines are considered dead-end hosts and are recognized as sentinels of the virus in many countries.

This work presents a WNV seroprevalence study in 40 horses from the Castelo Branco county. Breed, gender, age and geographical area were analyzed. An enzyme-linked immunosorbent assay (ELISA) was performed to detect total antibodies against WNV, using a commercial kit, according to the manufacturer's instructions. Twelve animals were found seropositive (30%; 95% CI: 18.1-45.4%) and 1 (2.5%) was regarded as doubtful.

We truly believe there is much more to study in Portugal regarding the epidemiology WNV, and an active surveillance plan is needed in the short term.

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P27

Seroprevalence of West Nile Virus in Vultures in the Centre region of Portugal

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Keywords: ELISA, seroprevalence, vultures, West Nile Virus

West Nile Virus (WNV) is a vector-borne pathogen with zoonotic potential widely spread throughout the world. The transmission cycle is maintained by mosquitoes (mainly *Culex* spp.) and wild birds. Humans, equines and other mammals are incidental hosts, which are unable to amplify the agent in their blood circulation.

Two lineages have been identified as disease-causing in humans and other animals: WNV-1 and WNV-2, being the latter more common in Europe.

Symptoms in humans from WNV are mainly neurological, which can include severe encephalitis and death. Nevertheless, most of the infections are subclinical, and the true occurrence of the disease remains highly underestimated in Portugal.

We aim to contribute to study the seroprevalence of WNV in wild birds in Portugal. In this work we present the seroprevalence of antibodies to WNV in 27 vultures which were admitted at a wildlife recovery centre in the Centre region of Portugal (CERAS). Collected serum samples were from 2 different species, the Eurasian griffon vulture (*Gyps fulvus*) and the cinereous vulture (*Aegypius monachus*). A commercial enzyme-linked immunosorbent assay (ELISA) was performed and results have shown a high prevalence of seropositivity to WNV (n=14, 51.9%; 95% CI: 33.9-69.3%).

Occurrence of WNV antibodies was high in this study. This is a preliminary investigation that will be continued in other geographical areas and with a larger sample size.

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P28

"Application of the comet assay in studying of the genomic instability in blood cells from bovines with spontaneous tumours and healthy animals"- A preliminary study

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Keywords: comet assay, neoplasic disease, DNA damage, instability

The appearance of cancer is intricately related to genetic pathways, principally genomic instability. This DNA alteration disturbs the structure of the double helix, and normal cells may become malignant cells. A vast variety of methods allow the detection of the damage; the comet assay is one of them. This technique consists in embedding the cells in agarose, then passes through the lysis phase (to remove the proteins and form the nucleoid) and electrophoresis. When there is an occurrence of a break the loops relax and this allows them to move towards the anode, forming the typical "comet" shape. The aim of this study is to evaluate DNA damage in spontaneous tumours, as well as establish a correlation between malignity and the DNA damage itself using blood samples from 6 animals with neoplastic disease (2 cutaneous melanomas, 1 cutaneous papilloma, 1 vesical hemangiosarcoma and 1 mesothelioma) and 5 animals without any type of apparent illness, considered healthy. The results were obtained from DNA damage in blood lymphocytes (in arbitrary units 0-400). The animals without tumoural disease presented values of 14±16.4, 14± 40.7, 15±27.1, 16,5±27.6, 21±10.4, with a total mean of 15± 11.5 associated to non-ill animals. The blood of the animals with tumours presented in the cutaneous melanomas 22±8.50, 34±10.12 and 63,5±15.6, in the cutaneous papilloma 20±5.34, in the vesical hemangiosarcoma 89±19.12, in the mesothelioma 168±5.5, with a total mean o 65,25±4.9 relative to the animals that have a tumour associated. This preliminary study presents that the samples of the animals with neoplasia that have higher values than the animals without neoplasia, reflecting greater damage. Also analysing the total mean of the two groups reflect a larger difference between the healthy animals and the ones that have tumors. This results allowed to conclude that the comet assay may be a suitable method to understand and detect this instability relative to the appearance of spontaneous tumours in bovines.

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P29

Beyond toxicity: exploring the antibacterial properties of tryptamine

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Keywords: Tryptamine, Biogenic amine, *Pseudomonas putida* U, Antimicrobial therapy

Tryptamine, a biogenic amine derived from tryptophan, is found in various food sources. High concentrations of tryptamine can cause mild to severe toxicity in humans. Interestingly, it has been suggested that tryptamine may also has antibacterial properties, inhibiting the growth of certain bacteria, such as *Escherichia coli* and *Listeria monocytogenes*. However, some bacteria can degrade tryptamine through specialized enzymes. *Pseudomonas putida* U is unable to catabolize tryptamine. Moreover, tryptamine causes a deleterious effect over the growth of *P. putida* U when using other carbon sources. Thus, this study aimed to investigate the toxic effects of increasing concentrations of tryptamine when *P. putida* U is cultured in media containing unrelated carbon sources to support growing. Our findings may contribute to a better understanding of the potential use of tryptamine as a potential biocontrol molecule.

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P30

Repositioning of drugs to treat intracellular infections of *Staphylococcus aureus*

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Keywords: *Staphylococcus aureus,* intracellular, infection, antibiotic resistance, drug repositioning

Staphylococcus aureus is a pathogenic bacterium with great impact worldwide. Although, *S. aureus* was considered an extracellular microorganism, it has been found that it has the ability to enter cells and replicate in them. This capacity, together with the resistance to multiple antibiotics that it presents, is becoming a problem in order to treat the infections that it produces.

In this work, the objective of finding effective compounds against *in vitro* intracellular infections of *S. aureus* was set. Lung epithelial cells from the A549 line were infected with *S. aureus* strain USA300 and a screening of different drugs was performed. This was based on the repositioning of drugs, which consists of giving new uses to drugs that are approved for the treatment of other diseases. In this way, it was possible to verify that some compounds were capable of increasing cell survival, which led to the conclusion that there are possible effective alternatives to treat diseases caused by *S. aureus*.

P31

Marker Chromossomes and FISH technique: still a winning duo

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Keywords: Marker chromosomes, FISH, molecular cytogenetics, karyotype, diagnosis

Small supernumerary marker chromosomes (sSMCs) are abnormal structures of unknown origin, usually consisting of centromeric heterochromatin and resulting from one or two chromosome arms. The presence of marker chromosomes can interfere with the meiotic process being important in these cases the genetic study of the progenitors to determine the origin of these chromosomes, whether *de novo* or inherited events. In humans, the severity of phenotypes depends on the size, the genes and involved chromosomes. In most cases (70%-80%) is derived from an acrocentric chromosome. In general, the identification of sSMCs is not possible by conventional cytogenetic techniques due to small chromosomal size and structural unidentifiable shape, and resolution limitations of the technique itself. Fluorescence *in situ* hybridization (FISH) is the recommended molecular cytogenetics technique to identify these rare sSMCs. The authors present successful results in the application of this technique in various postnatal samples, obtained during the MSc training period in the CGM/CHUdSA Cytogenetics Unit. Using specific probes of centromeric regions, FISH proved to be effective in the characterization of sSMCs chromosomes that otherwise, by karyotype or aCGH, was not possible. These results emphasize the continuous importance of targeted molecular cytogenetic techniques in genetic diagnosis.

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P32

Antitumor Potential of Small RNAs from Mushrooms

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Keywords: Mushrooms, antitumor, small RNAs

Edible mushrooms have been considered as functional foods and an excellent source of nutraceuticals with high potential for prevention of diseases, such as cancer. Even if mushrooms are a rich source of bioactive compounds, our knowledge in this field is still scarce. Recently, ethanol-insoluble and water-soluble small RNA (sRNA) fraction purified from co-extracted polysaccharides by anion-exchange chromatography from some edible mushrooms species as *Cantharellus cibarius* and *Boletus edulis* shown to have antiproliferative activity without cytotoxicity to normal cells. Small RNAs have the ability to regulate gene expression by binding to messenger RNA (mRNA), inhibiting its translation into protein. Several studies have reported that sRNAs from mushrooms can inhibit the growth and proliferation of cancer cells by targeting genes involved in cell proliferation, angiogenesis, and apoptosis.

Although research in this area is still in its early stages, data suggest that mushrooms sRNAs may have therapeutic potential as agents for the treatment of cancer and other diseases. Here, we intend to summarize the potential that sRNAs from mushrooms have as antitumor agents, isolation techniques and methodologies that have been implemented alongside major outcomes and findings, to highlight active research on the new generation of anticancer biotherapy.

P33

Characterization of the fungal biodiversity on surfaces in veterinary facilities

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Keywords: Fungal; Zoonotic; *Aspergillus* spp.

The proximity between animals and humans in veterinary hospitals is a risk factor for exposure to diverse pathogens with zoonotic potential, such as fungal agents.
The aim of this study was to evaluate the fungal biodiversity in a veterinary teaching hospital (VTH).
Seventy-one samples were collected using swabs with peptone water, in different infrastructural surfaces of one VTH. This procedure was carried out in all services of the VTH before the major rate of movement and worktime.
All the collected samples were inoculated in Sabouraud Dextrose Agar® culture medium and incubated at 27°C for 5 days. After laboratory processing, the morphological identification of fungal genera was achieved through macro and microscopic characteristics.
Fungal growth was observed in 83,1% of the samples. After isolation methods, it was possible to identify 10 different genera of molds, mainly *Aspergillus* spp. (34,1%), *Penicillium* spp. (23,0%) and *Mucor* spp. (18,3%). Other genera were identified in lower percentages, as *Cladosporium* spp., *Alternaria* spp., *Acremonium* spp., *Fusarium* spp., *Verticillium* spp. and *Scopolariopsis* spp.
Regarding to the predominant genera, *Aspergillus* spp. was obtained from 74.4% samples of the floor, and 25.6% were from other surfaces. However, 43.8% of the identified *Aspergillus* spp. observed in floor samples were from the companion animals' sector and 28.1% from the equines and production animals' sector of the VTH.
Considering the fact that veterinary hospitals might constitute potential reservoirs of zoonotic agents, these results may represent a concern due to the proximity of animals and the contaminated surfaces and after with their tutors.

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P34

Production and evaluation of aptamer-functionalized liposomes for oral cancer therapy

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Keywords: G-quadruplex, aptamer, Human Papilloma virus, liposomes

Conventional anticancer therapies present low specificity, leading to several secondary effects. To improve these drawbacks, aptamers able to fold into G-quadruplex (G4) are being used to promote drug accumulation in cancer cells. AS1411 is a G4 aptamer able to recognize nucleolin, a protein overexpressed in cancer cells' surface. This aptamer was tested in phase II clinical trials but showed low response rates and suboptimal pharmacokinetics. Nevertheless, AS1411 is being used as targeting agent.

Herein, AT11 (a AS1411 derivative with improved activity) was functionalized with liposomes produced by ethanol injection method with the aim to improve the selectivity of a potential anticancer drug, the acridine orange derivative (C8) towards oral cancer cells.

The resulting liposomes were characterized by dynamic light scattering. The effect of the produced liposomes on oral cancer (SCC154) and healthy (Het1A) cells' viability was determined by MTT assay and its internalization was visualized by confocal microscopy. When cells were treated with C8-liposomes, a dose-response effect was observed on both cell lines. However, when conjugated with AT11 a clear selectivity of the liposomes towards the SCC154 cell line was observed. Moreover, we demonstrated, by confocal microscopy analysis, that the AT11 conjugated liposomes are efficiently internalized and can reach the cytoplasm of the treated cells.

Overall, these findings suggest that the designed liposomal formulation represent a promising drug carrier for oral cancer cell line.

