XX ENCONTRO NACIONAL DE BIOLOGIA EVOLUTIVA

19-20 Dezembro 2024, Vila do Conde

19 – 20 December 2024
BIOPOLIS | Vila do Conde, Portugal

ABSTRACT BOOK

BIOPOLIS

cibio

CÂMARA MUNICIPAL VILA DO CONDE

InBIO RESEARCH NETWORK
Biodiversity and Evolutionary Biology

apbe

Ciências ULisboa

U.PORTO

Initiative supported by the European Union's Horizon 2020 Research and Innovation Programme under the Grant Agreement Number 857251

Organizing Committee

Ana M Lopes (CIBIO, Vairão, Portugal)

Angelica Crottini (CIBIO, Vairão, Portugal)

Diana Lobo (CIBIO, Vairão, Portugal)

Diogo Lima (CIBIO, Vairão, Portugal)

João P Marques (CIBIO, Vairão, Portugal)

Manuel Curto (CIBIO, Vairão, Portugal)

Raquel Tavares (CIBIO, Vairão, Portugal)

Roberto Arbore (CIBIO, Vairão, Portugal)

Rui Faria (CIBIO, Vairão, Portugal)

Silvia Carvalho (CIBIO, Vairão, Portugal)

Vítor Sousa (CE3C and FCUL, Lisbon, Portugal)

Scientific Committee

Alice Pinto (CIMO e IPB, Bragança, Portugal

Filipe Castro (CIIMAR, U. Porto, Portugal)

Filipe Pereira (UTAD, Vila Real, Portugal)

Isabel Amorim (CE3C, U. Açores, Portugal)

Lara Baptista (NIOZ, Netherlands e CIBIO-Açores, Portugal)

Maria Romeiras (ISA, U. Lisbon, Portugal)

Martim Melo (CIBIO, Vairão, Portugal)

Patrícia Beldade (CE3C and FCUL, Lisbon, Portugal)

Pedro Soares (CMBA e U. Minho, Braga, Portugal)

Raquel Tavares (CIBIO, Vairão, Portugal)

Ricardo Pereira (SMNS, Stuttgart, Germany)

Rui Faria (CIBIO, Vairão, Portugal)

Sara Santos (MED, U. Évora, Portugal)

Sónia Cruz (CESAM, U. Aveiro, Portugal)

Susana Varela (IGC e ISPA, Oeiras-Lisboa, Portugal)

Organization

Institutional Support

Funding

Initiative supported by the European Union's Horizon 2020 Research and Innovation Programme under the Grant Agreement Number 857251

Apoio no âmbito do Acordo de Mecenato entre a Universidade do Porto e a Caixa Geral de Depósitos

Sponsors

Short Programme

Centro Municipal da Juventude, Vila do Conde [Directions]

DAY 1: 19th December 2024

11:00: Registration and Poster placement

13:30: Opening Ceremony

14:00: Plenary Talk | José Cerca

14:40: Talks 1 to 7

16:04: Coffee Break & Posters

16:34: Plenary Talk | Allowen Evin

17:14: Talks 8 to 14

18:38: Homage to Prof. Amorim

19:15: End of first day

9:00: Plenary Talk | Margarida Moreira

9:40: Talks 15 to 20

10:52: Coffee Break & Posters

11:22: Talks 21 to 26

12:35: Group Photo

12:40: Lunch Break

14:00: Plenary Talk | Rebecca Mead

14:40: Talks 27 to 34

16:16: Coffee Break

16:45: Poster Session

17:45: Closing Ceremony

18:00: General Assembly of APBE

DAY 2: 20th December 2024

Detailed Programme

Centro Municipal da Juventude, Vila do Conde [Directions]

DAY 1: 19th December 2024

11:00: Registration and Poster placement

13:30: **Opening Ceremony**

14:00: Plenary Talk | **José Cerca** | **Adaptive radiation on Oceanic Islands; Tales of ecological niches and ecological generalists)**

14:40: Talk 1 | Bárbara Freitas | Does song act as a behavioural barrier to gene flow? Evidence from divergent populations of La Palma's Canary Islands Chaffinch

14:52: Talk 2 | Lara Almeida | Evolutionary history of the Mus musculus in Cabo Verde

15:04: Talk 3 | Maria Romeiras | Patterns of diversification and colonization in Macaronesian Apiaceae lineages

15:16: Talk 4 | Estêvão Faustino | Hybridization between currently allopatric species at the root of speciation? The case of Iberian chubs (Genus Squalius)

15:28: Talk 5 | Tristan Bertrand | Comparative Population Genomics Illuminates Species Boundaries and Symbiotic Disruption in *Eunicella Octocorals*

15:40: Talk 6 | Salomé Barreto | Ecological differentiation influences the evolution of pre-zygotic isolation in spider-mite species

15:52: Talk 7 | Ivo Chelo | Can reproductive isolation arise from epistatic deleterious and compensatory mutations in small populations: an experimental test with *C. elegans*

16:04: Coffee Break & Posters

16:34: Plenary Talk | **Allowen Evin** | **8,000 years of domestic plants and animals evolution : understanding local adaptation under socio-economic and climatic fluctuations**

17:14: Talk 8 | Pedro Sousa | Understanding hybridization between alpine hares in a context of climate-induced range shifts

17:26: Talk 9 | Carlos Yadro | Population structure and environmental adaptation in two honey bee subspecies from southern European refugia: *A. M. ligustica* and *A. M. iberiensis*

17:38: Talk 10 | José Costa | Genetic divergence and local adaptations in endemic hares: Insights from the Italian and Broom hares

17:50: Talk 11 | Dora Henriques | population structure and conservation status of middle eastern honey bee subspecies

18:02: Talk 12 | Matthew Moreira | Distribution of intraspecific diversity across species ranges: insights from herptiles in six global regions

18:14: Talk 13 | Susana Almeida | Phylo-transcriptomics of the giant kelp *Macrocystis pyrifera* reveals multiple trans-oceanic dispersal events across the Southern Ocean

18:26: Talk 14 | Maria Carolina Matos | FCRL ancestry: can we truly find parallels between placental mammals, marsupials and monotremes?

18:38: Homage to Prof. Amorim | **Sandra Martins** | **New paralogs of ataxin-3 identified in primates and the constraint evolution of *ATXN3L* retrocopy**

19:00: Homage to Prof. Amorim | **Maria João Prata** | **António Amorim: a singular professor**

19:15: End of first day

DAY 2: 20 December 2024

9:00: Plenary Talk | **Margarida Cardoso Moreira** | **Origins of cells and organs – the view from the placenta**

9:40: Talk 15 | Inês Miranda | The origin and maintenance of adaptive seasonal camouflage in the least weasel

9:52: Talk 16 | Diogo Berjano | How do biochemical and environmental factors shape the fitness landscape of synonymous mutations on Hsp90?

10:04: Talk 17 | Joel Laia | Monoterpene synthase ligand preference: a case of both divergent and convergent evolution in Menthinae, Lamiaceae

10:16: Talk 18 | Beatriz Sousa | Evolutionary and functional study of the thyroid Sodium/Iodide Symporter homologs in plants

10:28: Talk 19 | Ana Serra Silva | Very Weak Support for Deuterostome Monophyly is Boosted by Long Branch Attraction Artefacts

10:40: Talk 20 | Bruno Nevado | Phylogenomics of Lupinus reveals strong geographic structuring and multiple instances of phenotypic convergent evolution

10:52: Coffee Break & Poster Session

11:22: Talk 21 | Sandra Estela Moreno Fernández | Lineage diversification of Acrocephalidae family (Reed Warblers) and its relationship with migratory behaviour and islands colonisation

11:34: Talk 22 | Carolina Peralta | Characterization of polymorphic inversions in locally adapted populations of *Clunio marinus*

11:46: Talk 23 | Carla Gonçalves | A turbulent evolutionary history involving massive gene gains and losses has shaped the genome and metabolism of a fungal lineage

11:58: Talk 24 | Mario Torralba Sáez | A shark's eye view on genome size evolution: phylogenetic patterns, molecular causes, and phenotypic consequences

12:10: Talk 25 | Daniel García Souto | Phenotypic and genetic differentiation between two chromosomal taxa of the gastropod Nucella lapillus at Galician rocky shores (NW Spain)

12:22: Talk 26 | Catarina Branco | Identifying the sex chromosomes of *Laurus azorica*

12:35: Group Photo

12:40: Lunch Break

14:00: Plenary Talk | **Rebecca Mead | The Sandwalk: Time for Thinking about Evolution Education**

14:40: Talk 27 | Xana Sá-Pinto | Stronger together: a revision of stakeholder impact of NEDE.APBE's 12 years of education and outreach in evolution

14:52: Talk 28 | Javier Oñate Casado | When individuality obscures geographic song variation: a comparison of two passerine sister species with different migratory strategies

15:04: Talk 29 | Gabriel Munar Delgado | Popular birds live longer: The association of social bonds and survival in a social bird

15:16: Talk 30 | Miguel Cruz | A genetic trade-off between intrinsic growth and sensitivity to competition, but not with reproductive interference, in spider mites

15:28: Talk 31 | Clara Pidner | Is the devil in the details? Investigating the role of the body spot in male-female interactions of the fish Poecilia vivipara's

15:40: Talk 32 | Rita Melo-Miranda | The role of exercise in mitigating inflammaging and gut dysbiosis

15:52: Talk 33 | Ana Sousa | Aging drives gut bacteria toward pathoadaptation

16:04: Talk 34 | Lekshimi B Sreelatha | Environmental drivers of colouration in Lusitanian wall lizards: Testing key ecogeographical hypotheses

16:16: Coffee Break

16:45: Poster Session

17:45: Closing Ceremony

18:00: General Assembly of APBE

Posters

Poster 1 | Sofia J Alves | The adaptive potential of sharks in a changing ocean: insights from comparative and population genomics

Poster 2 | Sofia J Alves | From darkness to data: genomic insights into the evolution and conservation of the Azorean Cave Ground Beetle *Trechus terceiranus*

Poster 3 | Inês Amorim | From molecules to conservation: PXR as a tool for ecological impact assessment

Poster 4 | Marta Antunes | Thermal Evolution and Plasticity: Insights from Gene Expression in *Drosophila subobscura*

Poster 5 | Rui Faria | Different genetic patterns of introgression across multiple hybrid zones of *Littorina saxatilis* in the Iberia Peninsula

Poster 6 | Ricardo Borges | Evolutionary dynamics of immune gene families in Lagomorphs: A comparative genomics approach

Poster 7 | Sofia Coimbra | Contribution to the Dairy Cattle Resistome

Poster 8 | Maíra Costa | Distribution of Alleles Linked to Pyrethroid and Amitraz Resistance in Varroa *destructor* across Portugal

Poster 9 | Sofia Costa | Evolving tensions: The role of stressful environments in shaping sexual conflict in a haplodiploid species

Poster 10 | Diogo Coutinho-Lima | Historical human practices have shaped the genomic diversity and structure of Iberian livestock dogs

Poster 11 | João Ramos Pacheco | A comparative test of social signalling with barred and spotted plumage in estrildid finches

Poster 12 | Varvara Fazalova | Variable mutation rates in lupins

Poster 13 | Guilherme Ferreira | Genetic architecture of pathogen avoidance

Poster 14 | Orlando Gallo | Population genetics of Argentine pumas (*Puma concolor*) suggest a broader scale for management and conservation strategies

Poster 15 | Catarina Garcês | The evolutionary history of the yellow coat colour in the Iberian wolf

Poster 16 | Raquel Gaspar | Adapt, adjust or move: can the spider mites *Tetranychus cinnabarinus* do it all in response to heat?

Poster 17 | Lurdes Gomes | Temperature-adaptative phenotypic plasticity – stressful temperatures induce different transcriptomic profiles in the genomically homogeneous worm pipefish

Poster 18 | Pilar Jurado-Angulo | Intra-archipelago dispersal patterns in oceanic islands based on phylogeography of darkling beetles (Oxycara, Tenebrionidae) in Cabo Verde

Poster 19 | Mariya Kozak | Inducible tomato defences persist in detached leaves, but are differentially affected by plant integrity and genotype

Poster 20 | Fernanda Li | An unprecedented large-scale survey of honey bee mitochondrial diversity in Europe: c-lineage dominance and the need for conservation efforts

Poster 21 | Ana Lima | A Multi-Omics and Primer Database for Virus Identification: Focus on HIV, Ebola, and SARS-CoV-2

Poster 22 | Diana Lourenço | Exploring Microalgae-Enzymes for Sustainable Plastic Biodegradation

Poster 23 | Adam Marques | Urbanization shapes dietary composition and head morphology of salamanders

Poster 24 | Rui Faria | Understanding the evolutionary history of inversions and their role in speciation and adaptation across the Littorina marine snails genus

Poster 25 | Victor Martinez-Mariño | Determination of barriers to gene flow between Galician *Littorina saxatilis* ecotypes

Poster 26 | Tiago Nogueira | Comparing demographic trajectories of Iberian carnivores and scavenges using a population genomics approach

Poster 27 | Maria João Paúl | Predictors of intraspecific diversity in herptiles using machine learning

Poster 28 | João Pimenta | The genetic landscape and the adaptation of the bumblebee *Bombus terrestris* to distinct Portuguese biomes

Poster 29 | Lorena Pinilla | Advancing methodological approaches applied to the conservation and management of wolves

Poster 30 | Laura Regalado | Genomics insights of fire blight: deciphering *Erwinia amylovora* for improved on-field disease control

Poster 31 | Mariana Ribeiro | Wild vs. Captive: a comparative study of genetic and genomic approaches across continents

Poster 32 | Ana Rocha | Investigating Mammalian Tissue Expression Patterns Using Public Transcriptomic Data

Poster 33 | Guilherme Roxo | The Azorean Daucus: a morphological, cytogenomic, phylogenetic and ecological study

Poster 34 | João Souto | Incomplete Lineage Sorting and Its Role in the Recurrent Evolution of Parthenogenesis in Stick Insects

Poster 35 | Gonçalo Themudo | DNA from cetariae fish remains confirms sardine (*Sardina pilchardus*) use and local population continuity in Northwestern Iberia since Roman times

Poster 36 | Raquel Tomé | Estimating trait heritability and variation in predator-prey dynamics

Poster 37 | Ana Carolina Varela-Pereira | Escaping from a stressor: evolution of larval development under nutritional constraints

Poster 38 | Rui Faria | Evolution on our heads: disentangling mechanisms of diversification in human lice

Poster 39 | André Vidal-Capón | Chromosomal rearrangements and the evolution of reproductive barriers in a marine snail

Poster 40 | Inês Vieira | Phylogenomic analysis of evolutionary radiations: dealing with reference bias

Poster 41 | Ramón Vigo | A marine snail color cline under study: the role of crypsis from a field tethering experiment

Poster 42 | Joana Vilas Boas | Uncovering Mechanisms of Behavioural Developmental Bias

Poster 43 | Tamara Wonner | Tipping the scale: investigating the feedback between evolution and stability in a predator-prey system

Poster 44 | Maryane Wielewski | From Ship to Shore: The Genetic Legacy of Invasive Rats in Atlantic Islands

Poster 45 | Ana Barbosa | The evolution and functional convergence of iodine assimilation genes in brown algae and animals

Poster 46 | Diana Simões | An integrated bioinformatics approach to study the Transgenerational Impact of Simvastatin's on the crustacean *Gammarus locusta*

Plenary Lecture I

Adaptive radiation on Oceanic Islands; Tales of ecological niches and ecological generalists)

José Cerca

Centre for Ecological and Evolutionary Synthesis, University of Oslo, Oslo, Norway

Adaptive radiations occur when a single lineage diversifies to exploit a broad range of ecological niches. However, the sequence of ecological occupation remains poorly understood. Two potential scenarios are herein proposed: random occupation, where species fill niches without a discernible pattern, and progressive occupation, where species track specific environmental gradients in a systematic manner. In this study, I examined four well-known adaptive radiations: Galápagos finches (Galápagos), Descurainia (Macaronesia), Aeonium (Macaronesia), and Anoles (West Indies). By analyzing phylogenetic and ecological distances between species, I found evidence supporting the hypothesis of progressive ecological occupation. This suggests that species diversify along a structured niche axis, implying that adaptive radiations may follow a more predictable, directional pattern. These findings are framed within the context of the 'least action principle' described by Gavrilets and Vose (2005). I will also focus on the continuum of generalist vs specialists in the context of insular diversification, offering examples from the Cabo Verde iago sparrow.

Plenary Lecture II

8,000 years of domestic plants and animals evolution: understanding local adaptation under socio-economic and climatic fluctuations

Allowen Evin

Institute of Evolutionary Science of Montpellier, University of Montpellier, Montpellier, France

The development of agricultural societies is closely entangled with that of domestic animals and plants. Local and traditional domestic breeds and varieties are the result of millennia of selection of landraces by farmers.

The north-western Mediterranean basin is home to numerous archaeological sites and provides a unique repository of archaeozoological and archaeobotanical remains. The DEMETER project (ERC-St Grant) is using plant and animal remains from 586 sites to trace the long-term biological and cultural evolution of domestic species since their introduction to the region 8,000 years ago. The project focuses primarily on morphometric variation to explore how and why the diversity of domestic populations evolved into the multitude of breeds and varieties we see today. We have primarily analysed geometric morphometric data from animal teeth (sheep, pigs and goats) and cereal grains (barley), as well as linear biometric data from both wild and domestic species.

The trajectories of morphometric change revealed similarities and divergences between species that were interpreted using archaeological, available and newly established palaeoclimatic and palaeoenvironmental reconstructions specific to the region. Overall, our analyses revealed a complex interplay between anthropogenic and environmental factors over the last 8 millennia, highlighting a stronger influence of environmental conditions on domestic populations than previously thought.

Plenary Lecture III

Origins of cells and organs – the view from the placenta

Margarida Cardoso Moreira

Evolutionary Developmental Biology Lab, The Francis Crick Institute, London, United Kingdom

My group's research centres on understanding the molecular and developmental bases of organ evolution. We aim to understand how new cells, new tissues, and entire new organs arise during evolution. To do so, we study an organ that has originated multiple times independently and exhibits an extraordinary phenotypic diversity: the placenta. In mammals, we have found that the placenta evolves faster than other mammalian organs. This fast evolution is driven by new expression patterns, the addition of newly evolved genes, protein adaptation, and – remarkably – the evolution of new cell types. The level of cell-type innovation that we found makes the placenta an outlier among mammalian organs. We are using the newly evolved placental cell types as a model to understand, more generally, how new cell types originate. In addition to our work with mammals, we also study a family of fish (Poeciliidae) where complex placentas have evolved independently multiple times. Whereas the mammalian placenta is an exceptional model for studying the evolution of new cell types, the placentas of Poeciliids offer a unique system to study how new complex organs arise during evolution. In Poeciliids, we combine evolutionary genomics, single-cell transcriptomics, spatial transcriptomics, and imaging approaches to determine the identity and developmental origins of the cells that comprise four independent origins of a placenta. This unique system allows us to establish the commonalities and differences in cell-type composition between the independently originated placentas and identify their molecular and developmental basis.

Plenary Lecture IV

The Sandwalk: Time for Thinking about Evolution Education

Rebecca Mead

Milner Centre for Evolution, University of Bath, United Kingdom

What is the best way to teach evolution? Despite its importance, evolution is widely recognised as one of the most misunderstood topics in biology. Its eminent position within science is not fully recognised in schools and there are concerns over its unsatisfactory teaching. Many students have difficulty in accepting evidence for evolution and debate is currently in progress about how best to teach evolution in schools.

Over the last 12 years, researchers at the Milner Centre for Evolution have aimed to understand how best to teach evolution to school-aged children. The “GEVOteach” project was the first of our school-based research studies. This investigated how and why acceptance of evolution may alter for secondary school students during their education. In particular, we were keen to understand how knowledge of genetics may contribute to understanding and acceptance of evolution. In the UK, genetics and evolution are typically taught to 14- to 16-year-old secondary school students as separate topics with few links. Here, we report the results of this large trial into teaching order of evolution and genetics. We modified extant questionnaires to ascertain students’ understanding of evolution and genetics along with acceptance of evolution. Students were assessed prior to teaching, immediately post teaching and again after several months. We report that teaching genetics first has a marked and significant impact on both evolution and genetics knowledge. Qualitative focus group data highlights the role of authority figures in determination of acceptance.

Dissemination of research findings and outreach is an important aspect of our work. To build teacher confidence and understanding of evolution, we have developed “Exploring Evolution” teacher training workshops. This collaboration with the Darwin Correspondence Project and Galapagos Conservation Trust has been successfully running for eight years, during which time we have worked with over 500 trainee and practising teachers across England.

Talk 1

Does song act as a behavioural barrier to gene flow? Evidence from divergent populations of La Palma's Canary Islands Chaffinch

Freitas, B (1,2,3); Gil, D (1); Thébaud, C (2); Milá, B (1)

(1) MNCN-CSIC – Museo Nacional de Ciencias Naturales, Consejo Superior de Investigaciones Científicas, Madrid, Spain; (2) CRBE - Centre de Recherche sur la Biodiversité et l'Environnement (UMR 5300 CNRS-IRD-TINPT-UPS), Université Paul Sabatier, Toulouse, France; (3) Facultad de Ciencias, Universidad Autónoma de Madrid, Madrid, Spain

Bioacoustics; pre-mating barrier; reproductive isolation; speciation; local adaptation

Understanding the speciation process is one of the most pressing questions in evolutionary biology. In songbirds, vocalisations are important in individual and species recognition and are crucial in sexual and social communication. Populations that diverge in song may fail to recognise each other, leading to assortative mating and, potentially, speciation. Therefore, song may constitute a premating barrier to reproduction. The chaffinches from La Palma (Canary Islands, Spain) constitute a clear case of incipient lineage formation, with populations that inhabit pine forests being genetically and morphologically differentiated from cloud forest populations. However, song characteristics of these populations have not been studied, so the role of song in their divergence into different lineages that have remained distinct in spite of being in close proximity from one another remains unclear. We aimed to characterise and quantitatively compare acoustic traits between populations from the two habitats, focusing on song composition, syllable diversity, and shared syllables, and we also tested experimentally whether their song differences could act as a behavioural isolating mechanism. By recording songs of both populations across their distribution ranges and using a novel approach to directly capture the frequency modulation of syllables, we found differences in the songs of individuals that were associated with forest type, with results revealing three well-differentiated groups: pine, cloud, and ecotone between these two. Our playback experiments used songs from the same and different habitats, as well as songs from a close relative, the Common Chaffinch from mainland Europe, and the reaction of the territorial individuals was registered. Both probability and intensity of response to songs of the close relative species and to the different-habitat birds were weaker than to songs of the same-habitat birds, but differences observed in the second case were not statistically significant. Although song differences seem not to be strong enough to elicit marked reactions between territorial males from opposite habitat types, further experiments will be necessary to test female response to these songs in order to determine if they convey lineage-specific information or not. This study adds then to the debate over the role of song differentiation in driving the evolution of reproductive isolation, and whether song acts as a barrier only when other phenotypic differences are already well established.

Talk 2

Evolutionary history of the *Mus musculus* in Cabo Verde

Almeida, L (1,2), Pinho, CJ (1,2), Lopes, EP (1,3), Boratynski, Z (1), Gabriel, SI (4), Vasconcelos, R (1)

(1) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, BIOPOLIS Program in Genomics, Biodiversity and Land Planning, Campus de Vairão, Universidade do Porto, Portugal; (2) FCUP - Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Portugal; (3) ISECMAR - Instituto de Engenharia e Ciências do Mar, UTA, Universidade Técnica do Atlântico, Ribeira de Julião, São Vicente, Cabo Verde; (4) CESAM - Centro de Estudos do Ambiente e do Mar, Departamento de Biologia Animal, Faculdade de Ciências da Universidade de Lisboa, Portugal.

Evolution; Invasive species; Island Biogeography; Population Ecology; Rodents

Biological invasions on islands provide unique opportunities to study evolutionary processes. Additionally, understanding the colonisation and radiation of introduced species is a priority for biodiversity conservation. Given that the Cabo Verde Archipelago is located in a biodiversity hotspot, it is important to study the impact of the introduced Mus musculus in more detail and to understand its colonisation history, diversification patterns and adaptation to these new environments. In Cabo Verde, the origin of this alien species and its adaptation to the islands are still unknown, even with preliminary genetic studies. Therefore, this work aims to provide information on the colonisation and diversification patterns of house mice in Cabo Verde as a model for other more complex systems. Phylogenetic and population analyses were performed by amplifying mitochondrial DNA, D-loop, from stomach tissues from seven islands and one islet. Additional morphological analyses were carried out using photographs of more than 100 individuals, to test if coats are different across islands. The results showed the recent colonisation of the archipelago by Mus musculus of different origins. Also different morphological groups were identified, probably due to the isolation of the islands and the rapid adaptation capacity of this species.

Talk 3

Patterns of diversification and colonization in Macaronesian Apiaceae lineages

Romeiras, M.M. (1,2); Roxo, G. (3); Silva, L. (3); Menezes-Sequeira, M. (4); Brilhante, M. (1); Costa, J.C. (1); Moura, M. (3); Talhinhas P. (1); Sousa, VC (1,2); Fragata, I (1); Parreira, B (2,3); Rodrigues, L (1)

(1) Linking Landscape, Environment, Agriculture and Food (LEAF), Instituto Superior de Agronomia (ISA), Universidade de Lisboa, Tapada da Ajuda, 1340-017 Lisbon, Portugal; (2) CE3C - centre for ecology, evolution and environmental changes, FCUL University of Lisbon, Lisbon, Portugal; (3) CIBIO-Açores, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO, Faculdade de Ciêcnias e Tecnologias, Universidade dos Açores, Campus Ponta Delgada, Rua Mãe de Deus 58, Apartado 1422, 9501-801 Ponta Delgada, Portugal; (4) Madeira Botanical Group, Faculty of Life Sciences, University of Madeira, 9020-105 Funchal, Portugal

The Macaronesian region comprises five archipelagos (i.e., Azores, Madeira, Selvagens, Canary Islands, and Cabo Verde) and harbours a rich endemic flora of approximately 900 vascular plant species. For this reason, it is considered a biodiversity hotspot and an ideal model to study evolutionary processes involved in flora diversification. Apiaceae is a family of aromatic plants, and 70 taxa occur in Macaronesia (including naturalized non-native taxa), of which 35 are archipelago-endemics. The Daucus complex (Daucus – Melanoselinum – Monizia – Tornabenea) represents a cladogenesis speciation model, whereas *Crithmum maritimum (*a monotypic genus) represents an anagenesis speciation model. This study aimed to investigate the cytogenomic patterns of two Apiaceae lineages with different speciation models, analyzing this along with ecological and morphological, and comparing it with mainland Portugal populations. The study of different species belonging to the Daucinae subtribe revealed that the taxon is a good predictor of genome size, however, only at a species level, as it is unable to clearly discriminate the *Daucus carota* subspecies. *Crithmum maritimum* presented a high cytogenomic variation despite belonging to a monotypic genus. Thus, to determine which environmental factors better explain this cytogenomic variation, we calculated different generalized linear models, revealing a tendency for an increase in genome size along the Portuguese coast, from south to north, in association with lower temperatures, higher precipitation, and lower precipitation seasonality. This gradient might result from historical phylogeographical events associated with previous dispersal and extinction of local populations. Results seem to support the tendency for smaller genomes to occur in islands.

