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5th EUGLOH PLANT SCIENCE MEETING

9th Meeting in Functional Biology and Biotechnology of Plants

BOOK OF ABSTRACTS

Oral Presentations & Poster Presentations

Book of abstracts

5th EUGLOH Plant Science Meeting

9th Meeting in Functional Biology and Biotechnology of Plants

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Book of abstracts

Oral communications

Session 1: Plant functional genomics

OP1

Environment perception at the local chromatin structure

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Temperature influences nucleosome dynamics, and thus chromatin, to regulate gene expression. Such mechanisms underlie the epigenetic silencing of the Arabidopsis *FLOWERING LOCUS C (FLC)* by prolonged cold. We show a temperature-dependent transition in local chromatin structure at the H3K27me3 nucleation region, from a modality active for transcription to a state that can be Polycomb silenced. In vivo chromatin measurements and coarse-grained simulations at near-atomistic resolution show that the active transcription state is characterised by a highly dynamic nucleosome arrangement that exposes the *FLC* transcription start site (TSS). Cold exposure then changes the chromatin by reducing nucleosome dynamics and repositioning the + 1 nucleosome, leading to transcriptional repression. This local chromatin transition partially depends on VERNALIZATION1 (VRN1), a non-sequence-specific DNA-binding protein. Loss of VRN1 results in hyperaccumulation of H2A.Z, more dynamic nucleosomes and an inability to accumulate H2Aub and H3K27me3. Our work highlights how local nucleosome dynamics link to chromatin structure transitions to integrate temperature inputs into epigenetic switching mechanisms in plants.

OP2

The PSI lab's research @GreenUPorto. Exploring vacuolar targeting approaches of Arabidopsis thaliana's type 2b metallothionein in tobacco leaves

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Nowadays, anthropogenic activities are one of the main sources of heavy metals (HM) release to soils. This poses a major issue since HM can induce toxicity that risks the overall fitness of the plants when in excess. Plants possess several strategies to deal with HM toxicity, one being the use of metallothioneins (MTs), which are small Cys-rich proteins capable of binding HMs and scavenging Reactive Oxygen Species, the other being the sequestration and accumulation of HM into the vacuole. The plant-specific insert (PSI) domains of the aspartic proteinases cardosin A and B from *Cynara cardunculus* possess vacuolar targeting functions. In this work, an attempt was made to target *Arabidopsis thaliana* MT2b to the vacuole through its fusion to PSI A and to mNeonGreen (MT2bΩ-6xHisPSIA-mNeonGreen) as a strategy to increase the HM sequestration capacity. Its cellular location was visualised *in vivo* through confocal microscopy in *Nicotiana tabacum* leaves after *Agrobacterium* infiltration. The results showed no accumulation of this MT in the vacuole, as it was only found in the cytosol, revealing new insights regarding the PSI A function as a Vacuolar Sorting Determinant (VSD), as other factors may be at play.

OP3

Exploring plant reproductive biology to secure future food production

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Plant reproduction lies at the heart of global food security, yet reproductive processes are among the most sensitive to environmental change. At SPReDLab, we explore the molecular and cellular mechanisms underlying plant reproduction, with particular focus on how plants communicate, adapt, and ensure fertilization. Our research integrates molecular genetics, transcriptomics, and cell biology to uncover the dialogue between male and female tissues during the progamic phase and identify key determinants of reproductive success.

Several projects are now active in our lab, most of them focusing on improving the reproductive process under heat stress conditions. Through the PrOryza project, we merge molecular analyses to classical breeding, to develop thermotolerant Portuguese rice varieties, contributing to sustainable agriculture in the face of climate change. The European CRISPit project leverages genome editing tools to uncover regulatory networks controlling fertility and to generate new genetic resources for crop improvement. Meanwhile, the LoveTalk project investigates the molecular "language" of love and death between pollen tubes and the transmitting tract in Arabidopsis, revealing how programmed cell death and extracellular signalling coordinate successful fertilization.

Together, these lines of research provide a holistic understanding of reproductive resilience, from the molecular mechanisms in model species to translational applications in crops. By bridging fundamental discovery and applied innovation, SPReDLab aims to strengthen the foundations of plant reproductive biology and contribute to the development of climate-resilient crops, ensuring stable food production for future generations.

