XII SIBV

(Italian Society of Plant Biology)

CONGRESS

BARI, 11-14 SEPTEMBER 2023

CONGRESS LOCATION:
Department of Biosciences, Biotechnologies and Environment
Campus "E. Quagliariello" via Orabona 4, Bari
UNIVERSITY OF BARI ALDO MORO

S.I.B.V.
SOCIETA' ITALIANA di BIOLOGIA VEGETALE

UNIVERSITÀ DEGLI STUDI DI BARI ALDO MORO

ANTONIO FIORE

VITO LORUSSO LIFE SCIENCE SALES

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PICCIN

Committees

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Laura De Gara, Campus Bio-Medico University of Rome
Fiorella Lo Schiavo, University of Padua
Matteo Ballottari, University of Verona
Sara Cimini, Campus Bio-Medico University of Rome
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Endorsers

XII National Conference of the Italian Society of Plant Biology

Dipartimento di Bioscienze, Biotecnologie e Ambiente

Università degli Studi di Bari Aldo Moro

11-14 September 2023

PROGRAMME

11 September 2023

12:30-14:00 Registration and poster installation

14:00-14:30 **Opening ceremony**

Laura De Gara

Chiara Tonelli

Symposium 1

PLANT IMMUNITY AND PLANT-MICROBE INTERACTION

Chairs: **Michela Zottini**, *University of Padua*

Roberto De Michele, *CNR-IBBR of Palermo*

14:30-15:10 **Tina Romeis**

Leibniz Institute of Plant Biochemistry

Calcium signalling in plant immunity: how to vaccinate a plant?

15:10-15:40 **Giulia De Lorenzo**

Sapienza University of Rome

The evolution of plant immunity at the crossroad between defense and development: the role of damage-associated molecular patterns (DAMPs) in tissue injury

15:40-15:55 **Manuel Benedetti, University of L'Aquila**

Identification of a novel cellodextrin-dependent degrading activity against plant polyphenols

15:55-16:10 **Valentina Bigini, University of Tuscia**

Engineering danger sensing and signaling in plant immunity: use of oligosaccharins to enhance durum wheat resistance to fusariosis

16:10-16:25 **Sara Paola Nastasi, University of Milan**

Enhancing drought tolerance in Arabidopsis thaliana plants through beneficial grapevine endophytes: preliminary findings

16:25-17:00 **COFFEE BREAK**

17:00-17:15 **Eva Maria Gòmez Alvarez, Scuola Superiore Sant'Anna of Pisa**

Barley germination tolerance to submergence stress: a genetic and a microbiome approach

17:15-17:30 **Raffaella Balestrini, CNR, IPSP, Turin**

Combination of arbuscular mycorrhizal fungi and natural compounds for improving tomato resilience to environmental stresses

17:30-17:45 **Michele Perazzoli, University of Trento**

Psychrotolerant endophytic bacteria of wild alpine plants can improve cold tolerance in crops

17:45-19:00 **Round Table on "The state of the art of innovation in Agriculture"**

Moderator: **Giovanni Matera** Journalist for RAI3

Prof. Chiara Tonelli President of Italian Federation of Life Sciences

Prof. Piero Morandini, University of Milan

On. Paolo De Castro, Member of the Committee on Agriculture and Rural Development of the European Parliament

Dott.ssa Elena Sgaravatti, VicePresident of Assobiotech and President of Planta Rei Biotech Srl

Deborah Piovan, Agricultural Entrepreneur and Scientific Popularizer

19:00-20:30 **WELCOME PARTY**

12 September 2023

Symposium 2

FROM SIGNALLING TO DEVELOPMENT

Chairs: **Francesca Secchi**, *University of Turin*

Alex Costa, *University of Milan*

09:00-09:40 **Lieven De Veylder**

Ghent University

Restoring a damaged root stem cell niche

09:40-10:10 **Andrea Schubert, University of Turin**

Strigolactone signaling at the interface of plant development and a changing environment

10:10-10:25 **Ilaria Fraudentali, Roma Tre University**

Distinct roles of AtCuAOβ and RBOHD in wound-induced local and systemic leaf-to-leaf and root-to-leaf stomatal closure in Arabidopsis

10:25-10:40 **Riccardo Lorrai, Sapienza University of Rome**

Turgor-sensitive responses link cell wall integrity to a signalling module promoting apical hook formation in *Arabidopsis thaliana*

10:40-10:55 **Vladimir Valkov Totev, Institute of Bioscience and BioResources, CNR, Naples**