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P35

The effect of resistance training on hematological parameters: data from an animal model of mammary cancer

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Keywords: breast cancer, physical exercise, serum parameters

Breast cancer continues to be one of the main causes of death from cancer worldwide. Haematological analysis is not only useful to evaluate the animals' health status, but also for the diagnosis and to study the progression of several diseases, including cancer. This study aimed to evaluate the effects of resistance training on haematological parameters in a rat model of mammary cancer. Twenty-eight female rats were divided into four groups (n=7): Sedentary (SED); SED+N-methyl-N-nitrosourea (MNU); Exercised (EX); and EX+MNU. SED+MNU and EX+MNU animals received an intraperitoneal injection of the carcinogen MNU (50mg/Kg), at seven weeks of age. Exercised animals were trained 3 days/week for 18 weeks, by climbing a 1-meter-high homemade ladder, 8-12 dynamic movements/climb and 4-8 climbs/session. At necropsy, blood was collected by intracardiac puncture into an EDTA tube and kept at 4°C until the analysis. The following haematological parameters were evaluated using a haematology analyser (IDEXX ProCyte Dx Haematology system): red blood cells (RBC), haematocrit (HCT), haemoglobin (HGB), reticulocytes, leucocytes, neutrophils, lymphocytes, monocytes, eosinophils, basophils, and platelets. Data were analysed using SPSS and values were considered statistically significant at $p<0.05$. Some significative differences in the haematological parameters were found among groups ($p<0.05$). RBC (M/µL), HCT (%) and HGB (g/dL) were significantly higher in MNU+EX group when compared with the remaining groups ($p<0.05$). Inversely, in the leukocyte formula, we noticed a lower percentage of neutrophils and lymphocytes in MNU+EX group when compared with non-exercised groups (SED and SED+MNU) ($p<0.05$). The leucocytes were higher in MNU+EX group when compared with MNU group ($p>0.05$). Reticulocytes, monocytes, eosinophils, and basophils were slightly lower in both EX groups, when compared with SD groups, but the difference did not reach the level of statistically significance ($p>0.05$). Despite the increase in RBC, there was no decrease in HGB, suggesting that the animals, were not anaemic, even though the haematological effort caused by exercise and the tumour environment. Consistent with this information, no alterations were observed in the reticulocytes, indicating that immature RBCs were not being sent to the bloodstream. The leucocytosis observed in EX+MNU animals corresponds to a typical inflammatory response. The absence of neutropenia is suggestive of positive response of animals to exercise. These results suggest that resistance training can lessen the negative effects caused by mammary cancer.

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P36

Putrescine-mediated induction of alternative genes to gabD and gabT in polyamine metabolism in *Pseudomonas putida* U.

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Keywords: *Pseudomonas putida*, putrescine, cadaverine, GABA, 5-AV

It has been suggested in different bacteria that 4-aminobutyric acid (GABA) and 5-aminovaleric acid (5-AV) are produced by deamination of putrescine and cadaverine, respectively. These compounds can be used as carbon and nitrogen sources supporting bacterial growth.

In *Pseudomonas putida* U, *gabD* and *gabT* genes, coding for a semialdehyde dehydrogenase and an aminotransferase, have been identified as involved in the catabolism of 5-AV and GABA. The growth of strains knocked out on one or the other of these genes has been analyzed in minimal media with cadaverine, putrescine, GABA, or 5-AV as sole carbon sources. Small concentration of putrescine (1 mM) in these media allows the growth of the strains, or at least decreases the lag phase. In conclusion, the results suggest that the presence of putrescine in the media could induce genes replacing the functions of GabD and GabT in *P. putida* U.

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P37

Influence of biostimulants and a boron-based fertiliser in the nucleolar activity and protein content of *Juglans regia* L. (walnut) trees

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Keywords: Biostimulants, interphase anomalies, nucleolus, protein content, walnuts

Walnut is an important economic and nutritional crop. Biostimulants and boron can improve yields, quality, and stress tolerance. While typical, there needs to be more studies on the effects of those at the cytological level. Methods that analyse the more suited treatment for each condition are valuable. Studying the nucleolar activity and total protein content can provide clues about the treatments that improve the protein synthesis necessary for vital biological processes. This work studied the effects of five treatments based on seaweed extracts (from two different species, SWE_{EM} and SWE_{AN}); free amino acids (AA); a mixture of a seaweed extract and free amino acids (SWE-AA), and boron ethanolamine (BE), on the nucleolar activity and total protein content of *Juglans regia* individuals of cv. “Franquette” grown in an organic orchard in the NE of Portugal by compared with untreated trees (C). The cytogenetic and biochemical data integration allowed us to suggest that the treatments BE, AA, and SWE_{AN} were the most suited to the cv. “Franquette”.

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P38

Fungal biodiversity on bird feathers

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Keywords: Fungi, Biodiversity, Health

Analysing fungi is important for our health. Although some are beneficial, many can cause harm to humans and other animals. This study focuses on what fungi can be found on captive birds. By collecting samples from the feathers of different birds in different environments, growing the fungi, isolating, and, finally, through phenotypic analysis we were able to identify several different fungi living among these birds, such as *Alternaria, Aspergillus, Aureobasidium pullulans, Cladosporium, Fusarium, Mucor, Penicillium, Rhodotorula, Sarocladium, Trichoderma* and various yeasts. Some of the fungi collected may cause disease and further genetic testing is required to access their pathogenicity. We concluded that potentially dangerous fungi can be found on bird feathers and this analysis helps us prepare better against these diseases.

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Oncolytic Virotherapy: The Future of Cancer Treatment

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Keywords: Cancer, Virus, Virotherapy, Treatment

The idea that some viruses are more aggressive towards cancer cells than others is not new. Ever since the 19th century scientists have dwelled on the possibility of oncolytic virotherapy.[1] In the last few years, the advancements in the fields of molecular biology and virology have made oncolytic virotherapy a reality.[2] Oncolytic viruses are defined as genetically engineered or naturally occurring viruses that selectively replicate in and kill cancer cells without harming the normal tissues.[3,4] Most oncolytic viruses in use nowadays are genetically engineered by being armed with transgenes that can have different objectives.[3,5,6] A few viruses have been selected and are currently being used in clinical trials, the most common being the adenovirus.[5,6] The first and only oncolytic virotherapy to complete clinical trials and be approved for medical use was the talimogene laherparepvec (T-VEC), a herpes simplex virus type 1 (HSV-1) armed with GM-CSF, being approved by the Food and Drug Administration (FDA) in the USA.[1,2,3,5] Many studies have shown promising results, reducing the side effects of treatment, when compared to chemotherapies, but with improvements to be made when it comes to efficacy, particularly with cell penetration, tumour targeting and the body's immune responses.[1,2,6] However, with these improvements, oncolytic virotherapy can become a viable and efficient cancer treatment option with reduced discomfort to patients.

P40

Novel plant protection compounds derived from macroalgal cell walls

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Keywords: cell wall, plant defense, macroalgae, subcritical water extraction

Marine macroalgae are a very diverse group of organisms often used for industrial purposes. The diversity of compounds they harbour is so great that the residues after their use in these industrial processes still contain compounds that are active on plants, many of them in their cell walls.

The main objective of this work was to determine the cell wall composition of selected representative species from different taxonomic groups with industrial interest. Cell walls were isolated and fractionated following both classical chemical methods and the novel and greener subcritical water extraction technology. The purified cell walls and their fractions were analysed by a wide array of analytical techniques. Once extracted, these fractions were tested for their capacity to trigger immune response in plants.

In this way, macroalgal residues from industrial processes could be revalorized through efficient and environmental-friendly processes in the context of the circular economy.

P41

Carbon Dots *in vivo* Evaluation in *Drosophila Melanogaster*

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Keywords: Carbon Dots, Toxicity, Transgenerational effects, Reactive Oxygen Species, *Drosophila melanogaster*

In recent years there has been an increasing interest in the use of nanoparticles. Carbon Dots, Cdots, are one of the most appealing alternatives to the traditional semiconductor nanoparticles used for confocal microscopy assisted studies and surgeries. However, it is necessary to evaluate every possible cytotoxic effect that may emerge from their use in biological media. An *in vivo* model that has been quite used due to its availability and neurological resemblance to humans is the *Drosophila melanogaster*. Here we evaluated the oviposition and transgenerational effect of the nanoparticles, along with the overall survival of the parental generation. Moreover, since these nanoparticles can also induce the generation reactive oxygen species (ROS) when exposed to ultraviolet radiation, we have also evaluated the previous parameters in ROS exposed Drosophilas. The second generation of the ROS exposed group and the non-exposed where further analysed in terms of resistance and recovery after the exposition to a stress situation. This is preliminary studies that help us determine the overall effect of Cdots on *Drosophila melanogaster*.

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Evaluation and Characterization of Molds and Yeasts in Meat

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Keywords: molds, yeast, meat

During the meat maturation process, the development of molds and yeasts occurs, which can contribute to organoleptic modifications of the meat.
This study aimed to quantify and characterize the fungal genera that predominate during the dry-aging process, comparing their occurrence on the external surface and inside the muscle. The samples were collected from 3 different animals and stored for 90 days. During seven times of storage, a total of 126 samples were collected (84 samples from external surface and 42 from muscle). The molds and yeasts were isolated on Chloramphenicol Glucose Agar® and Sabouraud Dextrose Agar® culture medium, according to ISO methods. The identification was performed following macro and microscopic parameters. Fungal growth was observed, with molds observed in 57,1% of the total samples. The most isolated genera was *Penicillium* spp. (45,3%), followed by *Aspergillus* spp. (18%) and *Cladosporium* (7,1%). Yeasts were developed in 84,1% of the samples. The microbiology analyses showed a decrease in the development of yeast during the process. In time 0 of storage, 4,48 $\log_{10}$ CFU/g was observed on the surface and 3,78 $\log_{10}$ CFU/g in the muscle. At 90 days of storage, the value on the surface was 3,15 $\log_{10}$ CFU/g, and in the muscle was 0,90 $\log_{10}$ CFU/g. Contrasting to these results, the molds development increased during the dry-aging process. In the first time, only 0,6 $\log_{10}$ CFU/g levels were observed on the surface and no molds were detected inside the muscle. Although in the last day of storage (90) values of 2,35 $\log_{10}$ CFU/g were observed in the surface and of 0,97 $\log_{10}$ CFU/g inside the muscle.
Recognizing the importance of fungal development to the dry-aging process, further research should be focus on the application of newly identified fungi strains to reduce the aging time and achieve desirable features.

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Analysis of the antiviral activity of fusaricidin against SARS-CoV-2

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Keywords: Fusaricidin, SARS-CoV-2, *Paenibacillus polymyxa*

Soil *Paenibacillus* species are microorganisms able to produce a wide variety of antibiotic compounds. Among the producing species, *Paenibacillus polymyxa*, a non-pathogenic, endospore-forming from the *Bacillus* group that occurs naturally in the soil, rhizosphere, and roots of cultivated plants, being one of the most industrially important facultative anaerobic bacteria.

Fusaricidins are cyclic hexapeptide antibiotics which are known to have antimicrobial activities against a wide diversity of fungi and gram-positive bacteria.

Similar compounds like surfactin, also produced by *P. polymyxa*, have shown as well, an interesting activity against viruses like SARS-CoV-2.

In this work, fusaricidin antiviral properties were evaluated, especially against SARS-CoV-2; as well as its cytotoxic effects on mammalian cells, with the aim of determine if this compound could be used as an effective agent against the virus.

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The use of mitotic stimulants to diagnose B-cell hematologic neoplasms

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Keywords: Cytogenetic, B-cells, TPA, DSP30, IL2

Conventional cytogenetics (CC) and Fluorescent *in situ* Hybridization (FISH) play a very important role in the diagnosis of B-cell disorders, namely chronic lymphocytic leukaemia (CLL) and non-Hodgkin lymphomas (NHL). Due to low proliferation rate of mature B-cells *in vitro*, the use of mitotic stimulants in cell cultures, such as 12-0-Tetradodecanoylphorbol-13-acetate (TPA) and CpG Oligonucleotide, DSP30 + Interleukin 2 (IL2), is required.