OP4

RiceItUp : Understanding the heat stress mechanisms of *Oryza sativa*

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Plants are fundamental to humans, being a source of food, pharmaceuticals, among other things. Rice (*Oryza sativa*) provides about 35-60% of global daily calories to humans worldwide.
However, the quantity and quality of rice are heavily affected by heat. For every 1ºC increase in temperature, there's a decline in rice production of 6.2-10%, being the reproductive period the most sensitive one. The consequences of heat stress in rice inflorescences include abnormal pollen development, lack of anther dehiscence, low stigmatic receptivity, defects in germination and elongation of the pollen tube, among others. Due to global warming, these problems will become increasingly frequent, leading to major impacts on food production.

Therefore, this project aimed to study the *O. sativa*'s heat stress tolerance molecular mechanisms at the reproductive level. A previous RNA-seq performed on pistils from a heat-tolerant rice variety, Nagina22 was used to select a list of candidate genes related to this mechanism in *O. sativa*. Their orthologues in *Arabidopsis thaliana* were identified. The expression level of the selected genes in control and heat stress conditions was validated by RT-qPCR. A T-DNA insertion line in *A. thaliana* was selected and analysed for phenotypic characterization in both conditions. Constructs for reporter and overexpression lines in Arabidopsis, and overexpression and prime editing lines in rice were generated.

The analysis of the T-DNA insertion line revealed a significant delay in the pollen tube growth and a lower ovule fertilization ratio under heat stress conditions, although the establishment of seed set was not compromised.

OP5

Unravelling cell identity acquisition in the brown alga *Ectocarpus*

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Brown algae are among the few eukaryotic lineages to have evolved complex multicellularity, which entails intricate biological processes such as cell identity acquisition. While some of these processes are homologous across lineages, brown algae's early divergence may have led to unique molecular processes, many of which are still unknown due to limited research. This study aimed to understand cell identity acquisition in the brown alga *Ectocarpus* by analysing mutants lacking basal cells with various microscopy techniques, including brightfield, fluorescence, expansion, and electron microscopy. Most of these mutants lacked a basal system and developed as upright filaments with a single undifferentiated cell on one tip. Compared with the wild-type Ec32, mutants released smaller mitospores, grew mainly in one direction after the first division, branched rarely, lacked secondary rhizoids, ended up smaller with fewer cells and presented altered reproductive structures. Preliminary intracellular data suggested differences in microtubule organisation, centriole and nuclear positioning, and cell wall structure. Together, these data link defects in early division and intracellular organisation to failed basal cell-type specification. Additionally, this extensive phenotypic data will be relevant for the functional characterisation of the still-unknown genes involved in the acquisition of basal cell identity. By focusing on *Ectocarpus*, the present work provides new insights regarding cell-type determination and the evolution of complex multicellularity in brown algae. Additionally, the use of cutting-edge techniques, such as expansion microscopy, expanded the toolbox of methods available for *Ectocarpus* and may be extendable to other brown algae.

OP6

Discovery of the interaction between SUMOylation and the functioning of telomeric complexes in plants

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Background & Aim: The small ubiquitin-like modifier (SUMO) interacts covalently with different proteins through a process known as SUMOylation. SUMOylation has been implicated in regulating various biological functions in different organisms by specifically controlling the activity of central proteins. In non-plant systems, it was found that SUMO contributes to telomere stability by interacting with telomeric proteins, namely members of the Telomere Repeat-binding Protein (TRP) and Telomere Repeat Binding (TRB) families. The reconstruction of a network mega-database of SUMO targets, showed that these two families are likely targeted for SUMOylation in plants. This project aimed to confirm that TRBs and TRPs are regulated by SUMO in plants with impact on telomere functioning.

Methods: *Arabidopsis thaliana* (Arabidopsis) was used as a model organism. A fine phenotypic analysis was performed along plant development using T-DNA insertion Arabidopsis mutants for SUMO machinery components and TRP/TRB families' members. SUMO association to telomere and resident proteins was inspected by colocalization and bimolecular fluorescence complementation (BiFC) assays in transient expression in *Nicotiana benthamiana*.

Results: Single mutants for *TRP2* and *TRBs* displayed no striking phenotype, whereas the triple *trb1/2/3* revealed drastic defects from early developmental stages. The BiFC assays showed that SUMO interacts with TRP2 and TRB2, whereas colocalization confirmed that SUMO was in the nucleus and co-localized with the telomeres.