Talk 4

Hybridization between currently allopatric species at the root of speciation? The case of Iberian chubs (Genus Squalius)

Faustino, E (1); Mendes, SL (1); Carvalho, J (1,2,3); Perea, S (4); Doadrio, I (4); Sousa-Santos, C (5); Sousa, VC (1)

(1) cE3c – Centre for Ecology, Evolution and Environmental Changes & CHANGE-Global Change and Sustainability Institute, FCUL, Lisboa, Portugal; (2) CIBIO – Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO, Laboratório Associado, Universidade do Porto, Vairão, Portugal; (3) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal; (4) Museo Nacional de Ciencias Naturales, Madrid, Spain; (5) MARE –Marine and Environmental Sciences Centre/ARNET - Aquatic Research Network, ISPA – Instituto Universitário de Ciências Psicológicas, Sociais e da Vida, Lisboa, Portugal.

Population genetics; hybridization; freshwater fish

Hybridization, i.e., the mating between distinct species or genetically distinct lineages, has been shown to be widespread throughout the evolutionary tree. Our understanding of hybridization and its prevalence has been made possible due to the generation of population genomics data for increasingly higher numbers of species, coupled with the development of different methods, such as demographic modelling. Freshwater fish stand out amongst animal groups with some of the highest rates of hybridization. Yet, if and how hybridization can lead to speciation remains an open debate. Here, we investigate a potential case of ancient hybridization in Iberian chubs, previously described using genotyping by sequencing (GBS) data. By using whole genome population genomic data, we aim to uncover the origin of the Iberian chub *Squalius pyrenaicus*. This species has been hypothesized to result from hybridization between its sibling species *Squalius carolitertii* and *Squalius tartessicus*, with a major (~80%) contribution from *S. carolitertii*. We generated high-coverage (>17x) whole-genome re-sequencing data for 37 individuals comprising *S. pyrenaicus*, *S. carolitertii*, *S. tartessicus* and two outgroup species, *S. torgalensis* and *S. aradensis*. Based on estimates of individual ancestry we found no evidence of an admixed ancestry in S. pyrenaicus individuals, consistent with an ancient hybridization. We used D-statistics to distinguish incomplete lineage sorting from gene flow and found that the relationship between *S. pyrenaicus*, *S. carolitertii* and *S. tartessicus* cannot be explained by simple a bifurcating tree, with evidence of gene flow, in agreement with previous results using GBS data. We finally assessed region-specific genomic ancestry patterns with chromosome painting, as a proxy for selection detection. Our results provide further support for the hypothesis of hybridization being involved in the evolutionary history of *S. pyrenaicus*. Furthermore, our results highlight how gene flow can be differently recognized by different metrics, especially in cases involving ancient hybridization. This is particularly relevant in cases where there is a significant imbalance between the contributions of each parental species.

Talk 5

Comparative Population Genomics Illuminates Species Boundaries and Symbiotic Disruption in *Eunicella* Octocorals

Bertand T (1); Sarropoulou X (2); Manousaki T (2); Tsigenopoulos C (2); Babe O (1); Meyer HS (1); Coelho M (3); Stevens J (4); Xavier JR (1); Sartoretto S (5); Antunes A (1); Gut M (6); Gomes-Garrido J (6); Cruz F (6); Alioto T (6); Garrabou J (7); Viard F (8); Aurelle D (9); Ledoux J-B (1);

1- CIIMAR CIIMAR/CIMAR, Centro Interdisciplinar de Investigação Marinha e Ambiental, Universidade do Porto, Porto, Portugal. 2-HCMR Hellenic Centre for Marine Research, IMBBC Institute of Marine Biology, Biotechnology & Aquaculture, Heraklion, Greece 3-CCMAR Centre for Marine Sciences (CCMAR), University of Algarve, Faro, Portugal 4-Department of Biosciences, Faculty of Health and Life Sciences, University of Exeter, Exeter, United Kingdom 5-Ifremer, LITTORAL, La Seyne-sur-Mer, France 6-CNAG Centro Nacional de Análisis Genómico, Baldiri Reixac 4, 08028 Barcelona, Spain. / Universitat de Barcelona (UB), Barcelona, Spain. 7-Departament de Biologia Marina, Institut de Ciències del Mar (CSIC), Barcelona, Spain. 8-ISEM, Institut des Sciences de l'Evolution, Univ Montpellier, CNRS, IRD, Montpellier, France 9-Aix Marseille Univ, Université de Toulon, CNRS, IRD, MIO, Marseille, France / ISYEB Institut Systématique Evolution Biodiversité, Muséum national d'Histoire naturelle, CNRS, Sorbonne Université, EPHE, Université des Antilles, Paris, France

The process of speciation is a central topic in evolutionary biology due to its role in shaping biodiversity patterns. Yet, the study of speciation is challenging particularly in recently diverged species, in which reproductive isolation is not complete and gene flow and hybridization can still occur. Here, we investigate species boundaries in three accepted octocoral species: *Eunicella verrucosa* (Pallas, 1766), *E. cavolini* (Koch, 1887), and *E. singularis* (Esper, 1791). *E. verrucosa* is widely distributed in the Eastern Atlantic and Mediterranean sea *while E. cavolini* and *E. singularis* are only found in the Mediterranean. In spite of morphological differences, the three species are considered as habitat-forming species and support biodiversity rich communities. E. singularis hosts a photosynthetic Philozoon species (previously classified as Symbiodiniaceae temperate clade A) contrary to the two other species. Noteworthy, E. cavolini and E. singularis are under strong conservation concerns owing to recurrent mass mortality events linked to anthropogenic climate change. Using whole genome re-sequencing data from 20 individuals from 13 localities, we i) characterized the genetic relationships between individuals of the three species; ii) looked for the genomic landscapes of differentiation; iii) inferred demographic history using the PSMC model; and iv) provided first insights into the associated microbiome using a metagenomics framework. Analyses of genome-wide diversity and divergence reveal clear genetic differentiation among the three recognized species; *E. verrucosa* being the most divergent one. However, a triangular hybrid plot based on ancestry-informative markers revealed two potential hybrids and one backcrossed individual between *E. cavolini* and *E. singularis*. Genome scan analyses did not reveal distinct genomic islands of divergence between *E. cavolini* and *E. singularis* in line with a scenario of recent divergence with ongoing gene flow. Comparing the genomic landscape of the potential hybrids, we identified distinct genomic regions with ancestry from either *E. singularis* or *E. cavolini* supporting different hybridization events. We revealed a pronounced and similar demographic decline in all the individuals, including the potential hybrids, from the Middle Pleistocene through to the Holocene. This result suggests a shared demographic history among the three species in spite of their contrasting distribution ranges.The metagenomic analysis focused on the occurrence of Philozoon (previously classified as Symbiodiniaceae temperate clade A) supported the absence of the symbiont in *E. cavolini* and *E. verrucosa* while it was found in all *E. singularis* individuals. Noteworthy, the symbiont was not detected in the potential *E. cavolini / E. singularis* hybrids suggesting a disruption in the evolutionary stability of the symbiotic association following the hybridization. This study provides an example of how gene flow and hybridization influence species boundaries and biodiversity patterns in octocorals. The observed hybridization raises important questions about the stability of symbiotic relationships in *E. singularis* and its effects on hybrid fitness and adaptability. Further analysis of the impact of this process on the microbiome, alongside detailed characterization of hybrid zones and scenario-based demographic inferences will deepen our understanding on the evolution of octocorals. This knowledge is required to contribute to ongoing conservation and restoration efforts.

Talk 6

Ecological differentiation influences the evolution of pre-zygotic isolation in spider-mite species

Barreto, S (1); Grandela, A (1); Henriques, S (1); Cruz, MA (1); Sousa, VC (1); Magalhães, S (1,2); Zélé, F (3); Fragata, I (1); Blanckaert, A (1);

(1) cE3c - Centre for Ecology, Evolution and Environmental Changes & CHANGE - Global Change and Sustainability Institute, FCUL University of Lisbon, Lisbon, Portugal; (2) Wissenschaftkolleg zu Berlin, Berlin, Germany; (3) ISEM - Institut des Sciences de l'Évolution de Montpellier, University of Montpellier, CNRS, IRD, EPHE, Montpellier, France;

Ecological speciation; Hybridization; Experimental evolution; Mate choice; Sperm precedence;

Understanding how speciation occurs requires the study of reproductive isolation and its evolution. In particular, it is important to characterize the role of the different reproductive barriers (e.g., local adaptation, mate choice, hybrid load), and how they arise and are strengthened. Although these barriers have been previously studied, the evolution of their interactions remains somewhat elusive. Using experimental evolution, we investigated how pre-zygotic barriers may evolve in the presence of ecological differentiation and with pre-existing post-zygotic isolation. We used the two sister species of spider mites, *Tetranychus urticae* (Tu) and *T. cinnabarinus* (Tc). This system displays incomplete reproductive isolation, with F1 hybrids being generally infertile. In addition, males of both species mate preferentially with Tc females and both species exhibit strong first male sperm precedence. We created three isolated regimes adapted to their host plants: Tu on bean, Tu on tomato, and Tc on bean; and two connected regimes formed by pairs of populations exchanging individuals once every generation. The two connected regimes differed only in plant hosts: Tu on bean connected to Tc on bean, and Tc on bean connected to Tu on tomato. After 10 generations, we quantified changes in assortative mating by measuring male preference (first choice, copulation duration) and changes in the ability of females to remate successfully. Tu males showed stronger preference for mating with conspecific females when coming from the connected regimes with different plant hosts, than males coming from the isolated populations. Additionally, Tu females from this same regime produced more non-hybrid daughters when mating first with heterospecific males and then with a conspecific. This suggests a shift in the known first male sperm precedence, allowing for a rescue of the production of fertile daughters. Altogether, our results show both a shift towards conspecific assortative mating, and a weakening of the impact of heterospecific matings by changes in male sperm precedence. Overall, contact between populations in the presence of ecological differentiation enabled the evolution of stronger pre-zygotic barriers, a step towards the completion of the speciation process.

Talk 7

Can reproductive isolation arise from epistatic deleterious and compensatory mutations in small populations: an experimental test with *C. elegans*

Chelo, IM (1); Morais (1), D; Nevado, B (1)

(1) CE3C - centre for ecology, evolution and environmental changes, FCUL University of Lisbon, Lisbon, Portugal

Reproductive isolation; speciation; experimental evolution; *C. elegans*

The tendency of natural populations to undergo periodic fluctuations in size raises an important question: can deleterious mutations that fix during bottlenecks interact with epistatic compensatory mutations, which appear during population recovery, and thus contribute to reproductive isolation and species diversification?

In this study, we use *Caenorhabditis elegans* as a model organism to observe and validate these evolutionary processes in real time. Mutation accumulation (MA) lines were established from five distinct genotypes over twenty generations, with mutation rates intentionally amplified up to 100-fold. Populations were maintained with either two or thirty individuals to compare the evolution of populations with different deleterious mutational loads. Following this mutation accumulation period, experimental lines were evolved over 60 generations under conditions of high population size (~3,000 individuals), which provide an opportunity for the fixation of new beneficial mutations.

We measured population growth and fertility of experimental lines at three stages: the ancestral populations, MA derivatives, and recovery populations. Observed fitness dynamics indicate a general decrease in fitness during the MA period, and that some – but not all – populations underwent compensatory adaptation during the final period with large population size.

We discuss these results in relation to potential differences in ancestral fitness, mutational input, and the adaptive roles of compensatory mutations versus unconditionally beneficial mutations. Additionally, we explore recent findings on reproductive isolation in some of these populations and consider their implications for speciation under natural conditions with similar demographic histories.

Talk 8

Understanding hybridization between alpine hares in a context of climate-induced range shifts

Sousa, P (1,2,3); Costa, J (1,2,3,4); Farelo, L (1,3); Letty, J (5); Kaerle, C (6); Queney, G (6); Alves, PC (1,2,3,7); Pimenta, J (1,3); Melo-Ferreira, J (1,2,3); Marques, JP (1,3)

(1) CIBIO, Research Centre in Biodiversity and Genetic Resources, Universidade do Porto, Vairão, Portugal; (2) DBIO-FCUP - Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto, Portugal; (3) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal; (4) ISEM - Institut des Sciences de l'Évolution Montpellier, Université Montpellier, Montpellier, France; (5) ONCFS - Research Department, National Hunting and Wildlife Agency, Juvignac, France; (6) ANTAGENE - Animal Genomics Laboratory, Lyon, France; (7) EBM - Estação Biológica de Mértola, CIBIO, Mértola, Portugal.

Genomics; biodiversity conservation; climate change; range-shifts; introgression

Climate change poses significant challenges to biodiversity, forcing species and natural populations to move, adapt or face extinction. Historically, natural climate oscillations have influenced species' ranges, leaving distinct imprints that shaped their genomes and improving their ability to adapt to changing conditions. However, anthropogenic-driven environmental changes have intensified these effects, leading to habitat loss and fragmentation, range contractions of vulnerable species and expansion of favoured species. These contrasting demographic trends often lead to species replacements in shared habitats, increasing opportunities for hybridization and gene flow between species that are not fully reproductively isolated. Understanding how interspecific gene flow shapes the genetic variation of affected species is therefore crucial for conservation, as it may impact genetic integrity by introducing harmful or maladaptive traits. However, interspecific gene flow can also introduce potentially adaptive genetic variants, enhancing species resilience by increasing their adaptive potential. Dissecting these genetic dynamics is essential to evaluate and forecast the influence of climate change on species, particularly those with contracting or expanding ranges. In this study, we investigated the genetic interactions between the Mountain hare (*Lepus timidus*) and the European hare (*Lepus europaeus*), two species with opposing demographic trends influenced by climate change. The Mountain hare, adapted to alpine-arctic climates, is currently facing range contraction, while, the European hare, a temperate species, is expanding its distribution northwards and upwards. Both species are generally parapatric, but when their distributions overlap, hybridization occurs. This project focused on the contact zone between these two species across the French Alps, where their distributions partly overlap and where climate change, hybridization and gene introgression are thought to intervene, with synergic adverse effects on the Mountain hare. Using ddRADseq (double digest Restriction-site Associated DNA sequencing) data, we explored the patterns of species hybridization, and our analysis indicated gene flow between European and Mountain hare populations in the Alps, suggesting hybridization across the contact zone. We also identified genetic structure among Eurasian Mountain hare populations, while European hare populations appear genetically more homogeneous. Overall, this research emphasizes the importance of genomics in biodiversity conservation strategies to better predict and mitigate the impacts of climate change on species with overlapping ranges.

Talk 9

Population structure and environmental adaptation in two honey bee subspecies from southern European refugia: *A. M. ligustica* and *A. M. iberiensis*

Yadró, C. A. (1,2,3,4,5); Henriques D. (1); Cilia G. (6); Rufino J. (7); Nanetti A. (6); Martín-Hernández R. (8); Pinto M. A. (1)

(1) CIMO– Centro de Investigação de Montanha , LA SusTEC, Instituto Politécnico de Bragança, Campus de Santa Apolónia, 5300-253 Bragança, Portugal; (2) Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Porto, Portugal; (3) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, Centro de Investigação em Biodiversidade e Recursos Genéticos; (4) InBIO - Research Network in Biodiversity and Evolutionary Biology Laboratório Associado, Vila do Conde, Portugal; (5) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, Vila do Conde, Portugal; (6) CREA-AA - Centro di Ricerca Agricoltura e Ambiente, Consiglio per la Ricerca in Agricoltura e l'Analisi dell'Economia Agraria, Bologna, Italy; (7) CeDRI - Research Centre in Digitalization and Intelligent Robotics, LA SusTEC, Instituto Politécnico de Bragança,Campus de Santa Apolónia, Portugal; (8) CIAPA - Centro de Investigación Apícola y Agroambiental, IRIAF - Instituto Regional de Investigación y Desarrollo Agroalimentario y Forestal, 19180 Marchamalo, Spain

Population genetics; *Apis mellifera*; WGS; genotype-environment association; local adaptation

The Iberian and Italian peninsulas served as refugia to honey bees (*Apis mellifera*) during glacial periods and are home to two subspecies: *A. m. iberiensis* (M-lineage) and *A. m. ligustica* (C-lineage), respectively. Here, we analyzed 311 (86 A. m. iberiensis; 225 A. m. ligustica) whole genomes generated from drones (haploid males) to investigate population structure, genetic diversity, and the molecular basis of local adaptation in both subspecies. For A. m. iberiensis, Admixture analysis revealed a Northeastern-Southwestern cline between two genetic backgrounds and no introgression from other subspecies. For A. m. ligustica, geographically extended introgression from its neighbouring sister subspecies, A. m. carnica, was detected even in areas distant from their natural contact zone. Compared to A. m. iberiensis, A. m. ligustica showed lower levels of genetic diversity, as estimated by nucleotide diversity, expected heterozygosity, LD-based effective population size (Ne) and higher inbreeding. The analysis of recent historical Ne for A. m. iberiensis revealed a strong bottleneck between the 1940s and 1980s, after which a slight recovery occurred. In the case of A. m. ligustica, it always showed lower Ne than A. m. iberiensis and remained stable until the 1990s, when it experienced a massive drop that lasted until today. Selection signatures were detected and cross-validated using PCAdapt, SAMBADA, and RDA. SNPs with q-adjusted p-values < 0.05 detected by at least two selection methods were considered strong candidates. For A. m. iberiensis, we detected 279 SNPs, and these were mainly associated with Longitude, Precipitation Seasonality, Mean Temperature and Precipitation of the Driest Quarter. Among other functional annotations, a subset of candidate SNPs were annotated to genes involved in cold and heat response and cuticle assembly. For A. m. ligustica, we detected only 37 SNPs, and these were mainly associated with Latitude and Precipitation of the Warmest Quarter. A subset of these SNPs were also annotated to genes involved in heat and cold response. While we did not find overlapping SNPs between the two subspecies, we found 20 common genes. In addition to the aforementioned annotations, we also detected SNPs in genes associated with olfaction, ultraviolet and spectral perception and circadian clocks. Our findings will be discussed in light of modern apicultural practices. This project is part of the PRIMA programme supported by the European Union. This work was supported by national funds through FCT/MCTES (PIDDAC): CIMO, UIDB/00690/2020 (DOI: 10.54499/UIDB/00690/2020) and UIDP/00690/2020 (DOI: 10.54499/UIDP/00690/2020); and SusTEC, LA/P/0007/2020 (DOI: 10.54499/LA/P/0007/2020). National funding by FCT, Foundation for Science and Technology, through the individual research grant 2021.06948.BD of Carlos A. Yadró.

Talk 10

Genetic divergence and local adaptations in endemic hares: Insights from the Italian and Broom hares

Costa J (1,2,3,4); Marques, J.P (1,3); Farelo L; Boursot, P (4); José Melo-Ferreira (1,2,3)

(1) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Universidade do Porto, Campus de Vairão, 4485-661 Vairão, Portugal; (2) DBIO-FCUP - Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, 4099-002 Porto, Portugal; (3) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, 4485-661 Vairão, Portugal; (4) ISEM - Institut des Sciences de l'Évolution Montpellier (ISEM), Université de Montpellier, CNRS, IRD, Montpellier, France

Genomics, biodiversity, introgression, range-shifts

The Italian hare (*Lepus corsicanus*) and the broom hare (*Lepus castroviejoi*) are two genetically similar yet distinct species endemic to the Italian and Iberian Peninsulas, respectively. Unlike other European hares, both species exhibit unusually low genetic diversity, making them particularly vulnerable to environmental changes. Despite this, we uncovered strong population structure in the Italian hare, with distinct northern and southern genetic clusters shaped by unique selective pressures that suggest possible localized adaptations. The divergence of these sister species dates back roughly 50,000 years, a time frame that does not align with the cooling events of the Pleistocene, hinting at other influences, such as inter-species competition, driving their split. Notably, both species retain introgressed genetic regions from the mountain hare, indicating a history of allele sharing that may have equipped them with adaptive traits for survival in their distinct ranges. To deepen our understanding of the evolutionary and ecological factors shaping these species, we are integrating ecological niche and demographic modelling. This dual approach seeks to reconstruct historical distributions and reveal how past environmental shifts and species interactions have influenced their current genetic and geographic profiles. Our findings contribute to the broader understanding of genetic resilience and adaptation in endemic species, offering insights that could inform future conservation strategies.

Talk 11

Population structure and conservation status of middle eastern honey bee subspecies

Henriques D(1); Yadró CA (1,2,3,4,5); Honrado M(1,6); Amaral JS(1); Muz MN(7); Muz D(7); Haddad N(8); Al-Shagour B(8); Obeidat W(9); Hosri C(10); Arab A(11); Arruda J(12); Lamghari F(12); Rufino J(13); Martín-Hernández R(14); Nanetti A (15); Pinto MA(1)

(1) CIMO, LA SusTEC, Instituto Politécnico de Bragança, Campus de Santa Apolónia, 5300-253 Bragança, Portugal.; (2) Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Porto, Portugal; (3) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, Centro de Investigação em Biodiversidade e Recursos Genéticos; (4) InBIO - Research Network in Biodiversity and Evolutionary Biology Laboratório Associado, Vila do Conde, Portugal; (5) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, Vila do Conde, Portugal;(6) LAQV-REQUIMTE – Laboratório Associado para a Química Verde-Rede de Química e Tecnologia, Departamento de Química, Universidade de Aveiro, Aveiro, Portugal; (7) University of Namık Kemal, Faculty of Veterinary Medicine. Department of Virology, Tekirdağ Türkiye; (8) NARC- National Agricultural Research Center, Bee Research Directorate, Jordan; (9) The University of Jordan, Jordan; (10) UL Lebanese University, Faculty of Agriculture, Beirut, Lebanon; (11) Department of Animal Science, College of Agriculture and Natural Resources, University of Tehran, Iran; (12) Fujairah Research Centre, Fujairah, UAE; (13) CeDRI - Research Centre in Digitalization and Intelligent Robotics, LA SusTEC, Instituto Politécnico de Bragança, Campus de Santa Apolónia, Portugal; (14) CIAPA - Centro de Investigación Apícola y Agroambiental, IRIAF - Instituto Regional de Investigación y Desarrollo Agroalimentario y Forestal, 19180 Marchamalo, Spain; (15) CREA-AA - Centro di Ricerca Agricoltura e Ambiente, Consiglio per la Ricerca in Agricoltura e l'Analisi dell'Economia Agraria, Bologna, Italy

Population genetics; Apis mellifera; whole-genome sequencing; Middle Eastern

The honey bee *Apis mellifera* evolved into at least 31 subspecies in its native range in the Middle East, Africa, and Europe. The Middle Eastern subspecies have been understudied, hindering a comprehensive understanding of honey bee evolutionary history and the conservation status of native diversity patterns. Since ancient times, humans have assisted gene flow within each subspecies native range. However, in the last decades, globalization and intensification of queen trading have facilitated the contact between previously allopatric subspecies (especially involving the beekeeper-favoured Eastern European *A. m. ligustica* and *A. m. carnica* from Eastern Europe), leading to introgressive hybridization. This poses a problem as it may lead to the breakdown of gene complexes adapted to local environmental conditions, particularly to the dry and hot summers of the Middle East. In this study, we used whole-genome sequence data to investigate the genetic integrity and population structure of five Middle Eastern subspecies across a broad geographical range: Turkey (*A. m. anatoliaca*, N=97; *A. m. caucasia*, N=75; *A. m. meda*, N=18), Jordan and Lebanon (*A. m. syriaca*, N=238 and N=29), Iran (*A. m. meda*, N=75), Oman, and the UAE (*A. m. jemenitica*, N=13 and N=10). Our findings reveal concerning conservation status for many populations and subspecies. In Turkey, three distinct subspecies have traditionally been recognized: *A. m. anatoliaca, A. m. caucasia*, and *A. m. syriaca*. However, our analysis indicates that the populations previously assumed to be *A. m. syriaca* are in fact *A. m. meda*. Moreover, while three subspecies still exist in Turkey, populations are highly admixed. In Jordan and Lebanon, where *A. m. syriaca* is native, we observed variable levels of introgression from *A. m. ligustica*. Similarly, in Iran, where *A. m. meda* is found, extensive introgression was detected, primarily involving *A. m. ligustica* and *A. m. caucasia*. In the UAE, two main groups were identified: the first containing hybrids of *A. m. jemenitica*, *A. m. lamarckii*, and *A. m. ligustica*, while the second group predominantly consisted of hybrids of *A. m. lamarckii* and *A. m. ligustica*. Oman was the only location within the native range of *A. m. jemenitica* where all samples were classified as pure. This study highlights widespread hybridization across the Middle East and underscores the urgent need for targeted conservation efforts to preserve the genetic diversity of Middle Eastern A. mellifera subspecies.

Talk 12

Distribution of intraspecific diversity across species ranges: insights from herptiles in six global regions

Moreira, MO (1,2); Paúl, MJ (1,2,3); Liz, AV (1,2,3); Carvalho, SB (1,2)

(1) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Universidade do Porto, Vairão, Portugal; (2) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal; (3) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto, Portugal

Genetic diversity; central-peripheral hypothesis; geographic range; climatic niche

Understanding the distribution of intraspecific diversity (ISD) across species' geographic ranges is fundamental to evolutionary biology and conservation. However, the distribution of genetic diversity within species remains poorly understood at broad spatial scales. This study explores patterns of ISD in herptiles (amphibians and reptiles) across six regions, examining how genetic diversity and differentiation vary relative to species' geographic and climatic-niche centroids. Specifically, we test wether ISD declines from the center towards the periphery of species ranges or from the species climatic-niche optimum towards marginal conditions. We leverage an extensive dataset encompassing molecular data for over 600 herptile species and, for each candidate species, we i) mapped ISD across species ranges, ii) described the centrality and marginality gradients of the climatic niche and calculated both geographic and climatic niche distances, and iii) calculated regression coefficients (Pearson's r) between genetic diversity and distance metrics. By employing species-specific sequence alignments, species distribution modelling and comparative analyses, we identify whether populations closer to current optimal niche conditions exhibit greater genetic diversity, reflecting enhanced stability and reproductive success. Our results provide novel insights into the processes shaping ISD, highlighting the interplay between ecological, geographic, and evolutionary factors. This study underscores the importance of integrating climatic-niche models with genetic data to understand biodiversity patterns and their drivers. By identifying regions and populations critical for maintaining genetic diversity, our findings have significant implications for the conservation of herptile species in the face of global environmental change.