In this work, with the objective of analysing the efficacy of the mitotic stimulants, in thirty-eight samples (twenty-one of peripheral blood (PB) and seventeen of bone marrow (BM)) two cultures were performed: one with DSP30 plus IL2 (ID) and the other with TPA (T), both for 72 hours. FISH was performed using specific probes, depending on the clinical indication, in thirty samples. From all samples, we successfully obtained metaphases in thirty-seven (there were no metaphases in one sample of BM, for both cultures). By CC, twenty cases with chromosomal alterations were found (fourteen samples of PB and six samples of BM). In those twenty chromosomal anomalies, twelve were observed in both ID and T cultures (eight samples of PB and four of BM), six only in ID cultures (four samples of PB and two of BM), and two only in T cultures (two samples of PB). Regarding FISH analysis, fifteen samples were positive in ID cultures, and one in T culture.

Although the sampling was small, we could verify that both stimulators remain important in samples with suspected lymphoid neoplasia: with DSP30+IL2 the detection rate of the abnormal clone was higher, but TPA allowed detection of these abnormal clones in two samples. FISH remains important, as complement of CC and especially in cases where metaphases are not obtained.

P45

Assessment of genetic relationships in *Vigna* germplasm

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Keywords: Genetic diversity, microsatellites, legumes

The genus *Vigna* belongs to the family Leguminosae and has a high economic and social importance worldwide, particularly in the developing countries. This genus has a great variability and comprises more than 100 cultivated and wild species, containing some of the most relevant grain legumes species, such as cowpea (*V. unguiculata* L. Walp.), mung bean (*V. radiata* L. Walp.) and black gram (*V. mungo* L. Hepper).

Climate changes will be responsible for several losses in crops' production, leading to the emergence of new pest and diseases. The development of new varieties, with greater tolerance to high temperatures and drought and resistance to pests and diseases, exploiting underused cultivated germplasm and wild relative crops, will be crucial. However, the information available on the whole genetic diversity in the genus *Vigna* is limited. Microsatellites (SSRs) are an excellent marker system for accessions' DNA fingerprinting and genetic diversity studies.

To evaluate the genetic diversity in the genus *Vigna*, a set of four SSR loci was analysed in 31 accessions, including four different *Vigna* species (*Vigna mungo*, *V. racemosa*, *V. radiata* and *V. unguiculata*) and six subspecies of *Vigna unguiculata* (spp. *alba, pubescens, spontanea, sesquipedalis, unguiculata* and *tenuis*). The four loci revealed to be polymorphic and a total of 26 alleles and 25 genotypes were detected. This study allowed to establish a *Vigna* species identifier genotype at only four SSR loci. Nevertheless, it was not possible to distinguish some of the subspecies compromising the establishment of the phylogenetic relationships between all the accessions studied. In the future, the study will be complement analysing a greater number of SSR loci to provide full assessment of *Vigna*'s genetic diversity to be to be exploited by plant breeders.

P46

Suitable conditions *in vitro* to cowpea root phenotyping

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Keywords: *Vigna unguiculata* L. Walp., phenotyping, root system architecture

Root traits are pivotal for plant productivity and responsible for its resilience to various stresses. Root system architecture (RSA) is an underexplored trait on plants probably due to the dense nature of soils that makes its phenotyping in situ challenging compared to aerial part of plant. The selection of simples and cost-effective root phenotyping methodologies to identify genotypes resilient to drought stress is fundamental. Cowpea (*Vigna unguiculata* L. Walp.) is a grain legume considered as one of the most legume crops adapt to high temperatures and drought having potential to be used as model to drought stress studies. This study intends to test different gelling agents and concentrations that will be useful to growth cowpea genotypes under in vitro conditions.

Three different concentrations (low, medium and high) of gelling agents (agar, Phytagel and Gelrite) were evaluated in Murashige and Skoog (MS) medium. In this experiment, three cowpea genotypes (‘Bambey 21’, ‘IT97K-499-35’ and ‘Fradel’) were submitted to these conditions during two weeks, in four replications. During this experiment, germination data and root and plant length were registered.

The three gelling agents did not revealed significant differences on germination rate. The highest concentration of each gelling agent indicated a slight decrease on roots length in all cowpea genotypes. The Phytagel and Gelrite agents appear as an alternative advantageous to agar for exploitation the cowpea root phenotyping due to its transparent colour allowing a best digital imaging acquisition.

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P47

Novel oligosaccharides that trigger immune responses in plants

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Keywords: pattern-triggered immunity, microbe-associated molecular patterns, β-glucans

Immune responses in plants can be triggered by damage-/microbe-associated molecular patterns (DAMPs/MAMPs), which are recognized by plant pattern recognition receptors (PRRs). The response cascade activated after the DAMPs/MAMPs perception is known as pattern-triggered immunity (PTI). Since there is limited knowledge on PTI-triggering elicitors based on glycans, this work focused on the identification of novel β-glucan oligosaccharides which act as MAMPs. Therefore, the ability of different β-glucans to prompt PTI responses was tested. It has been seen that a specific group of β-glucans unleash this response in plants by increasing the production of reactive oxygen species, phosphorylation of MAP kinases and gene transcriptional reprogramming. In order to determine which PRRs are involved in the recognition of these carbohydrates, we studied PTI responses in PRR mutant plants. The results obtained pave the way to develop products to enhance plant resistance against pathogens.

P48

Determinants of oxidative stress, assessed from the trypanothione reductase activity, in the trypanosomatid *Leishmania infantum*

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Keywords: Leishmaniasis, NTD, $T[S]_2$, TryR, ROS

Leishmaniasis is a neglected tropical disease (NTD) spread to animals and humans and transmitted by the bite of a female sand fly. It is caused by the protozoan parasite *Leishmania*, which has a redox metabolism based on trypanothione ($T[S]_2$) and trypanothione reductase (TryR). This system is crucial for survival inside the host macrophage phagolysosome, allowing the elimination of reactive oxygen species (ROS). The purpose of this study is to determine some factors that induce oxidative stress in *Leishmania*. With this purpose, the activity of trypanothione reductase will be analysed, as a possible therapeutic target, after induction of oxidative stress in *Leishmania infantum* promastigotes/amastigotes under different conditions, such as the overexpression of heterologous reporter proteins in amastigotes used in HTS screening platforms, the addition of increasing concentrations of antileishmanial drugs, changes in temperature and pH, etc.

P49

A phylogenetic and evolutionary analysis of transposable elements in the wolf (*Canis lupus*, *Canidae*, *Carnivora*) genome

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Keywords: Transposable Elements (TEs), Long interspersed nuclear element 1 (LINE-1), *Canis lupus* (CLU), *Canis lupus familiaris* (CLUfa), Karyotype

Transposable Elements (TEs) are DNA sequences that can move within species' genomes and influence the appearance, regulation, and evolution of genomes. One of these elements, Long Interspersed Nuclear Element 1 (LINE-1) is highly present in most mammalian genomes, but there aren't studies on the Canis genome. We used two cell lines (male and female) from a European Wolf (*Canis lupus lupus*) stipulated in the host lab. Cytogenetics characterization through G- and C-banding revealed a karyotype of 78 chromosomes, with 38 pairs of acrocentric autosomes and 2 sex chromosomes, with a submetacentric X and acrocentric Y, identified by chromosome painting. *Canis lupus* is accepted as the *Canis lupus familiaris* (dog) ancestor that also harbours 78 chromosomes, but little is known about the differences of the two related genomes. LINE-1 elements were isolated from genomic DNA of the wolf cell lines using specific primers and used as probes in FISH (Fluorescence *in situ* Hybridization) to physically map these TEs in wolf chromosomes. This analysis was extended to the dog, allowing a qualitative and semi quantitative analysis and comparison between these two phylogenetically related genomes. An interspersed pattern distribution throughout the chromosomes was observed with an apparent higher LINE-1 content in the wolf genome. An in depth *in silico* analysis was performed on available Genome project sequences from both species, allowing to search for the presence and exact location of LINE-1 in the chromosomes and to perform a phylogenetic analysis on the sequences intra and inter genomes. A combined analysis of the *in silico* and cellular analysis of LINE-1 and the syntenies' analysis of the dog chromosomes with the reference genome of Carnivora (*Felis catus*) allowed to disclose the possible involvement of these TEs in the evolutionary pathway of the dog genome, and allowed, as well, to extrapolate it to the wolf genome for the first time. *Acknowledgements: The Author Maria Gaspar thanks Maris Hindrikson, from the Department of Zoology of the University of Tartu in Estonia, for collecting the muscle and skin biopsies of Canis lupus lupus.*

P50

Molecular varietal identification of grapevines in an old vineyard of Quinta Monte Travesso

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Keywords: grapevines genetic resources, autochthonous varieties, grapevine varietal diversity, SSR, microsatellites

The grapevine (*Vitis vinifera* L.) is the best-known species of the genus *Vitis* and there are thousands of varieties of grapevines wildly dispersed all over the world. The great number of varieties identified made that there are different names for the same variety and even different varieties with the same denomination.

Microsatellites, also called Single Sequence Repeats (SSRs), are nucleotide sequences where a short base-pair motif is repeated in tandem. These sequences can be used as molecular markers and are recommended to be used in the genotyping of *Vitis vinifera* by the International Organization of Vine and Wine (OIV). The existence of databases with the results of these analysis allows for the identification of a sample from *Vitis vinifera*.

In this work, 82 plants from a parcel of old vineyard, planted around 90 years ago, in the "Quinta do Monte Travesso", were studied. The amplification of the nine different loci of SSRs (VVS2, VVMD5, VVMD7, VVMD25, VVMD27, VVMD28, VVMD32, VrZAG62, VrZAG79), recommended by the International Organization of Vine and Wine for the identification of the *Vitis* species, was performed. The results from this analysis, when compared with the information present in the database *Vitis* International Variety Catalogue (*V*IVC), allowed the identification of the 82 plants. It was possible to identify 22 different varieties of *Vitis vinifera*, where only six of them have a representation in Portugal higher than 2%. The preservation and valorisation of old vineyards is of utmost importance given the high varietal diversity that they encompass.

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P51

In Silico Analyses for Subsequent Genotyping and Sequencing *TPMT* and *NUDT15* Genes in Clinical Pharmacogenetics

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Keywords: Genotyping, Sanger Sequencing, HRM, Pharmacogenetics, *In Silico*, Clinical

Pharmacogenetics/pharmacogenomics are emergent fields that aim to elucidate genetic basis for inter-individual differences in drug response. Genotype analysis of *TPMT* and of *NUDT15* gene is relevant in cancer and inflammatory bowel disease treatments, providing information on the risk of toxicity or effectiveness of treatment. Outcomes are improved by testing the variants presented in *CPI Consortium* and *Dutch PWG* guidelines. *In Silico* studies have been conducted to determine and confirm primers/probes, and to standardize genotyping using different protocols (e.g. hydrolysis, HRM and Sanger Sequencing). We have compiled the whole sequence of these genes and organized information from different genomic databases, together with 'primer/probes' design from different scientific companies.

Preliminary *in silico* analysis is germane to obtain precise data of the genes of interest and check for inconsistencies, providing reliable interpretation of results for appropriate management of patients.