Conclusions: This study identified telomere-associated components important for plant development, particularly TRPs and TRBs, as SUMO targets in Arabidopsis, indicating that SUMOylation plays a crucial role in telomere regulation in eukaryotes.

Session 2: Plant Microbe Interactions

OP7

INIAV - Integrating Science and Practice. Tackling Shoot Blight Disease in Stone Pine

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The National Institute for Agrarian and Veterinary Research (INIAV, I.P.) is the Portuguese public research institute dedicated to supporting national policies in agriculture, forestry, veterinary and food systems through scientific research, innovation, and technical services. Within INIAV, the Mycology Laboratory of the Plant Health Unit (SAFSV) integrates the National Reference Laboratory (LNR) for fungal plant pathogens, providing official diagnostic services, developing detection and identification methods, and leading research on plant-pathogenic fungi affecting both crops and forest species.

Stone pine (*Pinus pinea*) is one of the most important forest species in Portugal, special due to the high value of the pine nuts, which are the main resource of this forest industry. This pine tree is considered a robust species affected only by a few pathogens. Nevertheless, in recent years, shoot blight has been frequently reported in Portugal, causing serious concerns for the pine nut producers and industry. By hindering the development of cones, this disease might affect the production of pine nuts, bringing economical constraints. In order to identify the associated fungal pathogens, symptomatic plant material from stone pine orchards has been collected and analysed. Isolated fungi were identified based on morphological and phylogenetic analyses. Pathogenicity was tested on artificially inoculated plant material. Fungi belonging to *Pestalotiopsis spp*. and *Sydowia polyspora* were identified in association with these symptoms. *Pe. pini* was hable to cause disease on artificial inocculated seedlings. *S. polyspora* caused lesions on excised *P. pinea* branches. Henceforth, these fungi represent pathogens causing shoot blight on stone pine. Understanding the biotic interactions among the fungi colonizing stone pine shoots, as well as the abiotic factors influencing disease, may provide valuable clues for developing effective management strategies. Hence new research lines focus on identifying additional pathogens, unravelling possible synergistic effects, and developing rapid diagnostic tools and biocontrol solutions towards an integrated approach against shoot blight disease in stone pine.

OP8

Harnessing Bacteria to Boost Eucalyptus Growth and Immunity

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Forests occupy 39% of Portugal’s total area, predominantly with maritime pine (23%), eucalyptus (26%), and cork oak (23%) dominating [1]. *Eucalyptus globulus'* importance in the pulp and essential oil industry [2-3], has intensified plantation forestry and fertilizer use [4], leading to environmental impacts [5]. Therefore, it is necessary to find sustainable alternatives. Microbial fertilizers, including PGPRs, mycorrhizal fungi, and algae, can enhance yields and plant health when integrated with conventional methods [6-9].

Thus, this study aims to evaluate the influence of selected microorganisms on *E. globulus* growth, in order to establish a consortium with biofertilizer properties. Due to the bactericidal and fungicidal properties of *E. globulus*, bacteria and fungi from leaves and surface-disinfected roots were isolated in different media and subsequently identified by 16S rRNA gene analysis. Afterward, to assess the species' biofertilizer potential, *E. globulus* seeds were sown in Petri dishes containing agar-solidified Hoagland medium supplemented with bacterial extracts. Each bacterial strain was cultured in 10 mL of LB broth for 72 h, sonicated for 1.5 min, centrifuged at 4,000 rpm for 20 min, and the filtered supernatant was applied to the growth medium.

While these results are preliminary, some isolates have demonstrated the ability to stimulate plant growth. Isolates 76, 88, and 102 notably enhanced shoot development, with isolate 83 (*Pseudomonas canadensis*) showing particular effectiveness by promoting both shoot and root growth.

Therefore, this study is a significant step forward in developing a biofertilizer consortium for forest management to enhance *E. globulus* fertilization and support more sustainable production.

OP9

The efficiency of microorganisms in protecting Eucalyptus plants against *Neopestalotiopsis*

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Eucalyptus globulus is a forest species of great economic and ecological relevance, whose cultivation requires the optimisation of nutritional processes, the adoption of sustainable management practices, and the development of more effective pest control strategies to increase productivity. Among the main pathogenic agents affecting this species, the fungus *Neopestalotiopsis* stands out, as it causes necrosis, leaf loss, and a reduction in the quality and productivity of plantations. Its rapid spread and resistance to some forest protection alternatives have significant economic impacts on the pulp and paper industry — one of the main uses of eucalyptus' wood — and compromise plantation sustainability.