The *Lotus japonicus* NPF4.6 is a KNO3 and ABA transporter involved in the lateral root elongation process

10:55-11:30 **COFFEE BREAK**

11:30-11:45 **Matteo Pivato, University of Verona**

Compartment-specific Ca^{2+} imaging in the green alga *Chlamydomonas reinhardtii* reveals high light-induced chloroplast Ca^{2+} signatures

11:45-12:00 **Alberto Tamborrino, Università of Padua**

Unveiling the mitochondrial unfolded protein response (UPRmt) in *Arabidopsis thaliana*

12:00-12:15 **Nicolaj Jeran, University of Milan**

Study of three putative Plastid Peptide Transporters mediating chloroplast-to-nucleus signalling in response to folding stress in *Arabidopsis thaliana* chloroplasts

12:15-13:30 **POSTER SESSION A**

13:30-14:30 **LUNCH**

Symposium 3

BIOTECHNOLOGICAL APPROACHES FOR CIRCULAR BIO-ECONOMY AND SUSTAINABILITY

Chairs: **Maria Manuela Rigano**, *University of Naples Federico II*

Giuseppe Forlani, *University of Ferrara*

14:30-15:10 **Vassilis Fotopoulos**

Cyprus University of Technology

Next generation chemical priming as a green strategy for sustainable agriculture

15:10-15:40 **Tomas Morosinotto**

University of Padua

Genetic engineering approaches to produce proteins, lipids and bioplastics from algae.

15:40-15:55 **Daniel Savatin, University of Tuscia – DAFNE**

Re-Waste: a green, sustainable and circular strategy for crop resilience

15:55-16:10 **Stefano Cazzaniga, University of Verona**

Enhancing biomass productivity and mitigating photoinhibition under high light through biotechnological engineering of astaxanthin accumulation in *Chlamydomonas reinhardtii*

16:10-16:25 **Ilaria Colzi, University of Florence**

Improving chromium removal from wastewater: the use of natural biostimulants towards circular economy

16:25-17:00 **COFFEE BREAK**

17:00-17:15 **Enrico Doria, University of Pavia**

A sustainable biotech system to extract bioactive compounds from vegetable waste and by-products

17:15-17:30 **Moira Giovannoni, University of L’Aquila**

Investigating the potential of microalgae-microbe interactions for efficient extraction of algal metabolites

17:30-19:00 **ELEVATOR PITCHES**

13 September 2023

Symposium 4

PLANT ADAPTATION TO ENVIRONMENTAL CONDITIONS

Chairs: **Fiorella Lo Schiavo**, *University of Padua*

Matteo Ballottari, *University of Verona*

09:00-09:40 **Frank Van Breusegem**

Ghent University

Hydrogen peroxide signalling in plants

09:40-10:10 **Maria Concetta de Pinto**

University of Bari

Redox signalling in plant heat stress response

10:10-10:25 **Ambra Selene Parmagnani, University of Turin**

The geomagnetic field (GMF) modulates *Arabidopsis thaliana* ROS metabolomics and transcriptomics

10:25-10:40 **Marco Dainelli, University of Florence**

Disturbance of nitrogen-fixing symbiotic interactions by PET micro/nanoplastics: the case of *Azolla filiculoides* Lam. and *Anabaena azollae*

10:40-10:55 **Sara Cimini, Campus Bio-Medico University of Rome**

New insights into durum wheat salt stress tolerance

10:55-11:30 **COFFEE BREAK**

11:30-11:45 **Gianpiero Vigani, University of Turin**

Exploring durum wheat germplasm to minimize drought impact on the nutritional status of plants

11:45-12:00 **Francesca Silvana, University of Naples "Federico II"**

Dissection of heat and drought tolerance traits in a *Solanum pennellii* introgression tomato line

12:00-12:15 **Yuri Telara, Sant'Anna School of Advanced Studies**

Plant responses to complex environmental stresses: investigation of molecular crosstalk between low oxygen and iron deficiency

12:15-12:30 **Guido Domingo, University of Insubria**

Phosphorylation-mediated regulation of alternative splicing in plant heat stress response with a focus on the role of cAMP