Talk 13

Phylo-transcriptomics of the giant kelp *Macrocystis pyrifera* reveals multiple trans-oceanic dispersal events across the Southern Ocean

Almeida, SA (1,2); Jacinto R (1); Serrão EA (1); Faugeron S (1,2); Pearson GA (1)

(1) CCMAR, CIMAR, Universidade do Algarve, Faro, Portugal; (2) Núcleo Milenio MASH, Facultad de Ciencias Biológicas, Pontificia Universidad Católica de Chile, Santiago, Chile

Phylogenomics, giant kelp, microevolution, historical biogeography

The giant kelp *Macrocystis pyrifera* is a foundational species of cold-temperate coasts worldwide. Its extensive distribution is thought to rely heavily on its dispersal capacity, which enables the (re) colonization of remote habitats and maintains genetic connectivity through gene flow. However, the interplay between the dispersal ability of this cosmopolitan species, past climate dynamics, and the ecological factors driving its evolutionary history remains poorly understood. Here, we aim to elucidate the evolutionary history and diversification of *M. pyrifera* through multilocus RNA-seq phylogenomics, using populations across its global distribution range. Our results support a recent origin for the species in the Northern Hemisphere, followed by trans-equatorial dispersal and sequential colonization across both hemispheres, likely shaped by significant climatic and oceanographic changes. Additionally, we found previously unrecognized patterns of genetic structure and shared lineages across the Southern Ocean, suggesting multiple independent colonization events. These findings provide new insights into the biogeography and colonization dynamics of this key habitat-forming species. As climate change accelerates, *M. pyrifera* populations face increasing threats, underscoring the importance of understanding how historical climate shifts have shaped species evolution to inform conservation strategies and predict future responses of this foundational species.

Talk 14

FCRL ancestry: can we truly find parallels between placental mammals, marsupials and monotremes?

Matos MC (1,2,3); Davis RS (4) and Esteves PJ (1,2,3)

(1) CIBIO-UP, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Campus Agrário de Vairão, Universidade do Porto, 4485-661 Vairão, Portugal; (2) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, 4485-661 Vairão, Portugal; (3) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, 4169-007 Porto, Portugal; (4) Departments of Medicine, Microbiology, and Biochemistry & Molecular Genetics, Comprehensive Cancer Center, University of Alabama at Birmingham, Birmingham, AL 35294, USA

Marsupials; Placental mammals; Monotremes; Evolution; Immunoreceptors

Fc receptor-like (FCRL) molecules constitute a diverse family of immunoregulatory receptors that share homology with Fc receptors (FCR) and play a role in regulating immune responses, contributing to the pathogenicity of various diseases. In mammals, this family includes nine identified genes: FCRL1-6, FCRLA, FCRLB, and FCRLS. To explore the evolutionary history of these receptors, we analyzed FCRL genes across marsupial species (*Monodelphis domestica*, *Gracilinanus agilis, Trichosurus vulpecula, Phascolarctos cinereus, Vombatus ursinus, Sarcophilus harrisii, Antechinus flavipes and Dromiciops gliroides*) and Monotremata species (*Ornithorhynchus anatinus* and *Tachyglossus aculeatus*) currently available in the NCBI database. Our analysis uncovered multiple copies of the FCLR2-5 genes in the marsupials, whereas the monotremes appear to retain only the FCRL6 and an FCRL6-like gene. Through phylogenetic analysis, we were able to determine that the FCRL genes present in marsupials are not true orthologs of the FCRL genes present in Eutheria, with only the FCRL6 appearing to be conserved throughout evolution. This suggests that marsupial FCRL diversity originated from duplication of an ancestral FCRL4, leading us to believe that originally a mammalian ancestral for both FCRL6 and FCRL4 existed. While marsupials appear to be more conserved and have a closer relationship with the ancestral FCRL4, in placental mammals we have a higher mutation rate and the appearance of highly diverse FCRL members. In addition to this, our results show that the NCBI annotation of the marsupials FCRL's is wrong since these receptors do not have a close phylogenetic relationship with their supposed orthologs.

Talk 15

The origin and maintenance of adaptive seasonal camouflage in the least weasel

Miranda, I (1,2,3); Ruivo, R (4); Farelo, L (1,3); Alvarenga, M (1,2,3); Borowski, Z (5); Elmeros, M (6); Kalthoff, DC (7); Merilä, J (8,9); Müller, JP (10); Schley, L (11); Suchentrunk, F (12); Sundell, J (13); Rodrigues, M (14); Santos-Reis, M (14); Fernandes, CR (14,15); Zub, K (16); Good, JM (17); Mills, LS (18,19); Castro, LFC (2,4); Dalén, L (20,21,22); Melo-Ferreira, J (1,2,3)

(1) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Universidade do Porto, Vairão, Portugal; (2) Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Porto, Portugal; (3) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal; (4) CIIMAR – Interdisciplinary Centre of Marine and Environmental Research, Universidade do Porto, Matosinhos, Portugal; (5) Department of Forest Ecology, Forest Research Institute, Raszyn, Poland; (6) Department of Ecoscience, Aarhus University, Aarhus, Denmark; (7) Department of Zoology, Swedish Museum of Natural History, Stockholm, Sweden; (8) Organismal and Evolutionary Biology Research Programme, Faculty of Biological and Environmental Sciences, University of Helsinki, Helsinki, Finland; (9) Area of Ecology and Biodiversity, School Biological Sciences, The University of Hong Kong, HKSAR; (10) Bündner Naturmuseum, Chur, Switzerland; (11) Administration de la Nature et des Forêts, Diekirch, Luxembourg; (12) Research Institute of Wildlife Ecology, University of Veterinary Medicine Vienna, Vienna, Austria; (13) Lammi Biological Station, University of Helsinki, Hämeenlinna, Finland; (14) CE3C – Centre for Ecology, Evolution and Environmental Changes & CHANGE – Global Change and Sustainability Institute, Faculdade de Ciências, Universidade de Lisboa, Lisboa, Portugal; (15) Faculdade de Psicologia, Universidade de Lisboa, Lisboa, Portugal; (16) Mammal Research Institute, Polish Academy of Sciences, Białowieża, Poland; (17) Division of Biological Sciences, University of Montana, Missoula, MT, USA; (18) Wildlife Biology Program, University of Montana, Missoula, MT, USA; (19) Office of Research and Creative Scholarship, University of Montana, Missoula, MT, USA; (20) Centre for Palaeogenetics, Stockholm, Sweden; (21) Department of Bioinformatics and Genetics, Swedish Museum of Natural History, Stockholm, Sweden; (22) Department of Zoology, Stockholm University, Stockholm, Sweden

Adaptive evolution, population genomics, phylogenomics, functional assays

Studying the evolution of locally adaptive traits is fundamental to understand the evolutionary mechanisms underlying natural diversity and species' adaptive potential to changing climates. Seasonal coat colour change, from summer-brown to winter-white, is a key phenological adaptation of several mammal and bird species, to maintain camouflage year-round in environments with seasonal snow. Locally adapted winter-brown and winter-white morphs occur across species' distribution ranges, allowing adaptation to environments with distinct winter snow conditions. In European least weasels (*Mustela nivalis*), polymorphism of the winter colour is known to map to the MC1R gene, a key component in the melanin-production pathway. In this work, we used in-vitro functional assays that showed how non-synonymous variation in MC1R alters the protein function, explaining the generation of alternative winter phenotypes. We then used a custom targeted-enrichment assay, targeting MC1R and anonymous genome-wide regions, to explore the origin and maintenance of this adaptive polymorphism. First, we applied this assay to a dataset representative of 11 Mustelidae species to investigate the evolutionary origin of MC1R variation in least weasels, in the context of the diversification of the genus. Second, we applied the capture assay to more than 240 least weasel specimens, representative of the species distribution range in Europe, including three regions polymorphic for winter colour. We used this dataset to i) characterise neutral population structure and demographic history, ii) detail patterns of genetic and haplotypic diversity within the MC1R adaptive locus, and iii) explore the relative role of neutral demography and selection in maintaining this adaptive genetic polymorphism through time. Our results highlighted a species-specific origin for the polymorphism and supported its long-term maintenance, likely due to snow-driven spatially varying selection, throughout the species diversification in Europe. Our work sheds light on the evolutionary processes generating and maintaining key adaptive variation throughout the evolution of species, which may be fundamental to promote rapid adaptation to future environmental change.

Talk 16

How do biochemical and environmental factors shape the fitness landscape of synonymous mutations on Hsp90?

Berjano, D (1); Bank, C (2,3); Fragata, I (1)

(1) cE3c - Centre for Ecology, Evolution and Environmental Changes & CHANGE - Global Change and Sustainability Institute, Faculdade de Ciências, Universidade de Lisboa; (2) IEE, University of Bern, Bern, Switzerland; (3) SIB-Swiss Institute of Bioinformatics, Lausanne, Switzerland

Synonymous mutations; Distribution of Fitness Effects; Deep Mutational Scanning

Synonymous mutations, i.e., nucleotide mutations that do not change the amino acid in a protein sequence, have been traditionally considered of negligible effect on fitness or phenotype. However, accumulating evidence suggests that some synonymous mutations matter for fitness. Codon bias is thought to be one of the main mechanisms through which synonymous mutations affect organismal fitness by changing the amount and quality of protein produced. The impact of codon bias is known to vary with the location of the mutation and expression level of the gene (by differentially affecting the elongation rate, co-translational folding process, and mRNA structure). Another factor important in determining the fitness effect of synonymous mutations is the presence of an environmental stress, by imposing additional need for specific protein quantity, conformation, translational speed or accuracy. Systematic quantifications of the prevalence of fitness effects of synonymous mutations and which factors shape this effect are rare.

To fill this gap, we reanalyzed published deep mutational scanning data from the chaperone Hsp90 of *Saccharomyces cerevisiae* at two expression levels. We estimated the fitness effects of every possible synonymous mutation when exposed to different environmental stresses and identified candidate mutations that showed the most extreme and consistent effects on fitness. We found that the expression level of Hsp90 strongly affected the effect of synonymous mutations, with only 9 candidate mutations identified at normal expression and 495 at low expression. We also observed a strong effect of the environment on the low expression candidate mutations. Heat stress produced on average many mildly deleterious mutations, whereas oxidative and osmotic stress led to stronger, but fewer deleterious mutations. Additionally, we observed that normal expression levels led to a more leptokurtic distribution of fitness effects, whereas low expression levels showed more non-neutral synonymous mutations. This result, together with testing of the relation between codon frequency and fitness, suggests that codon usage bias may not be the main driver of synonymous fitness effects in our data.. Overall, we find that synonymous mutations can show a wide range of fitness effects, which are altered by the presence of abiotic stresses and changes in gene expression. This suggests that synonymous mutations might have a greater impact on adaptation to novel conditions than traditionally thought.

Talk 17

Monoterpene synthase ligand preference: a case of both divergent and convergent evolution in Menthinae, Lamiaceae

Laia, J (1,2); Rodríguez-Fernandes, C (1); Trindade, H (1,2)

(1) CE3C - Centre for Ecology, Evolution and Environmental Changes & CHANGE - Global Change and Sustainability Institute, Faculdade de Ciências, Universidade de Lisboa, Campo Grande, Lisboa, Portugal; (2) Faculdade de Ciências da Universidade de Lisboa (FCUL)

Specialized metabolism; terpenoids; positive selection; plant communication; sesquiterpenes

Plant functional evolution relies on selection on phenotypes involved in plant growth and development, physiology and interactions with biotic and abiotic external environmental elements. For these purposes, plants synthesize and produce metabolites: (1) primary metabolites, crucial for vital processes of development and physiology, (2) phytohormones, small compounds that interact with receptor proteins to regulate the production of other metabolites and (3) specialized metabolites, which are involved in communication and stress resistance. Specialized metabolites show low evolutionary conservation and a high variation, especially because they are implicated in defence towards different organisms. The most abundant and diverse group of secondary metabolites is terpenoids, which include monoterpenes (10-carbon chain), sesquiterpenes (15-carbon chain) and diterpenes (20 carbon-chain). Terpenoids can be part of the primary metabolism (e.g. phytol, present in chlorophylls and crucial for photosynthesis) and be important phytohormones. However, the largest fraction of terpenoids is involved in specialized metabolism. Functions of specialized terpenoids include attracting pollinators, deterring herbivores, attracting parasitoids, and sending signals to neighbouring plants for cold and drought resistance. The subtribe Menthinae, family Lamiacae, not only has played an invaluable role in human culture as food, industrial products and medicines thanks to terpenoids, as it is a taxon that shows a vast chemical diversity of specialized metabolism, especially terpenoid diversity. Specific terpene synthases (TPS) recruited to the cellular compartments synthesize sesquiterpenes from farnesyl pyrophosphate (FPP) in the cytosol, and monoterpenes from geranyl pyrophosphate (GPP) and diterpenes from geranylgeranyl pyrophosphate (GGPP) in the plasts. However, terpene synthases are promiscuous, which means that they can use several ligands as substrate, not only their specific ligand. The vast terpenoid diversity in plants is the result of: (i) TPS plasticity dependent on metal cofactors, pH and substrate geometry; (ii) genome duplications; and (iii) the effect of single-residue changes in the TPSs resulting from mutations and selection on genes. In this study, we aimed to characterize the variability and function of TPS within the Menthinae subtribe. For that, we assessed the impact of lineage-specific mutations on positively selected sites in TPS within the subtribe Menthinae on the affinity of binding to their ligands. First, we addressed which sites of a reference protein sequence from each orthogroup are under positive selection. Second, we tested the impact of the SNPs present for each of those sites on the binding preference to GPP, FPP and GGPP. We found evidence for both divergent and convergent evolution taking place in monoterpene synthase function within the Menthinae subtribe. While there is divergence in substrate preference between the Mentha genus and the Micromeria group, we found evidence of convergence in FPP preference in several Micromeria lineages. JL is funded by the PhD research fellowship BD05097 from Fundação para a Ciência e Tecnologia (FCT). Support was given by FCT for Portuguese National Funds attributed to CE3C within the strategic project UID/BIA/00329/2020 (DOI: 10.54499/UIDB/00329/2020).

Talk 18

Evolutionary and functional study of the thyroid Sodium/Iodide Symporter homologs in plants

Sousa, BS (1,2,3); Lourenço, T (1,2,3,4); Pires, L (1,2,3); Azevedo, H (1,2,3); Castro, PH (1,2,5)

(1) CIBIO-InBIO, Universidade do Porto, Vairão, Portugal; (2) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal; (3) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto, Portugal; (4) IPSiM, Université de Montpellier, CNRS, INRAE, Institut Agro, Montpellier, France; (5) Departamento de Biologia, Escola de Ciências, Universidade do Minho, Campus de Gualtar, Braga, Portugal

Iodine homeostasis, comparative genomics, gene expression, Arabidopsis

In the human diet there are several essential elements involved in the proper development of the human organism. A particularly fundamental element for brain development is iodine and, consequently, its deficiency leads to several neurological complications, among others serious symptoms. In an attempt to overcome iodine deficiency, an important strategy was to reinforce the use of iodised salt supplementation. Although this solution achieved positive results, it has been proven that this strategy is not sufficient to mitigate iodine deficiency. Therefore, other approaches are also being explored, such as improving iodine content in crops through biofortification strategies. However, knowledge about iodine regulation in plants is very limited. Hence, this project aimed to investigate possible conserved iodine uptake systems through the evolutionary study of putative iodine transporters in plants and using *Arabidopsis thaliana* as a model organism for gene functional characterization. The present work was based on a previous comparative genomics approach that identified the homologous genes in eukaryotes of well-known iodine transporter in animals, including the sodium-iodine symporter (NIS). A phylogenetic analysis of NIS orthologous genes, anchored in representative plants species, revealed the presence and conservation of a NIS-like family in plants with a unique member in Arabidopsis thaliana, named AtNIS-like. We subsequently performed several in silico analyses, including an AtNIS-like gene expression pattern of plants exposed to various abiotic stresses. Additionally, AtNIS-like transcript levels in response to different potassium iodine (KI) concentrations were accessed by quantitative RT-PCR. Results showed that the expression of AtNIS-like increased during seed germination under osmotic-related stresses, and a similar result was observed for plants grown in media with iodine supplementation. Moreover, an AtNIS-like tagged with Green Fluorescent Protein construct, 35S::AtNIS:GFP, was transiently overexpressed in N. benthamiana leaves. This allowed us to confirm that the subcellular localization of AtNIS-like:GFP is mainly at the plasma membrane. Finally, we performed an iodine sensitivity assay by exposing Arabidopsis Atnis-like mutant lines to increasing doses of different iodine salts. We observed that Atnis-like mutants were less sensitive to higher doses of iodine salt supplementation. In conclusion, the NIS-like gene family is conserved in plants and evidences suggest that the AtNIS-like likely functions as a cell membrane-localised iodine transporter.

Acknowledgments: Work supported by National Funds through FCT-Fundação para a Ciência e a Tecnologia in the scope of the project UIDB/50027/2020. Work co-funded by the project NORTE-01-0246-FEDER-000063, supported by Norte Portugal Regional Operational Programme (NORTE2020), under the PORTUGAL 2020 Partnership Agreement, through the European Regional Development Fund (ERDF).

Talk 19

Very Weak Support for Deuterostome Monophyly is Boosted by Long Branch Attraction Artefacts

Serra Silva, A (1); Kapli, P (1,2); Natsidis, P (1); Telford, MJ (1)

(1) CLOE - Centre for Life's Origins and Evolution, Department of Genetics, Evolution and Environment, University College London, London, UK; (2) Science Group, Natural History Museum, London, UK

Deuterostomia; Phylogenomics; Systematic errors

Much of our understanding of early animal evolution rests on the existence of two bilaterian clades, Deuterostomia and Protostomia. An exhaustive comparison of multiple independent phylogenomic datasets revealed disparate levels of support for these clades; however, while strong support for protostome monophyly is widespread across datasets, support for deuterostomes is equivocal and linked to conditions known to lead to systematic errors in tree inference (inadequate substitution models, presence of long-branches and short-internal branches).

We generated a new dataset with over 180 loci and 300 metazoan species to systematically explore the sources of error suspected to underpin support for Deuterostomia. To parse the effects of long-branch artefacts and inadequate substitution models, we compared the support for Deuterostomia under site-homogeneous and site-heterogeneous substitution models on two sets of taxon jackknifed alignments – including or excluding long-branched taxa. This systematic approach confirmed that, when sources of error are mitigated, it is nearly impossible to distinguish between monophyletic Deuterostomia and its paraphyletic alternatives, and that long-branch artefacts have a higher impact on support for monophyletic deuterostomes than model inadequacy. Our results suggest that even if Deuterostomia is monophyletic, many of its purported synapomorphies were probably present in the last common ancestor of Bilateria. These results have implications for our understanding of bilaterian relationships and evolution.

Talk 20

Phylogenomics of Lupinus reveals strong geographic structuring and multiple instances of phenotypic convergent evolution

Nevado, B (1); Wahlert, G (2); Wong, E (3); Carona, S (1); Hughes, CE (4)
(1) CE3C - centre for ecology, evolution and environmental changes, FCUL University of Lisbon, Lisbon, Portugal; (2) Cheadle Center for Biodiversity and Ecological Restoration, University of California, Santa Barbara, USA. (3) Senckenberg Biodiversity and Climate Research Center, Frankfurt am Main, Germany. (4) Department of Systematic and Evolutionary Botany, University of Zurich, Zurich, Switzerland.

Speciation; phylogenomics; evolutionary radiations

The plant genus Lupinus encompasses over 300 species, the vast majority belonging to three clades that have diversified over the past ca. 5Myr along the western regions of North, Central and South America. Rapid diversification in these clades was accompanied by phenotypic divergence and invasion of different habitats. Indeed, Lupinus species exhibit an array of phenotypes ranging from small herbaceous annuals to woody shrubs, subshrubs, prostrate mat-forming species, small trees or acaulescent rosettes; and are found in various habitats including the arctic tundra, sand dunes, forests, prairies, deserts, or the high elevation Paramo regions of the Andean Mountain range. Due to its young age and large number of species, previous phylogenetic analyses using Sanger-sequencing and multi-locus approaches were unable to robustly resolve phylogenetic relationships among western New World Lupinus spp. In this work, we collected and analysed genomic data (Illumina re-sequencing) from the vast majority of western New World species of Lupinus – ca. 450 individuals representing 190+ taxa. Phylogenomic relationships inferred with this dataset were highly supported, and most species represented by more than 1 individual were resolved as monophyletic. We find that phylogenetic diversity in Lupinus spp. exhibits a strong geographic signal, at both large (i.e., continental) and medium (e.g., within the Andean mountain range) scales. Such strong geographic signal suggests limited dispersal, with younger “geographic clades” founded by single long-range dispersals events. Yet, we find that within each clade a largely overlapping set of plant habits evolved, and that these can sometimes be linked to invasion of specific habitats. Such convergent evolution of phenotypes is a strong indicator of the role of natural selection driving diversification, as described in other iconic evolutionary radiations such as the Anolis lizards from the Greater Antilles, or the East African Cichlids from Lakes Tanganyika, Malawi and Victoria. How the genetic diversity underpinning such convergent phenotypic evolution survives the bottlenecks associated with long-range dispersal remains unclear, and is the focus of ongoing work.

Talk 21

Lineage diversification of Acrocephalidae family (Reed Warblers) and its relationship with migratory behaviour and islands colonisation

Moreno Fernández, SE (1,2); Sannolo, M (3); Ponti, R (2)

(1) FCUP – Faculty of Sciences, University of Porto, Porto, Portugal; (2) BIOPOLIS-CIBIO – Center of Research in Biodiversity and Genetic Resources, Vairão, Portugal; (3) MNCN-CSIC - Museo Nacional de Ciencias Naturales, Madrid, España.

Lineage diversification, Passeriformes, biogeography, phylogenetics, ancestral state reconstruction

Avian diversification processes and the factors involved have been subject of debate in the last decades. Migratory behaviour was proved to affect speciation and extinction rates in birds, but how it could have influenced Old World Reed Warblers (Acrocephalidae, Aves) remained unclear. In this study we aimed to a) understand the phylogenetic relationships within Acrocephalidae; b) unravel if migratory behaviour or island colonisation had an effect in the family diversification processes; and c) disentangle migratory behaviour in the basal ancestor and reconstruct the biogeographic history of the family. We built the most comprehensive and up-to-date phylogeny of Acrocephalidae that gave light to previously unsolved nodes and that dated the origin of the family 10 million years ago (Mya). Based on it, we carried out comparative analyses to study the tempo and mode of diversification of the family associated with the place where they live (mainland or island), and migratory behaviour. Results suggest that migratory behaviour, more concretely, the loss of migration, played a key role in the evolutionary history of Acrocephalidae, influencing speciation and extinction rates in continental and insular taxa. In addition, ancestral states and biogeographic reconstructions were built to elucidate that the basal ancestor of the family was migratory and lived among Central Eurasia, the Middle East and the Arabian region. Therefore, migratory behaviour is a cornerstone in Acrocephalidae diversification patterns.

Talk 22

Characterization of polymorphic inversions in locally adapted populations of *Clunio marinus*

Peralta, CM (1); Kaiser, TS (1)

(1) MPI - Max Planck Institute for Evolutionary Biology, Ploen, Germany

Population genetics; local adaptation; chromosomal inversions; recombination; chronobiology

Chromosomal inversions can play an important role in local adaptation, especially when local adaptation is being mediated by multiple and/or polygenic traits. By reducing recombination in heterozygotic state, inversions allow for the maintenance of co-adapted alleles. Even though there is a recent increase in studies focusing on inversions, the processes by which they arise, their role in maintaining multiple locally adaptive traits and how they are influenced by selection still requires further studies. In the marine midge *Clunio marinus*, locally adapted timing phenotypes can be observed in the intertidal zone of the European coast. As tidal regimes differ along the coastline, Clunio populations differ in daily and lunar emergence time, and these differences are genetically determined. Here, we explore the role of inversions in local adaptation in 15 *C. marinus* populations. Using SNP data, we detect nine large genomic regions in high linkage disequilibrium, and with long-read sequencing data we confirmed that at least five of these correspond to inversions. We characterized the inversion breakpoints and found that the breakpoint positions in the inverted haplotypes often co-occur with repeated regions. Interestingly, all the inversions are paracentric (i.e. they do not include the centromeres). After genotyping the inversions across all populations, we tested if inversion frequencies are correlated with emergence timing or geographical latitude. Our results shed light on the direct effects of inversions by gene disruption and indirect effects via recombination reduction in local adaptation of *C. marinus* populations.

Talk 23

A turbulent evolutionary history involving massive gene gains and losses has shaped the genome and metabolism of a fungal lineage

Carla Gonçalves (1,2), Ana Pontes (1,2), Patrícia Brito (1,2), Jacob L. Steenwyk (3,4), Antonis Rokas (3,4), José Paulo Sampaio (1,2) and Paula Gonçalves (1,2)

(1) UCIBIO, Department of Life Sciences, NOVA School of Science and Technology, Universidade NOVA de Lisboa, Caparica, Portugal; (2) Associate Laboratory i4HB, NOVA School of Science and Technology, Universidade NOVA de Lisboa, Caparica, Portugal; (3) Department of Biological Sciences, Vanderbilt University, Nashville, TN 37235, USA; (4) Evolutionary Studies Initiative, Vanderbilt University, Nashville, TN 37235, USA.

Genome evolution, horizontal gene transfer, yeast metabolism, ecology

In yeasts (kingdom Fungi, subphylum Saccharomycotina), genes involved in essential metabolic pathways are generally well-conserved.

However, in a specific lineage of floral yeasts, known as the Wickerhamiella and Starmerella clade (W/S clade), we observed a unique pattern of central metabolic pathway modifications driven by extensive gene loss and frequent horizontal gene transfer (HGT).

Although HGT is a rare mechanism of genetic exchange in eukaryotes, it appeared to have been exceptionally frequent in the W/S clade, which is currently recognized as the yeast lineage with the highest rate of HGT events, specifically from bacteria. This raises several questions:

(i) what are the functions of these HGT-acquired genes?

(ii) how have these genes integrated and adapted to the eukaryotic setting?

(iii) why so much HGT in this lineage?

Our findings suggest that both gene loss and HGT have significantly reshaped the metabolic architecture of species within the W/S clade, contributing to their remarkable diversity in sugar preference, sugar utilization rates or fermentation profiles. Here, we will examine possible links between gene loss/acquisition events and metabolic evolution within an ecological framework. Furthermore, we will discuss hypotheses to explain the unusual prevalence of HGT in this clade, drawing on recent genomic and phenotypic data from over 50 W/S yeast species.