In response to these challenges, this work had the objective of evaluate the effectiveness of microorganism-based biofertilizers in the antifungal protection of *Eucalyptus globulus* plants inoculated with *Neopestalotiopsis*, testing formulations containing *Trichoderma asperellum*, *Trichoderma gamsii*, and *Bacillus pumilus* in plant genotypes with different susceptibilities. Growth, physiological, and biochemical parameters were assessed, as well as lesion severity and plant mortality.

The results showed that the effectiveness of biofertilizers depends strongly on genotype and application method. Both treatment with *Bacillus pumilus* and *Trichoderma* sp gave positive results in the more resistant genotype, when applied to the soil, reducing stem necrosis, visual symptoms, and mortality, while no beneficial effects were observed in the more susceptible genotype. Foliar application of *B. pumilus* fertilizer had no effect in protecting neither of the two genotypes.

Overall, the results indicate that biofertilizers based on *Trichoderma* and *Bacillus* have potential for integration into sustainable protection strategies for *E. globulus*.

OP10

Increased resistance to bacterial pathogens in winter forms of SWD is independent of stronger immune status

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Drosophila suzukii, commonly known as the spotted-wing drosophila (SWD), is a major invasive pest that originated in Asia but has now spread worldwide, posing a significant threat to soft fruit agriculture. A key factor contributing to its ecological success is its pronounced seasonal plasticity, characterized by distinct summer (SM) and winter morphotypes (WM) that exhibit different metabolic and phenotypic traits adapted to seasonal conditions. Winter morphotypes display increased melanization, stress tolerance, and longevity. However, their immune function remains poorly understood.

Drosophilids are highly responsive to temperature fluctuations, and exposure to cold can prime immune pathways by upregulating stress and defense-related genes, even in the absence of pathogens. In *D. melanogaster*, cold-induced immune priming has been documented, though its effects on pathogen resistance remain variable. For SWD, evidence for morphotype-specific immune responses is limited, despite its ecological and agricultural significance.

Here, we investigated immune variation between SWD morphotypes using a multifaceted approach that combined RNA sequencing, quantitative PCR, and survival assays following infection with the entomopathogenic bacterium *Pseudomonas entomophila*. Our results revealed marked transcriptional differences between morphotypes but only minimal variation in immune-related pathways. Nonetheless, WM individuals were less susceptible to infection, suggesting that enhanced tolerance, rather than differential immune activation, underlies their increased survival.

This study provides the first direct comparison of bacterial infection responses between SWD morphotypes, highlighting that seasonal adaptation extends to pathogen resistance and contributes to overwintering success, which has implications for understanding population dynamics and optimizing biological control strategies.

Session 3: Plant Physiology

OP11

The life of a stressed plant - *Plant Stress lab* hits the stage

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Climate change is altering temperature and precipitation patterns, increasing the frequency of droughts, floods, and heatwaves, which disrupt plant growth and reduce yields. Besides, soil salinization and contamination bring further challenges to plants, threatening agroecosystem dynamics. Developing climate-resilient crops and mitigation strategies is, therefore, essential to sustain global food production. The *Plant Stress lab*, a research group from GreenUPorto (FCUP), aims to tackle these issues by investigating how plants respond to abiotic stress caused by climate change and soil degradation, while also addressing the need to develop green mitigation tools. Integrating morpho-physiological, biochemical, and molecular approaches, our topics of action are categorized into three main research lines: i) unravelling the physiological and molecular basis of plant responses to combined stress; ii) testing of stress-alleviation tools; iii) developing new sources of biopesticides and valorization of organic residues. Interinstitutional R&D projects are tackling these priorities in multiple crops under single or combined drought, heat, salinity, and soil contaminant stress. Within the scope of stress-mitigation strategies, *CC&NUTS* aims to validate mycorrhization and stress-priming to enhance young chestnut trees' resilience to heat and drought. Also addressing climate change, the goal of *endoLEGUME* is to link the seed microbiome of local chickpea varieties to their stress susceptibility and to develop a microbial consortium that can promote this species' stress resilience. In terms of residue valorization, *Olive4Cereal* seeks to transform olive mill wastewater, a discarded by-product, into a sustainable biostimulant to improve drought resilience of cereal crops. Moreover, addressing soil degradation issues, the *epiAGRO_2.0* will explore how glyphosate affects non-target tomato plants at the transcriptomic and epigenetic levels, uncovering potential transgenerational effects that could compromise future plant resilience and agricultural sustainability.