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P52

Unveiling the role of the Ferric Uptake Regulator FurA from *Anabaena* sp. PCC 7120 in biofilm formation

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Keywords: cyanobacteria, biofilm, exopolysaccharides, ferric uptake regulator, biofertilization

Cyanobacteria are photosynthetic microorganisms able to fix atmospheric N_2 and CO_2. Understanding biofilm formation in these organisms is of special interest given the numerous biotechnological applications of cyanobacterial biofilms in the protection against environmental stresses, biofertilization or biorremediation. The Ferric Uptake Regulator FurA from the cyanobacterium *Anabaena* sp. PCC 7120 is a global transcriptional regulator which is thought to be involved in biofilm formation. For this reason, in this work we have created a strain of *Anabaena* that overexpresses FurA and we have analysed biofilm formation and exopolysaccharide biosynthesis in this strain, unveiling that FurA plays an important role in biofilm production in *Anabaena* sp. PCC 7120. These results shed new light on the understanding of biofilm formation in cyanobacteria, laying the foundations for future studies aimed at implementing biotechnological applications of cyanobacterial biofilms such as biofertilization.

P53

Genome and terroir: how climate might affect berry bioactive compounds and antioxidant activity

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Keywords: *Vitis vinifera*, climate change, varietal selection, phenolic compounds, antioxidant activity

Portugal is a major wine-producing country, with viticulture and winemaking playing a significant role in its economy. However, climate change (CC) threatens the national viticulture, especially in the warmer and dryer regions such as 'Douro Superior' of the Douro Demarcated Region. Grapevine physiology, development, and berry quality are expected to be severely affected by CC, so adaptation strategies have been the focus of research in the past years. Portugal's grapevine inter-varietal diversity offers potential adaptation to CC, but there is still a need for research on how different varieties perform under adverse abiotic conditions. The main objective of this work was to study berry quality parameters of 28 different red grapevine varieties grown under the same *terroir*. These varieties were located in the ampelographic grapevine collection of Quinta do Ataíde (Symington Family Estates), in the 'Douro Superior' sub-region (Vila Flor, Portugal). Berry sampling occurred at the harvest stage during the years 2021 and 2022, and phytochemical analysis of total phenolic content (TPC), flavonoids, *ortho*-diphenols and antioxidant activity (ABTS $^{\bullet+}$, DPPH and FRAP) were performed. Meteorological conditions for the year 2022 were considered warmer and dryer when compared to 2021. Statistically significant differences ($p < 0.05$) were observed among varieties for all parameters, as well as between the years 2021 and 2022. Data related to the berry phytochemical parameters and the antioxidant activity were shown to be significantly different between both years for most varieties, with a general increase from 2021 to 2022, especially in the varieties "Cornifesto", "Donzelinho Tinto", "Tinta da Barca" and "Casculho". However, other varieties such as "Aragonez", "Cabernet Sauvignon", "Touriga Franca", "Trincadeira" and "Vinhão" presented little variation. In sum, the warmer and dryer conditions of the year 2022 led to an accumulation of phytochemical compounds in the berry, as well as increased antioxidant activity in most varieties.

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P54

The potential of *in vitro* cellular models in neurodegenerative diseases: insights from a promising therapeutic strategy for Alzheimer's disease

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Keywords: Cellular models; Animal in vitro cell culture; Neurodegenerative diseases

With the rise of the life expectancy, we have witnessed an increase in the prevalence of age-related neurodegenerative disorders both in the populations of developed and developing countries. Neurodegenerative diseases are characterized by the progressive degeneration and loss of function of nerve cells in the brain and spinal cord, which leads to selective static neuronal loss. This means that populations of neurons, which are particularly vulnerable, experience a progressive decline in function and ultimately degenerate over time. Consequently, these diseases have a negative impact on people's quality of life, as they cause dementia. One of the most common causes of dementia and of top care concern worldwide is Alzheimer disease (AD). It is characterized by the accumulation of amyloid-β (Aβ) plaques, tau tangles, neuroinflammation, oxidative stress, and progressive memory deficits. *In vitro* cellular models have proven to be crucial and extremely valuable in the understanding of these complex diseases and in the finding of therapeutic targets and therapies. In a recent research conducted by Xiaonan Wang, Bei Li, Xiaohong Yu, Ye Zhou, Yue (2022), the authors used the PC12 cell line, a cellular model of AD, obtained from a rat pheochromocytoma tumor that mimics the pathological process of AD *in vitro*. The authors treated the cells with an amyloid β- peptide, which inhibited cell viability and facilitated cell apoptosis, as expected. And after that, they discovered that GM-1 (one of the most abundant gangliosides in the brain) activated the signaling pathway of the Nuclear factor erythroid 2-related factor (Nrf-2) and reduce the accumulation of beta-amyloid plaques in the brain. Nrf-2 seem to have prevented PC-12 cells from the damage induced by amyloid-β and reduce the accumulation of beta-amyloid plaques in the brain. This suggests that GM-1 treatment may be a valuable therapeutic strategy for Alzheimer disease and for others related with amyloid accumulation. These new findings are only possible due to the availability and thorough characterization of these *in vitro* cellular models, especially for diseases of difficult access tissues and organs, such as brain.

P55

How to micropropagate olive *in vitro*

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Keywords: *Olea europaea,* micropropagation, sterilization, nodal segments

Olive (Olea europaea L.), belongs to the genus Olea of the Oleaceae family and is one of the most ancient domesticated fruit trees and recently its cultivation has expanded worldwide and also into some non-traditional areas.

Usually, olive trees are propagated vegetatively by roots, leaves, stems, or softwood cuttings. To overcome the difficulties connected to the conventional propagation methods, micropropagation was proposed for olive propagation. Micropropagation is used worldwide as an essential tool for the large-scale production of clonal plants since it has several advantages compared to conventional agamic propagation systems.

Micropropagation of olive is not easy mainly due to explant oxidation, difficulties in explant disinfection and establishment of in vitro shoot cultures.

The nodal segments have to be surface sterilized and to do that they must be treated with 70% ethanol for 1 to 2 min and then dipped in 15% sodium hypochlorite solution for 10 min. Finally, the nodal segments are going to be rinsed four times using sterilized distilled water for 5 min each. All sterilization steps are carried out under laminar air flow hood by sustaining sterilized atmosphere. Both ends of explants are cut off and transferred in culture vessels containing solid medium. This solid medium can be olive medium or woody plant medium both supplemented with various concentrations of sucrose (15, 20, and 30 g l^{-1}), zeatin (0.1, 0.5, and 4 mg l^{-1}) that can be replaced by neem oil or coconut oil and solidified with 6 g l^{-1} agar. The pH is adjusted to 5.8 before autoclaving at 110°C for 30 min. Explants are incubated at 22.5°C in the dark for the first 7 days and then under a 16-h photoperiod (40 mmol m^{-2} s^{-1}) for the following 21 days. Other approaches have been made using other types of explants and different growth conditions. This work aims to combine the study of olive in vitro propagation performed by different scientists worldwide.

P56

Natural compounds for the treatment of *Rhodococcus equi* infections

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Keywords: bacteria, pathogen, *Rhodococcus equi*, natural compounds

Antibiotic resistance has become a significant problem not only in human health but also in animal health. The recent pandemic has highlighted the importance of keeping livestock in good health as zoonotic diseases can cause problems for society. It is therefore necessary to prevent the emergence of potential new pandemics that could involve multidrug-resistant bacteria.

Rhodococcus equi is a type of bacteria that can cause serious infections in both humans and animals. While traditional antibiotics are commonly used to treat these infections, the overuse of these drugs has led to the development of antibiotic-resistant strains of the bacteria. As such, we aim to find new alternatives to replace traditional antibiotics or to combine them with new compounds.

To achieve this, we exposed *Rhodococcus equi* to a library of natural compounds and measured whether these compounds could inhibit the proliferation of the pathogen. These natural compounds may offer a safer and more sustainable alternative to traditional antibiotics, particularly in cases where antibiotic resistance is a concern. Some of these compounds, such as reactive oxygen species generators or those belonging to the steroid family, have shown a significant effect against the equine pathogen.

P57

Bioactive compounds and antioxidant activity of different apple cultivars

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Keywords: antioxidants; food quality; *Malus domestica*; polyphenols

Apple (*Malus domestica*) is a sweet, edible fruit and it is among the most widely consumed fruits in the world. Apples are a rich source of antioxidants, e.g., vitamins and phenolic compounds, minerals, and also a good source of soluble carbohydrates, reducing the cholesterol and blood glucose levels. Additionally, apples are associated to reduce risk of developing some types of cancer, Alzheimer's and cardiovascular disease, and asthma. Several factors influence the composition of biologically active compounds in apples and other quality attributes, such as the genotype. In this sense, the aim of this study was to determine the phenolic composition and the antioxidant activity (AA, by the iron reduction method – FRAP) in apples from three different cultivars: Golden, Reineta and Starking. There were no significant differences between fruit weight of different cultivars. Nevertheless, the fruits of the cv. Reineta were flatter, which is characteristic of this cultivar, presenting, however, larger diameters than the other cultivars. Significant differences were observed for the phenolics, flavonoids and *ortho*-diphenols, with cv. Reineta presenting the highest contents and cv. Golden presenting the lowest ones. Regarding the AA, it was possible to observe a greater AA in the Reineta fruits, in comparison with the other two cultivars. On the other hand, the cv. Golden presented the lowest AA. These results were corroborated by the higher levels of phenolics, *ortho*-diphenols and flavonoids found in the cv. Reineta, which consequently translated into a greater AA.

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P58

The role of maize cell wall in stalk rot disease

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Keywords: maize inbred lines, *Fusarium graminearum*, matrix polysaccharides, cellulose

Maize is essential and highly produced crops in the world. One of the most important factors affecting maize production is stalk rot disease, which is caused by *Fusarium graminearum*. This fungus infects maize across the stalk pith tissue, and thus the composition and structure of its cell walls (CWs) could be determinant. In order to gain knowledge about this disease, two maize inbred lines with different stalk strength resistance were analyzed for CW and molecular parameters.

Firstly, maize plants were grown in field and were infected or not with *F. graminearum*. At maturity stage, pith tissues of these plants were collected, and their CWs were extracted and fractionated. After a CW FTIR monitorization, cellulose and matrix polysaccharide content was evaluated for both inbred lines.

In addition, these inbred were grown under controlled conditions and, after testing different infection methods, an effective protocol for RNA extraction from the pith tissue is being developed, and gene expression analyses will be carried out in the future. The production of reactive oxygen species and the progression of the fungus through the pith were also evaluated in these plants. All these parameters will be useful for breeding programs in the search for new *Fusarium* resistant genotypes.

P59

Nanoparticle´s effect on *Drosophila melanogaster* life span

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Keywords: Nanoparticles, *Drosophila melanogaster*, toxicity, longevity

Nanoparticles are used in several fields of application, such as cosmetics and medicine. However, despite their unique properties they may have potential biological toxicity, thus the importance of studying each nanoparticle's particular effect.

The use of animal models can help to understand the toxicity effect, and *Drosophila melanogaster* is due to ethical and biological reasons an interesting model for such purpose. In this study, *D. melanogaster* was used to infer the effect of nanoparticle's toxicity regarding longevity.

Nanoparticles were incorporated in the nutritional medium (0; 0,4; 0.6 µM) for the F1 generation.

The male F1 were transferred to a medium without nanoparticles, and the survival rate was registered for a 6 week period. The lifespan of the 0.4 µM group when compared to 0.6 µM was 1 week longer, having an average of the original population of 0.1% remaining, with a similar behavior of the control group.