Talk 24

A shark's eye view on genome size evolution: phylogenetic patterns, molecular causes, and phenotypic consequences

Torralba Sáez, M (1); Hofreiter, M (2); Straube, N (3)

(1) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, FCUP University of Porto, Porto, Portugal; (2) IBB - Institute for Biochemistry and Biology, University of Potsdam, Potsdam, Germany; (3) Department of Natural History, University Museum Bergen, University of Bergen, Bergen, Norway

Genome size; genome evolution; karyotype evolution; ancestral reconstruction; phylogenetic comparative methods

Genome size, the amount of DNA contained in a haploid genome, varies extensively across organisms irrespective of their complexity. The current state of research in the evolution of genome size addresses questions concerning its mode and rate of evolution, the molecular processes underlying its variability, as well as the selective forces driving it. The observation that these trends often manifest variably across different taxa renders genome size evolution one of the most enduring puzzles in genome biology. Within vertebrates, the large and heterogeneous genome sizes of sharks are only exceeded by those of lungfishes and salamanders. However, previous research on the evolution of fish genome size has been either entirely focused on ray-finned fishes or strongly biased towards them, leaving sharks heavily understudied. Aiming for a better understanding of the patterns and causalities of shark genome size evolution, we applied phylogenetic comparative methods to published genome-size estimates for 71 species representing the main phylogenetic lineages, life-histories, and ecological traits. Our study revealed that the sixfold range of genome size variation was strongly traceable throughout the phylogeny, with a major expansion preceding shark diversification during the late Paleozoic and an ancestral state (6.33 pg) close to the present-day average (6.72 pg). Subsequent deviations from this average occurred at higher rates in squalomorph than in galeomorph sharks and were unconnected to evolutionary changes in the karyotype, which were dominated by descending disploidy events with no polyploidizations involved. Genome size was strongly positively correlated with cell and nucleus sizes and negatively with metabolic rate. The metabolic constraints on increasing genome size manifested at higher phenotypic scales: active swimming behaviours were accompanied by small genome sizes, as were the rapid early developments of placental viviparous species. Moreover, the prevalence of smaller genomes in marine-brackish and amphidromous species relative to purely marine ones suggests that metabolic constraints related to osmoregulatory costs in fluctuating salinities may play a role. Contrary to other vertebrates, we found no support for a maladaptive nature of large DNA accumulations, as they did not affect the diversification of affected clades (evolvability) nor were influenced by neutral processes, like population size.

Talk 25

Phenotypic and genetic differentiation between two chromosomal taxa of the gastropod *Nucella lapillus* at Galician rocky shores (NW Spain)

García-Souto, D (1); Fernández-Rodríguez, J (1); Pasantes JJ (1); Rolán-Álvarez, E (1); Galindo, J (1)

(1) Centro de Investigación Mariña, Universidade de Vigo, Departamento de Bioquímica, Genética e Inmunología, Campus Universitario Lagoas-Marcosende, 36310 Vigo, Spain.

Evolution; Cytogenetics; Marine gastropods; Karyotyping

In certain North Atlantic rocky shores, two chromosomal lineages of *Nucella lapillus* have been observed at different extremes of an environmental gradient, and supposedly detected hybridization between them. Here, we have determined the presence of this chromosomal polymorphism, resulting from Robertsonian translocations, in populations of the Iberian Peninsula spawning a similar gradient. Interestingly, we have found monomorphic populations (2n = 26) across the gradient while we only found polymorphic populations (2n = 27-36) in certain exposed microhabitats, and never both types of populations together. These chromosomal lineages differ morphologically and genetically in a manner that can be successfully discriminated, based in their morphology (95 % of success) and molecular variation (99.9 % of success), in the studied set of samples, and so attending to this data this could potentially represent isolated evolutionary lineages or taxa. This situation is discussed in relation to the data available in previous studies in this and other geographical areas for this species. The new findings do not solve the problem of the Robertsonian polymorphism known to exist in this species, but it could perhaps suggest a new approach to solve it, suggesting a wide geographical comparison between the two taxa in order to confirm if they represent evolutionary isolated lineages or maintain certain degree of hybridization.

Talk 26

Identifying the sex chromosomes of *Laurus azorica*

Branco, C (1,2,3,4); Roxo, G (3,5,6,7); Chapoix, I (2,3,4); Rego, MCR (3,5,6,7); Tessarotto, H (8); Santoni, S (8); Tavares, R (1,2,3,4); Linheiro, R (1,2,3,4); Iannucci, A (9,10); Ciofi, C (9); Diroma, MA (9); Natali, C (9,11); Morselli, M (12); Resendes, R (2); Sim-Sim, M (13,14,15); Trindade, H (13,15); Vicente, S (13,16); Marques, JP (2,3); Sousa, V (13,17); Melo-Ferreira, J (2,3,4); Cartney, AM (18); Mouton, A (9,19); Formenti, G (20); Marais, G (1, 21); Moura, M (3,5,6,7)

(1) LBBE, Laboratoire de Biométrie et Biologie Evolutive, Université de Lyon,Villeurbanne, France; (2) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Universidade do Porto, Vila do Conde, Portugal; (3) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal; (4) Faculdade de Ciências, Universidade do Porto, Porto, Portugal; (5) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Pólo dos Açores – Faculdade de Ciências e Tecnologia, Universidade dos Açores, Ponta Delgada, Açores, Portugal; (6) UNESCO Chair – Land Within Sea: Biodiversity & Sustainability in Atlantic Islands; (7) Departamento de Biologia, Faculdade de Ciências e Tecnologia, Universidade dos Açores, Ponta; (8) DIADE, Université de Montpellier, IRD, CIRAD; Montpellier, France; (9) University of Florence, Sesto Fiorentino, Italy; (10) National Biodiversity Future Center (NBFC), Palermo, Italy; (11) Okinawa Institute of Science and Technology, Okinawa, Japan; (12) University of Parma, Parma, Italy; (13) Faculdade de Ciências, Universidade de Lisboa, Lisboa, Portugal; (14) Museu Nacional de História Natural e da Ciência (MUHNAC), Universidade de Lisboa, Lisboa, Portugal; (15) Faculdade de Ciências, Universidade de Lisboa, Lisboa, Portugal; (16) Escola Superior de Saúde Ribeiro Sanches (ERISA), Instituto Politécnico da Lusofonia (IPLUSO), Lisboa, Portugal; (17) Faculdade de Ciências, Universidade de Lisboa, Lisboa, Portugal; (18) Genomics Institute, University of California, Santa Cruz, USA; (19) University of Liege, Liege, Belgium; (20) The Rockefeller University, New York City, USA; (21) Department of Biology & GreenUP, Faculty of Science, University of Porto, Porto, Portugal

Evolution of sexual systems; Colonization; Oceanic islands; Dioecy

The Laurus genus exhibits a current distribution encompassing the southern Black Sea region, the Mediterranean Basin, Morocco, and the Macaronesian islands (i.e., the Azores, Madeira, and Canaries archipelagos). Despite numerous attempts, previous studies faced challenges in elucidating the taxonomy, ploidy, and phylogenetic relationships within this genus. Concerning the taxonomy, two species are traditionally recognized: *L. nobilis* and *L. azorica*. This study delves into *L. azorica*, a dioecious tree endemic to the Azores archipelago that falls within the European Reference Genome Atlas (ERGA) initiative to ensure high-quality reference genomes of the European biodiversity. Despite the well-established dioecy of this species, its sex-determination system remains elusive. Here, we aim (i) to characterize *L. azorica* sex chromosomes without a reference genome, and (ii) generate a reference genome. Firstly, long-read sequencing data from Pacific Biosciences (PacBio) were acquired to ensure a robust genomic dataset. Additionally, chromatin conformation capture data (Hi-C) were incorporated to facilitate the construction of a chromosome-level assembly. Then, Illumina short reads for 32 individuals were obtained, ensuring an even representation of phenotypic males and females. SDPOP (Käfer et al. 2021) analysis were applied to infer the genes' sex-linkage by modelling populations' allele and genotypic frequencies. This framework allows statistical testing of alternative sex-determination systems (i.e., presence or absence of sex chromosomes, distinguishing between XY and ZW systems). Our results indicate that this species has XY sex chromosomes that stopped recombining 10 million years ago. Also, *L. azorica* is tetraploid, consistent with prior studies on this genus. Ongoing research is expected to yield contribute valuable information to broader studies on dioecy and sex-determination mechanisms in plant species. This study serves as a cornerstone for further exploration into the evolutionary dynamics within the Laurus genus.

Talk 27

Stronger together: a revision of stakeholder impact of NEDE.APBE's 12 years of education and outreach in evolution

Ponce, R. (1,2); Laurentino, TG (N.A); Cavadas, B (3); Fonseca, M.J.(4,5); Campos, R (6); Alpedrinha, J. (7); Nogueira, T (7,8,9); Pessoa, P. (10, 11); Sousa, B. (12); Varela, S.(13,14); Rodrigues, A. (NA); Aboim, S. (15); Branquinho, R. (16,17); Sá-Pinto, X (10, 18).

(N.A.) Not affiliated (1) ESE-IPS School of Education, Polytechnic Institute of Setúbal, Setúbal, Portugal (2) ICNOVA, NOVA University Lisbon, Lisbon, Portugal (3) Polytechnic University of Santarém; CeiEd, Lusofona University, Lisbon, Portugal. (4) Natural History and Science Museum of the University of Porto (MHNC-UP) (5) Faculty of Sciences of the University of Porto (6) CES - Centre for Social Studies, University of Coimbra, Coimbra, Portugal (7) cE3c—Center for Ecology, Evolution and Environmental Changes (8) INIAV—Instituto Nacional de Investigação Agrária e Veterinária, I.P., 4485-655 Vairão, Portugal (9) CHANGE—Global Change and Sustainability Institute, 1749-016 Lisbon, Portugal (10) CIDTFF.UA, Research Centre on Didactics and Technology in the Education of Trainers, Department of Education and Psychology, University of Aveiro;Aveiro, Portugal (11) University of Trás-os-Montes and Alto Douro, Vila Real, Portugal (12) Agrupamento de Escolas de Albufeira Poente (13) WJCR - William James Center for Research, ISPA-Instituto Universitário, Lisboa, Portugal. (14) GIMM - Gulbenkian Institute for Molecular Medicine, Oeiras, Lisboa, Portugal. (15) ESE-IPS School of Education, Polytechnic Institute of Porto, Porto, Portugal. inED - Centre for Research and Innovation in Education (16) Faculty of Arts and Humanities of the University of Porto, Porto, Portugal. (17) Arma-Sci/GOMA (Rede de Promoção do Capital Científico de Armamar), Armamar, Portugal. (18) EvoKE, Evolutionary knowledge for Everyone Association, Portugal.

Evolution education and outreach; evolution literacy; transdisciplinary research network; public engagement in science; Community approach

Understanding evolutionary processes is crucial not only for grasping the systemic impacts of anthropogenic changes on ecosystems and biodiversity but also for addressing pressing challenges such as drug resistance, emerging pandemic diseases, and food security. Thus, knowledge of Evolution is important for scientists, educators, policymakers, conservationists, and society. Despite its intersectional relevance, evolution remains scarcely understood by many. Science curricula and textbooks across Europe, including Portugal, still fail to bridge the scientific knowledge on evolution with its impacts on daily life issues and to properly address widespread misconceptions about evolution. This results in a lack of public evolutionary literacy, which ultimately affects short- and long-term decision- and policy-making. To address this issue, in 2012 the Portuguese Association of Evolutionary Biology (APBE) created the Evolution Education and Outreach group (NEDE.APBE), a transdisciplinary research network (TdRN) aiming to promote public evolutionary literacy in Portugal. Here we report on NEDE.APBE's activities since its foundation. Specifically, we will focus our presentation in: i) actions to co-construct and disseminate scientific knowledge and science-based solutions for evolution education, in formal and non-formal contexts; ii) outreach activities and their outcomes for education research; iii) the professional development of teachers, evolutionary biologists and NEDE.APBE community members; and iv) the main limitations identified. Our results show that the actions of NEDE.APBE engaged diverse stakeholders and resulted in an important community that includes teachers, researchers and science communicators thus highlighting the potential of TdRN's social impacts across stakeholders. The actions of this community resulted in important research results and changes in curricula and textbooks. In the forthcoming period, NEDE.APBE intends to consolidate its collaborative relationships and extend its influence to reinforce the understanding of evolution in Portugal and beyond.

Talk 28

When individuality obscures geographic song variation: a comparison of two passerine sister species with different migratory strategies

Oñate-Casado, J (1); Porteš, M(1,2); Beran, V(3,4); Pérez-Granados, C(5); Traba, J(6,7); Barrero, A(6,7); Sakhalkar, SP(1); Petrusek, A(1); Petrusková, T(1)

(1) Department of Ecology, Faculty of Science, Charles University, Prague, Czechia; (2) Regional Office České středohoří Protected Landscape Area Administration, Nature Conservation Agency of the Czech Republic, Litoměřice, Czechia; (3) Municipal Museum of Ústí nad Labem, Ústí nad Labem, Czechia; (4) ALKA Wildlife o.p.s., Lidéřovice, Czechia; (5) Ecology Department, Alicante University, Alicante, Spain; (6) Terrestrial Ecology Group, Department of Ecology, Universidad Autónoma de Madrid, Madrid, Spain; (7) Centro de Investigación en Biodiversidad y Cambio Global, Universidad Autónoma de Madrid, Madrid, Spain

Bioacoustics; bird ecology; behavioural ecology; steppe birds

Songbirds are a well-established model group to study cultural evolution as their songs are learnt, i.e. culturally transmitted, which may lead to divergence in song characteristics between populations. Song variation may be affected by various factors, including geographic isolation, timing and duration of learning period, and life strategies such as migratory behaviour. In this study, we explored geographic song variation in two congeneric sister species of songbirds with a simple song: a Palaearctic migrant Tawny Pipit (*Anthus campestris*), and a sedentary Berthelot's Pipit (*A. berthelotii*) endemic to Macaronesian archipelagos. We recorded songs of territorial males from six Tawny Pipit populations in the European mainland, and of Berthelot's Pipits in three Canary and two Madeira islands. We confirmed that both species have individually unique repertoires, usually consisting of a single song type per male. The structural characteristics of song types within each species were compared by dynamic time warp analysis, allowing for quantifying song dissimilarity and analysing its geographic patterns. Contrary to our expectations, we did not find any regionally specific song characteristics that would allow assessing the origin of recorded songs recording in either species, not even between the two Berthelot's Pipit subspecies from different Macaronesian archipelagos. In both species, we observed very high within-population variation of song types, possibly reflecting rapid cultural evolution of song driven by the constant introduction of novel, although minor, individually-specific differences. Mean song dissimilarity was higher when song types were compared between different regions (mainland localities or islands) than within them. Only in Berthelot's Pipits, however, we observed significant differences in geographic song variation at the smallest and largest spatial scales, which might reflect Berthelot's Pipits' sedentarity and the isolation of Macaronesian archipelagos, respectively.

Talk 29

Popular birds live longer: The association of social bonds and survival in a social bird

Munar-Delgado, G (1); Paquet, M. (2); Ferreira, A. (3); Doutrelant, C. (4); Covas, R. (1)

(1) CIBIO-InBio, Research Centre in Biodiversity and Genetic Resources, Vairão, Portugal; (2) CNRSTheoretical and Experimental Ecology Station, Moulis, France; (3) Department of Evolutionary Biology and Environmental Studies, University of Zurich, Zurich, Switzerland; (4) CNRS - Centre d'Ecologie Fonctionnelle et Evolutive, University Montpellier, Montpellier, France

evolutionary ecology; social networks, capture-recatpure

Understanding how social structure influence individual fitness is key to uncovering the ecological and evolutionary costs and benefits of social behavior. In many species, social interactions provide critical advantages, such as improved access to resources and increased protection against environmental challenges. Our research focuses on sociable weavers (*Philetairus socius*), a highly social and cooperative breeding species and aimed to investigate how the number and strength of social bonds associate to survival during the winter months, a period that corresponds to harsher conditions. Using long-term data of birds marked with transponders which visited automatic feeders equipped with RFID technology, we recorded individuals' association and built social networks to estimate the number and strength of each bird's social bonds. We then used capture-recapture analysis to assess how the number of strong social bonds an individual is associated to survival. We found that individuals with a higher number of strong social bonds had a greater probability of surviving through winter. These results are consistent with previous studies showing that sociable weavers that roost in chambers with a higher number of conspecifics at their communal nests experience less time below their critical thermal temperature. In addition, birds with higher number of associations may have better foraging success or improved predator detection. While the actual mechanisms require further study, these results emphasize the broader significance of social bonds in enhancing individual survival under environmental stress and contribute to a deeper understanding of the adaptive value of social behavior.

Talk 30

A genetic trade-off between intrinsic growth and sensitivity to competition, but not with reproductive interference, in spider mites

Cruz, MA (1); Zélé, F (2); Sousa, VC (1,3); Magalhães, S (1,3)

(1) cE3c – Centre for Ecology, Evolution and Environmental Changes & CHANGE – Global Change and Sustainability Institute, Faculdade de Ciências da Universidade de Lisboa, Portugal; (2) ISEM – Institut des Sciences de l'Évolution de Montpellier, Université de Montpellier, CNRS, IRD, EPHE, Montpellier, France; (3) Departamento de Biologia Animal, Faculdade de Ciências da Universidade de Lisboa, Portugal.

Genetic variation; genetic correlations; coexistence theory; trophic interactions; sexual interactions

Although studies investigating competitive interactions have typically viewed species as homogenous units, a framework addressing the effects of individual variation on species coexistence has recently emerged. However, whether such variation has a genetic basis and is constrained by genetic correlations among traits remains elusive. We addressed this using two closely-related spider mite species, *Tetranychus cinnabarinus* (Tc) and *T. urticae* (Tu), which hamper each other's growth through both food competition and reproductive interference (costly sexual interactions due to partial reproductive isolation), with the former affecting the strength of the latter. Here, in 29 inbred Tc lines derived from a genetically diverse population, we measured (a) the intrinsic growth rate in the absence of any interaction; (b) food competition against a gradient density of intra- or interspecific competitors (i.e., from the same Tc line or from a reference Tu line), while sexual interactions were prevented; and (c) reproductive interference with the Tu line, while food competition was prevented. We found significant genetic variance (i.e., broad-sense heritability) among Tc lines for all estimated demographic or interaction variables (growth rate, food competition and reproductive interference strength), except for the negative effect that they have on Tu via food competition. Moreover, we document significant positive correlations between the intrinsic growth and sensitivity to competition of Tc lines, such that genotypes with higher offspring production also produced offspring that were more sensitive to food competition. This is evidence for a growth-competition genetic trade-off. The intrinsic growth of Tc also correlated with their negative effect on Tu through reproductive interference. However, no correlations between food competition and reproductive interference were found, suggesting that their interplay is not genetically determined. Our results thus highlight that variation in traits involved in species interactions have a genetic basis and can thus change during evolution. Moreover, we show that traits underlying different types of interactions can be under genetic linkage, and this, in turn, may constrain evolutionary responses to heterospecifics.

Talk 31

Is the devil in the details? Investigating the role of the body spot in male-female interactions of the fish Poecilia vivipara’s

Massote, C (1); Leitão, RF (2); Motta, TRP (3); Peixoto, PEC (2)

(1) CE3C - centre for ecology, evolution and environmental changes, FCUL University of Lisbon, Lisbon, Portugal; (2) 1Departamento de Ecologia, Conservação e Manejo da vida silvestre, Universidade Federal de Minas Gerais, Brasil; (3) 2Departamento de Fisiologia e Biofísica, Universidade Federal de Minas Gerais, Brasil

Sexual selection, mate choice, monomorphism

Sexually selected traits serve as indicators of individual quality to potential mates. Much of this understanding comes from studies on traits that are perceived as exaggerated by humans and in sexually dimorphic species. A problem with this approach is that traits may appear exaggerated or dimorphic to the human sensory system but not to the sensory system of the species itself. To broaden our understanding of sexual selection on individual traits, it is essential to investigate non-exaggerated traits in apparently monomorphic species. For this purpose, we investigated the potential role in mate selection of the dark spot present on the sides of both males and females of the fish Poecilia vivípara, an apparently monomorphic species. Since the dark spot manifests in sexually mature individuals and is costly to produce, we tested the hypotheses that the dark spot 1) indicates individual nutritional quality and 2) plays a role in mate choice in both sexes. To do so, we investigated the reflectance spectrum of the dark spot, correlated the spot area with morphophysiological measures that may indicate individual quality, and conducted a mate choice experiment. We found that males on average have larger spots than females and that it indicates individual quality for both females and males. However, the correlation for females is isometric, while for males it is a negative allometry. From this, we can conclude that the spot is actually dimorphic. Additionally, the spot is not used in mate choice. Given that it is costly to produce and sexually dimorphic, we suggest that the dark spot may play a role in post-copulatory processes.

Talk 32

The role of exercise in mitigating inflammaging and gut dysbiosis

Melo-Miranda, R (1); Silva, C (1); R Almeida, C (1); Sousa, A (1)

(1) Institute of Biomedicine (iBiMED), Department of Medical Sciences, University of Aveiro, Aveiro, Portugal

Aging; Escherichia coli; exercise; gut microbiota; microbial evolution

Aging is marked by a general decline in physiological functions, several of which are strongly linked to increased chronic low-grade inflammation (inflammaging), as well as dysbiotic changes in the gut microbiota. Given that voluntary exercise has been shown to slow inflammaging and extend healthspan, we aimed to explore its effects on the gut microbiota. For this purpose, middle-aged mice were enrolled in voluntary exercise and followed until old age, while a control group without access to exercise was monitored in parallel. We assessed systemic and intestinal inflammation, several health indicators and the gut microbiota composition. Next, we colonized animals with a commensal gut strain of Escherichia coli and followed its adaptation as it colonized both environments.

Exercised mice displayed delayed sarcopenia, evidenced by increased soleus muscle mass and improved forelimb grip strength compared to sedentary mice. Although body weight and non-fasted glucose levels were similar between groups, exercised mice exhibited a significant reduction in visceral fat, suggestive of enhanced metabolic health. Exercise also delayed the onset of inflammaging, mitigating the increase in the levels of pro-inflammatory cytokines such as IL-6, IL-17A, and IL-27.

Following antibiotic treatment – essential for colonization with Escherichia coli – exercise was associated with a smaller reduction in microbiota diversity and with the prevention of a bloom in Allobaculum sp., a bacterial genus with pro-inflammatory potential. Additionally, exercised mice showed a distinct evolutionary trajectory of E. coli populations, suggestive of a potentially more anerobic environment. Interestingly, the gat-negative phenotype, previously identified as highly advantageous in mice, regardless of age, emerged tendentially more slowly in exercised animals, with alternative metabolic mutations gaining prominence.

These results suggest that the senolytic effect of voluntary exercise is associated with alterations in gut microbiota dynamics. Moreover, exercise-induced shifts in microbial evolution highlight the complex interplay between physical activity and microbial adaptation, providing new insights into lifestyle interventions that may help delay age-related diseases.

Talk 33

Aging drives gut bacteria toward pathoadaptation

Melo-Miranda R (1); Pinto A (1); Barreto H (2); Jesus C (3); Gordo I (4); Duarte I (3); Sousa A (1)

(1) Institute of Biomedicine (iBiMED), Department of Medical Sciences, University of Aveiro, Aveiro, Portugal; (2) Université Paris Cité, CNRS, Inserm, Institut Cochin, F-75014 Paris, France ; (3) CICECO – Aveiro Institute of Materials, Department of Chemistry, University of Aveiro, Aveiro, Portugal; (4) Gulbenkian Institute for Molecular Medicine (GIMM), Pólo de Oeiras, Oeiras, Portugal;

Microbial adaptation; gut microbiota; experimental evolution

Laboratory-raised mice live approximately seven times longer and healthier lives than their wild counterparts, primarily due to a standardized healthy diet and minimal exposure to environmental stressors. Aging is linked with increased inflammation, gut permeability, and gut microbial dysbiosis. Collectively, these shape microbiota evolution and may contribute to the enrichment in pathobiont frequency observed in old age. Alternatively, another explanation for this increase in pathobionts might be a decline in colonization resistance due to age-associated immunosenescence, creating favorable conditions for pathobiont invasion. In this study, we aimed to test whether aging under controlled, healthy conditions with restricted exposure to external microorganisms could prevent the occurrence of age-related pathobionts.

We examined the adaptive evolution of a commensal strain of *Escherichia coli* in the guts of mice of advanced age (25 months old) and found that it acquired several mutations typical of bacteria colonizing young (6-8 weeks old) mice, which were not selected in a similar experiment in old (19 months old) animals. This, together with the observed increase in *Akkermansia muciniphila* and *Oscillospira sp.* in mice of advanced age, may be an indication of healthy aging. However, mutations acquired exclusively in the old and very old were mainly pathoadaptive, tuning the metabolism to oxygen and iron availability, hypermotility, and biofilm formation.

In conclusion, while the evolutionary patterns in the guts of very old mice suggest youth-like features that could relate to longevity, there is also an increased selection for pathoadaptive traits in very old age. These findings suggest that a decrease in colonization resistance alone does not explain the higher abundance of age-related pathobionts. Future research should determine whether these observations extend broadly across gut commensals or predominantly reflect the high metabolic flexibility characteristic of bacteria like *E. coli*.

Talk 34

Environmental drivers of colouration in Lusitanian wall lizards: Testing key ecogeographical hypotheses

Sreelatha, LB (1,2,3); Tarroso P (1,2,3) ; Nokelainen O (4,5) ; Boratyński Z (1,3) ; Carretero MA (1,2,3)

(1) CIBIO-InBIO, University of Porto, Vairão, Portugal; (2) Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Porto, Portugal; (3) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Vairão, Portugal; (4) Open Science Centre, University of Jyväskylä, Jyväskylä, Finland; (5) Department of Biological and Environmental Science, University of Jyväskylä, Jyväskylä, Finland.

Colouration; wall lizards; melanism; ecology; ecophysiology

The environment is a key driver of animal phenotypes, shaping adaptations to local conditions. However, how individual coloration varies across the climatic gradients of a species' range remains unclear. We investigated whether dorsal coloration in Lusitanian wall lizards (*Podarcis lusitanicus*) is associated with environmental gradients across the north-western Iberian Peninsula. We analyzed multispectral photographs of 463 lizards, from 21 locations to measure their dorsal lightness. Using piecewise structural equation modeling approach, we tested the links among lightness, environmental factors and body size to test four ecogeographical hypotheses: Gloger's rule (darker coloration in warm and humid), thermal melanism (darker coloration in cold), photoprotection (darker coloration with greater solar radiation) and Bergmann's rule (larger body size in colder climates). Our results support Gloger's hypothesis; humidity covaried with lightness regardless of individual's ancestry or geographic location. Temperature influenced color lightness, as predicted by thermal melanism hypothesis, but only indirectly, through covariation with body size, consistent with Bergmann's rule. Solar radiation did not covary with the animals' lightness, thus not supporting the photoprotection hypothesis. Our findings demonstrate that multiple climatic factors contribute to variations in the lizard phenotypes. To fully understand the functional mechanisms underlying these broad correlations, further experimental studies are needed.