OP12

Melatonin-treated tomato plants – a shield against glyphosate and copper toxicity?

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As the accumulation of pesticide residues in agricultural soils continues to pose a growing environmental challenge, compromising plant growth and development, the finding of sustainable tools to mitigate pesticide toxicity in crops is essential. Therefore, this study evaluated the potential of melatonin (MT), a plant growth regulator, to reduce the phytotoxic effects of copper (Cu) and glyphosate (GLY) co-exposure on tomato plants (*Solanum lycopersicum* L.). A preliminary seed priming assay (16h overnight) with increasing MT concentrations (0–100 µM) revealed a dose-dependent effect on germination and early plant growth, with 25 and 50 µM showing the highest biostimulant potential. Based on these results, 25 µM MT was selected for further studies, accompanied by a weekly foliar spray reinforcement (100 µM). Tomato plants were grown for 35 days in soil contaminated with Cu (75 mg kg^{-1}) and/or GLY (10 mg kg^{-1}). Results showed that while GLY exposure reduced plant biomass without significantly impacting the overall redox homeostasis, Cu, either single or combined with GLY, induced a stronger growth inhibition and oxidative damage, particularly in roots. MT supplementation (seed priming and foliar spray) promoted growth under optimal conditions and alleviated GLY-induced toxicity, restoring biomass and maintaining the redox balance. Still, MT's protective role was less evident in Cu-exposed plants, especially in those co-treated with GLY. Overall, MT acted as an effective growth biostimulant and a moderate stress mitigator, particularly under herbicide exposure. However, its capacity to counteract metal toxicity appears limited, emphasizing the need for integrated soil management approaches.

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OP13

Consequences of the exposure to silver on the *Arabidopsis thaliana*’s metallothionein gene family expression

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Silver (Ag) has been accumulating in the biosphere due to anthropogenic activities. Ag is a non-essential heavy metal, and exposure to Ag may cause stress, negatively influencing plant growth. This work aimed to study the effects of high concentrations of Ag on *Arabidopsis thaliana*’s expression rates of several metallothionein-encoding (MT) genes (type 1 (MT1 a-c), type 2 (MT2 a and b) and type 3 (MT3)). *A. thaliana*’s wild-type plants were exposed to 3 different Ag concentrations (0, 6.1 and 25.6 mg/L) in MS medium for 21 days after germination. Plants were separated into shoots and roots for RNA extraction, cDNA synthesis and RT-qPCR analysis. Biomass was also collected to quantify the Ag bioaccumulated. A one-way analysis of variance (One-way ANOVA) was performed, followed by Dunnett’s multiple comparison tests. The results with a *p*-value lower than 0.05 were considered statistically significant. In the shoots, only the highest Ag concentration results were statistically significant, with all genes showing an increased expression. In the roots, this same increase was only statistically significant for MT1a and c, while MT1b and MT2a decreased, although not significantly. The bioaccumulated Ag was higher in roots and increased with the added Ag to the growth medium. Still, no difference was seen in shoots in the lowest concentration, but a greater accumulation was observed with the higher concentration. These results hint at a relevant participation of MT for the bioaccumulation of Ag in an organ-specific way.

OP14

The Effect of Nisin Z On the Shelf-life of Strawberries in Post-Harvest Conditions

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Strawberries are non-climacteric fruits with an important content of sugar, fiber and bioactive compounds that benefit the health of the consumer. Together with their notable organoleptic properties, these berries are highly associated with a healthy lifestyle, an aspect that has led to an increase in their production over the last years. However, the commercialization of strawberries is hindered due to their short shelf-life, mainly caused by fungal infections, provoking post-harvest losses of 35%-50%.