12:30-13:30 **POSTER SESSION B**

13:30-14:30 **LUNCH**

Symposium 5

IMPROVING CARBON ASSIMILATION FOR PLANT PRODUCTION

Chairs: **Mirko Zaffagnini**, *University of Bologna*

Andrea Nardini, *University of Trieste*

14:30-15:10 **Johannes Kromdijk**

University of Cambridge

Genetic determinants of photosynthetic variation in field-grown MAGIC maize

15:10-15:40 **Paolo Trost**

University of Bologna

Shedding light on dark complexes of the Calvin-Benson cycle

15:40:15:55 **Emily Rose Palm, University of University of Milan-Bicocca**

Constitutive sodium compartmentalization patterns differentially affect biomass production of two salt-treated edible halophytes

15:55:16:10 **Claudia Beraldo, University of Padua**

Physcomitrium patens flavodiiron proteins for the adaptation to changing environmental conditions

16:10:16:25 **Simone Barera, University of Ferrara**

Evidence from an *Arabidopsis thaliana* p5cdh mutant strengthening the occurrence of a proline-P5C cycle in plants

16:25-17:00 **COFFEE BREAK**

17:00-17:15 **Manuel Bellucci, Campus Bio-Medico University of Rome**

Novel function of isoprene in root physiology and salt stress tolerance

17:15-17:30 **Libero Gurrieri, University of Bologna**

Increasing phosphoribulokinase affinity to improve CO_2 flux: testing the approach and first results

17:30-17:45 **Sara Gargiulo, University of Udine**

Krebs cycle substrates starvation explains the decrease of O_2 consumption induced by hexanoic acid in pea roots

17:45-19:00 **General Assembly of the Italian Society of Plant Biology**

20:30-23:30 **Social Dinner**

14 September 2023

Symposium 6

NOVEL TECHNOLOGIES FOR MULTISCALE LEVELS OF INVESTIGATION IN PLANT PHYSIOLOGY

Chairs: **Laura De Gara**, *University of Rome Campus Bio-Medico*

Anca Macovei, *University of Pavia*

09:00-09:40 **Manuel Rodriguez-Concepcion**

Spanish National Research Council

Coloring games: new ways of making and storing health-promoting carotenoid pigments in plant cells

09:40-10:10 **Francesco Loreto**

University of Naples Federico II

Plant phenotyping: past achievements, state of the art and future perspectives

10:10-10:25 **Davide Patono, University of Turin**

Effects of light quality on carbon assimilation/emission and growth of lettuce: use of an upgraded prototype-platform to unravel mechanisms of photosynthesis/respiration control

10:25-10:40 **Gjata Isidora, University of Bari**

Physiological response of Lens culinaris Medik. to cerium and neodymium chlorides exposure

10:40-10:55 **Brunetti Cecilia, National Research Council, Institute for Sustainable Plant Protection, Sesto Fiorentino**

Investigating holm oak forest dieback through a multiscale approach: physiological measurements and visual assessment data with remote sensing

10:55-11:30 **COFFEE BREAK**

11:30-11:45 **Tricerri Niccolò, University of Turin**

Does slow and fast wilting rates affect the recovery from embolism in poplar seedlings? New insights from micro-CT analysis

11:45-12:00 **Francesca Resentini, University of Milan**

New tools to study in vivo the dynamics of Ca^{2+} in plant mitochondria

12:00-12:15 **Antony Surano, National Research Council, Institute for Sustainable Plant Protection, Bari**

Exploring different phenotypic and molecular approaches to unravel early events in olive–Xylella interactions

12:15-13:00 **CONGRESS CLOSURE**

Elevator Pitches

Plant Immunity and Plant Microbe Interaction

German Dario Ahumada, Scuola Superiore Sant'Anna, Pisa

Bacterial endophytes contribute to rice seedling establishment under submergence.

Andrea Tonanzi, Sapienza University of Rome

Elicitor-Induced Transgenerational Priming of Defense Responses in *Arabidopsis thaliana*

From signaling to development

Laila Moubayidin, John Innes Centre, Norwich, United Kingdom

Post-translational modification of SPATULA by SECRENT AGENT and SPINDLY promotes organ symmetry transition at the gynoecium apex

Sri Amarnadh Gupta Tondepu, University of Pavia

Evidence of miRNA regulation on the DNA damage response during germination of the irradiated wheat seeds

Paolo Maria Triozzi, Scuola Superiore Sant'Anna

Spatiotemporal internal oxygen dynamics define cyclic hypoxia in plants

Biotechnological Approaches for Circular BIO-Economy

Rachele Ingrisano, University of Bologna

Effect of the trophic regime on the growth of microalgae

Antonella Gori, University of Florence

Nutritional and nutraceutical composition of italian wild pear (*Pyrus communis* var. zingaro) at different maturation stages: a comparative analysis of peel and pulp