Poster 1

The adaptive potential of sharks in a changing ocean: insights from comparative and population genomics

Alves, JS [1,2,3]; Marques, JP [1,3]; Farelo, L [1,3]; Arnaud-Haond, S [4]; Queiroz, N [1, 3]; Melo-Ferreira, J [1,2,3]

(1) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Campus de Vairão, Universidade do Porto, 4485-661 Vairão, Portugal;
(2) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, 4099-002 Porto, Portugal;
(3) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, 4485-661 Vairão, Portugal;
(4) UMR248 MARBEC, Univ. Montpellier, Ifremer, IRD, CNRS, Sète, France

The loss of biodiversity worldwide is an escalating concern affecting both terrestrial and marine environments. In the ocean, climate change is driving deoxygenation (a decrease in dissolved oxygen concentration) and warming, which pose important challenges for the survival of marine species. Rising sea temperatures pose a particular threat to ectothermic species, which depend on external conditions for temperature regulation, while deoxygenation affects species that expend large amounts of energy and have high oxygen requirements, such as top predators. Together, these stressors increase oxygen demands while simultaneously limiting oxygen availability. Given this context, the blue shark (*Prionace glauca*) – a cosmopolitan, migratory predator known for its ability to tolerate oxygen-poor environments - serves as a suitable model for studying the adaptive potential of marine species under climate change. However, despite its ecological importance and substantial population declines due to overfishing, few studies have examined its genomic diversity and capacity to adapt to environmental change. This project aims to leverage whole genome sequencing to investigate key evolutionary processes shaping the blue shark's responses to climate change. We are currently generating a complete and annotated genome assembly for the blue shark, which will serve as an anchor for subsequent analyses across two evolutionary scales. Firstly, we will undertake a macroevolutionary analysis to assess its adaptive potential in comparison to other shark species. To do so, we'll explore the deep evolution of traits affected by climate change, such as thermal tolerance, metabolic rates, and deep diving, using the wealth of genomic resources available for shark species along with those generated during the project, to conduct comparative genomic analyses. Next, we will focus on the blue shark as the model species to comprehensively characterize its genetic diversity and population history throughout its range. To achieve this, we will build our analysis on preliminary evidence of population structure and investigate its association with markers of local adaptation. Furthermore, to examine the diving behaviour observed in oxygen minimum zones (OMZs), we will collect genetic data alongside satellite tracking information on individual movements, aiming to identify genetic traits linked to diving capacity and to pinpoint specific adaptations to OMZ conditions. Finally, the identification of correlations between abiotic parameters and genotypes potentially revealed in our previous analyses will enable the modelling of these relationships and the development of predictive models using seascape genomics. Projections under future conditions will facilitate the assessment of the genetic and geographic changes necessary for adaptation to the rapid pace of climate change, as well as the potential for adaptive responses. Ultimately, this approach is expected to quantify the predicted impact of climate change on the species, considering its genetic diversity and adaptive capacities.

Poster 2

From darkness to data: genomic insights into the evolution and conservation of the Azorean Cave Ground Beetle *Trechus terceiranus*

Alves, JS [1,2,3]; Farelo, L [1,3]; Melo-Ferreira, J [1, 2, 3]; Salces-Ortiz, J [4]; Fernández, R [4]; Ciofi, C [5]; Natali, C [5]; Morselli, M [6]; Fernandes, CR [7]; Sousa, V [7]; Pereira, F [8]; Borges, Paulo AV [8]; Marques, JP [1, 3]; Amorim, IR [9]

(1) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Campus de Vairão, Universidade do Porto, 4485-661 Vairão, Portugal; (2) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, 4099-002 Porto, Portugal; (3) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, 4485-661 Vairão, Portugal; (4) Institute of Evolutionary Biology (CSIC-Universitat Pompeu Fabra), Passeig Marítim de la Barceloneta 37-49, 08003 Barcelona, Spain; (5) Department of Biology, University of Florence, Sesto Fiorentino, Italy; (6) Department of Chemistry, Life Sciences and Environmental Sustainability, University of Parma, Parco Area delle Scienze 23A, 43124 Parma, Italy;(7) CE3C - Centre for Ecology, Evolution and Environmental Changes & CHANGE - Global Change and Sustainability Institute, Departamento de Biologia Animal, Faculdade de Ciências, Universidade de Lisboa, 1749-016 Lisboa, Portugal; (8) University of the Azores, cE3c—Centre for Ecology, Evolution and Environmental Changes, Azorean Biodiversity Group, CHANGE—Global Change and Sustainability Institute, Faculty of Agricultural and Environmental Sciences, Rua Capitão João d'Ávila, Pico da Urze, 9700-042 Angra do Heroísmo, Azores, Portugal; (9) University of the Azores, Azorean Biodiversity Group/CE3C- Centre for Ecology, Evolution and Environmental Changes & CHANGE Global Change and Sustainability Institute, Rua Capitão João d´Ávila, Pico da Urze, 9700-042, Angra do Heroísmo, Portugal.

The Azorean archipelago, located in the mid-Atlantic, is a biodiversity hotspot renowned for its exceptional endemism. Among its unique inhabitants are nine species of *Trechus* ground beetles: seven of these species are adapted to subterranean habitats existing in lava tubes, volcanic pits and the MSS (mesovoid shallow substratum), while two occupy surface habitats. One notable example is *Trechus terceiranus*, a ground beetle endemic to Terceira Island (single island endemic), which exhibits adaptations to subterranean life, such as depigmentation, reduced eyes, and reliance on specialized ecological strategies. Classified as Vulnerable by the IUCN, *T. terceiranus* faces threats from habitat degradation caused by agricultural and tourism expansion, infrastructure development, and climate change necessitating urgent conservation efforts supported by genomic insights. This study, a collaboration fostered through the Portuguese node of the European Reference Genome Atlas (ERGA-PT), has produced the first high-quality, haplotype-resolved reference genome for *T. terceiranus*, adhering to stringent Vertebrate Genomes Project (VGP) standards. This genomic resource not only establishes a robust foundation for evolutionary studies but also enables population-genomic analyses to uncover the genetic underpinnings of subterranean adaptations. By comparing *T. terceiranus* with its surface-dwelling relative, *T. terrabravensis*, also endemic to Terceira Island (single island endemic), we aim to identify genetic markers and mechanisms linked to traits involved in adaptation to the subterranean habitat, such as depigmentation, reduced ocular structures, and the regulation of biological rhythms. Additionally, molecular data reveal that *T. terceiranus* shares closer evolutionary affinities with other subterranean Trechus species across the Azores than with *T. terrabravensis*, underscoring the complex biogeographic and evolutionary history of this genus in the archipelago. This work exemplifies the integration of evolutionary biology and conservation genomics. By leveraging cutting-edge genomic methodologies and fostering international collaboration through ERGA-PT, we contribute to the preservation of island endemics, namely of *T. terceiranus* and its volcanic cave ecosystem. These insights also provide a framework for addressing broader questions about adaptation and resilience in biodiversity hotspots facing increasing anthropogenic pressures.

Poster 3

From molecules to conservation: PXR as a tool for ecological impact assessment

Amorim, I [1,2,3]; Oliveira, D [1,3]; Castro, F [1,3]; Ruivo, R [1], Lopes-Marques, M [1]

(1) CIIMAR- Interdisciplinary Centre of Marine and Environmental Research, University of Porto, Portugal;
(2) ICBAS-School of Medicine and Biomedical Sciences, University of Porto, Portugal;
(3) FCUP- Faculty of Sciences of the University of Porto, Portugal

Pinnipeds, a clade of aquatic mammals, are endangered due to anthropogenic pressures (e.g. water pollution)[1]. Many chemical compounds have been identified as concerning pollutants, or xenobiotics, in aquatic systems leading to several negative effects on marine mammals' health (e.g. impaired immune responses and reproduction). Pinnipeds, as one of the top predators in the trophic chain, are exposed to higher concentrations of these chemical substances1. Xenobiotic metabolism is a physiological process that results in the biotransformation of chemicals to facilitate their excretion from the organism. Pregnane X Receptor (PXR) is a nuclear receptor responsible for the transcriptional regulation of genes encoding enzymes involved in this process[2]. In this study, we investigate the PXR gene in pinnipeds. Results show high degree of conservation of pinniped PXR when compared to other mammals', apart from one seal, *Neomonachus schauinslandi*, which presents a frameshift mutation in the C-terminal region of PXR. Despite being protected today, this species experienced extensive hunting in the past. This likely contributed to its low genetic diversity, serving as a possible explanation as to why this mutation is present in all available genomes of this species[3]. This work aims to investigate N. schauinslandi PXR and assess the impact of this mutation in the binding of several known ligands and pollutants e.g: Clotrimazole, Estradiol, Rifampicin and Bisphenol A-BPA using comparative homology modelling and molecular docking tools. Docking results show that the amino acid residues responsible for xenobiotic binding in the PXR of *N. schauinslandi* are different when compared to *Homo sapiens*. This study will contribute with new information regarding the susceptibility of pinnipeds to xenobiotics and offer insights into how genetic bottlenecks increase conservation risks for endangered species.

Acknowledgements: This work was supported by Portuguese Foundation for Science and Technology (projects UIDB/ 04423/2020). FCT supports MLM and RR under the Scientific Employment Stimulus program MLM-2022.00397.CEECIND/CP1728/CT0006 and RR-2022.03697.CEECIND

[1] Desforges, J.-P. W., Sonne, C., Levin, M., Siebert, U., Sylvain De Guise, & Dietz, R. (2016). Immunotoxic effects of environmental pollutants in marine mammals. Environment International, 86, 126–139. https://doi.org/10.1016/j.envint.2015.10.007

[2] Timsit, Y. E., & Negishi, M. (2007). CAR and PXR: the xenobiotic-sensing receptors. Steroids, 72(3), 231–246. https://doi.org/10.1016/j.steroids.2006.12.006

[3] Schultz, J. K., Marshall, A. J., & Pfunder, M. (2010). Genome-Wide Loss of Diversity in the Critically Endangered Hawaiian Monk Seal. Diversity, 2(6), 863–880. https://doi.org/10.3390/d2060863

Poster 4

Thermal Evolution and Plasticity: Insights from Gene Expression in *Drosophila subobscura*

Antunes, MA [1,2]; Santos, MA [1,2]; Quina, AS [2,3]; Santos, M [1,4]; Matos, M [1,2]; Simões, P [1,2]

(1) CE3C - Centre for Ecology, Evolution and Environmental Changes and CHANGE - Global Change and Sustainability Institute, Lisbon, Portugal;
(2) Departamento de Biologia Animal, Faculdade de Ciências, University of Lisbon, Lisbon, Portugal;
(3) CiiEM - Egas Moniz Center for Interdisciplinary Research, Egas Moniz School of Health and Science, Caparica, Portugal;
(4) Departament de Genètica i de Microbiologia, Universitat Autònoma de Barcelona, Bellaterra, Spain

Understanding how organisms adapt to new thermal conditions at the molecular level is crucial to predict the impacts of global warming. However, there is still a lack of research on the effects of rising temperatures over time and of studies involving different populations from the same species. This study addresses these gaps, investigating the impact of global warming on the gene expression patterns of *Drosophila subobscura* populations from two different latitudinal locations after 23 generations of evolution.

Our results indicate that evolutionary changes depend on the genetic background of the populations, with different starting points for thermal evolution, and that high-latitude populations show more pronounced evolutionary changes, with some evidence of convergence towards low-latitude populations. The study highlights an interplay between plasticity and selection, with the high-latitude population showing fewer initial plastic genes and lower levels of adaptive plasticity, but a greater magnitude of change in both plastic and selective responses during evolution under warming conditions compared with its low-latitude counterpart.

A substantial proportion of the transcriptome evolved, despite the lack of observable response at higher-order phenotypic traits. The interplay between plasticity and selection may prove to be an essential component in shaping species' evolutionary responses to climate change. Furthermore, the value of conducting studies on multiple populations of the same species is emphasized, given the identification of differences between populations with different backgrounds in several contexts.

Poster 5

Different genetic patterns of introgression across multiple hybrid zones of *Littorina saxatilis* in the Iberia Peninsula

Balmori-De-La-Puente, A [1-3]; De Jode, A [4-6]; Galindo, J [7]; Rólan-Alvares, E [7]; Westram, A [9]; Johannesson, K [4]; Butlin, R [4,10]; Faria, R [1,2,11]

(1) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório As-sociado, Campus de Vairão, Universidade do Porto, Vairão, Portugal;
(2) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Portugal;
(3) University of Salamanca;
(4) Department of Marine Sciences-Tjärnö, University of Gothenburg, Strömstad, Sweden;
(5) Department of Biological Sciences, University of Alabama, Tuscaloosa, Alabama USA;
(6) Dauphin Island Sea Lab, Dauphin Island, Alabama, USA;
(7) Centro de Investigación Mariña (CIM-UVIGO), Universidad de Vigo, Vigo Spain;
(8) Faculty of Biosciences and Aquaculture, Nord University, Bodø, Norway;
(10) Ecology and Evolutionary Biology, School of Biosciences, The University of Sheffield, Sheffield, UK;
(11) Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Porto, Portugal

The rough periwinkle (*Littorina saxatilis*) shows a high phenotypic variability (e.g., ‘wave’ and ‘crab’ ecotypes), in part resulting from parallel divergence associated to similar environmental pressures in the intertidal across multiple locations in European Atlantic shores. Populations in the Iberian Peninsula, in particular, present distinct genetic and phenotypic diversity, which may be key for adapting to varying environmental conditions and diversify at the southernmost range of the species. Recent studies based on whole-genome sequencing (WGS) and phenotypic data revealed i) stronger reproduction isolation between Iberian ecotypes (when compared with Sweden), consistent with late stages of speciation; and that ii) many of the 20 chromosomal inversions detected de novo in the northernmost range are also present in the Iberian Peninsula. However, this is largely based on the analysis of a single location. Thus, we still need to extract the full potential from the genomic information gathered across Iberian *L. saxatilis* populations to reconstruct the history of the genetic variation underlying ecotype divergence and speciation in this (and other) areas of the species distribution range. In order to reach this goal, a SNP genotyping assay consisting of ~200 SNPs divided in three different categories (i- diagnostic (or nearly) between ecotypes; ii) linked to inversions; iii) randomly selected from collinear regions) was designed based on WGS data from a single Spanish site. The analysis of for SNP set across samples collected across three transects from low (Wave) to high (Crab) shore in Iberia Peninsula revealed: i) overall strong reproductive isolation across transects; ii) individuals with phenotypic and genetic ancestry consistent with recent hybridization (despite rare) and pst introgression but not F1s; iii) variable patterns of introgression across hybrid zones, suggesting and environmental influence on the observed patterns.

Poster 6

Evolutionary dynamics of immune gene families in Lagomorphs: A comparative genomics approach

Borges, JR [1,2,3]; Esteves, PJ [1,2,3]; Melo-Ferreira, J [1,2,3]; Marques, JP [1,3]

(1) CIBIO-InBIO, Research Center in Biodiversity and Genetic Resources, University of Porto, Vairão, Portugal;
(2) Department of Biology, Faculty of Sciences, University of Porto, Porto, Portugal;
(3) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal.

The diversification of immune system gene families in response to selective pressures has been essential to the adaptive response of organisms to diseases throughout evolution. In particular, multigene families evolve through a dynamic process of birth-and-death, characterized by frequent duplications, deletions, pseudogenizations and neofunctionalization events. In Lagomorphs, the immune system is known to have played an important role driving organismal diversification, indicative of unique evolutionary responses to selective pressures. This order, which includes pikas, rabbits and hares, represents 25-30 million years of evolutionary history. Research on Lagomorph immunity has so far been predominantly focused on the European rabbit (*Oryctolagus cuniculus*). However, the recent increase in available chromosome-level reference genomes across Lagomorph species, has opened novel opportunities for comparative genomics studies, enabling detailed comparisons of genomes and gene structures.

This work is part of a master thesis project, in which we plan to apply a two-stage comparative genomics approach to characterize the evolution of genome structures and immune gene families in Lagomorphs. The first stage will involve comparing the genome structure of different genus and species, using synteny analyses and inferences of phylogenetic orthology, revealing insights into structural rearrangements and genomic architectures. Tools such as SyMAP are currently being used for alignment and synteny analysis, with future plans to incorporate software like SVbyEye for more thorough analysis. In the second stage, we will focus on cataloguing immune system multigene families, using synteny analysis, phylogenetic gene trees and measures of genetic distances.

This two-stage approach is expected to offer valuable insights into the evolution of genome structure and immune system adaptation in Lagomorphs. Preliminary results suggest structural variation among genus in Lagomorph, which we plan to further investigate in the coming months. Furthermore, it may uncover correlations between genetic factors and disease susceptibility or resistance, offering potential applications in disease prevention and management.

Poster 7

Contribution to the Dairy Cattle Resistome

Coimbra, S [1,2,4]; Beja-Pereira, A [1,2,3]; Pérez-Pardal, L [1,2,4]

(1) Centro de Investigação em Biodiversidade e Recursos Genéticos (CIBIO), InBIO Laboratório Associado, Universidade do Porto, Campus de Vairão;
(2) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, Campus de Vairão, Vairão;
(3) DGAOT, Faculdade de Ciências, Universidade do Porto, Porto;
(4) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto.

The dairy holobiome refers to the community of microorganisms present in the ecosystem of a dairy farm, incorporating the animal, cohabiting human, and environmental microbiomes. As part of the MICRODAI project, this study aims to characterize the farm holobiome ecosystem and assess its potential as a reservoir for antibiotic resistance genes. This knowledge can serve as a biosafety indicator, enhancing the well-being of animals on dairy farms and improving One health. Fecal samples were collected from 18 dairy farms in the northern region of Portugal, selected based on biosafety surveys and with equitable representation of small, medium, and large farms. Total DNA was extracted from each sample and sequenced using long-read Nanopore technology, employing the PromethION equipment. Bioinformatic analysis identified 107 taxa, with the most abundant phyla being Bacillota, Pseudomonadota, and Cyanobacteriota. The predominant species included *Clostridium botulinum*, *Escherichia coli*, and *Pelosinus fermentans*. *C. botulinum* is a pathogenic bacterium known to cause botulism. It is also a typical inhabitant of the intestinal tract of many healthy horses, cattle, and poultry. Furthermore, we identified the TetQ and TetW genes, which confer resistance to tetracycline, a commonly used antibiotic in veterinary medicine to treat hoof dermatitis in dairy cattle. These findings indicate the dairy farm environment is a significant reservoir for antibiotic resistance, highlighting the need for improved biosecurity measures to mitigate public health risks linked to the spread of antibiotic resistance genes. Although these results are still preliminary, they demonstrate the potential of third-generation sequencing to identify bacterial genomes, including genomic regions associated with resistance to antibiotics, disinfectants, and detergents.

Poster 8

Distribution of Alleles Linked to Pyrethroid and Amitraz Resistance in *Varroa destructor* across Portugal

Costa, M [1]; Yadró, CA [1]; Lopes, AR [1]; Quaresma, A [1]; Rodrigues, C [1]; Pinto, MA [1]; Henriques, D[1]

(1) CIMO, LA SusTEC, Instituto Politécnico de Bragança, Campus de Santa Apolónia, 5300-253 Bragança, Portugal

Varroa destructor, an ectoparasitic mite of honey bees (*Apis mellifera*), is a significant threat to apiculture by causing varroosis and transmitting dangerous viruses such as the deformed wings virus. This mite can be controlled by the use of synthetic or organic acaricides. Currently, in Portugal, two classes of synthetic compounds are used: pyrethroids (fluvalinate and flumethrin), and formamidines (amitraz). However, excessive and repeated use of acaricides has led to resistance in mite populations. Resistance to pyrethroids is primarily associated with mutations in the voltage-gated sodium channel gene, and involves an amino acid change at position 925, where a leucine (L) is found in the wild type. At this position, three alleles have been described that confer Varroa resistance to pyrethroids, where leucine is replaced by valine (L925V), isoleucine (L925I), or methionine (L925M). A novel mutation at position 918 was described in a population in the Valencian Community in Spain, where the amino acid methionine, generally found at position 918, was replaced by leucine (M918L). For amitraz, resistance is linked to mutations in the Octβ2R gene, involving a substitution of asparagine by serine at position 87 (N87S), found in France, and a substitution of tyrosine by histidine at position 215 (Y215H), found in the USA. However, the distribution of these resistance-associated alleles remains unexplored in Portugal. To address this gap, we sampled Varroa mites from honey bee colonies from different locations in Portugal, and used a PCR assay with mutation-specific primers, followed by Sanger sequencing of the amplicons. The results confirmed pyrethroid resistance alleles at positions 925 and 918 in the Portuguese populations. In contrast, amitraz-resistance alleles remained undetected, although this result should be interpreted with caution as the sample size was reduced. These findings represent a step forward for understanding the status of Varroa resistance in Portugal, providing baseline data for the development of more effective, region-specific management strategies. Further research with larger sample sizes is essential to confirm these preliminary observations.

Acknowledgements: This work was supported by the project "MITE- Varroa e vírus transmitidos: MonItorização de muTações e dEsenvolvimento de ferramentas moleculares inovadoras"funded by National Beekeeping Programme 2023-2027. FCT/MCTES (PIDDAC): CIMO, UIDB/00690/2020 (DOI: 10.54499/UIDB/00690/2020) and UIDP/00690/2020 (DOI: 10.54499/UIDP/00690/2020); and SusTEC, LA/P/0007/2020 (DOI: 10.54499/LA/P/0007/2020).

Poster 9

Evolving tensions: The role of stressful environments in shaping sexual conflict in a haplodiploid species

Costa, SG [1]; Mira, AF [1,2,3]; Alpedrinha, J [1]; Santos, I [1]; Eira, C [1]; Magalhães, S [1,4]; Rodrigues, LR[1]

(1) CE3C - centre for ecology, evolution and environmental changes, FCUL University of Lisbon, Lisbon, Portugal;
(2) Department of Biogeography and Global Change, Museo Nacional de Ciencias Naturales, Madrid, Spain;
(3) Department of Biology, Universidad Autónoma de Madrid, Madrid, Spain;
(4) Wissenschaftskolleg zu Berlin, Institute for Advanced Study, Berlin, Germany.

Adaptation to new environments may reshape ecological interactions within populations. One such interaction is sexual conflict, resulting from different optima in reproductive traits of males and females. Here, we addressed how evolving in novel environments affects sexual conflict on the haplodiploid spider mite *Tetranychus urticae*. We used populations that evolved for 23 generations in different intensities of two stressful environments: cadmium, a non-essential heavy metal that accumulates in tomato plant shoots and is highly toxic to spider mites, and interspecific competition. After 2 generations of common garden to equalize environmental effects, males and females from each regime were crossed with each other or with individuals from the base population, which allows disentangling the response of each sex and hence detect sexual conflict. These crosses were done under non-stressful conditions and measures of fecundity, offspring sex ratio, and female survival were taken. Results showed no effect of evolving in the presence of cadmium or competitors on fecundity and female survival, indicating no sexual conflict expressed via these traits. However, females evolving under high cadmium concentration produced a higher proportion of sons when mating with males from the base population than with their evolved counterpart. Males from the same selection regime, on the other hand, sired a similar proportion of sons regardless of the evolutionary status of the female they mated with. An increase in the proportion of sons might allow females to increase genetic diversity in the offspring, which would be beneficial under the low population densities typical of the environments with high cadmium concentration. Males in haplodiploid systems benefit solely from the production of daughters, the diploid sex, meaning male and female sex allocation optima differ, giving rise to sexual conflict over sex-ratio. The fact that females only successfully manipulated the offspring sex ratio when paired with non-evolved males suggests that males are ahead of the arms race. All in all, these findings contribute to a deeper understanding of the interplay between natural and sexual selection, highlighting how stressful environments influence reproductive strategies and conflict between sexes.

Poster 10

Historical human practices have shaped the genomic diversity and structure of Iberian livestock dogs

Coutinho-Lima, D [1,2,3]; Doadrio, I [4]; Godinho, R [1,2,3]

(1) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO, Laboratório Associado, Campus de Vairão, Universidade do Porto, Vairão, Portugal ;
(2) Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Porto, Portugal;
(3) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal;
(4) Museo Nacional de Ciencias Naturales, Consejo Superior de Investigaciones Científicas, Madrid, Spain

In Iberia, where livestock practices have long been essential to the people's livelihood, genetic diversity and relationships between native livestock guarding and herding dogs remain poorly understood. We genotyped over 300 Iberian dogs to address this gap using 39 autosomal, 6 Y-chromosome microsatellite markers, and over 170k SNPs. Our findings reveal high levels of genetic diversity, without substantial differences between the two dog types. This likely reflects the long history of pastoralism in Iberia and the absence of modern breed management in working dogs. We also identified pervasive signals of historical gene flow and a clear absence of genomic differentiation between functionally distinct breeds. We attribute this to human and livestock practices, particularly transhumance, a powerful driver of gene flow in the past. Overall, this work highlights the complex interplay between livestock practices and breeding dynamics in shaping functionally distinct dogs.

Poster 11

A comparative test of social signalling with barred and spotted plumage in estrildid finches

Pacheco, JR [1,2]; Cardoso, GC [1,2]; Gomes, ACR [1,2,3]

(1) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Campus de Vairão, Universidade do Porto, Vairão, Portugal;
(2) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Vairão, Portugal;
(3) Department of Zoology, Stockholm University, Stockholm, Sweden

Animal pigmentation patterns can provide camouflage, but some patterns may also have a signalling function. Barred and spotted plumage are common in birds, and were suggested to serve a dual function: camouflage and signalling. We used the bird family Estrildidae to test whether barred and spotted plumage evolved associated with ecological traits that influence the need for camouflage, visual communication or both, while controlling for phylogenetic relatedness. For this, we examined the relation between the extent of barred and spotted plumage, separately for males and females, with traits associated with sexual selection or the need for camouflage. We also tested for correlated evolution between the two patterns and, for each pattern, between the sexes. The extent of barred plumage tended to be associated with social and ecological traits suggesting either a need for camouflage or less sexual or social signalling, whereas spotted plumage was linked to traits indicative of signalling functions. Barred and spotted plumage appear to have evolved independently of each other. We found support for correlated evolution of each pattern between the sexes. Our findings suggest that the evolution of barred and spotted plumage may have partly different functions, but we do not rule out a possible dual function of barred plumage, as previously reported in the literature.

Poster 12

Variable mutation rates in lupins

Fazalova, V [1]; Nevado, B [1]

(1) CE3C - centre for ecology, evolution and environmental changes, FCUL University of Lisbon, Lisbon, Portugal

There is growing evidence that mutation rates vary among different taxa, yet how mutation rates differ among closely related species remains unclear. In this work, we performed mutation rate estimation for two species from the plant legume genus Lupinus.

We carried out single-generation mutation accumulation experiments for two annual Lupinus species: *L. mutabilis*, a domesticated species from the Andean mountains; and *L. bicolor*, a wild species from North America. For each of the two species, we performed self-fertilisation of a single parental individual to obtain F1 seeds, and re-sequenced six complete genomes: the parental individual and five of its offspring. We employed stringent filters and performed pairwise parent-offspring comparisons to identify candidate de novo mutations.

Our preliminary results show that mutation rate in *L. mutabilis* is about five times higher compared to *L. bicolor*. The difference in mutation rates between these species could be due to relaxed selection during domestication of *L. mutabilis*, which could have resulted in loss or degeneration of genes involved in DNA replication and DNA repair mechanisms. Alternatively, high mutation rates may be a feature of the Andean clade of Lupinus, an extremely species-rich and fast diversifying clade with 130+ species that emerged over the past 2 Myr. Moreover, we found that in *Lupinus mutabilis* about one quarter of mutations are present in more than one offspring, suggesting that these are heritable somatic mutations. This result is surprising as most studies thus far have identified heritable somatic mutations in long-lived trees, not annual herbaceous species. Sanger validation of the candidate mutations and other analyses focused on understanding the variation in the mutation rate in Lupinus are ongoing.