Hence, in this work, strawberries of different varieties were treated with Nisin Z, an antimicrobial peptide with a preservative effect in dairy products but seldom studied in fruits. Various peptide concentrations were applied using spray and immersion techniques, determining that the immersion in 1 mg/mL Nisin Z significantly reduced the incidence and severity of fungal infections, preserving the strawberries for 7–15 days under cold storage. Isolates of *Alternaria* spp. and *Aspergillus* spp. were obtained from the treated strawberries, being identified through the amplification of their ITS region followed by Sanger sequencing. Spot-on-Lawn assays applying concentrations of 1 mg/mL–100 mg/mL on these isolates showed no inhibition halos. This result was confirmed with higher concentrations (12.5 mg/mL–500 mg/mL) tested against *Botrytis cinerea*, *Aspergillus fumigatus, Penicillium spp.,* and *Candida albicans*, suggesting that Nisin Z does not exert a direct antifungal effect but may instead induce priming mechanisms in the treated fruit, a hypothesis under investigation using omics, and biochemical studies.

OP15

Exploring the Potential of Induced Tetraploidy: Characterization of *Cannabis sativa* Cultivars

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This research project investigates the induction and characterization of polyploidy in a high-CBD, low-THC cultivar of *Cannabis sativa* named “CBD Zero”. The study was divided into two phases: the first phase focused on the induction and selection of tetraploid plants, while the second phase aimed to compare the morphological and chemical variations resulting from polyploidization. In the initial phase of the experiment, germinated *Cannabis sativa* seeds were treated with different concentrations of colchicine to induce polyploidy. The resulting plants were subjected to morphological and nuclear analyses to identify and select the most promising specimens to be used in the subsequent phase. During the second experimental phase, the selected plants were induced to flower, and volatile organic compounds (VOCs) were monitored *in vivo* both during the terminal stage of the vegetative phase and throughout the entire flowering period, up to harvest. After harvesting, the biomass was dried and separated into different tissues for ionomic analysis. The inflorescences were analyzed using HPLC to quantify the major cannabinoids. The results confirm that colchicine is an effective agent for inducing polyploidy in *Cannabis sativa*, with the most successful outcome observed at a treatment concentration of 0.05%. The polyploid plants exhibited significant differences in both morphological traits and biochemical profiles, including variations in terpene and cannabinoid content, compounds that play a key role in the medicinal properties of *Cannabis*.

Keywords: *Cannabis sativa*, Polyploidy, Colchicine, Cannabinoids, Terpenes

OP16

Kiwi-Kiss: Paving the way for awakening sleeping beauty

Searching for hallmarks of bud developmental transitions towards a proxy of the kiss of cold to break bud dormancy in kiwifruit

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Climate change, through warm winters, poses an increasing challenge to Portuguese kiwifruit growers in the rate and synchronization of budbreak, negatively impacting fruit yield and quality. In autumn, kiwifruit buds enter an endogenously regulated endodormancy state that is only released upon accumulation of over 700 hours below 7°C. Therefore, it's increasingly necessary to develop an efficient, environmentally friendly protocol for inducing synchronous budbreak in the field, replacing chilling requirements. To achieve this, several experimental strategies were designed and optimized for the kiwifruit, a woody species presenting specific challenges. Concerning budbreak regulation, key transcription factors were identified, and a ranked list of candidate inducers was built. RNA extraction methods were optimized, and RT-qPCR conditions were tested. The conditions for the study of anatomical developmental hallmarks were also established. Lugol staining revealed a starch accumulation pattern in the buds that changes throughout chilling, presenting a potential budbreak hallmark. Toluidine blue enabled the identification of many tissues, such as an idioblast-rich protective parenchyma surrounding the bud. Aniline blue staining of callose in phloem sieve plates, confirming its value to investigate callose dynamics during dormancy. Finally, a budbreak assay was successfully conducted, showing H_2O_2 to be promising for inducing both budbreak and flower bud differentiation. Overall, strategies to determine molecular and anatomical developmental hallmarks were optimized, and candidate budbreak inducers were assayed. Ultimately, we hope to provide kiwifruit growers with insights to mitigate the climatic impact, fostering stable and consistent productivity.