P60

Antifungal activity of *Origanum vulgare* against *Aspergillus fumigatus, Aspergillus niger and Talaromyces marneffei*

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Keywords: Microbiology; *Origanum vulgare, Aspergillus fumigarus, Aspergillus niger, Talaromyces marneffei*

Origanum vulgare (i.e. oregano) is an aromatic herb commonly used in Mediterranean, besides that it's also known for its medicinal properties. In this experiment we tested its antifungal properties against 2 different species of *Aspergillus (niger and fumigatus)* and the emerging *Talaromyces marneffei.*

We tested the plants antifungal activity in the following concentrations (5, 10, 20 and 30 mg /mL) in which the plant was dissolved and mixed with the growth medium used (PDA) and afterwards we proceeded to inoculate the plates with the fungi, the percentage of inhibition was measured throughout 7 days with data being collected on the 3rd, 5th and 7th days, high percentages of fungal activity inhibition were achieved ranging from 30 to 100% against *Aspergillus niger* and *Aspergillus fumigatus* and from 50 to 100% against *Talaromyces marneffei.*

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P61

Genomic analysis of the human HSAT1 satellite DNA sequence in primates

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Keywords: Satellite DNA, Next Generation Sequencing, HSAT1B, *N. leucogenys*, *In silico* tools

Satellite DNA sequences (satDNA) vary among species (at the nucleotide sequence or the array size, for example) and follow the processes of evolution and speciation. Within the multiple sequences of satDNA shared among humans and non-human primates, HSAT1B is present in multiple autosomic locations in several primates, and it is largely present in the human Y chromosome.

In this work we isolated and cloned the HSAT1B satellite DNA in both the human and the gibbon (*Nomascus leucogenys*) genomes. To the best of our knowledge, HSAT1B presence in the gibbon genome is reported here for the first time. We then compared and analyzed the obtained HSAT1B sequences of human and gibbon. Representative clones from human and gibbon were physically mapped by Fluorescent *in situ* Hybridization (FISH) to assess HSAT1B chromosomal locations. Subsequently, we performed an *in silico* analysis of HSAT1B in the available genome assemblies for the two species. With the obtained results we can deduce about the evolution of this sequence both on humans and primates.

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P62

In vitro germination of *Cucumis sativus* and *Raphanus sativus* seeds

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Keywords: *cucumis sativus,* radish, *in vitro,* germination

Cucumber is the fruit of the cucumber plant *(Cucumis sativus),* it belongs to cucumis genus, they are annual lianas (creepers) with lobed leaves and yellow flowers. Cucumber is a natural diuretic and a great help in dissolving kidney stones.

Radish (*Raphanus sativus* L.), from Cruciferae family, is an important crop, cultivated worldwide for its nutritional values and also as a source of the peroxidases and isothiocyanates used in medicine.

This work aimed to determine the best medium culture for in vitro germination of cucumber and radish seeds. Three Murashige & Skoog media were tested, differing in salts concentration: one with 100% of salts (MS), another with 50% (MS/2) and the last with 25% (MS/4). The seeds were immersed in a solution of gibberellic acid (3g/L), in order to accelerate their germination. Then, disinfection was performed with 70% ethanol and a aquosus solution of NaClO. Finally, the seeds were placed in the respective culture media. Cucumber seeds were placed in a growth chamber at a constant temperature of 24°C and kept in the dark. The radish seeds were also placed in the same temperature conditions but with a photoperiod of 16 hours of light and 8 hours of darkness.

Over the course of 1 week, the germination of the plants under study was observed in the different culture media. The best medium culture is MS/4 for cucumber, and MS/2 for radish.

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P63

Production and quantification of coagulase-negative *staphylococci* biofilms isolated from healthy quails

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Keywords: public health, CoNS, staphylococci, biofilms

Coagulase-negative *staphylococci* (CoNS) biofilms are a threat to public health, due to their presence in animals of consumption such as quails and chicken. *Staphylococci* can cause severe food poisoning and they are also very frequent in cases of hospital-acquired infections. Biofilms are bacterial cell structures clustered in a self-produced matrix attached to biotic or abiotic surfaces. Therefore, we aimed to study the biofilm formation capacity of CoNS isolated from quails for human consumption. The biofilm formation species of CoNS was evaluated, including *S. sciuri, S. lentus, S. urealyticus* and *S. haemolyticus*. The biofilm formation was investigated by the microtiter assay and the biofilm biomass was quantified by the Crystal Violet assay in an ELISA microplate reader at 570 nm. All isolates tested formed biofilm. *S.lentus* was the species that showed the highest biofilm formation with an average above 100% while *S. sciuri* was the one that produced the lowest biofilm biomass with an average around 100%. Some isolates of *S.lentus* and *S. urealyticus* formed more biofilm mass than the control strain. Statistically, there were some differences in terms of biofilm formation between the different species of CoNS, but these were not significant. All of this research was carried out to study the resistance and multiplication capacity of staphylococci, samples were taken from cordons, in order to know their environmental adaptive capacity, such as their percentage capacity for biofilm formation. If there is a strong capacity for biofilm formation, it could be an alarming sign if its consumption occurs, mainly in humans, where this bacterium is difficult to treat due to its resistance to antibiotics and its ability to form dense biofilms, thus being a great danger to the public health, and it is necessary to be very careful in evaluating the meat before going on sale.

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P64

Study of the role of volatile organic compounds emitted by *Trichoderma* on the immune response of *Arabidopsis thaliana*

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Keywords: *Arabidopsis thaliana*, coculture chamber, immune response, *Trichoderma harzianum*, VOCs (volatile organic compounds)

Previous studies on plant-microorganism interaction mediated by volatile organic compounds (VOCs) have shown that certain fungi are able to promote the development and the defensive response of plants to various stresses. The importance in agricultural production of studying this interaction lies in the potential development of biological control methods which limit the use of current chemical products that are harmful for the environment. Thus, the effect of VOCs emitted by *Trichoderma harzianum* on *Arabidopsis thaliana* was studied using a novel coculture system. Coculture caused a delay in germination and a decrement in seedling development. The analysis of reactive oxygen species (ROS) production as a marker of innate immunity showed no differences in plants cocultured with fungus. However, coculture increased the expression of an innate immunity marker gene in plants. These results may be useful as a basis for further research to better understand the effects of VOCs on plants.

P65

Characterization of the mouse embryonic stem cell line

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Keywords: mouse embryonic stem cells (mESCs), pluripotency, self-renewal, 2D differentiation models

Embryonic Stem cells (ESCs), derived from the inner cell mass of the blastocyst, can proliferate indefinitely while maintaining the undifferentiated state (self-renewal) and have ability to give rise to all cell types of the adult body (pluripotency). These features make them ideal to generate clinically relevant models. ESCs-derived 2D and 3D platforms constitute robust cellular models to explore development and disease, and their use is expected to improve the cost-effectiveness and the success rate of drug discovery.
The aim of this study was to test if the ESC line E14Tg2α fulfilled the features of pluripotent stem cells. To this end, several assays, including growth curves, immunofluorescence and qPCR, were performed to evaluate their growth kinetics and confirm their self-renewal and pluripotency capacity.
Our data show that cultured E14Tg2α cells maintain ESC features and thus, they represent a valuable model to elucidate the molecular mechanisms underpinning mouse development.

P66

Epidermal Growth Factor Receptor: variant gene characterization and *in silico* analysis in canine mammary cancer – a comparative genomic approach

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Keywords: *EGFG*; *in silico*; mammary cancer; *Canis familiaris*

EGFR is a transmembrane receptor involved in the regulation of cell proliferation, differentiation, motility and survival. Searching for inhibitors of this protein binding is the aim of multiple studies. Using databases as STRING and KEGG can be an interesting in silico approach. This strategy has been recently described in humans. This same approach can be applied to other species, namely the dog, based on comparative genomics. *Canis familiaris* is a natural and spontaneous model for different diseases, including neoplasia and inflammatory diseases. A review publication from our group present the advantageous of the dog comparing with other animal models.

Mammary tumours are the most frequent neoplasm diagnosed in intact female dogs. Although frequently studied in human breast cancer, few studies have been devoted to analyse EGFR in canine mammary tumours, particularly concerning the search for *EGFR* gene variants using in silico strategies.

The aim of the present study is to describe *EGFR* gene variants in this animal model. In silico search is being used in order to answer the interrogation: what is the real role of *EGFR* gene and its variants in both human and dog mammary tumours? The comparative genomic approach is our first point for contributing to this scientific question.

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Evaluation of disinfection methods for *in vitro* propagation of *Vaccinium corymbosum*

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Keywords: *Vaccinium corymbosum*, disinfection, seeds, buds, contamination

Vaccinium is a genus of terrestrial shrubs in the Ericaceae family and consists of approximately 450 species, of which *Vaccinium corymbosum* is one of the three main species of blueberries domesticated in the 20th century. This culture is polyploid and highly heterozygous. The demand for blueberry has been increasing in recent years, as it is a plant that has advantages for the health of its consumers, such as reducing blood sugar levels, improving vision and anti-inflammatory effects. This small fruit is at the top of foods with the highest content of antioxidants, surpassing the advantages of other vegetables such as cabbage, spinach, and broccoli, in addition to being the red fruit richest in anthocyanins. *In vitro* propagation of *Vaccinium corymbosum* will allow to produce plants on a large scale, in a short time and in a controlled environment, allowing to have the plant available all year round and facilitating future studies.

This work aimed to determine the best disinfection method for *in vitro* propagation of blueberry. Different methods were carried out and were evaluated for each part of the plant, namely seeds and buds. For the seeds, 70% ethanol, NaClO diluted in water and three water washes were carried out. With regard to the buds, three methods were carried out that differed in the concentration of NaClO. In addition, a passage through hydrogen peroxide diluted in water was carried out. Over four weeks bacterial and/or fungal contamination was evaluated. The method used for the seeds was successful. The best disinfection method for the buds was proven to be the one with higher concentration of NaClO.

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FA-SAT ncRNA functional profile in different passages of a feline mammary cancer cell line

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Keywords: satellite DNA, *FA-SAT* ncRNA, FkMTp

In the last years, several evidence have recognized tandem repeat sequences, especially satellite DNA (satDNA), as being associated with biological process and diseases such as cancer.
FA-SAT was firstly described as the major satellite DNA sequence of the domestic cat. Yet, it is known now that this satDNA is conserved and transcribed in several Bilateria species, including human. Functional assays performed in normal cat and human cancer cells demonstrated that *FA-SAT* non-coding RNA (ncRNA) interacts with PKM2 protein and that it is a key player in cellular proliferation. Moreover, *FA-SAT* ncRNA silencing has shown to result in apoptosis.
In order to understand the *FA-SAT* ncRNA influence in tumor cells, we performed a functional study in several passages of a cat mammary tumor cell line – FkMTp. For that, *FA-SAT* antisense LNA GapmeR was used to deplete this ncRNA. Next, we analyzed its cellular phenotype and the transcription levels of PKM2 and MYC (as previously demonstrated their relation with *FA-SAT* levels).
Our results point to a conserved function in cat tumor and normal cells, being the phenotype of *FA-SAT* knock-down the same as previously reported for normal cells, that is, cell death. However, to determine the influence of this ncRNA in tumorigenesis further studies are needed.

P69

Hepatitis E Virus seroprevalence in wild boar (*Sus scrofa*) in Portugal

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Keywords: ELISA, Hepatitis E virus, seroprevalence, wild boar

Hepatitis E virus (HEV) infection is recognised as one of the primary causes of acute viral hepatitis in humans worldwide. A number of animal species, including wild boar, pigs, camels, and deer are reservoirs for HEV.

In order to investigate the importance of wild boar *(Sus scrofa)* in the epidemiology of HEV infection in Portugal, a serological survey was performed on samples from 351 wild boar from the Centre region of Portugal. Specific antibodies to HEV were detected with a commercial enzyme-linked immunosorbent assay (ELISA) (IDVet®, Montpellier, France). HEV-specific antibodies were detected in sera from five of 351 (1.4%; 95% CI: 0.6-3.3%) wild boar. Wild boar from the Centre of Portugal are exposed to HEV. The study demonstrates that wild boar could be reservoirs of infection for both livestock and humans.