Poster 13

Genetic architecture of pathogen avoidance

Ferreira, G [1]; Vilas Boas, J [1]; Picão-Osório, J [1]

(1) CE3C - Centre for Ecology, Evolution and Environmental Changes, Department of Animal Biology, Faculty of Sciences, University of Lisbon, Lisbon, Portugal

Many organisms fight infectious diseases through three main mechanisms: avoidance, resistance and tolerance. While the latter two usually work after contact with a pathogen, the avoidance strategy tries to prevent this contact, saving energy and resources that would otherwise be spent fighting it. Although, this behavioural avoidance to pathogens is a widespread strategy for survival, its wild genetic basis remains largely unknown.

In this project, we will investigate the natural genetic basis of behavioural pathogen avoidance to ecologically-associated bacteria. For this, we will use a resource of whole-genome sequenced wild isolates of the nematode *Caenorhabditis elegans* (CaeNDR, the Caenorhabditis Natural Diversity Resource) together with a collection of naturally associated bacteria (CeMbio, the *C. elegans* Microbiome Resource). *C. elegans* serves as an excellent model for studying this behaviour due to its simplicity, the immense biological tools available, and extensive knowledge on the genetics of pathogen avoidance in the laboratory-adapted reference strain N2. Furthermore, this nematode's bactivorous nature makes this behaviour ecologically relevant: it needs to distinguish between bacteria that are pathogenic and that are food. Previous studies have found wild genetic variation for pathogen avoidance behaviour, but the specific loci involved in this response are elusive.

We have three main aims to uncover the genetic architecture of wild *C. elegans* variation for avoidance behaviour. First, we will phenotype about 100 wild isolates of *C. elegans* for the avoidance behaviour to 10 ecologically associated bacteria. Second, we will associate the phenotypic variation of the wild isolates to their genotypic diversity through genome-wide association study (GWAS). Third, we will validate the significant quantitative trait loci of the GWAS by linkage-mapping introgression. To assess the genetic architecture of such a complex trait, we will study it by partitioning it into sub-traits that will be analysed in different assays, namely: the ability to detect volatiles emitted by the bacteria, chemotaxis assay; their ability to process the bacteria as a bad food source, bacterial lawn avoidance assay; and a fitness assay to find out how pathogenic the bacteria are, fitness assay.

Currently, we are conducting a pilot test to select the set of 10 bacteria that we will use. For this, we are conducting fitness assays in the 34 bacteria that we have at our disposal. We will test two strains of *C. elegans*, the lab adapted N2 and the highly divergent wild isolate XZ1516. Also, we will choose a group of bacteria that represents a gradient of pathogenicity, since avoidance behaviour is expected to be correlated with how detrimental the bacteria is to the worms.

In summary, we expect to uncover the wild genetic architecture of pathogen avoidance behaviour in *C. elegans* using a quantitative genetics approach. Our main goal is to identify candidate genes that will serve as a starting point for future research aiming to understand how complex behaviours are genetically and neuronally encoded.

Poster 14

Population genetics of Argentine pumas (*Puma concolor*) suggest a broader scale for management and conservation strategies

Gallo, O [1,2,3]; Godinho, R [1,2,3]; Casanave, EB [4]; Castillo, DF [4]

(1) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, InBio Laboratório Associado, Campus de Vairão, Universidade do Porto, Vairão, Portugal;
(2) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto, Portugal;
(3) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Vairão, Portugal;
(4) GENCON - Laboratorio de Genética para la Conservación, Instituto de Ciencias Biológicas y Biomédicas del Sur, Dpto. de Biología, Bioquímica y Farmacia, Universidad Nacional del Sur, Bahía Blanca, Argentina

The knowledge of population genetics (e.g. genetic structure and connectivity patterns) is essential for planning conservation management strategies aiming to long-term persistence of populations and species, particularly when those patterns are human induced. Large carnivores' conservation is threatened by depleted levels of gene flow due to habitat fragmentation and human-related mortality, which affect functional connectivity by influencing dispersal behavior and population density. *Puma concolor* has suffered a reduction in its distribution range mainly due to deforestation, habitat fragmentation, and direct persecution. Despite being protected by national law, pumas in Argentina are facing high hunting pressure due to the conflict, with individual-rewarded legal harvesting plans implemented by Patagonian provincial governments as a preventive population control to mitigate livestock loss. In this work, we perform the first comprehensive assessment of puma genetics in Argentina, associating genetic diversity and structure, gene flow and demographic patterns to landscape connectivity and management plans. By genotyping 199 pumas at 25 nuclear loci, we show the occurrence of four genetically distinct populations, exhibiting similar diversity (Ho range: 0.607-0.618, He range: 0.697-0.738) but considerably variable effective population size (Ne range: 35.2-94.7). Puma populations were genetically depauperated, showing signs of bottlenecks, and had minimum effective population sizes below the suggested threshold to prevent genetic diversity loss from inbreeding. Pumas from eastern central-north Argentina formed the largest source population (net migration rate = +0.291), while western pumas showed evidence of being the main sink (-0.210). Southernmost and central-eastern provinces provided for most of the migrants, comprehending the management units with lowest diversity and immigration rates, confirmed by their highest isolation. This study indicates that genetic structure and gene flow are likely associated with landscape connectivity and hunting pressure. Legal harvesting seems to be driving gene flow particularly among Patagonian management units, where the largest sink appears to be attractive for dispersers. Our results start to fill the lack of genetic data for this top predator in Argentina, providing baseline information for the conservation of the species, as well as highlighting the importance of taking into account the population-level and connectivity pattern for planning management strategies at a broader scale (i.e. inter-provincial).

Poster 15

The evolutionary history of the yellow coat colour in the Iberian wolf

Garcês, C [1,2,3]; Lobo, D [1,3]; Godinho, R [1,2,3]

(1) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, InBio Laboratório Associado, Campus de Vairão, Universidade do Porto, Vairão, Portugal;
(2) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto, Portugal;
(3) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Vairão, Portugal;

In Galicia (NW Spain), an atypical uniform yellow coat colour has been observed in Iberian wolves (*Canis lupus signatus*). Individuals with the yellow phenotype have been observed living in packs alongside wildtype individuals, but the genetic basis of this atypical coat colour had not yet been determined. In the domestic dog, the yellow coat colour is regulated by structural variation at the ventral and hair-cycle promoters of the ASIP gene (VP1-HCP1), responsible for regulating the temporal and spatial distribution of pigments. Given that hybridization between wolves and dogs has been reported in the area, we hypothesized whether this colour phenotype in Iberian wolves could result from the introgression of dog ASIP promoters. To address this, we designed a target enrichment approach to capture a 64-kb region encompassing ASIP and its 5' promoters in the Iberian wolf genome and applied it to high- and low-quality DNA samples known to carry the dog yellow genotype. We found that the yellow coat colour in Iberian wolves is regulated by the same VP1-HCP1 structural variants described in yellow dogs. Phylogenetic reconstruction and haplotype analysis indicate that this haplotype in Iberian wolves was acquired through introgression from dogs in a likely single hybridization event.

Poster 16

Adapt, adjust or move: can the spider mites *Tetranychus cinnabarinus* do it all in response to heat?

Gaspar, R [1]; Costa, S [1]; Nunes, S [1]; Magalhães, S [1,2]; Rodrigues, L [1]

(1) cE3c-Centre for Ecology, Evolution and Environmental Changes & CHANGE-Global Change and Sustainability Institute, Faculdade de Ciências, Universidade de Lisboa, 1749-016 Lisbon, Portugal;
(2) Wissenschaftkolleg zu Berlin, Wallotstraße 19, 14193 Berlin, Deutschland.

In recent years, organisms have been exposed to increasingly frequent heat waves and higher long-lasting local temperatures, conditions that can ultimately lead to extinction. However, several strategies may allow individuals to cope with heat: they can adjust their response plastically, adapt to these new conditions or move to cooler places. Importantly, these are non-exclusive hypotheses, and they can affect each other, which justifies a transversal assessment of all strategies in the same model system. I will do such an assessment using the carmine spider mite, *Tetranychus cinnabarinus*, a cosmopolitan pest that infests various important crops.

To assess if there is a plastic response to high temperatures, I will measure the body size and fertility of individuals exposed to 25°C, 33°C and 36°C. As herbivores, spider mites may respond to the effect of temperature both directly and indirectly, via the response of their host plants to heat. Hence, I will also compare leaf consumption by mites and plant stress at different high temperatures on both bean and tomato. Next, to determine if temperature promotes movement to cooler places, I will measure the degree of dispersal of individuals under control and high temperatures. Finally, I will assess the spider mites' evolutionary response to heat and determine whether their adaptation is contingent upon the simultaneous exposure to a novel host plant. This will be done by exposing *T. cinnabarinus* populations for ca. 10 generations to heat and/or new host plant (populations reared on bean will evolve on tomato). Importantly, the plasticity and dispersal capacity of the evolved individuals will be among the traits assessed by the end of the experimental evolution.

With this work, I hope to shed light on the ability of a crop pest to respond to the harsh conditions predicted under global warming, an important piece of information when considering what to expect and which management strategies to employ in future agricultural fields.

Poster 17

Temperature-adaptative phenotypic plasticity – stressful temperatures induce different transcriptomic profiles in the genomically homogeneous worm pipefish

Gomes, LP [1]; Roth, O [2]; Monteiro, N [1,3]

(1) BIOPOLIS-CIBIO - Biopolis Program in Genomics, Biodiversity and Land Planning, Research Center in Biodiversity and Genetic Resources (CIBIO), and CIBIO, InBio Associate Laboratory, University of Porto, Vairão, Portugal;
(2) MarEvol - Marine Evolutionary Biology, Zoological Institute, Kiel University, Kiel, Germany;
(3) IUCN SSC Seahorse, Pipefish and Seadragon Specialist Group, Vancouver, Canada.

In the worm pipefish, *Nerophis lumbriciformis*, a species with a wide latitudinal distribution, Monteiro and colleagues (2017) revealed marked phenotypical variation along the species range. For instance, pipefish size linearly increased towards the colder North (adhering to Bergmann's rule) while size dimorphism decreased (following Rensch's rule). Also, females showed a parabolic expression of secondary sexual traits, similarly expanded at both edges of distribution and indicating higher reproductive aptitude attested by the production of more and bigger eggs. It was then hypothesized that females at the fringes of distribution experience stronger sexual selection pressure resulting from sub-optimal temperature regimes that constrain the breadth of the breeding season or the time window for gamete production. Surprisingly, as sexual selection often promotes divergence, an apparent absence of population structure was observed by Mendes et al. (2020) when using mainly mitochondrial markers. Here, we searched for signs of differentiation along the species range by employing a two-omics approach. With the information gained from genome resequencing, we first aimed to confirm the lack of population structure while also describing genomic diversity and looking for genes under positive selection. With transcriptomic information, we aimed to understand which genes were being differentially expressed between the distribution edges. Our results showed that the worm pipefish genome is highly diverse and confirmed that this pipefish seems to be indeed panmictic. Also, our selection analysis didn't return positively selected genes associated with the hypothesized drivers of differentiation (e.g., temperature and sexual selection). Nevertheless, at the transcriptomic level, the North and South distributional edges could be easily distinguished. The differential gene expression analysis suggested that this clustering was mainly influenced by the stressful temperatures experienced at the range limits. Since the worm pipefish seems to be genomically homogeneous, it seems that the most likely driver of phenotypic variation is gene expression patterns modulated by thermal stress. Thus, we tentatively suggest that, along the species' distribution, the observed morphological variation, especially that related with the expression of secondary sexual traits, results from a response to climate conditions, fully displaying the species' phenotypic plasticity (a process known to uphold mean fitness in marginal populations).

Poster 18

Intra-archipelago dispersal patterns in oceanic islands based on phylogeography of darkling beetles (Oxycara, Tenebrionidae) in Cabo Verde

Jurado-Angulo, P [1,2,3,4,5]; García-París, M [5]; Gonçalves, H [1,6]; Lopes, EP [4]; Vasconcelos, R [1,3]

(1) CIBIO-InBIO – Centro de Investigação em Biodiversidade e Recursos Genéticos - Laboratório Associado, Campus de Vairão, Universidade do Porto, Vairão, Portugal;
(2) FCUP – Faculdade de Ciências da Universidade do Porto, Departamento de Biologia, Porto, Portugal, Portugal;
(3) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal;
(4) ISECMAR-UTA – Instituto de Engenharias e Ciências do Mar - Universidade Técnica do Atlântico, Mindelo, São Vicente, Cabo Verde;
(5) MNCN-CSIC – Department of Biodiversity and Evolutionary Biology, Museo Nacional de Ciencias Naturales, Madrid, Spain;
(6) MHNC-UP – Museu de História Natural e da Ciência - Universidade do Porto, Porto, Portugal

Evolutionary radiation occurs when there is a proliferation of taxonomic diversity in a clade caused by higher rates of speciation. The colonisation of isolated regions, such as islands, can release species from the biotic pressures experienced on the mainland and provide them with novel ecological opportunities. This can enable them to diversify into a range of ecological niches that were previously unavailable. One of these radiations has occurred in Cabo Verde, specifically with a genus of tenebrionid beetles Oxycara. Although Oxycara predominantly occurs in continental Africa, Cabo Verde hosts 16 endemic species, making it an area of interest for studying the evolutionary and biogeographical dynamics of this genus within the archipelago. This study will employ both mitochondrial and nuclear markers to elucidate the phylogenetic relationships among the species and to infer patterns of colonisation. By examining the distribution of genetic diversity among species, it aims to shed light on the evolutionary processes that have shaped the diversity of Oxycara in Cabo Verde. Results may guide conservation management, as will allow to identify populations that are evolutionarily significant units to protect. In this conference, the preliminary results are presented and discussed.

Poster 19

Inducible tomato defences persist in detached leaves, but are differentially affected by plant integrity and genotype

Kozak, M [1]; Godinho, DP [1,2]; Yayla, E [1,3]; Fragata, I [1]; Magalhães, S [1,4]; Rodrigues, LR [1]

(1) CE3C - Centre for Ecology, Evolution, and Environmental Changes & CHANGE – Global Change and Sustainability Institute, Faculty of Sciences, University of Lisbon, Lisbon, Portugal;
(2) GIMM - Gulbenkian Institute for Molecular Medicine, Oeiras, Portugal;
(3) Istanbul Technical University, İstanbul, Turkey;
(4) Wissenschaftskolleg zu Berlin, Institute for Advanced Study, Berlin, Germany.

Herbivore and pathogen attacks often lead to the induction of plant defences. Given the intricate nature of such response, plant integrity is assumed to be a pre-requisite for the successful mounting of defences, a hypothesis supported by work done in plant-pathogen interactions. However, the relevance of plant integrity in plant-herbivore interactions, particularly in direct plant defences, is unclear and lacks empirical molecular validation. To test this, we measured gene expression of plant defence-related genes (PPO-D, PI-IIc and PR-1a) in intact plants or detached leaves of two tomato varieties (Castlemart and Moneymaker) infested with the herbivorous spider mite *Tetranychus urticae*. We also tested whether defences persisted in leaves after one, four, seven or fourteen days of infestation of detached leaves. We found that tomato defences were induced in both intact plants and detached leaves. Moreover, the expression of all three genes in detached leaves increased over time, with the two late-expression genes, PI-IIc and PR-1a, displaying a higher increase. However, differences in induction levels between intact plants and detached leaves varied with plant variety and the gene assessed. Our results indicate that detached leaves mount a persistent defensive response, which suggests that both intact plants and detached leaves can be used in studies addressing herbivore interactions with plant defences. However, studies aiming at identifying the role of specific genes should account for the differential, variety-dependent, role of plant integrity in their expression.

Poster 20

An unprecedented large-scale survey of honey bee mitochondrial diversity in Europe: c-lineage dominance and the need for conservation efforts

Li, F [1,2]; Costa, M [1,2]; Lopes, AR [1,2]; Gonçalves, T [1,2]; Henriques, D [1,2]; Quaresma, A [1,2]; Yadró, C [1,2]; Albo, A [3]; Blažytė-Čereškienė, L [4]; Brodschneider, R [5]; Brusbardis, V [6]; Carreck, N [7]; Charistos, L [8]; Chlebo, R [9]; Coffey, MF [10]; Dahle, B [11]; Danneels, E [12]; Dobrescu, C [13]; Dupleix-Marchal, A [14]; Filipi, J [15]; Gajda, A [16]; Gratzer, K [5]; Groeneveld, L [11]; Hatjina, F [8]; Johannesen, J [17]; Kolasa, M [18]; Körmendy-Rácz, J [19]; Kovačić, M [20]; Kristiansen, P [21]; Martikkala, M [22]; McCormack, G [23]; Martín-Hernández, R [24]; Pavlov, B [25]; Pietropaoli, M [26]; Poirot, B [3]; Radev, Z [27]; Raudmets, A [28]; René-Douarre, V [29]; Roessink, I [30]; Škerl, MIS [31]; Soland, G [32]; Titera, D [33]; van der Steen, J [34]; Varnava, A [35]; Vejsnæs, F [36]; Webster, M [37]; Fedoriak, M [38]; Zarochentseva, O [39]; de Graaf, DC [12]; Pinto, MA [1,2]

(1) CIMO - Centro de Investigação de Montanha, Instituto Politécnico de Bragança, Bragança, Portugal;
(2) SusTEC - Laboratório Associado para a Sustentabilidade e Tecnologia em Regiões de Montanha, Instituto Politécnico de Bragança, Bragança, Portugal; (3) APINOV – Centre de Recherche & Formations Apicoles, France; (4) Institute of Ecology, Nature Research Centre, Vilnius, Lithuania; (5) University of Graz, Austria; (6) Latvian Beekeepers Association, Latvia; (7) Carreck Consultancy Ltd; (8) Institute of Animal Science & Department of Apiculture, Greece; (9) Slovak University of Agriculture, Nitra, Slovakia; (10) Department of Agriculture, Food and the Marine, Ireland; (11) Norwegian Beekeepers Association, Norway; (12) University of Ghent, Belgium; (13) ROMAPIS, Romania; (14) CoActions/Alt-RD, Bègles, France; CRISES, EA 4424, Université Paul Valéry, Montpellier, France; (15) University of Zadar, Croatia; (16) Warsaw University of Life Sciences, Poland; (17) DLR-Westerwald-Osteifel, Germany; (18) Jagiellonian University, Kraków, Poland; (19)Hungarian Apitherapy Association, Hungary; (20) Faculty of Agrobiotechnical Sciences Osijek, Croatia; (21) Apinordica AB, Sweden; (22) Finnish Beekeepers Association, Finland; (23) University of Galway, Ireland; (24) Centro de Investigación Apicola y Agroambiental Marchamalo, Spain; (25) Makedonska pcela- Negorci (MacBee) - Macedonian association for preservation of Macedonian native honey bee (Apis mellifera macedonica), Gevgelija, North Macedonia; (26) Istituto Zooprofilattico Sperimentale del Lazio e della Toscana, Italy; (27) Tobacco and tobacco products institute, Markovo, Bulgaria; (28) Estonian Beekeepers Association, Estonia; (29) FEdCAN – European federation for the conservation of dark bee, France; (30) Wageningen Environmental Research, Netherlands; (31) Agricultural Institute of Slovenia, Slovenia; (32) Apigenix, Institute for Beegenetics, Twann, Switzerland; (33) Bee Research Institute at Dol, Czech Republic; (34) Alveus AB Consultancy, Netherlands; (35) Department of Agricultural Sciences, Cyprus University of Technology, Cyprus; (36) Danish Beekeepers Association, Denmark; (37) Uppsala University, Sweden; (38) Educational and Scientific Institute of Biology, Chemistry and Bioresources, Yu. Fedkovych National University of Chernivtsi, Chernivtsi, Ukraine; (39) Department of Ecology and Biomonitoring, Chernivtsi National University, Chernivtsi, Ukraine.

Europe is home to ten *Apis mellifera* subspecies, which belong to three mitochondrial lineages: the Western European (M), Eastern European (C), and African (A). However, the long-standing human-mediated movement of queens, primarily of C-lineage ancestry, has threatened the genetic integrity of many of these native subspecies through introgression and replacement. This has led to the establishment of conservation programs to recover the native lines in some European countries. The maternally-inherited mitochondrial DNA (mtDNA), particularly the highly polymorphic intergenic region tRNAleu-cox2, has been the marker of choice for assessing honey bee variation and introgression at large geographical scales. Herein, we will show the results of the tRNAleu-cox2 variation obtained from over 1200 colonies sampled across the range of the ten subspecies and covering 33 European countries. These revealed that apart from a few countries (Portugal, Spain, and Ireland) and isolated protected populations, European populations are predominantly dominated by C-lineage haplotypes, and many native subspecies exhibit a signature of C-derived introgression. In conclusion, this unprecedented survey of honey bee diversity across Europe underscores the concerning dominance of C-lineage genetic variation, highlighting the urgent need for strategic conservation efforts to preserve the native genetic diversity of *Apis mellifera*.

This work was conducted in the framework of the project Better-B, funded by the European Union, the Swiss State Secretariat for Education, Research and Innovation, and UK Research and Innovation, under the UK government's Horizon Europe funding guarantee (grant number 10068544).

Poster 21

A Multi-Omics and Primer Database for Virus Identification: Focus on HIV, Ebola, and SARS-CoV-2

Lima, AS [1,2]; Carneiro, J [3,4]; Sousa, SF [5]; Sá, VJ [1, 9]; Pratas, D [6,7,8]

(1) LabRP/CIR, School of Health, Polytechnic of Porto, Porto, Portugal;
(2) CIIMAR – Interdisciplinary Centre of Marine and Environmental Research of the University of Porto, University of Porto, Matosinhos, Portugal;
(3) Centre of Molecular and Environmental Biology (CBMA), Department of Biology, University of Minho, Campus Gualtar, 4710-057 Braga, Portugal;
(4) Institute of Science and Innovation for Bio-Sustainability (IB-S), University of Minho, 4710-057 Braga, Portugal
(5) LAQV/REQUIMTE - BioSIM, Department of Biomedicine, Faculty of Medicine, University of Porto, Porto, Portugal;
(6) IEETA/LASI, Institute of Electronics and Informatics Engineering of Aveiro, University of Aveiro;
(7) DETI, Department of Electronics, Telecommunications and Informatics, University of Aveiro;
(8) DV, Department of Virology, University of Helsinki;
(9) ALGORITMI/LASI, University of Minho, Braga, Portugal

Highly infectious viruses such as HIV, Ebola, and SARS-CoV-2 have presented ongoing challenges to global health. Consequently, the optimization of rapid detection tests, including PCR, and the identification of new therapeutic targets remain of paramount importance. The development of genomic and proteomic databases like the HIV Oligonucleotide Database (HIVoligoDB), EbolaID, and CoV2ID has facilitated the accumulation and accessibility of knowledge through comprehensive, user-friendly, open-access platforms. This study aims to update, expand, and integrate these databases into a single resource, while conducting thorough analyses of informative genomic regions with the goal of enhancing viral detection methods and treatment strategies. Complete genomic sequence variants for each virus were compiled using Geneious Prime and NCBI Virus, followed by multiple sequence alignment via MAFFT within the Galaxy platform. The extraction of primers and probes from research articles was attempted using two approaches: Large Language Models (LLMs), specifically NotebookLM and DonutAI/OpenLLaMa-7b, and a classic method combining the Python package PyMuPDF4LLM for PDF data extraction with regular expressions (RegEx) for oligonucleotide identification. Preliminary testing revealed that DonutAI/OpenLLaMa-7b had the lowest accuracy, failing to correctly identify any primers. NotebookLM achieved an accuracy of 39%, while the PyMuPDF4LLM + RegEx method attained the highest accuracy at 71%, successfully identifying 85 out of 121 primers in the test batch of articles. Due to its superior performance and execution speed, the PyMuPDF4LLM + RegEx approach was selected for further refinement. This methodology improves upon previous RegEx-based techniques by eliminating the need for PDF preprocessing and refining the capture of relevant information while minimizing non-relevant captures. Future steps include cross-validation of the extracted primers against the reference genome to eliminate primers intended for other viruses and to accurately identify the binding regions of the identified oligonucleotides. Additionally, parameters such as percentage of identical sites and pairwise identity will be calculated to determine the optimal primer pairs for PCR optimization. Further structural analysis of the collected sequences will form the foundation for 3D modelling and molecular dynamics simulations.

This research was supported by Portuguese national funds through the Foundation for Science and Technology (FCT) within the scope of UIDB/04423/2020 (CIIMAR) and UIDP/04423/2020 (CIIMAR), 10.54499/LA/P/0008/2020,10.54499/UIDP/50006/2020, UIDB/04050/2020 (UMinho CBMA - https://doi.org/10.54499/UIDB/04050/2020), 10.54499/UIDB/50006/2020 (LAVQ), UIDB/00319/2020 (ALGORITMI/LASI) and UIDB/00127/2020 (IEETA/LASI - doi.org/10.54499/UIDB/00127/2020). JC acknowledges the FCT funding for his research contract established under the transitional rule of Decree Law 57/2016, amended by Law 57/2017. D.P. is funded by national funds through FCT–Fundação para a Ciência e a Tecnologia, I.P., under the Scientific Employment Stimulus—Institutional Call—reference CEECINST/00026/2018.

Poster 22

Exploring Microalgae-Enzymes for Sustainable Plastic Biodegradation

Lourenço, D [1,2]; Pratas, D [3,4,5]; Sousa, SF [6]; Carneiro, J [7,8]

(1) FCUP - Department of Computer Science, Faculty of Sciences, University of Porto, Porto, Portugal;
(2) CIIMAR - Interdisciplinary Centre of Marine and Environmental Research, University of Porto, Porto, Portugal;
(3) IEETA/LASI - Institute of Electronics and Informatics Engineering of Aveiro, University of Aveiro, Aveiro, Portugal;
(4) DETI - Department of Electronics, Telecommunications and Informatics, University of Aveiro, Aveiro, Portugal;
(5) DV - Department of Virology, University of Helsinki, Helsinki, Finland ;
(6) LAQV/REQUIMTE BioSIM - Department of Biomedicine, Faculty of Medicine, University of Porto, Porto, Portugal;
(7) CBMA - Centre of Molecular and Environmental Biology, Department of Biology, University of Minho, Campus Gualtar, 4710-057 Braga, Portugal;
(8) IB-S - Institute of Science and Innovation for Bio-Sustainability, University of Minho, 4710-057 Braga, Portugal

Plastic pollution is a significant environmental issue affecting ecosystems globally. Conventional mechanical and chemical methods have been used for plastic degradation, but they can be harmful to the environment. Biodegradation, in contrast, offers a more sustainable alternative. Although bacterial degradation has shown promise for plastic biodegradation, research on plastic-degrading enzymes from microalgae remains limited (Liu et al., 2022). Microalgae have the potential to naturally produce plastic-degrading enzymes, such as ligninolytic enzymes and exopolysaccharides (EPS), that could contribute to plastic waste remediation (Chia et al., 2020). This study examines enzymes from microalgae that interact with different plastic polymers, improving our knowledge of algae-based plastic degradation mechanisms. We reviewed existing literature on plastic-degrading enzymes and EPS across different microalgae species, collecting data on enzymes produced by algae with potential in plastic biodegradation. This established a dataset that serves as the basis for subsequent computational analysis.