Plant adaptation to climate changes

Lucia Biruk, University of Trieste

Better safe than sorry: the surprising drought tolerance of a wetland sedge (Cyperus alternifolius)

Ilva Licaj, University of Sannio

Digging up through Artificial intelligence how two wheat cultivars response to Polyethylene Glycol-Simulated Drought Stress by analyzing root morpho-anatomical traits

Cristina Pagliano, University of Piemonte Orientale

Physiological responses to salt stress in wild and domesticated rice

Chiara Pagliarani, National Research Council (CNR) of Turin, IPSP

Exploiting somaclonal variability to increase drought stress tolerance in grapevine

Improving carbon assimilation for plant production

Sara Natale, University of Padua

Biodiversity of photosynthetic response in various bryophyte accessions

Erika Bellini, Sapienza University of Rome

Efficient utilization of monosaccharides from agri-food by-products supports Chlorella vulgaris biomass production

Novel Technology for multiscale level of investigation in plant physiology

Bianca Maria Orlando Marchesano, University of Milan

New state-of-the-art imaging tools to study how crops adapt to environmental changes: *Lycopersicum esculentum* key study

Teodora Chiara Tonto, Campus Bio-Medico University of Rome

Methodological pipeline for monitoring post-harvest quality of leafy vegetables

ORAL COMMUNICATION

PLANT IMMUNITY AND PLANT-MICROBE INTERACTION

76 - Identification of a novel cellodextrin-dependent degrading activity against plant polyphenols

Manuel Benedetti [(1)] - Anna Scortica [(1)] - Valentina Scafati [(1)] - Moira Giovannoni [(1)] - Benedetta Mattei [(1)]

Università dell'Aquila, Dipartimento di Medicina clinica, sanità pubblica, scienze della vita e dell'ambiente, L'Aquila, Italy [(1)]

The Arabidopsis cellodextrin-oxidase (CELLOX) is a berberine bridge enzyme-like flavoenzyme that catalyses the oxidation of elicitor active-cellodextrins (CDs). The enzymatic oxidation of CDs suppresses their ability of inducing plant defences, pointing to CELLOX as a key enzyme in the modulation of growth defence trade-off. Kinetic studies showed that CELLOX is a versatile oxidoreductase that acts as oxidase or dehydrogenase depending on the presence of specific electron acceptor/donor combinations. In vitro, CELLOX receives the electrons from the reducing end of CDs and uses them either to reduce O2 to H2O2 by acting as oxidase or to reduce the synthetic radical cation 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)•+ (ABTS•+) to ABTS by acting as dehydrogenase. To identify the natural electron acceptors involved in the reaction, the activity of CELLOX was tested in the presence of different plant secondary metabolites. By using CDs as electron donors, CELLOX succeeded in depolymerizing “tetra-guaiacol”, a tetra-phenolic compound obtained by the oxidative polymerization of the coniferyl alcohol analogue “guaiacol”. This result clearly demonstrates that the reducing power of cell wall oligosaccharides can be used by oligosaccharide-oxidases to assist the dismantling of polyphenolic compounds. The involvement of CELLOX in the CD-dependent catabolism of polyphenols such as lignin provides novel notions for a better understanding of plant cell wall metabolism in the context of growth defence trade-off.

101 - Engineering danger sensing and signaling in plant immunity: use of oligosaccharins to enhance durum wheat resistance to fusariosis

Valentina Bigini (1) - Fabiano Sillo (2) - Sarah Giulietti (1) - Daniela Pontiggia (3) - Luca Giovannini (2) - Raffaella Balestrini (2) - Daniel V. Savatin (1)

University of Tuscia, Department of Agriculture and Forest Sciences, Viterbo, Italy (1) - Institute for Sustainable Plant Protection, National Research Council, Torino, Italy (2) - Sapienza University of Rome, Department of Biology and Biotechnology 'Charles Darwin', Roma, Italy (3)