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Seroprevalence of Hepatitis E virus in red deer (*Cervus elaphus*) in Portugal

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Keywords: ELISA, Hepatitis E virus, red deer, seroprevalence

Hepatitis E virus (HEV) is a causative agent of hepatitis. The disease is considered an emerging human viral disease in industrialized countries. Domestic pigs and wild boar are the main animal reservoir for HEV worldwide. Wild ruminants may be relevant in the epidemiology of HEV infection.

In order to investigate the significance of the wild red deer *(Cervus elaphus)* in the epidemiology of HEV infection, a serological survey was performed on samples from 297 wild red deer from the Centre region of Portugal. Specific antibodies to HEV were detected with a commercial enzyme-linked immunosorbent assay (ELISA) (IDVet®, Montpellier, France). We found that 9.1% of the samples (95% CI: 6.3-12.9%) were reactive for antibodies to HEV and regarded as seropositive. Red deer from the Centre of Portugal are exposed to HEV. The study demonstrates that red deer could be reservoirs of infection for both livestock and humans.

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Lactoferrin-loaded extracellular vesicles as a potential treatment against triple negative breast cancer

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Keywords: triple negative breast cancer, exosomes, lactoferrin, genetic engineering

Breast cancer is the most commonly diagnosed type of cancer worldwide. Triple negative breast cancer (TNBC) is a subtype of breast cancer that is more aggressive and has a poor prognosis, since it tests negative for epidermal growth factor receptor-2 (HER2), estrogen and progesterone receptors, thus not responding to conventional hormonal therapies. More selective tumor-specific treatments are thus urgently needed. Lactoferrin (Lf) is a natural milk-derived protein that has been demonstrated to be selectively cytotoxic against TNBC cells comparing to normal cells, in both *in vitro* and *in vivo* studies. Due to its selectivity, good tolerability by humans, effectiveness against TNBC and worldwide availability, Lf appears as an excellent alternative to fight TNBC. Though other routes for Lf administration can be considered, oral intake is by far the most suitable and commonly used. However, Lf passage through the digestive system can lead to its degradation, reducing the amount of Lf that reaches the target cells, and therefore decreasing its therapeutic effect. Protection of Lf by its encapsulation within nanocarriers is thus a promising alternative for its delivery. Exosomes are small extracellular natural vesicles released from cells that exhibit prominent features as nanocarriers including biocompatibility, inherent ability to transfer molecules between cells, high stability, and low toxicity. In this context, the aim of this work is to develop Lf-loaded exosomes decorated with a TNBC-targeting peptide to efficiently deliver Lf into TNBC cells potentiating its anticancer effect. The susceptibility of TNBC cells (MDA-MB-231 cells) to Lf has been studied and the strategy to produce the TNBC-targeting peptide and to load Lf into exosomes is currently being optimized.

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P72

The enemy is among us – isolation of *Trichoderma* spp. in the fur of our pets

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Keywords: cats; dogs; *Thricoderma* spp.; One Health

The genus *Trichoderma* are most commonly recovered from soil, but have also been isolated from air. Previously regarded as non-pathogenic to humans, *Trichoderma* spp. have emerged as new fungal pathogens in immunocompromised patients and peritoneal dialysis patients.
The aim of this study was to evaluate the occurrence of 341 in the fur of animals from private pet clinics and shelter animals in Northeast Portugal.
Samples were sent to the Medical Microbiology Laboratory of the University of Trás-os-Montes and Alto Douro, where cultures were performed in on Potato Dextrose Agar and Sabouraud Dextrose agar and incubated at 25 ºC and 37 ºC for 3 to 7 days.
Trichoderma were identified in culture from 45 animals. The occurrence in animals was 13.2% (CI 95%: 10.0-17.2%). Further research is required to better understanding the relevance of dogs and cats and their significance for public health in a One Health approach.

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Isolation of *Fusarium* spp. in Dogs and Cats in the Northeast of Portugal - A One Health Approach

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Keywords: cats; dogs; *Fusarium* spp.; One Health

Fusarium spp. are hyaline filamentous fungi that have become increasingly recognized as a cause of invasive fungal infections in neutropenic patients and in those undergoing transplantation. Fusarium species have long been associated with infections of the skin, nail and cornea but they seldom cause locally invasive disease in the immunocompetent patient. The aim of this study was to evaluate the occurrence of *Fusarium* in the fur of animals from private pet clinics and shelter animals in Northeast Portugal.

Samples were sent to the Medical Microbiology Laboratory of the University of Trás-os-Montes and Alto Douro, where cultures were performed in on Potato Dextrose Agar and Sabouraud Dextrose agar and incubated at 25 ºC and 37 ºC for 3 to 7 days.

Fusarium spp. were identified in culture from 27 animals. The occurrence in animals was 7.9% (CI 95%: 5.5-11.3%).

Since, *Fusarium* is an emerging agent in humans is required to better understanding the relevance of the isolation of the fungus in the fur and their significance for public health in a One Health approach.

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P74

Characterization of karyotypes of four species from Vespertilionidae family bats using classical cytogenetics

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Keywords: Bats; Cytogenetic; Karyotypes; Genomic diversity

Bats have been extensively studied in recent decades regarding the several species' karyotypes, which exhibit significant variability in terms of chromosome number, structure and genome organization. However, many bat species lack reported karyotypes, requiring this area further investigation. Cytogenetic studies in bats have proved to be important to understanding these species genetic diversity contributing to biodiversity knowledge, conservation, identification of isolated populations that may be at risk of extinction, among others.

Most bat species exhibit karyotypes with chromosome numbers ranging from 14 to 66, being the most common karyotype 2n=44. The great diversity of this group of mammals justifies the importance of the present work, that focuses on the cytogenetic characterization of four different species from *Myotis* and *Rhinolophus* genera (Vespertilionidae family), that inhabit the North of Portugal. Classical cytogenetic techniques, such as G- and C-banding were applied to enable the identification and pairing of the set of chromosomes of each species. These techniques were specific optimized to the different species chromosome preparations. The set of chromosomes of each species was analysed based on chromosome size, centromere position and banding pattern, resulting in the karyotypes organization So far, this study provides crucial insights into the karyotypic diversity of bat species, contributing as a foundation for future research on evolutionary relationships among this group of species and bringing insights at the taxonomy level.

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P75

DNA repair capacity and its relationship with variants in the *PARP1* gene

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Keywords: PARP1, repair, SNP, AS-PCR, primers

The *PARP1* [*Poly (ADP-ribose) polymerase 1*] gene, located at position 1q42.12 in *Homo sapiens*, encodes a protein that is involved in various nuclear processes. The PARP1 protein is one of the most studied enzymes due to its role in maintaining the genome's integrity by participating in different DNA repair pathways. This protein has a crucial role as a regulator of the process of identifying and processing single-strand breaks (SSB) of the DNA molecule through the base excision repair (BER) mechanism. Currently, there are more than 1000 SNPs of the *PARP1* gene reported, several in the coding region (cSNPs). This study focuses on the rs1805414 variant that is associated with the risk of several types of cancer, including breast cancer. This variant is characterized by the alteration of a thymine (wild type) by a cytosine (SNP) in exon 7, which corresponds to the codon that originates the amino acid at position 284, in the PADR1 domain of the PARP1 protein. This nucleotide alteration does not cause an amino acid change, keeping an alanine in that position. Genotyping will be performed using the Allele Specific PCR (AS-PCR) technique directed to the *PARP1* gene. There will be used primers that amplify fragments with 153 bp specific for the wild type allele and for a variant and a common reverse primer. In addition to these, an internal control primer for GAPDH with a product of 495 bp will be used. As the primers are allele-specific, we will only get results if that allele is present. In PCR1 we have the T forward primer, so we will only get results if thymine is present. In the case of PCR2 with the C forward primer, we will detect the presence of cytosine. For further confirmation of results, some samples will be sequenced. Through the in silico analysis, we were able to obtain the primers for sequencing.

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P76

A case report of dysplasia with der(11)t(11;18)(q13;q11.2),-18.

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Keywords: Dysplasia, Conventional Cytogenetics, FISH, Leukemia

Myelodysplastic syndromes (MDS) belong to the group of myeloid neoplasms, occur mainly, but not exclusively, in patients over 70 years of age, with an incidence of 4-5 cases per 100,000 inhabitants, and a predominance in males. Multiple myeloma originates from the lymphoid B cell lineage at the final stage of differentiation, has an incidence of 10% among all leukemia cases, with prevalence in individuals over 65 years old.

The authors present a case of an 89-year-old man with severe thrombocytopenia, anemia, and monoclonal gammopathy of undetermined significance. The clinical indication referred to suspicion of MDS or MM. Conventional cytogenetics and fluorescent *in situ* hybridization (FISH) were performed. Two probe panels were used for the most common chromosome alterations in MDS (5, 7, 8 and 20) and MM (13, 17, t(4;14) and t(11;14)). Twenty metaphases were analysed, seventeen of which showed a derivative of chromosome 11 resulting from a translocation between chromosome 11 (q13) and chromosome 18q, with monosomy of chromosome 18p and loss of the segment 11q13 to 11qter. FISH analysis detected only one signal for chromosome 11 in the t(11;14) probe, which confirms the 11q13 breakpoint. Region 11q13 to 11qter of chromosome 11 contains several genes in particularly the *KMT2A* (myeloid/lymphoid or mixed lineage leukemia) implicated at least 10% of acute leukemias leading to tumorigenesis and deregulation of hematopoiesis, and the *CBL* (Cas-Br-M (murine) ecotropic retroviral transforming sequence) gene found in MDS and myeloproliferative syndromes which undergo loss of ubiquitin ligase activity and an increase in cellular proliferation. Deletion of chromosome 18p is a rare alteration described in the literature reported in only 14 cases of myeloid lineage and associated with a complex karyotype. Chromosome 18p contains essential genes, including *SMCHD1* (Structural maientance of chromosomes flexible hinge domais containing 1) and *TGIF1* (TG-interacting factor 1), which, when lost, cause hematological changes and deregulation of hematopoiesis. In the presented case, conventional cytogenetics detecting a malignant clone in 85% of the analysed metaphases, was crucial for the diagnosis and to understand the development of the neoplasia.

P77

Antibiotic resistance in *Staphylococcus aureus* isolated from chicken bursitis for human consumption

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Keywords: *Staphylococcus aureus*, multidrug resistance, chickens

Antibiotic resistance is recognized as one of the greatest threats to global human health as modern medicine continues to rely heavily on the success of antimicrobial therapy. As microorganisms do not recognize the animal-human boundaries, human and animal medicine must be interconnected. *Staphylococcus aureus* is a gram-positive bacterium normally commensal, but can sometimes cause infections, as in the case of bursitis in chickens. Therefore, this study aimed to investigate the antimicrobial resistance in *S. aureus* strains isolated from chicken bursitis. In this study, 24 *S. aureus* strains previously isolated from chicken bursitis were used. The antimicrobial susceptibility of the isolates was tested using the Kirby-Bauer diffusion disc method against fourteen antibiotics. Eleven (45.8%) of the twenty-four strains tested showed resistance to more than three antimicrobial classes, being considered multidrug resistant. Furthermore, among the isolates, resistance to penicillin (n=2), cefoxitin (n=2), ciprofloxacin (n=2), tetracycline (n=2), chloramphenicol (n=2) and trimethoprim-sulfamethoxazole (n=2), were detected. None of the isolates showed resistance to linezolid nor to mupirocin. Subsequently, the genotyping of the isolates was performed to identify the genes that confer resistance to different types of antibiotics. The presence of four genes that confer resistance to tetracycline was tested, and only the presence of the tetK gene in strain number 11 has been proven. *S. aureus* may be an etiological agent of chicken bursitis and the isolates in this study carried resistance to several antimicrobials. Our findings are worrisome since *S. aureus* in chickens may be transmitted to humans through food or by direct contact. It can be concluded that antibiotics such as linezolid, mupirocin, penicillin, cefoxitin, ciprofloxin, chlorophenicol and trimethoprim-sulfamethoxazole may be a good choice for the control of these infections.