Our research identified several microalgae species producing diverse EPS and plastic-degrading enzymes. Among the enzymes identified were laccase, manganese peroxidase, and lignin peroxidase, found in species such as *Phormidium lucidum* and *Oscillatoria sp.*. These enzymes demonstrated degradation capabilities for multiple plastic types, including polyethylene (PE), low-density polyethylene (LDPE), and polyethylene terephthalate (PET).

Preliminary findings suggest that microalgae are a potential source of plastic-degrading enzymes. The compiled data on EPS composition and enzyme types provides a foundation for understanding algae-based biodegradation mechanisms. Future research will involve genomic, metagenomic, and computational studies to identify additional enzymes and explore their interactions with plastic polymers, aiming to characterize their biodegradation potential.

This research was supported by Portuguese national funds through the Foundation for Science and Technology (FCT) within the scope of UIDB/04423/2020 (CIIMAR) and UIDP/04423/2020 (CIIMAR), 10.54499/LA/P/0008/2020,10.54499/UIDP/50006/2020, JC acknowledges the FCT funding for his research contract established under the transitional rule of Decree Law 57/2016, amended by Law 57/2017. UIDB/04050/2020 (UMinho CBMA - https://doi.org/10.54499/UIDB/04050/2020) and 10.54499/UIDB/50006/2020 (LAVQV and UIDB/00127/2020 (doi.org/10.54499/UIDB/00127/2020). D.P. is funded by national funds through FCT–Fundação para a Ciência e a Tecnologia, I.P., under the Scientific Employment Stimulus—Institutional Call—reference CEECINST/00026/2018.

References: CHIA, Wen Yi et al. - Nature's fight against plastic pollution: Algae for plastic biodegradation and bioplastics production. Environmental Science and Ecotechnology. ISSN 26664984. 4:2020) 100065. doi: 10.1016/j.ese.2020.100065. LIU, Lingchen et al. - On the degradation of (micro)plastics: Degradation methods, influencing factors, environmental impacts. The Science of the Total Environment. ISSN 1879-1026. 806:Pt 3 (2022) 151312. doi: 10.1016/j.scitotenv.2021.151312.

Poster 23

Urbanization shapes dietary composition and head morphology of salamanders

Marques, AJD [1,2]; Alarcón-Ríos, L [1-4]; Mata, VA [1,2] ;Velo-Antón, G [1-3]

(1) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Universidade do Porto, Campus de Vairão, Vairão, Portugal;
(2) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Vairão, Portugal;
(3) Universidad de Vigo, ECOEVO Lab, EE Forestal, Campus Universitario A Xunqueira, Pontevedra, Spain;
(4) Departamento de Biología de Organismos y Sistemas, Universidad de Oviedo, Oviedo, Spain

Urbanization rapidly alters landscapes, creating a mosaic of habitats typified by increased impervious surface, decreasing soil humidity, and loss of canopy cover. Recurring urban environmental features are expected to reduce community richness, favouring urban-tolerant species and leading to a process of urban biotic homogenization. Changes in invertebrate community assemblages have been reported, but less is known about how this influences proximal trophic levels. Here we assessed how urbanization corresponds to variation in the diet of an amphibian species, the fire salamander (*Salamandra salamandra gallaica*), along the complex urban matrix of Porto, Portugal, which has experienced a period of rapid contemporary urban expansion. Prey richness estimates were determined through metabarcoding of fecal pellets. A complimentary examination of morphological geometries of dorsal head shapes was also included to observe whether dietary homogenization corresponds to reduced phenotypic variance. Results suggest impervious surface corresponds to a decline in annelid prey richness, as well as an increase in the prevalence of isopods. Diet and morphological characteristics indicate a convergence among urbanized sites with regard to predator niche and head shape morphology.

Poster 24

Understanding the evolutionary history of inversions and their role in speciation and adaptation across the Littorina marine snails genus

Barry, P [1,2]; Choo, LQ [3]; Le Moan, A [4,5]; Reeve, J [5]; Stankowski, S [6]; Westram, AM [7]; Johannesson K [5]; Butlin, RK [3,5]; Faria, R [1,2,8]

(1) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório As-sociado, Campus de Vairão, Universidade do Porto, Vairão, Portugal;
(2) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Portugal;
(3) Ecology and Evolutionary Biology, School of Biosciences, The University of Sheffield, Sheffield, UK;
(4) Adaptation et Diversité en Milieu Marin, Station Biologique de Roscoff, SorbonneUniversité, Roscoff, France;
(5) Department of Marine Sciences-Tjärnö, University of Gothenburg, Strömstad, Sweden;
(6) Department of Ecology and Evolution, University of Sussex, Brighton, UK;
(7) Faculty of Biosciences and Aquaculture, Nord University, Bodø, Norway;
(8) Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Porto, Portugal

How reproductive isolation barriers accumulate between nascent species remains a long-standing question in speciation research. Recently, chromosomal inversions, have been put forward as good candidates to generate strong barriers to gene flow. However, little is known about the time when inversions became established, and whether they can be re-used multiple times in the build-up of reproductive isolation in separate lineages. Here, we investigate the evolutionary history of inversions and their impact on speciation between ecotypes of different species of Littorina marine snails. We gathered 166 whole-genome sequences from 8 different species of Littorina and, when present, different ecotypes. We first confirmed the presence of multiple previously described inversions separating species' ecotypes, some of these inverted regions overlapping on the same genomic positions. Preliminary results suggest an old and complex history of these inversions shaped by selection, gene flux and geographical structure. We argue that this is a powerful system to improve our understanding about the evolutionary history of chromosomal inversions and their role in the evolution of barriers to gene flow in multiple closely related species.

Poster 25

Determination of barriers to gene flow between Galician *Littorina saxatilis* ecotypes

Martinez-Mariño, V [1]; Vidal-Capon, A [1]; Carvajal-Rodriguez, A [1]; Galindo, J [1]

(1) CIM - Centro de Investigaciones Mariñas, University of Vigo/Department of Biochemistry, Genetics and Immunology, Vigo, Spain

Genomic advances have contributed to the understanding of barriers to inter- and intra-species gene flow, improving our insight regarding the speciation process. The transition from such partial or weak barriers to gene flow between two groups to complete barriers contributes to the inevitable split into two new species. These barriers could be prezygotic, preventing fertilisation, or postzygotic, preventing the individual from successfully developing until its reproductive prime. A potential case can be observed in the marine gastropod *Littorina saxatilis*, which shows two ecotypes in NW Iberian Peninsula: RB, adapted to a habitat whose main selective pressure is predation by crabs, and SU, native to habitats whose main selective pressure is wave action. Both ecotypes overlap in a contact zone where restricted gene flow occurs. This PhD project will focus on identifying possible pre- and postzygotic barriers in this species, under the hypothesis that both positive associative mating and genomic architecture (possibly linked to inversions) constitute a crucial element in mate choice and the establishment of reproductive barriers. For prezygotic barriers, copulations in natural populations will be analysed as an indication of sexual selection in situ. Additionally, these analyses will be extended by laboratory experiments, including the study of the mucus tracking behaviour of males within different ecotypes. Finally, correlations between mate selection behaviour and genetic background will be sought to determine the potential genetic basis of these barriers. In the other hand, postzygotic barriers will be assessed according to potential differences in the physiological response against abiotic factors, such as heating, between hybrid individuals and the parental ecotypes. Furthermore, the reproductive patterns of the hybrids will be monitored in situ, by collecting copulations involving at least one hybrid. Also, the pattern of mate choice between pure ecotypes and the F1 generation resulting from a targeted cross among RB and SU ecotypes bred from immaturity will be analysed. Individuals found in hybrid matings will be sequenced. Lastly, hybrids and parents will be obtained from different areas of the intertidal and the females will be dissected to characterise the embryos. Virgin females will be maintained in aquariums where test crosses will be performed. Once embryos appear, these females will be dissected for embryo description. Both viable and aborted embryos will be sequenced by pool-seq to explore genetic differences.

Poster 26

Comparing demographic trajectories of Iberian carnivores and scavenges using a population genomics approach

Nogueira, T [1]; Lobo, D [2,3]; Godinho, R [1,2,3]

(1) FCUP - Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Porto, Portugal;
(2) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, Campus de Vairão, Universidade do Porto, Vairão, Portugal;
(3) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Vairão, Portugal;

During the Pleistocene transition to the Holocene, the Iberian Peninsula suffered climatic changes that affected many species harboured during the glacial era. Additionally, anthropogenic pressures are drastically affecting animal populations by destroying their habitat and reducing prey availability. In this context, large carnivores are especially vulnerable due to their specific ecological requirements. In the Iberian Peninsula, two endemic large carnivores, the Iberian wolf (*Canis lupus signatus*) and the Cantabrian brown bear (*Ursus arctos*) have experienced severe demographic decreases in the last millennia. On the other hand, the generalist red fox (*Vulpes vulpes*) has a widespread distribution throughout the Iberian Peninsula and seems less vulnerable to humaninduced changes. Furthermore, the Eurasian griffon vulture (*Gyps fulvus*) and the Cinereous vulture (*Aegypius monachus*) are two scavenger species distributed across the Iberian Peninsula that have historically depended directly on the feeding habits of carnivorous species to survive. However, the past demographic histories of these species, as well as the drivers behind their current status, remain vastly unknown. By employing a comparative genomic approach using at least five high coverage genomes of each species, this project aims to reconstruct the demographic trends of these five species over the past 30,000 years. We will dedicate particularly attention to recent times (last 10,000 years), to unveil the effects induced by human activities on species' distribution. Furthermore, we will test for convergent demographic trends among our five case studies and correlate with specific climate, ecological, and human-related events. The results of this study are expected to provide important information regarding the impact of natural and anthropogenic factors on the distribution of carnivores in the Iberian Peninsula, and also on the changes in our ecosystems given that these animals play a crucial role in trophic chains.

Poster 27

Predictors of intraspecific diversity in herptiles using machine learning

Paúl, MJ [1,2,3]; Moreira, M [1,3]; Liz, AV [1,2,3]; Decker, SK [4]; Moritz, C [5]; Carstens, B [4]; Carvalho, SB [1,3]

(1) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Universidade do Porto, Vairão, Portugal;
(2) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto, Portugal;
(3) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal;
(4) Department of Evolution, Ecology and Organismal Biology, The Ohio State University, Columbus OH, USA;
(5) Research School of Biology, The Australian National University, Canberra, Australia.

Given that an important element of variation is distributed within species, inferring intraspecific genetic diversity and identifying its predictors is of utmost importance so that we can develop effective conservation strategies. Here, we focus on the genetic diversity of amphibians and reptiles across six regions: the Iberian Peninsula, Northwestern Africa, the Arabian Peninsula, Madagascar, Northern Australia, and the Brazilian Atlantic Forest. We compiled mitochondrial DNA sequences from GenBank and respective geographic locations from literature. For each species, we also compiled primary data from open-source repositories (i.e. geographic, climatic and life history), as well as secondary data from species distribution modeling (i.e. niche breadth, hypervolume, marginality). We then tested for differences in genetic diversity among regions and used machine learning to identify its predictors. Our results reveal regional variations in genetic diversity and underscore the importance of life history traits, species range and land characteristics as key predictors of genetic diversity.

Poster 28

The genetic landscape and the adaptation of the bumblebee *Bombus terrestris* to distinct Portuguese biomes

Nunes, J [1,2,3]; Pinto, J [1,2]; Ferreira, S [1,2]; Paulo, O [4,5]; Rego, C [5]; Boieiro, M [4]; Amorim, I [6]; Melo-Ferreira, J [1,2,3]; Pimenta, J [1,2]

(1) CIBIO, Research Centre in Biodiversity and Genetic Resources, Universidade do Porto, Vairão, Portugal;
(2) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Vairão, Portugal;
(3) DBIO-FCUP - Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto, Portugal;
(4) cE3c—Center for Ecology, Evolution and Environmental Changes and CHANGE—Global Change and Sustainability Institute;
(5) DBA – Departamento de Biologia Animal, Faculdade de Ciências, Universidade de Lisboa;
(6) FCAA - Faculdade de Ciências Agrárias e do Ambiente, Universidade dos Açores

Human-induced environmental changes, including habitat destruction, urbanization, intensive agriculture, and pollution, pose a profound threat to Earth's biodiversity. Species are under unprecedented pressure due to environmental changes, forcing them to adapt, migrate, or face extinction. Among these, insect pollinators are one of the most affected group of species, with strong impact on ecosystems. This group of organisms is vital for pollination, being responsible for the pollination of around 80% of the flowering plants and 75% of the worldwide crops, making them crucial for ecosystem and biodiversity protection, and food security. The disappearance or severe reduction of abundance of insect pollinators would disrupt plant reproduction processes, potentially triggering a catastrophic cascade of extinctions across plants and animals. Therefore, understanding the population structure, evolutionary history, and adaptive mechanisms of insect pollinators is of paramount importance. These insights are essential for devising strategies to safeguard biodiversity and ecosystem services in the face of accelerating climate change.

Portugal, with its diverse climatic regions, spanning coastal zones, lowlands, and island ecosystems, offers an exceptional natural laboratory for studying how insect pollinators adapt to different environmental conditions. This BSc project will use whole genome sequencing (WGS) data to investigate the population structure and potentially detect signatures of adaptation of a bumblebee, *Bombus terrestris*, across four Portuguese biomes, in Porto, Lisbon, the Azores, and Madeira. The genetic data obtained will be mapped to high-quality reference genome and analysed to explore the demographic history, gene flow, and look for signatures of selective pressures in the genetic make-up of the populations.

The findings of this study will be a first step towards understanding how insect pollinators, particularly bumblebees, respond to climatic variability and environmental stressors. Ultimately, this research aims to contribute to more informed and effective conservation strategies, fostering the resilience of pollinators and their habitats, and ensuring their indispensable role in pollination for future generations.

Poster 29

Advancing methodological approaches applied to the conservation and management of wolves

Pinilla, L [1,2]; Lobo, D [1,2]; Godinho, R [1,2,3,4]

(1) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Campus de Vairão, Universidade do Porto, 4485-661 Vairão, Portugal;
(2) BIOPOLIS, Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, 4485-661 Vairão, Portugal;
(3) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, 4169-007 Porto, Portugal;
(4) Centre for Ecological Genomics and Wildlife Conservation, Department of Zoology, University of Johannesburg, 2006 South Africa

Non-invasive genetic sampling is widely used in wildlife management and conservation. However, its use in high-throughput sequencing approaches is often limited by low genotyping success and high error rates due to the low quality and quantity of DNA from such samples. Despite these challenges, non-invasive molecular approaches for monitoring mammalian carnivores have been advancing, particularly towards the development of Single Nucleotide Polymorphism (SNP) panels. Recently, SNP panels comprising up to 200 SNPs have been developed aiming to identify species, individuals, and hybrids (wild x domestic hybrids, when relevant) in various carnivores, such as the grey wolf. However, genotyping these SNP panels currently depend on Fluidigm Dynamic Array technology, which restricts its broader use. A promising alternative is amplicon sequencing, which involves amplifying SNP sets in multiplexed PCR followed by high-throughput sequencing. This approach offers greater flexibility in the number of samples and SNPs processed simultaneously can be used at any high-throughput sequencing platform while requiring only standard laboratory equipment. Nonetheless, implementing amplicon sequencing for SNP panels while ensuring high genotyping success rates in non-invasive samples remains technically challenging.

In this study, we aim i) establish an amplicon sequencing protocol to genotype existing SNP panels for the wolves and optimize it for non-invasive DNA samples, and ii) genotype non-invasive DNA wolf samples collected across the Iberian Peninsula to assess the prevalence of wolf-dog hybrids. Optimizing this methodology with non-invasive DNA samples will support wolf conservation and management by providing direct insights into wolf-dog hybridization across Europe.

Poster 30

Genomics insights of fire blight: deciphering *Erwinia amylovora* for improved on-field disease control

Regalado, L [1,2,3,4,5]; Marques, J [3,4]; Amorim, C [5]; Montenegro, MCBSM [5]; Santos, C [1,2]; Mendes, R [1,2]; Melo-Ferreira, J [1,3,4]

(1) FCUP – Faculty of Sciences, Biology Department, University of Porto, Porto, Portugal;
(2) LAQV-REQUIMTE, Faculty of Sciences, Biology Department, University of Porto, Porto, Portugal;
(3) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Campus de Vairão, University of Porto, Vairão, Portugal;
(4) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Vairão, Portugal;
(5) LAQV-REQUIMTE, Faculty of Pharmacy, Chemical Sciences Department, University of Porto, Porto, Portugal

Fire blight disease, caused by the bacteria *Erwinia amylovora*, is a global threat to pome fruit orchards, particularly apples and pears. Since its first detection in Portugal, the disease has become a pressing agricultural challenge, with considerable macroeconomic implications for one of the country's most valuable horticultural sectors. This bacterial disease infects a wide range of hosts within the Rosaceae family, causing severe damage through necrosis of blossoms, shoots, branches, and fruits, often leading to the loss of entire trees or even orchards. The rapid dissemination of the disease is exacerbated by favorable climatic conditions and the spread of contaminated plant material, but also due to the limited effective mitigation strategies.

The lack of comprehensive epidemiological data poses a significant barrier to effective management strategies. Additionally, the interplay between local environmental factors and pathogen evolution remains poorly understood, limiting the ability to predict outbreaks and mitigate their impact.

In this study, we will employ whole-genome sequencing (WGS) data to trace the pathogen's introduction routes and invasion dynamics in Portuguese agroecosystems. In addition, our work aims to pinpoint genetic variants possibly associated with key virulence traits, antibiotic susceptibility, and metabolic responses under different chemical environments. Key methodologies include the identification of single nucleotide polymorphisms (SNPs) linked to phenotypic traits, functional annotation of candidate genes, and the exploration of horizontal gene transfer events that may enhance *E. amylovora*'s virulence.

These genomic insights will advance our understanding of the evolutionary mechanisms driving fire blight's adaptation and success. Furthermore, this research is expected to contribute to outbreak mitigation by supporting the development of a biosensor based on an *E. amylovora*-specific molecular marker. The findings will provide a crucial framework for designing precision tools to improve disease management and reduce the impact of fire blight on Portuguese and global agricultural systems.

Poster 31

Wild vs. Captive: a comparative study of genetic and genomic approaches across continents

Meneses-Ribeiro, M [1, 2, 3]; Koepfli, KP [4, 5]; Jansen van Vuuren, B [6]; Godinho, R [1, 2, 7]

(1) CIBIO - Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Campus de Vairão, Universidade do Porto, 4485-661 Vairão, Portugal;
(2) BIOPOLIS - Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, 4485-661 Vairão, Portugal;
(3) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, 4099-002 Porto, Portugal;
(4) Smithsonian-Mason School of Conservation, George Mason University, Front Royal, VA 22630, USA;
(5) Center for Species Survival, Smithsonian's National Zoo and Conservation Biology Institute, Front Royal, VA 22630, USA;
(6) Centre for Evolutionary Genomics and Wildlife Conservation, University of Johannesburg, South Africa;
(7) Department of Zoology, Faculty of Sciences, University of Johannesburg, Auckland Park, South Africa

As defined by the IUCN, a species' conservation status considers population numbers in both wild and ex-situ captive populations. While genetic diversity is not formally included in these assessments, it plays a critical role in maintaining evolutionary potential and effective conservation management. Robust genetic monitoring in captive breeding programs is essential for sustaining biodiversity, particularly for species of economic relevance and conservation concerns. Traditional microsatellite analysis provides valuable insights into genetic structure, but advanced genomic techniques could offer deeper understanding of genetic diversity, inbreeding, and admixture in both captive and wild populations. The sable antelope (*Hippotragus niger*), a large African antelope with five distinct phylogroups, exemplifies the need for such thorough genetic assessments. Despite a decline in wild populations, the species is classified as "least concern" due to increasing numbers in captivity, driven primarily by its economic value for trophy hunting and farming in Southern Africa and North America.

This study examines the genetic diversity and structure of sable antelope populations in South African and North American game farms compared to their wild counterparts. Previous microsatellite data indicated a low prevalence of genetically pure individuals in South African farms, with only two of the five phylogroups represented in captivity, while the wild ancestry of North American stock remains largely unknown. We assessed whether North American farms might retain lineages now scarce in South Africa and compared traditional microsatellite analysis with a species-specific panel of 5,000 single nucleotide polymorphisms (SNPs). Our dataset included 54 wild samples, 85 from South African farms, and 76 from North America.

Our findings show that North American farms primarily house individuals from a single wild phylogroup, yet they exhibit greater genetic diversity than both South African captive and wild populations. Structure and differentiation analyses reveal that North American individuals form a distinct genetic group, separate from South African populations. SNPs and microsatellites demonstrated equally reliable assignments to wild groups and structure patterns, though SNP markers provided enhanced resolution. These results highlight how geographic isolation and management strategies influence genetic diversity and structure in captive populations. Incorporating comprehensive genetic analyses is crucial to preserve genetic health and ensure that reintroduction efforts support the long-term evolutionary potential and adaptability of species.

Poster 32

Investigating Mammalian Tissue Expression Patterns Using Public Transcriptomic Data

Rocha, AL [1,2]; Machado, AM [1]; Ruivo, R [1]; Carneiro, J [3,4]; Marques, ML [1]

(1) CIIMAR - Interdisciplinary Centre of Marine and Environmental Research, University of Porto, Portugal;
(2) FCUP - Faculty of Sciences of the University of Porto, Portugal;
(3) CBMA - Centre of Molecular and Environmental Biology, Department of Biology, University of Minho, Campus Gualtar, 4710-057 Braga, Portugal;
(4) IB-S - Institute of Science and Innovation for Bio-Sustainability, University of Minho, 4710-057 Braga, Portugal

Understanding gene expression patterns across different species within specific tissues is crucial for research focusing comparative genomics, evolutionary biology, and functional genomics. While many databases offer extensive gene expression data, this information is typically restricted to single-species contexts under specific experimental conditions.

Taking advantage of the immense amount of publicly available baseline transcriptomic data from multiple mammalian species, the aim of this work is to create a comprehensive tissue expression atlas. This atlas will facilitate comparisons of transcription profiles across the same tissues using multi-organism data, enabling the identification of molecular-level similarities and differences within a single tissue and create tissue-specific expression patterns. Gene expression changes are postulated to underline many phenotypic differences between species. Importantly, this comparative analysis can also bring new insights into interspecies differences which may be further linked to specific adaptations.

In this work, we have collected six baseline multi-tissue transcriptome datasets from the public database Expression Altas. After initial integration processing and normalization of the data we obtained comparable datasets for 3 tissues from six species. Our results reveal conserved patterns of genes expressed within these tissues, even when comparing transcriptomic data from different species. Additionally, we identified species-specific expression patterns in the analyzed tissues, which could provide insights into adaptive traits unique to each species.

Finally, this tissue expression panel will provide valuable insights into the genetic basis of physiological adaptations and provide detailed expression profiles for each tissue. This tool will be instrumental in evaluating transcriptome quality, selecting appropriate model organisms, and predicting expression profiles for species lacking available transcriptomic data.

Acknowledgements: This work was supported by Portuguese Foundation for Science and Technology (projects UIDB/ 04423/2020 and UIDP/04423/2020), UIDB/04050/2020 (UMinho CBMA - https://doi.org/10.54499/UIDB/04050/2020). JC acknowledges the FCT funding for his research contract established under the transitional rule of Decree Law 57/2016, amended by Law 57/2017. FCT supports MLM under the Scientific Employment Stimulus program MLM-2022.00397.CEECIND/CP1728/CT0006.

References

1-Sudmant, P.H., Alexis, M.S. & Burge, C.B. Meta-analysis of RNA-seq expression data across species, tissues and studies. Genome Biol 16, 287 (2015).

2-Brawand, D., Soumillon, M., Necsulea, A. et al. The evolution of gene expression levels in mammalian organs. Nature 478, 343–348 (2011)

3-Nancy George, et al. Expression Atlas update: insights from sequencing data at both bulk and single cell level, Nucleic Acids Research, Volume 52, Issue D1, 5 January 2024, Pages D107–D114

4-Cardoso-Moreira, M., Halbert, J., Valloton, D. et al. Gene expression across mammalian organ development. Nature 571, 505–509 (2019).

Poster 33

The Azorean Daucus: a morphological, cytogenomic, phylogenetic and ecological study

Roxo, G [1,2]; Vaz, A [1,2]; Dias, EF [1,2]; Silva, L [1,2]; Carine, M [3]; McCartney, AM [4]; Rego, RMC [1,2]; Talhinhas, P [5]; Romeiras, MM [5]; Zidorn, C [6]; Resendes, R [1,2]; Moura, M [1,2]

(1) CIBIO-Azores, Departamento de Biologia, Universidade dos Açores, Rua Mãe de Deus 58, Apartado 1422, 9501-801 Ponta Delgada, Portugal
(2) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO Centro de Investigação em Biodiversidade e Recursos Genéticos, Campus de Vairão, 4485-661 Vairão, Portugal
(3) Natural History Museum, London, UK
(4) Genomics Institute, University of California, Santa Cruz, California, 95060, USA
(5) Linking Landscape, Environment, Agriculture and Food (LEAF), Associate Laboratory TERRA, Instituto Superior de Agronomia (ISA), Universidade de Lisboa, Tapada da Ajuda, 1349-017 Lisboa, Portugal
(6) Pharmazeutisches Institut, Abteilung Pharmazeutische Biologie, Christian-Albrechts- Universität zu Kiel, Gutenbergstraße 76, 24118, Kiel, Germany

The *Daucus L.* genus contains approximately 45 species, the majority of which are found in the Mediterranean region. The *Daucus carota* complex includes ca. 25 infraspecific taxa, for which there are no or only poorly developed barriers to interbreeding between the wild and domesticated forms. In the Azores archipelago two sub-species occur: the endemic *Daucus carota subsp. azoricus Franco*, and the native *Daucus carota subsp. maritimus* (*Lam.*) *Batt*. Due to the phenotypic diversity observed across the Daucus carota complex, the distribution and diagnostic morphological characters of the endemic Azorean species remain unclear. This study aims to clarify the taxonomy of the *Daucus carota* complex in the Azores and determine if the *Daucus carota subsp. azoricus Franco* is in fact a distinct taxon. For this purpose, we used molecular, cytogenomic, morphological and ecological data. A total of forty-four specimens of *Daucus carota subsp. azoricus* were sampled across the nine islands of the archipelago and the ITS region and the COS region (CA7) sequenced. Estimations of DNA content were performed with flow cytometry to investigate variation between taxa (i.e., *Daucus carota subsp. azoricus* and *Daucus carota subsp. maritimus)* and populations. Moreover, we measured a total of 31 morphological characters from across the stem, inflorescences, flowers, and fruits. Finally, we performed germination tests in order understand the phenotypical diversity observed and cultivating *Daucus carota subsp. azoricus* under different climates and soils. Preliminary results indicate that there are no major morphological differences between the sampled sites. Similarly, with cytogenomic data it was not possible to discriminate between the two subspecies. However, the molecular data revealed variation across the archipelago with different haplotypes obtained from the screened regions.