Fusariosis causes substantial yield losses in wheat crop worldwide and compromises food safety because of the presence of toxins associated to fungal disease. Among the current approaches to crop protection, the use of elicitors able to activate natural defense mechanisms in plants represents a strategy gaining increasing attention. Several studies indicate that local application of plant cell wall-derived elicitors, such as oligogalacturonides (OGs), derived from partial degradation of pectin, induces systemic resistance against plant pathogens. The aim of this study was to establish the efficacy of OGs in protecting durum wheat, characterized by an extreme susceptibility to Fusarium graminearum (Fg). To evaluate the functionality of OGs, spikes and seedlings of cv. Svevo were inoculated with OGs, Fg spores and a co-treatment of both. Both bioassays and transcriptomic analyses demonstrated that relevant physiological processes in Svevo are differently regulated by diverse amounts of OGs. Moreover, results demonstrated that OGs are active elicitors of wheat defenses, triggering typical immune marker genes as well as a regulation of fungal genes. Furthermore, engineered durum wheat plants with potentially altered endogenous OG levels, i.e. OG-Machine lines, were generated, characterized and used to evaluate the possible OG involvement in orchestrating wheat responses to Fg infection. Interestingly, the OG-Machine lines displayed a higher resistance to Fg compared to cv. Svevo.

85 - Enhancing drought tolerance in Arabidopsis thaliana plants through beneficial grapevine endophytes: preliminary findings

Sara Paola Nastasi [(1)] - Francesca Resentini [(1)] - Davide Pacifico [(2)] - Roberto De Michele [(2)] - Michela Zottini [(3)] - Alex Costa [(1)]

Università di Milano, Dipartimento di Bioscienze, Milano, Italy [(1)] - Istituto di Bioscienze e BioRisorse, Dipartimento di Scienze Bio-Agroalimentari, Palermo, Italy [(2)] - Università di Padova, Dipartimento di Biologia, Padova, Italy [(3)]

In the last decades, the global temperature increase is causing dramatic changes in environmental conditions. The Mediterranean basin is one area subjected to warming and figures the growing of the most economically and culturally important crops including grapevine (Vitis vinifera sp.), a woody perennial plant which is particularly sensitive to arid conditions and water deficiency.

To limit damages on grapevine plant productivity, sustainable strategies applicable on field are needed. In this context, the European Project PROSIT (Plant micRObiome in Sustainable vITiculture) has the aim to exploit and identify beneficial microbial consortia to be used as environmental-friendly practices to overcome the problem of drought.

Drought stress causes reduction of plant yield and, in turn, plants limit the evapotranspiration by closing their stomata. One of the first physiological response of plants to drought stress is the biosynthesis of the phytohormone abscisic acid (ABA) which triggers stomatal closure.

We hypothesized that beneficial endophytes isolated from Sicilian grapevine varieties, resilient to arid conditions, could be employed to mitigate drought stress tolerance of plants potentially through the modulation of ABA levels. In order to test this hypothesis, we used the recent development of Arabidopsis thaliana plants expressing the genetically encoded ABA sensors ABACUS2.

Here, we report an initial characterization of Arabidopsis thaliana ABACUS2 lines to assess their functionality. Then, we started to test the impacts of the selected beneficial microbes on A. thaliana development and their possible alteration on ABA content in seedlings.

Drought stress experiments with plants expressing ABACUS2 biosensors inoculated or not with the selected endophytic consortia are ongoing.

25 - Barley germination tolerance to submergence stress: a genetic and a microbiome approach

Eva Maria Gómez-Álvarez (1) - Alessandro Tondelli (2) - Khac Nhu Nghi (1) - Viktoriia Voloboeva (1) - Guido Giordano (1) - Giampiero Valè (3) - Germán Darío Ahumada (1) - Monique Salardi-Jost (4) - Matteo Dell’Acqua (4) - Pierdomenico Perata (1) - Chiara Pucciariello (1)

Scuola Superiore Sant'Anna, PlantLab, Center of Plant Sciences, Pisa, Italy (1) - Council for Agricultural Research and Economics (CREA), Research Center for Genomics and Bioinformatics, Fiorenzuola d'Arda, Italy (2) - Università del Piemonte Orientale, Dipartimento per lo Sviluppo Sostenibile e la Transizione Ecologica, Vercelli, Italy (3) - Scuola Superiore Sant'Anna, Crop Genetics Lab, Center of Plant Sciences, Pisa, Italy (4)

Climate change has led to a dramatic increase in flooding, with a strong effect on plant productivity. Barley is one of the most important cereals and it is impacted considerably by flooding. At the initial growth stage, this can cause severe yield losses, which create the need for screening for flooding tolerance traits. To explore this aspect, we analysed a large panel of barley accessions for tolerance to submergence followed by recovery during germination, using Genome Wide Association Studies to identify candidate genes responsible for the trait. Among the loci identified, a laccase gene was found to be involved in tolerance, modifying seed permeability. Our hypothesis is that seed permeability plays a role in activating secondary dormancy under flooding, strongly influencing the capacity of barley to germinate promptly during the subsequent recovery period. Although plant genetic differences account for variations in the flooding-dependent phenotypes, peculiar aspects of the seed microbiota can also contribute to seedling establishment. We conducted a metagenomic analysis on tolerant and sensitive barley grains. Our results suggest the presence of a dissimilar microbiome availability in tolerant versus sensitive accessions of barley and support the hypothesis that this difference may be responsible of some activities involved in germination after submergence. Taken together, these results shed light on the complexity of the trait and enlarge the breeder’s toolbox.