P78

Mucor spp. in pet animals – a contribution to the study of mucormycosis

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Keywords: cats; dogs; *Mucor* spp.; COVID-19

Mucormycosis is a serious, but rare opportunistic fungal infection that spreads rapidly, and hence prompt diagnosis and treatment are necessary to avoid high rate of mortality and morbidity rates. Recent studies have documented alarming number of COVID-19 patients with mucormycosis infection associated with other comorbidities. Mucormycosis is mainly caused by *Rhizopus* and *Mucor* spp. The aim of this study was to evaluate the occurrence of *Mucor* spp. in the fur of animals from private pet clinics and shelter animals in Northeast Portugal. Samples were sent to the Medical Microbiology Laboratory of the University of Trás-os-Montes and Alto Douro, where cultures were performed in Potato Dextrose Agar and Sabouraud Dextrose agar and incubated at 25 ºC and 37 ºC for 3 to 7 days. *Mucor* spp. were isolated in culture from 50 animals. The occurrence in animals was 14.7% (CI 95%: 11.3-18.9%). Occurrence of *Mucor* spp. is high in this study. More studies are required to better understanding the relevance of the isolation of the fungus in the fur and their significance for humans in a One Health approach.

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P79

An immunohistochemistry study of MMP-9 in female dog mammary tumours

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Keywords: canine, metalloproteinase-9, immunohistochemistry, mammary

Dogs develop mammary tumours with similar biological and histopathological behaviour to humans makes them an excellent model for comparative medicine in studying human breast cancer in women. The metalloproteinase-9 (MMP-9) affects the growth and renewal of the matrix; it has become increasingly important due to their involvement tumour development and progression, and their overexpression has been frequently observed in several oncology studies. One of the principal stages of carcinogenesis is metastasis, for that to occur the cells must have a disruption in the base membrane and reach the blood stream. In this process its suggested that the MMP-9 as a leading part in the remodelling of the matrix, allowing the migration of the cells. Thus, the aim of this study was to evaluate the immunoexpression of MMP-9 in canine malignant mammary tumours by immunohistochemistry. In the studied tumours, increased expression of MMP-9 was significantly between the tumor size (p= 0,006), histological type (p=0,017), high nuclear grade (p=<0,001), low differentiation degree (p=<0, 001), high mitotic index (p=<0,001), high degree of malignancy (p=<0.001), presence of necrosis (p=0,0016), presence of lymph node metastases (p=0,001) and presence of plungers (p=<0,001). Our results are in agreement with certain studies that have been done in human medicine and veterinary medicine, suggesting an association between MMP-9 and the neoplastic progression of the tumour. Thus, the study allowed us to conclude that this marker may play a key role in tumour progression, suggesting the importance of MMP-9 in the clarification of human breast cancer and creation of new therapies.

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P80

Ketotifen DNA damage in the mouse model of HPV16

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Keywords: Antihistamine, HPV16, murine, disease, oxidative damage

Ketotifen is an antihistamine with mast cell stabilizing and degranulation-inhibiting properties. Mast cells trigger inflammatory response because when degranulating they release substances that take part in many biological processes such as angiogenesis. This cells´ infiltration was found in skin lesions identified in transgenic K14HPV16 mice.
Thus, it was our aim to evaluate effect of ketotifen administration on oxidative, total and basal DNA damage (NSS, NSS+GDI, GDI) via comet assay using lymphocytes. To achieve this goal, tree concentrations of ketotifen solutions (5mg/Kg, 7.5 mg/Kg and 10 mg/Kg) were administrated in the mice´s beverage for a 6-week period. There was evidence that the transgenesis influences the GDI. Also, we saw that the GDI lowers with the ketotifen intake. This is because this drug reduces inflammation and oxidative stress lowering the related DNA damage that happens when cells are exposed to this kind of environment. GDI+NSS and NSS did not show the same behaviour in all groups, but at the 5 mg/Kg concentration there was always an accentuated lowering of levels. However, on the higher concentration, 10mg/Kg, was observed a GDI increasement which indicates toxicity. So, ketotifen in moderate doses appeared to be safe since there was no negative influences in DNA damage.

P81

A preliminar study on *Brucella* spp. seroprevalence in wild boar (*Sus scrofa*) in Portugal

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Keywords: *Brucella* spp., ELISA, seroprevalence, wild boar

Various species of the genus *Brucella* are highly virulent zoonotic agents. *Brucella melitensis*, *B. abortus*, and *B. suis* are broadly spread worldwide and animal brucellosis has a significant economic impact. This zoonotic disease affects both domestic pigs and wild pigs such as wild boar (*Sus scrofa*).

In order to investigate the importance of wild boar *(Sus scrofa)* in the epidemiology of *Brucella* infection in Portugal, a serological survey was performed on samples from 184 wild boar from the Centre region of Portugal. Specific antibodies to *Brucella* spp. were detected with a commercial enzyme-linked immunosorbent assay (ELISA) (IDVet®, Montpellier, France). *Brucella*-specific antibodies were detected in sera from 74 of 184 wild boar (40.2%; 95% CI: 33.4-47.4%). Wild boar from the Centre of Portugal is exposed to *Brucella* spp. The risk of transmission of swine brucellosis to humans is regarded as minor due to low zoonotic potential, considerable awareness, and biosafety measures, but it should not be underestimated.

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A serological survey of *Brucella* spp. in red deer (*Cervus elaphus*) in Portugal

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Keywords: *Brucella* spp., ELISA, red deer, seroprevalence

Wildlife have been identified as important sentinels for the surveillance of zoonotic pathogens such as *Brucella* infection. Systematic brucellosis monitoring in wildlife is not demanded by regulatory acts, but several studies have reported the presence of this infection in European countries. In order to investigate the importance of red deer *(Cervus elaphus)* in the epidemiology of *Brucella* infection in Portugal, a serological survey was performed on samples from 276 hunted red deer from the centre of Portugal. Specific antibodies to *Brucella* spp. were detected with a commercial enzyme-linked immunosorbent assay (ELISA) (IDVet®, Montpellier, France). *Brucella*-specific antibodies were detected in sera from 18 of 276 (6.5%; 95% CI: 4.2-10.1%) red deer. The seroprevalence of *Brucella* spp. in Portuguese red deer seems to be relatively low, but the red deer population has still to be considered as potential reservoir for *Brucella* spp. transmission to domestic ruminants.

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How gene transcripts number affects the cracking index in sweet cherry

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Keywords: absolute quantification, cracking index, dPCR, gene expression, sweet cherry

Sweet cherry cracking, a physiological disorder that occurs during fruit growth and ripening, remains a great challenge to the producers, highly affecting the fruits commercial value and decreasing the orchard profitability. Actually, digital PCR (dPCR) emerges as novel and innovative approach to study gene expression, based on absolute quantification of transcripts number, making this technique more reliable than qPCR in gene expression studies. Thus, this study intended to quantify the absolute transcripts number of cracking related genes in sweet cherry, using cherry fruits collected from cultivar Burlat installed in an orchard located in Resende, where calcium (150 g/hL and 300 g/hL), seaweed (*Ascophyllum nodosum*) based-biostimulant (75 mL/hL and 150 mL/hL) and a combination of both nutrients (300 g/hL of calcium and 150 mL/hL of seaweed) were applied at foliar level during fruit development. From each treatment, RNA was extracted from fruit exocarp, cDNA was synthetized and then the absolute transcripts quantification of some cuticle genes, like *PaExp1*, *PaExp2*, *PaXTH*, *PaEG*, *Paβ-Gal* and *PaCYP78A9*, was done by dPCR. The results were normalized by housekeeping gene, *PaAct*, and correlated with cracking index (CI) rate. The preliminary results showed a negative correlation among gene expression based on transcripts number and CI in *PaExp1*, *PaExp2*, *PaEG* and *Paβ-Gal* genes, and a positive correlation in *PaXTH* and *PaCYP78A9* genes, suggesting that these nutrients have considerable effect in molecular mechanisms involved in cherry cracking.

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Can the aquaporin gene, *PaPIP1;4*, be involved in the molecular mechanisms of sweet cherry cracking?

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Keywords: aquaporin, cracking index, dPCR, gene expression, sweet cherry

Aquaporins (AQPs) play an important role in water transport in flesh fruits like sweet cherry and, thus, the expression of AQPs genes have been associated to the molecular mechanisms involved in cracking. Thus, this study intended to analyze a cherry aquaporin gene, *PaPIP1;4*, using fruits where different nutrients were applied at foliar level during the fruit development trying to reduce cherry cracking, namely calcium (150 g/hL and 300 g/hL), seaweed (*Ascophyllum nodosum*) based-biostimulant (75 mL/hL and 150 mL/hL) and a combination of both nutrients (300 g/hL of calcium and 150 mL/hL of seaweed). From each treatment, total RNA was extracted from fruit exocarp and then reverse transcribed into cDNA. A semiquantitative analysis of *PaPIP1;4* gene was done, enabling to observe differences among treatments. These results were complemented with an analysis of gene expression by dPCR and also correlated with cracking index. All results were normalized by housekeeping gene, *PaAct*, and a positive correlation was found among gene expression by dPCR and cracking index. Thus, cherries treated with 150 g/hL of calcium presented the lowest expression of *PaPIP1;4* gene and the lowest cracking index while cherries treated with a combination of both nutrients had the highest cracking index and the highest gene expression, suggesting a plausible involvement of *PaPIP1;4* gene in molecular mechanisms of sweet cherry cracking.

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P85

Optimisation of a Tetraplet-Primed PCR assay for the detection of the CCTG repeat in the *CNBP* gene

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Keywords: *CNBP,* $(CCTG)_n$ expansion, myotonic dystrophy type 2, Tetraplet-primed PCR

CNBP is a protein-coding gene located in the 3q21 chromosomal region. This gene contains a $[TG]_n[TCTG]_n[CCTG]_n$ motif complex, in which all three forming regions are highly susceptible to expansion. The increase in the number of (CCTG)n repeats is the cause of myotonic dystrophy type 2 (DM2; MIM #602668), an autosomal dominant degenerative disorder that affects muscles in multiple systems. Normal *CNBP* alleles have up to 30 repeats, and pathogenic alleles contain from 75 to up to 11 000 repeats. Tetraplet-Primed PCR (TTP-PCR) is a method which could be used to diagnose DM2 since it provides accurate and rapid identification of expanded CCTG tracts in pathogenic alleles by displaying a ladder pattern profile This study aimed to optimise the TTP-PCR, verify the validity of this technique as a diagnostic method for suspected cases of DM2 and simplify the testing procedure.

Acknowledgements: Grant References: UIDB/00215/2020; UIDP/00215/2020; LA/P/0064/2020.

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Evaluation of the genetic diversity of chestnut trees of the Martaínha variety with iPBS and ISSR markers

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Keywords: *Castanea sativa*, genetic diversity, Martaínha, molecular markers

Chestnut (*Castanea sativa* Mill.) culture is currently considered a strategic activity in the Portuguese economy, especially in the interior mountain regions. Among the several chestnut varieties on the market, the Martaínha variety is one of the first to be harvested, giving it a huge strategic advantage and excellent quality, with an enormous aptitude for fresh consumption. As a result of climate changes, chestnut trees face new challenges that weaken them and make them more prone to pests and diseases. As its cultivation has decreased genetic variation, the diversity of the European chestnut tree has been evaluated in order to maintain its conservation and better understand its adaptive potential.

This work was developed to evaluate the genetic diversity of the chestnut Martaínha variety from "Castanha dos Soutos da Lapa" using the molecular markers iPBS (inter-primer binding site) and ISSR (Inter-Simple Sequence Repeats). The obtained results showed a relatively high level of genetic similarity (51%) among the analysed samples, evidenced by similar iPBS and ISSR patterns produced by different primers. The verified low genetic variability of the selected individuals can be justified because they are all from the same variety and have the same origin. The preliminary results obtained in this work indicated that it is possible to analyse the genetic variability of chestnut trees through the molecular markers used. However, the sample size and the number of primers should be substantially increased.