Poster 34

Incomplete Lineage Sorting and Its Role in the Recurrent Evolution of Parthenogenesis in Stick Insects

Souto, J [1,2]; Mérel, V [1]; Goudet, J [1,2]; Schwander, T [1]

(1) Department of Ecology and Evolution, University of Lausanne, Lausanne, Switzerland
(2) Swiss Institute of Bioinformatics, University of Lausanne, Lausanne, Switzerland

The repeated evolution of similar traits in related species can occur through diverse mechanisms, including the independent selection of shared ancestral variants in different lineages. This mechanism can cause incongruences between gene and species trees, also known as incomplete lineage sorting (ILS). Here we study the impact that ILS could have in the parallel evolution of different traits in Timema stick insects, with a particular focus on transitions between sexual reproduction and parthenogenesis, which have occurred multiple times independently in this group. Given that Timema species diverged up to 30 mya, the traditional “one reference” approach – where all samples are mapped to a common reference genome - can introduce biases and mapping errors due to high divergence. Therefore, we tested a “liftover” approach in which samples are mapped to a reference from their own species. Inferred genotypes are then assigned to the homologous region of a different species by converting genome coordinates, whereby conversion settings are based on whole-genome alignments between species pairs. Our preliminary results show that, across varying levels of divergence, the “liftover” approach achieves higher mapping rates in gene regions compared to the “single reference” approach, supporting its utility for highly diverged genomes. Subsequently, to identify candidate regions for parthenogenesis in Timema, we will look for regions with excess shared variation between parthenogenetic lineages. By contrasting coalescence times along the genome, we will then infer whether candidate regions reflect ancestral polymorphisms (ILS; increased coalescence times) or introgression (recent coalescence times). Altogether we aim to quantify the impact of ILS on the evolution of highly diverged Timema lineages and its possible role in shaping adaptive traits.

Poster 35

DNA from cetariae fish remains confirms sardine (*Sardina pilchardus*) use and local population continuity in Northwestern Iberia since Roman times

Themudo, GE [1]; Fernández-Fernández, A [2]; Abad, PV [2]; Nóvoa, AAR [2]; Fernández-Rodriguez, C [3]; de Agüero, EG [3]; da Fonseca, RR [4]; Campos, PF [1]

(1) CIIMAR, Centro Interdisciplinar de Investigação Marinha e Ambiental, Universidade do Porto, Matosinhos, Portugal
(2) Faculty of History, Department of Art, Geography and History, GEAAT, University of Vigo, Ourense, Spain
(3) Department of History, University of Léon, León, Spain
(4) The Globe Institute, Faculty of Health and Medical Sciences, University of Copenhagen, Copenhagen, Denmark

The Romans were one of the first societies to fully exploit fish resources, creating large scale salting and preservation plants to prepare fish for storage and transportation across the empire. Small pelagic fish were fermented to make fish sauces, like garum. Here, for the first time, we recover DNA from 18 specimens retrieved from the bottom of fish salting vats from Adro Vello (O Grove), Galicia radiocarbon dated to the 1st to 4th century AD. Using whole genome resequencing, we reconstruct the mitogenomes and using phylogenetic analysis identify them as European sardines (*Sardina pilchardus*). Low coverage nuclear data assigns all samples to one of three previously described sardine populations, with a range from North Morocco to the Bay of Biscay. The low levels of admixture in these ancient samples compared to the modern samples of the same genetic group, and their larger differentiation from both the Mediterranean and the Macaronesia populations suggest lower connectivity for this species in Roman times. Exploring this, so far overlooked, archaeological material, either due to its fragmentary nature or difficult identification, will open a new window into the past for the understanding of subsistence economy, culture, and the diet of past peoples.

Poster 36

Estimating trait heritability and variation in predator-prey dynamics

Tomé, R [1]; Lima, A [1]; King, J [1]; Wonner, T [1]; Fragata, I [1]

(1) CE3C - centre for ecology, evolution and environmental changes, FCUL University of Lisbon, Lisbon, Portugal

Prey-predator interactions are prevalent in ecosystems, as most organisms are either consuming prey, being predated, or both. Theoretical and empirical studies have shown that different levels of genetic variation in the predator and/or prey population can determine the persistence of the system. However, most studies do not link the genetic variation observed in the system to traits involved in the predator-prey interactions. Thus, the phenotypic traits in which genetic variation is crucial to determine the persistence of a predator-prey system remain mostly elusive. However, this knowledge is key to determine the evolutionary potential and predict long-term eco-evolutionary dynamics in the system.

Here we aim to fill this gap by using a system composed of several isogenic lines of the predator *Amblyseius swirskii*, and its prey (*Tetranychus urticae*). Previous studies have produced mixed evidence of the ability of the predator to consume different prey stages (eggs, nymphs and adults), as well as its preference for each life-stage. However, preference and predation rate of different prey life-stages can have an important impact on prey population structure, potentially affecting population dynamics. Our aim is to quantify variation between isogenic lines for functional response, prey stage preference, predation rate, handling time and conversion efficiency and estimate the broad sense heritability associated with these traits to assess their evolutionary potential. We will then experimentally test how variation in the different traits tested affects predator-prey dynamics and the persistence of our system. Our results will contribute to elucidate the importance of individual variation in the persistence of predator-prey systems.

Poster 37

Escaping from a stressor: evolution of larval development under nutritional constraints

Varela-Pereira, AC [1]; Kawecki, TJ [1]

(1) UNIL - Department of Ecology and Evolution, Faculty of Biology and Medicine, University of Lausanne, Lausanne, Switzerland

In nature, organisms often face periods of food shortage, which can severely impact metabolism, fitness, and survival. Immature stages, like juvenile insects, are especially vulnerable due to limited mobility, restricting their ability to reach new nutrient sources and most of the time they have no other option but to continue development. Therefore, natural selection is expected to favor physiological, behavioral and life history adaptations that alleviate the negative consequences of nutrient scarcity for Darwinian fitness. In our lab, *Drosophila melanogaster* larvae evolved on a diet with only 25% of the standard nutrient content over 350 generations. As a result of this experimental evolution, these populations are now able to develop much faster in an undernutrition context than Control populations evolved on standard diet, and only emerge at a slightly smaller adult body weight. Such changes can be accomplished by faster growth throughout development but also by shortening the larval wandering period and other potential changes in relative duration of different larval stages. Here I will report on a study in which I characterized in detail the differences in growth (in terms of protein accumulation), accumulation of fat reserves and the timing and size at which Selected larvae achieve a growth advantage over Controls under nutrient-poor conditions. The project contributes to the understanding of mechanisms mediating the evolution of enhanced tolerance to juvenile organisms forced to develop despite nutritional constraints.

Poster 38

Evolution on our heads: disentangling mechanisms of diversification in human lice

Martinez, I [1,2]; Pierre, B [1,2]; Vasconcelos, D [1,2]; Campos, L [1,2,3]; Perera, A [1,2]; Sá-Pinto, A [4]; Xavier, R [1,2]; Faria, R [1,2,3]; Pinho, C [1,2]

(1) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório As-sociado, Campus de Vairão, Universidade do Porto, Vairão, Portugal
(2) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Portugal
(3) Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Porto, Portugal
(4) CIDTFF, University of Aveiro

Human lice (*Pediculus humanus*) are ectoparasitic insects that infest millions of people globally, posing significant public health challenges. Two forms of the human louse, currently regarded as subspecies, are known: the head louse, *Pediculus humanus capitis*, infesting the hair and scalp of its hosts, and the body louse, *P. h. humanus*, which lives on the hosts' clothes. However, there is still controversy about their process of divergence: while some studies suggest a single origin of body lice that started to diverge from head lice with the beginning of clothe wearing by humans, other studies rather suggest multiple origins of body lice associated with poor hygiene and intense parasitism in some human patients. Recently, we started research project EVOLICE with the main goal of testing these hypotheses as well as to understand the evolutionary processes shaping the diversity of human lice. Here we will show our first preliminary results based whole genome sequences from ~100 lice sampled in four different countries (Portugal, China, Nigeria and USA) from different continents, including body and head lice; as well as our ongoing efforts to better understand the evolutionary history of human lice and their potential as vectors of human diseases.

Poster 39

Chromosomal rearrangements and the evolution of reproductive barriers in a marine snail

Vidal-Capón, A [1]; García-Souto, D [1]; Pasantes, JJ [1]; Rolán-Álvarez, E [1]; Galindo, J [1]

(1) CIM - Centro de Investigación Mariña, Universidade de Vigo, Departamento de Bioquímica, Xenética e Inmunoloxía, Vigo, Spain

Genome-wide rearrangements act as powerful drivers of divergence and speciation across diverse organisms. In the case of Robertsonian translocations (Rb), their occurrence is typically associated with partial chromosomal homology, creating genomic instability that may result in variations in chromosome number. When such rearrangements become fixed, they contribute to the onset of distinct chromosomal races [1]. In the marine snail *Nucella lapillus*, five chromosomal pairs undergo Rb events, leading to metacentric or acrocentric forms. These rearrangements have given rise to two distinct chromosomal races, each adapted to specific habitats, exemplifying divergent evolution. One race, inhabiting sheltered shores, is thick-shelled and presents karyotypes within a chromosome range of 2n = 27–36, while the other, adapted to exposed shores, is thin-shelled with 2n = 26 chromosomes, characterized by the fixation of metacentric forms in five Rb-involved groups [2]. This differentiation has established a clear pattern of phenotypic, genotypic, and karyotypic divergence. Recent studies have reported significant genetic differentiation between these chromosomal races along the Galician Atlantic coast [3]. This Doctoral Thesis aims to integrate genomic and cytogenetic approaches to investigate the evolutionary trends between these races along the Atlantic coast. A central objective is to determine whether their divergence arose from a single origin or multiple independent events. Furthermore, we will examine the karyological traits of natural populations to evaluate the frequency and occurrence of hybridization events and identify barriers to gene flow. This work will also seek to describe the genomic basis underlying the observed polymorphisms, focusing on their maintenance, fixation, and role in race differentiation. Ultimately, this research will enhance our understanding of how barriers to gene flow emerge and evolve in a Robertsonian system, particularly under conditions of limited dispersal, high environmental heterogeneity, and strong selective pressures.

[1] Redi, C. A., Garagna, S., Zuccotti, M. (1990). Robertsonian chromosome formation and fixation: the genomic scenario. Biological Journal of the Linnean Society, 41(1-3), 235–255.

[2] Pascoe, P. L. (2006). Chromosomal polymorphism in the Atlantic dogwhelk, Nucella lapillus (Gastropoda: Muricidae): nomenclature, variation and biogeography. Biological Journal of the Linnean Society. 87(2), 195–210.

[3] García-Souto, D., Fernández-Rodríguez, J., Pasantes, J. J., Rolán-Alvarez, E., Galindo, J. (2024). Phenotypic and genetic differentiation between two chromosomal taxa of the gastropod Nucella lapillus at Galician rocky shores (NW Spain). In press.

Poster 40

Phylogenomic analysis of evolutionary radiations: dealing with reference bias

Vieira, I [1]; Nevado, B [1]

(1) CE3C - centre for ecology, evolution and environmental changes, FCUL University of Lisbon, Lisbon, Portugal

Second-generation sequencing (NGS) technologies allow for simultaneous sequencing of millions of short DNA fragments, which can be used for a wide array of evolutionary analyses. However, analysing these short sequences typically depends on the existence (or de novo assembly) of reference genomes for the target species, which remains a key limitation – particularly so for studies involving multiple species.

A particular challenge arises in the analysis of second-generation sequencing data in phylogenomic research. Such studies typically require mapping short-read data from multiple, closely related species to a shared reference genome, which can cause substantial biases. This study explores methods for characterising and correcting such biases, and is especially relevant for cross-species comparisons where traditional methods, aimed at intra-specific analyses, may fall short.

Using genomic data from multiple species of *Lupinus* as a test case, this study assessed the impact of using a single reference genome to analyse resequencing data from species with different levels of genetic divergence (from the reference genome). We evaluated several methodological strategies to minimize reference bias, including custom mapping parameters and constructing re-reference genomes incorporating single nucleotide polymorphisms (SNPs) from individual samples. Furthermore, to validate our analysis, we compared the experimental data with simulated datasets. Key metrics analyzed include mapping percentage, number of sites mapped, coverage, heterozygosity and missingness. Our findings show that the modifications and methods applied can enhance mapping percentages by up to 40%, increase read depth, and expand genome coverage. However, these improvements did not consistently translate to higher quality in downstream analyses, highlighting the complex nature of reducing reference bias in practical applications.

By advancing methodologies to mitigate reference bias, the results of this study contribute to more accurate evolutionary analysis of multiple-species genomic datasets. These improvements not only enhance mapping precision and data quality for species with incomplete or distant reference genomes but also provide a framework that can be adapted to other taxa facing similar genomic challenges.

Poster 41

A marine snail color cline under study: the role of crypsis from a field tethering experiment

Vigo, R [1]; Gefaell, J [1]; Galindo, J [1]; Rolán-Alvarez, E [1]

(1) CIM - Centro de Investigación Mariña, Universidade de Vigo, Vigo, Spain

How color polymorphism is maintained in spatially or temporally variable environments is a classic topic of study for evolutionary ecologists. A gradual variation shell color polymorphisms of the rough periwinkle (*Littorina saxatilis*) has been described in the Rias Baixas (NW Spain) and reported to remain relatively stable after forty years in at least one of them (Ria de Vigo). This color cline shows a dominance of plain light-colored shells in the inner, sheltered localities and a lineated morph in the outer and wave-exposed areas. One mechanism that has been experimentally tested for color polymorphism in the inner region is negative frequency-dependent selection via mate choice. For the outermost parts, the abundance of monomorphic populations leads to the hypothesis of differential visual predation, as the lineated morph may be highly cryptic against the characteristic barnacle-covered rock substrate of these areas. In this study, we conducted a tethering experiment at a representative wave-exposed locality from the Rias Baixas to explore a potential selection for crypsis, particularly by crabs. We compared three transplanted color morphs of different degrees of crypsis (plain yellow, plain black, and lineated shells) from a nearby polymorphic population with the resident lineated morph. Furthermore, complementary analyses were carried out within both populations to exclude the influence of other shell traits on predation vulnerability and confirm a degree of crypsis between color morphs. Although slight differences were observed in terms of shell scars frequency, the role of crypsis in this color cline remains unclear given that differential selection for shell color has only been observed in one out of two sessions when considering chipped shells as predation attempts. However, these results should be interpreted with caution due to its low statistical power resulting from the overall low predation rate.

Poster 42

Uncovering Mechanisms of Behavioural Developmental Bias

Vilas Boas, J [1]; Ferreira, G [1]; Picão-Osório, J [1]

(1) CE3C - centre for ecology, evolution and environmental changes, Department of Animal Biology, Faculty of Sciences, University of Lisbon, Lisbon, Portugal

Heritable phenotypic variation is essential for biodiversity, enabling organisms to adapt to different environmental conditions. This variation drives natural selection, evolutionary processes, and species persistence in changing ecosystems. Therefore, it is crucial to identify and understand mechanisms that define and influence the structure of phenotypic variability, as they shape not only the diversity of forms and behaviours observed in nature, but also the evolutionary potential of populations. Developmental mechanisms translate genotypes to phenotypes and can cause some phenotypic variants to arise more readily than others. This introduces a bias in the phenotypic variability available for selection, i.e. developmental bias. Despite its potential implications on evolutionary rates and trajectories, the specific developmental genetic mechanisms underlying developmental bias in organisms remain poorly understood. Here, we propose using the neurobehavioral system of *Caenorhabditis elegans* to uncover mechanisms of developmental bias at the whole-organism level in ecologically relevant scenarios. This involves a novel integrative approach that combines developmental biology, neuroscience, evolutionary ecology, and quantitative genetics. Moreover, the project will exploit the uniqueness of the model system *C. elegans* in combining remarkable biological features, tools and resources. *C. elegans* relies on its chemosensory and locomotory systems to detect and avoid pathogenic bacteria in its microbe-rich natural habitats. This avoidance behaviour shows significant natural variation, and the genes underlying the nematode's chemosensory system have strong signatures of positive selection within *C. elegans* and among *Caenorhabditis* species. This suggests that chemosensation might be a probable hotspot for behaviour evolution. This makes the ecologically relevant avoidance behaviour to pathogens an ideal trait to uncover mechanisms of developmental bias. Our objectives are twofold: (i) to quantify behavioural developmental bias and (ii) to identify the developmental mechanisms that drive this bias. To do this, we will induce random de novo mutations through chemical mutagenesis, generating isogenic random mutation lines (RMLs), that allow for a random and wide exploration of the genotypic space. These RMLs will be generated in the NeuroPal strain (Neuronal Polychromatic Atlas of Landmarks), which generates a stereotyped multicolour fluorescent map of the whole nervous system. This will later be crucial to uncovering organismal mechanisms of developmental bias. Next, we will perform high-throughput quantitative phenotyping and variance analysis of locomotory and pathogen avoidance behavioural traits among the RMLs. If developmental bias is present, we expect to observe a non-random distribution in phenotypic space upon de novo mutation, where some behaviours exhibit greater variability than others. Finally, we will correlate these patterns of phenotypic variability with changes in neuronal cell fate decisions during development using the NeuroPal transgenes. This will inform us on our ultimate goal of finding developmental pathways underlying traits with high variability. To start our project, we are currently characterising the pathogenicity of several ecological-associated bacterial strains to *C. elegans* in order to link fitness-related behavioural responses. These pathogenicity and avoidance assays are being conducted in the standard laboratory *C. elegans* strain N2 (Bristol) and its most divergent known wild strain, XZ1516 (Hawaii). In sum, we aim to uncover developmental processes that shape behavioural variability in *C. elegans*, providing insights into how biased phenotypic variability arises at the organismal level. Our findings will enhance our understanding of variation structure in populations and establish a foundation for future studies on how developmental bias impacts evolutionary rates and trajectories.

Poster 43

Tipping the scale: investigating the feedback between evolution and stability in a predator-prey system

Wonner, T [1]; López, J [1]; Fernandes, J [1]; Lima, A [1]; Magalhães, S [1,2]; Fragata, I [1]

(1) cE3c - Centre for Ecology, Evolution and Environmental Changes & CHANGE - Global Change and Sustainability Institute, FCUL University of Lisbon, Lisbon, Portugal; (2) Wissenschaftkolleg zu Berlin, Berlin, Germany

Understanding how ecology and evolution interactively affect predator-prey interactions is an important step in predicting their probability of extinction. Indeed, theoretical studies support the idea that the feedback between ecology and evolution can alter the stability of a system (defined as the ability to return towards the initial equilibrium after a small perturbation). In turn, system stability may affect how populations evolve. However, the extent to which the feedback between ecology and evolution affects population dynamics needs to be tested empirically. Here, we aim to address this gap using a predator-prey model system consisting of a crop pest (*Tetranychus urticae*) and its predator (*Ambluseus swirskii*). As a first step, we investigated which factors change the population dynamics and persistence of both species in our system. To this aim, we set up a system composed of bean plants (*Phaseolus vulgaris*) with a varying initial ratio of predator and prey (4%, 10% and 20% predator), replicated 10 times. All treatments started with 20 adult prey females, and predator females were added after one generation according to their respective predator-prey ratios. The number of adult females of both species was quantified twice a week for 12 weeks. Overall, we observed a higher rate of system collapse with higher initial predator-prey ratios. Thus, higher predator-prey ratios decrease the probability that evolution can operate in the system by increasing the probability of extinction. Our next step is to explore other factors that affect the stability of the system, i.e. spatial heterogeneity, nutrient enrichment of the plant and different host plants. In the future we plan to use more or less stable systems to investigate how evolution is affected by the stability of the system, and how stability is affected by evolutionary and eco-evolutionary dynamics.

Poster 44

From Ship to Shore: The Genetic Legacy of Invasive Rats in Atlantic Islands

Wielewski, M [1]; Almeida, L [1]; Alves, PC [1]; Gabriel, SI [2]; Vasconcelos, R [1]

(1) CIBIO-Research Centre in Biodiversity and Genetic Resources, FCUP University of Porto, Porto, Portugal; (2) Center of Studies of the Environment and the Sea, FCUP University of Lisbon, Lisbon, Portugal.

Islands offer unique ecological and evolutionary insights but are fragile ecosystems, particularly vulnerable to invasive species. This research focuses on the origin of invasive species in Macaronesia and other Atlantic islands: the black rat, Rattus rattus. This species thrived due to easy adaptation to new environments, omnivorous habits and a commensal relationship with humans. The goal of this study is to ascertain the origins of these insular rat populations through the analysis of molecular markers, examining their genetic diversity within the islands and provide information for their management and conservation of native species. The genomic DNA from these samples was extracted and the mitochondrial D-loop amplified by PCR and sequenced. The resulting data was compared with sequences from public databases to infer the most likely origin, elucidating the relationships among the rat invasive populations of Macaronesia with African and European populations, using phylogenetic and networks analyses. The understanding of the routes by which these populations arrived on the islands and their origins is crucial to understand the colonization patters of biological invasions. Additionally, the genetic data obtained can be used to monitor rat populations and evaluate the effectiveness of control measures.

Poster 45

The evolution and functional convergence of iodine assimilation genes in brown algae and animals

Barbosa, AN [1,2,3]; Pires, L [1,2,4]; Lourenço, T [1,2,4,5]; Monteiro, C [1,2]; Castro PH [1,2,6]

(1) CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Campus de Vairão, Universidade do Porto, Vairão, Portugal; (2) BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, Vairão, Portugal; (3) Escola de Ciências da Vida e do Ambiente,, Universidade de Trás-os-Montes e Alto Douro, Quinta de Prados, Vila Real, Portugal; (4) Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto, Portugal; (5) IPSiM, Université de Montpellier, CNRS, INRAE, Institut Agro, Montpellier, France; (6) Departamento de Biologia, Escola de Ciências, Universidade do Minho, Campus de Gualtar, Braga, Portugal

Iodine is a nutrient for many organisms. In humans and other vertebrates, iodine is a component of the hormones produced by the thyroid and its deficiency has a tremendous negative impact on human health. In brown algae, such as Laminaria species, iodine is not only an essential element, but also is greatly accumulated in the tissues. The impressive capacity of animals and brown algae to use and assimilate iodine relies greatly on the central role of iodide peroxidases (IPOs), such as thyroid peroxidase (TPO) and vanadium- dependent iodoperoxidase (VIPO), respectively. The VIPO enzymes have two important functions in brown algae, one is protection against stress caused by oxidative damage and, second, to act as a protective element against predators and diseases. Interestingly, iodine content quantification in brown algae revealed that there are differences in iodine accumulation between species. In particular, Laminaria hyperborea is generally more efficient at accumulating iodine than other species, including Laminaria digitata that is considered a reference in iodine accumulation studies. We hypothesize that these differences can be explained by variation in the activity of VIPO enzymes. As such, we initiated a cloning strategy for the Laminaria hyperborea VIPO gene using degenerated primers based on the L. digitata sequence. To understand the relationship and evolution of these important IPOs, we performed sequence comparisons and built a phylogenetic tree based on homologous IPO gene sequences present in different databases. From the analysis of the phylogenetic tree, we were able to suggest that the ability of brown algae to accumulate iodine may have been acquired through horizontal transference of early halide peroxidase genes from bacteria. These evidences are also indicative of a parallel origin of VIPO from animals TPOs, and a functional convergence for iodide assimilation should have occurred. Understanding the origin, conservation and function of IPOs will help explore iodine metabolism and its biological relevance. Most interestingly, deeper knowledge about VIPO enzymes can be used for genetically-based biofortification strategies.

Acknowledgments: Work supported by National Funds through FCT-Fundação para a Ciência e a Tecnologia in the scope of the project UIDB/50027/2020. Work co-funded by the project NORTE-01-0246-FEDER-000063, supported by Norte Portugal Regional Operational Programme (NORTE2020), under the PORTUGAL 2020 Partnership Agreement, through the European Regional Development Fund (ERDF).

Poster 46

An integrated bioinformatics approach to study the Transgenerational Impact of Simvastatin's on the crustacean *Gammarus locusta*

Simões, D [1,2]; Machado, A [1]; Lopes-Marques, M [1]; Carneiro, J [3,4]; Neuparth, T [1]

(1) Ciimar - Interdisciplinary Centre of Marine and Environmental Research, FCUP, University of Porto, Porto, Portugal; (2) FCUP - Faculty of Sciences of the University of Porto, Porto, Portugal; (3) CBMA - Centre of Molecular and Environmental Biology, Department of Biology, University of Minho, Campus Gualtar, Braga, Portugal; (4) IB-S - Institute of Science and Innovation for Bio-Sustainability, University of Minho, Braga, Portugal

Transgenerational effects induced by contaminants have become a topic of growing interest. Increasing evidence suggests that parental generation (F0) exposure to environmental contaminants can cause lasting effects that persist across unexposed generations. This study investigates this phenomenon using the arthropod Gammarus locusta as a model organism. Previous experiments showed that exposure to the hypocholesterolemic drug simvastatin (SIM) in the parental generation (F0) caused severe impairments in reproduction and growth, effects which were inherited by unexposed generations (F3), confirming true transgenerational inheritance. The primary aim of this study is to identify differentially expressed genes (DEGs) and proteins (DEPs) between the F0 and F3 generations, with a particular focus on the metabolic pathways affected by simvastatin.

To uncover the molecular basis of these effects, we are using a bioinformatics approach to compare gene expression and protein profiles between generations. Due to the lack of a reference genome, we are conducting de novo transcriptome assembly, followed by gene annotation and statistical analysis to identify DEGs in each generation. Additionally, proteomic analysis will be conducted to identify DEPs between generations.

Integrating these multi-omic data will illuminate key metabolic pathways disrupted by SIM and reveal potential epigenetic modifications driving these effects. Epigenetic changes are evolutionarily significant, as they can occur more rapidly than genetic mutations and may facilitate adaptive responses to environmental stressors. Given the ecological importance of Gammarus locusta as a bioindicator species and a key prey item for commercially important fish such as flatfish, this study highlights the need to assess the broader impacts of pharmaceutical contaminants on marine ecosystems. By focusing on transgenerational effects and potential epigenetic changes, we aim to understand how environmental pollutants like SIM influence the evolutionary trajectories of exposed populations. These insights are crucial for assessing the long-term effects of pharmaceutical pollutants on marine ecosystems and fisheries, highlighting the need to incorporate evolutionary perspectives into environmental risk assessments.