Book of abstracts

Poster Presentations

P1

Plant-Based Nature Solutions for Water Quality and Biodiversity

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Nature-based solutions (NbS) such as Floating Treatment Wetlands (FTWs) and Constructed Wetlands (CWs) are increasingly recognized for their capacity to enhance water quality while simultaneously delivering ecological and aesthetic benefits. This work integrates findings from experimental and bibliometric studies on FTWs and CWs, aiming to advance understanding of their multifunctional roles in water and landscape management. Field experiments demonstrated that FTWs established in rural stormwater ponds support diverse microbial and macroinvertebrate communities, notably promoting plant growth–promoting bacteria and Odonata species, which completed their full life cycles within the systems. Complementary bibliometric analysis identified three major research fronts on FTWs: nutrient and pollutant removal processes, functional mechanisms within system components, and the ecological role of plants. Despite growing field-scale evidence, standardization in design, operation, and maintenance remains a critical research gap. In parallel, the implementation of a horizontal subsurface flow CW treating variable sewage loads in a mountain tourism facility in Portugal confirmed the robustness of these systems, achieving high removal efficiencies. Moreover, the use of ornamental polycultures enhanced aesthetics and biodiversity, demonstrating additional socioeconomic value. Collectively, these studies reinforce the potential of NbSs as sustainable ecotechnologies for decentralized wastewater treatment, phytoremediation, and biodiversity enhancement. Future research should focus on integrating ecological engineering, local materials, and citizen science approaches to optimize performance and scalability of these systems.

P2

Exploring and Valorization of Vine Pruning Residues from *Vitis vinifera* cv.

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Viticulture generates large amounts of by-products, with vine pruning wood representing the main residue form vineyard management. Traditionally burned to prevent disease spread, these materials contribute to greenhouse gas emissions and environmental degradation. Their conversion into high-value, bio-based products aligns with the principles of sustainability and circular bioeconomy. This work aims to promote the valorization of pruning residues from *Vitis vinifera* transforming vineyard waste into biodegradable and multifunctional products, such as biostimulants, biochar, and geotextile materials, that can enhance plant growth, nutrition, and stress resilience while reducing the need for synthetic agrochemicals. Pruning residues were collected, dried, and characterized regarding physicochemical composition, including dry matter, ash, crude protein, nitrogen, and structural carbohydrates (NDF, ADF, ADL), as well as lignin, cellulose, and hemicellulose contents. Ethanolic extracts were prepared to evaluate their bioactive potential through total phenolic content and antioxidant activity assays (DPPH, reducing power, and TBARS). This approach promotes the use of vineyard residues as renewable sources of valuable compounds, reducing waste and fostering sustainable, circular, and eco-friendly viticulture.

Keywords: vine pruning residues, bio-based products, circular bioeconomy, sustainable viticulture.

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P3

Conservation of the menaced NW Portuguese populations of *Quercus lusitanica*

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Quercus lusitanica Lam. is a dwarf oak native to Portugal with remarkable conservation interest. The species has a sharp Atlantic distribution occurring in the extreme W areas of the Iberian Peninsula and Morocco. Predictive species distribution models, based on climate change scenarios, indicate significant loss of suitable areas and vast range shifts towards the scarce, threatened national NW communities.

Q. lusitanica forms small bushes and grows through rhizomes that confer an evolutionary-adaptive survival trait to recurrent fire cycles. The species has further ecological relevance since it acts as a pioneer species and is important in pre-climax stages of vegetation dynamics, possessing a unique capacity to provide the transitional conditions for progression into forest mature stages.

The overall goal of the work is to promote the intelligent restoration of the menaced *Q. lusitanica* populations from NW Portugal, by integrating population genomics, species distribution models, vegetative and seminal propagation, and landscape management. We want to engage public opinion and will encourage the use of the species in urban/rural landscaping, transforming *Q. lusitanica* in an iconic species representative of our natural heritage.

P4

Physiological and Biochemical Responses of Grapevine to Hydrogels and Endophytic Bacterial Inoculation

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Climate change in the last decades had a negative impact in Portuguese vineyards due to increase heat and drought stress. These changes threaten yield and quality, highlighting the need for innovative resilience strategies. The present study aimed to implement sustainable strategies in a commercial vineyard plot cv. Touriga Nacional, in Vila Nova de Foz Côa. The effect of hydrogels (HG) produced from vine pruning wood and two endophytic bacterial strains isolated from vineyard soils with grass cover, *Bacillus aryabhattai* (B140) and *Bacillus toyonensis* (B258), that can promote vegetative growth and increase tolerance to water deficit, was evaluated. Four treatments were tested: control, hydrogel application, and the two bacterial strains (B140 and B258). Afterwards, biochemical parameters (total phenols, flavonoids, *ortho*-diphenols, sugars, proteins, photosynthetic pigments, and proline) and physiological parameters (gas exchange and chlorophyll *a* fluorescence) were evaluated. The biochemical results revealed that B140 strains induced lower values of total chlorophyll and chlorophyll b, flavonoids, and *ortho*-diphenols, but significant increases in total phenols, proteins, proline, and sugars, reflecting defensive responses. Conversely, the HG treatment showed the lowest values of flavonoids, proline, proteins and total chlorophyll. Regarding fluorescence parameters, no significant differences were observed. However, plants with HG showed lower values of transpiration, stomatal conductance, and CO_2 assimilation, while those inoculated with bacteria recorded higher values, despite no significant differences were observed. This study demonstrates the potential of native endophytic microorganisms as effective complementary strategies to enhance grapevine resilience, supporting a more sustainable viticulture, adapted to future climate challenges in Portugal.