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In silico analyses for subsequent genotyping and sequencing of *DPYD* gene in Clinical Pharmacogenetics

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Keywords: Genotyping; Sanger Sequencing; HRM; Pharmacogenetics; *In Silico*; Clinical

Pharmacogenetics and pharmacogenomics are emergent fields that aim to elucidate genetic basis for inter-individual differences in drug response. Genotype analysis of *DPYD* gene is relevant in oncology, providing information on the risk of toxicity or effectiveness of treatment. Outcomes are improved by testing the variants presented in *CPI Consortium* and *Dutch PWG* guidelines. *In Silico* studies have been conducted to determine and confirm primers and probes, and to standardize genotyping using different protocols (e.g. hydrolysis, HRM and Sanger Sequencing). We have compiled the whole sequence of these genes and organized information from different genomic databases, together with 'primer/probes' design from different scientific companies.
Preliminary *in silico* analysis is germane to obtain precise data of the genes of interest and check for inconsistencies, providing reliable interpretation of results for appropriate management of patients.

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In silico analysis to optimize the genotyping of the rs17655 variant in the *ERCC5* gene

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Keywords: DNA repair, NER, ERCC5, Variants, rs17655

DNA is susceptible to damage and alterations from exogenous and endogenous processes that can interfere with essential cellular mechanisms. Identifying, signalling and repairing these damages depend on the type of damage and its origin. The NER (Nucleotide excision repair) mechanism is the main repair mechanism for DNA damage (bulky adducts) caused by pyrimidine dimers and photoproducts produced by the UV component of solar radiation, more specifically the UVc range. Several proteins are involved, and the malfunction of the mechanism causes diseases such as Cockayne Syndrome and Xeroderma Pigmentosum G (XP-G), as well as a higher predisposition for the development of cancer.
The *ERCC5* gene (Excision Repair, endonuclease) has 15 exons and encodes the XPG protein, a specific endonuclease involved in the 3' incision in the NER mechanism, which allows removing the damaged DNA sequence so that a new sequence can be synthesized in the same chain without the original damage. Variants of this gene have already been documented as possible predisposing factors for the development of various carcinomas, such as variants rs17655, rs751401, rs2094258, rs1047768. The rs17655 variant seems to be correlated with the development of cancer, with breast cancer being one of most documented. These studies are mostly carried out in Asian populations, with scarce references of the possible association in European populations. Genotyping the *ERCC5* gene for this variant in a European population would be a good method to assess the occurrence of this association. This variant is located at exon 15 and is a missense where an Aspartate (GAT) is replaced by a Histidine (CAT). The G allele is dominant and C allele is recessive. According to the "ALFA Allele Frequency" database the frequencies are 0.78 for the G allele and 0.22 for C allele in Europe. To carry out a genotyping an *in silico* approach is necessary in order to understand the characteristics of the variant, its location, the primers and the genotyping strategy to be implemented. The information at NCBI, Ensembl and UCSC databases was used and the conventional PCR technique with two primers were selected from the primer3plus software. The PCR-RFLP technique with the restriction enzyme *Hpy166II* was tested. The allele and genotype frequencies reported for the European population will be subsequently compared with the experimental result.

Acknowledgements: CITAB (Centre for the Research and Technology of Agro-environmental and Biological Sciences). This work is supported by National Funds from FCT - Portuguese Foundation for Science and Technology, under the project UIDB/04033/2020.

WORKSHOPS

DNA & RNA Biosensing platforms

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There are several biosensing platforms used in our daily lives, based on the detection of different types of analytes. In this workshop, we will focus on the platforms that aim to detect nucleic acids, and we will explore the various alternatives that have been reported in the literature. The different areas of applications will be addressed, as well as the potential that these systems have in each sector. We will end with the demonstration of a biosensor developed by a consortium between UTAD, REQUIMTE and ISEP in the context of the SARS-CoV2 testing.

Research funding: Cdots Biosensing COVID19, nº 041_596518523, financed by RESEARCH COVID-19 of the Portuguese Foundation for Science and Technology.

SSRs for genotyping plant genetic resources

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This workshop intends to provide an overview of the application of the SSR molecular marker for the plant genetic diversity assessment, specifically in species belonging to genus Vigna. During the workshop will be addressed the theoretical concepts of SSR molecular marker, its applications to plant breedings and management of germplasm collections. In the practical part, the participants will have the opportunity to analyze SSR genotyping data in species of the genus Vigna with specific softwares, identify varieties and determine relationships between them and prepare dendrograms.

Collection and preservation of samples in forensic science- from necropsy to lab

Isabel Pires and Anabela Alves

Keywords: forensic, necropsy, samples collection

A post-mortem examination is essential to identify the cause and circumstances of death. Even in the age of molecular pathology, necropsy remains the most valuable tool for understanding the organism and the disease. Forensic science is about using scientific knowledge to support the application of the law. However, for the evidence to be valid, the sampling and preserving samples must consider some rules. In fact, the forensic investigator's biggest nightmare is that the evidence will not be admissible in court.

In this workshop, we will perform necropsies of various animal species to collect samples for further forensic analysis: genetics, toxicology, ballistics and histopathology.

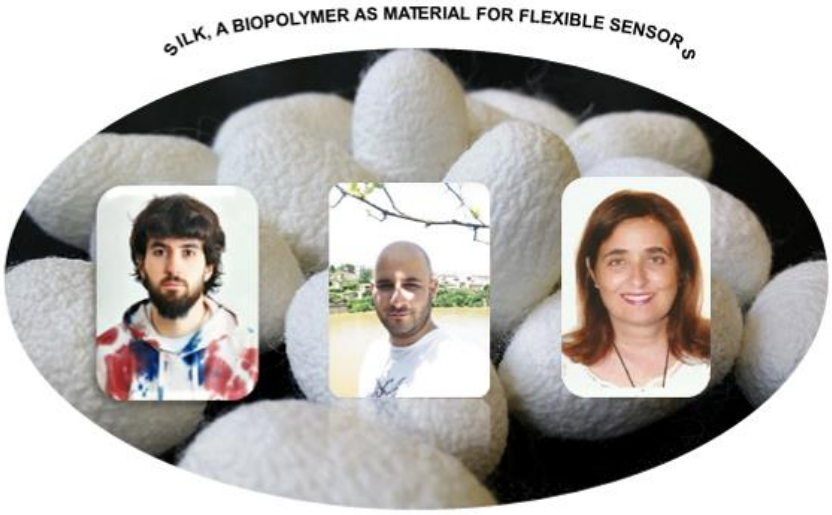
The Power of Microscopy in Research

Daniela Ferreira, Sandra Louzada and Mariana Lopes

Microscopy constitutes an important technique used to visualize and study cellular structures and their function. Over the years, several types of microscopes have been developed to meet investigative demands, allowing to observe fixed or live cells, chromosomes, and specific cellular components. One of the main advantages of such technique is the ability to provide single-cell analysis, essential for data validation and complementation of results obtained with other methods. Moreover, the detailed cell-to-cell analysis provided by microscopy, allied to the use of specific imaging softwares, has contributed to a great advance in biological research.

This workshop will consist in a brief theoretical introduction to some concepts of microscopy and its various applications in biological research, followed by a hands-on approach where the participants will have contact with different types of microscopes and applications as well as training in softwares for image analysis.

Silk, a biopolymer as material for flexible sensors



Guilherme J.G. Sousa, Tiago A.G. Duarte and V. de Zea Bermudez

Nature is an inexhaustible source of inspiration for the creation of innovative materials and devices with improved performance. In this context, silk appears, a biopolymer with a millennial history in the textile and biomedical industry. The extraordinary intrinsic characteristics of silk fibers, such as "self-assembly", biocompatibility, non-toxicity, superior mechanical properties and piezoresistivity (change of resistance that can be measured on physical contact), among others, offer a wide range of potential application in technological areas of the greatest relevance, such as optics, electronics and energy.

This workshop intends to provide an overview of silk science and highlight the tremendous technological potential, namely on sensors field. The basics of silk processing follow by a silk film synthesis and sensor circuit printing will be presented and explored in practical laboratory experiments. The participants will perform degumming of silk fibers harvested from Bombyx mori silkworm cocoons and obtain a silk-based film. The final step will consist of a demonstration on printing of the electronic sensor circuit on a silk film.

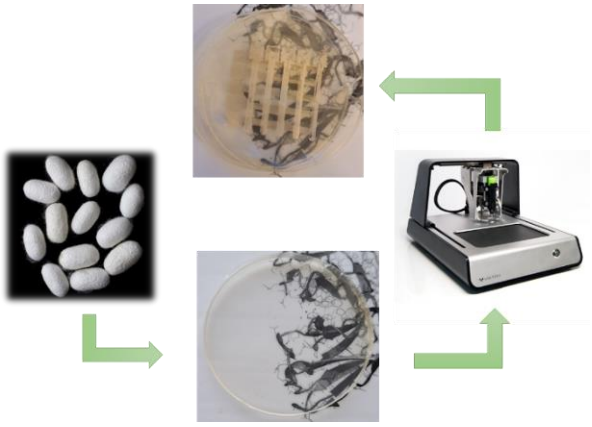


Figure 1. Illustration of the process from the silk cocoons until the printed silk film.

Breaking the code –bioinformatic tools in cancer research, diagnosis and treatment

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Keywords: Bioinformatics, Variant analysis, Diagnosis, Prognosis, Case studies

In recent years, scientific research has revealed an increasing number of variants associated to different diseases. Particularly in cancer, these mutations can be used in diagnosis, prognosis, therapeutic decision, follow up of patients and risk population assessment. Its identification is thus a very important and valuable tool in the fight against this group of diseases. Sequence technologies as Sanger and Next-generation sequencing are widely used for mutation detection in clinical settings. But the analysis relies on a strong bioinformatics approach, where the data generated from sequencing technologies is a critical process involving base calling, read alignment, variant identification, and annotation. During this process, the sequence information is compared to a reference to identify whether there are any variants in the targeted sequences. The annotation and interpretation processes are then set to identify and classify each variant and their clinical significance. In the present workshop, the participants will be challenged to solve clinical cases, identifying the presence/absence of genome mutations through the analysis of sequence data using bioinformatics' software and web-based tools. The exploration in databases and genome browsers will allow the interpretation of the detected mutations and the discussion of disease diagnosis, prognosis, and therapeutics.

Making bridges – solving problems. A Hands-on practical *in silico* workshop

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[5] MSc students, Post-Graduation in Molecular Comparative and Technological Genetics, Graduation in Genetics and Biotechnology

Keywords: Bioinformatics, diseases

Test you, in group, and solve a genetic enigma, using *in silico* tools!

We give you a code. You give us your result!

Finally, we evaluate the groups and congratulate the winner!

In 1h30m, the correct answer will be find with the help of our team.

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The speakers for their willingness to accept our invitation, to enrich this event with their conferences, and for the transmission of scientific knowledge.

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All the participants for demonstrating interest in joining this event and for some of them presenting their scientific work enriching greatly these days. Without you, this event would be meaningless.

The students of the Secondary Schools who participated in this event presented their Junior Posters.

The sponsors that support partially this event.

Overall, to all who have contributed to the success of the XV JGB | V JIGB.

Thank you very much.

The Organizing Committee of the XV JGB | V JIGB

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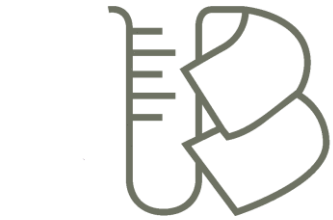
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