Keywords: Grapevine resilience, Climate change, Hydrogels, Endophytic strains.

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P5

Smart Tools for Sustainable Plant Health: The BIOMEHEALTH Approach

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BIOMEHEALTH is a Horizon Europe Twinning project that aims to reinforce the research and innovation capacities of the University of Porto (REQUIMTE/UP) in the field of plant pathogen detection and control under the pressures of climate change. In collaboration with Wageningen University & Research (The Netherlands) and the University of Bologna (Italy), the project addresses the urgent need for sustainable and data-driven approaches to safeguard plant health in an era of globalized agriculture and increasing climatic instability. The project integrates cutting-edge microbiome research, artificial intelligence, and synthetic microbial community (SynCom) engineering to study phytobiome–pathobiome interactions and develop innovative detection and control tools. Focusing on case studies in apple and pear crops affected by *Erwinia amylovora* and *Stemphylium vesicarium*, BIOMEHEALTH explores how climate change alters pathogen dynamics and host–microbiome relationships. Novel diagnostic strategies, including aerobiome-based detection, will be developed alongside sustainable biocontrol solutions to enhance crop resilience. Beyond scientific research, BIOMEHEALTH invests in capacity building, training, and knowledge transfer, fostering a new generation of researchers equipped to tackle emerging plant health challenges. The establishment of a Centre of Plant Pathogen Detection and Control in Portugal will ensure the long-term sustainability of these efforts and support dialogue among academia, industry, policymakers, and society. Aligned with the European Green Deal and Horizon Europe Cluster 6, BIOMEHEALTH contributes to strengthening European leadership in sustainable plant health, resilience, and food security under changing environmental conditions. Keywords: Plant health; Climate change; Microbiome; Pathogen detection; Sustainable agriculture.

P6

From the sea to the field, marine bacteria as potential plant bioherbicide

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With the world population now exceeding 8 billion, there is an urgent need to restructure, optimize and increase food production in the world. The agronomical sector worldwide faces numerous challenges, from low yields to the combat of pests, often relying on solutions that are neither eco-friendly nor sustainable. The ocean holds vast, untapped potential waiting to be explored. Research on its microorganisms has led to the discovery of a wide range of intriguing compounds. This study aims to explore another potential bioactivity of these intriguing microorganisms and their compounds, herbicidal activity. For this study, 3 strains of bacteria belonging to the Planctomycetota and Actinomycetota phyla (*Microbacterium oxydans* - Berg02_79, *Kocuria polarys* - PMIC_1H7A and *Rhodopirellula rubra* - LF2) were grown in three media (M600, M607 and 1:10M607), varying in concentrations of carbon and nitrogen sources. Crude compounds produced by these bacteria were extracted and applied to seeds of three model plants: *Arabidopsis thaliana* (a common weed), *Lactuca sativa* and *Lolium spp.*(two important crops). The morphological parameters of the plants were then analyzed. Additionally, the bacteria's ability to produce indole-acetic acid (IAA), was assessed. This study revealed that bacterial extracts from the strains LF2, PMIC_1H7A and Berg02_79 significantly affected the development of *A. thaliana*, particularly when grown in M607 and 1:10 M607 media. Extracts from LF2 extracts had no observable effect on L. sativa and *Lolium spp*. development. Regarding IAA production, strains Berg02_79 and PMIC_1H7A produced the highest levels among the tested bacteria, even in the absence of tryptophan. However, no correlation was found between IAA production and the biological effects of the extracts.

The bacterial strains tested in this study demonstrated great potential for producing bioherbicidal compounds, active over a common weed, *A. thaliana*. The inhibitory effects observed were not primarily due to IAA production, suggesting the presence of other bioactive substances. These findings highlight the potential for discovering novel compounds of agroeconomic interest.