7 - Combination of arbuscular mycorrhizal fungi and natural compounds for improving tomato resilience to environmental stresses

Luca Giovannini [(1)] - Eva CAÑIZARES [(2)] - Chiara Pagliarani [(1)] - Walter Chitarra [(3)] - Luca Nerva [(3)] - Elisa Zampieri [(1)] - Fabiano Sillo [(1)] - Andrea IOANNOU [(4)] - Alexandros SPANOS [(4)] - Federico Vita [(5)] - Miguel GONZÁLEZ-GUZMÁN [(2)] - Vasileos FOTOPOULOS [(4)] - Vicent Arbona [(2)] - Raffaella Balestrini [(1)]

CNR, IPSP, Torino, Italy [(1)] - Universitat Jaume I, Departament de Biologia, Bioquímica i Ciències Naturals, Castelló de la Plana, Spain [(2)] - CREA, Research Centre for Viticulture and Enology, Conegliano, Italy [(3)] - CUT, Department of Agricultural Sciences, Biotechnology and Food Science, Limassol, Cyprus [(4)] - Università di Bari, Department of Bioscience, Biotechnology and Environment, Bari, Italy [(5)]

The Mediterranean area is particularly susceptible to the incidence of adverse abiotic and biotic conditions derived from climate change that will severely impact agricultural production. Priming modulates plant stress responses before the stress factor appears, increasing the ability of the primed plant to endure the adverse conditions and thrive. This can be achieved by the application of different compounds and/or microorganisms (chemical and biological priming, respectively). In this context, we are investigating the priming effect of different natural compounds (i.e., chitosan, salicylic acid, melatonin, and their combinations) and biological (i.e., arbuscular mycorrhizal fungi, AMF) agent combinations against the concurrent incidence of water deficit and salinity on commercial tomato genotypes and local landraces to improve tolerance and resilience. To optimize the use of the considered priming agents, a seed priming protocol has been developed to generate more resilient and climate-flexible plants. As a first step, the effect of the chemical priming compounds on the AMF colonization has been evaluated, demonstrating the possibility of using them in combination. Next, plant responses to water and salt stress responses has been evaluated on primed seeds. The trials were carried out on potted plants by subjecting them to water deficit (by withholding irrigation up to 35% of water holding capacity) and salt stress (by exposing plants to progressively higher NaCl concentrations up to 200 mM). The impact on plant fitness of the several treatments has been studied using a combined approach, including eco-physiology, biochemical analysis, hormonal profiles, transcriptomics, and untargeted metabolomics.

4 - Psychrotolerant endophytic bacteria of wild alpine plants can improve cold tolerance in crops

Michele Perazzolli (1) - Irma Milanese (1) - Ahmad Alhariri (1) - Melissa Alussi (1) - Malek Marian (1)

University of Trento, Center Agriculture Food Environment, Trento, Italy (1)

Climate change is causing warmer winter and spring periods with increased frequency of chilling and frost damage on crop plants. Plant-associated bacteria are supposed to contribute to plant tolerance against abiotic stresses, but scarce information is available on the role of endophytic bacteria in the mitigation of cold stress. The aim of this project was to taxonomically and functionally characterize the endophytic bacterial communities associated with cold-adapted plants belonging to the Rosaceae family. The bacterial community structure associated with flowers, leaves, and roots of Alchemilla sp., Dryas octopetala, and Geum montanum differed according to the plant tissue and plant species, while it was scarcely affected by the collection site in alpine areas. Some psychrotolerant bacterial isolates were able to promote the growth of tomato seedlings at chilling temperatures (10°C and 15°C), reducing the accumulation of reactive oxygen species. Moreover, these psychrotolerant bacterial isolates reduced frost damage (-6°C) on strawberry and apple plantlets, limiting the electrolyte leakage of leaf tissues. These results provided better information on the structure and function of psychrotolerant endophytic bacteria associated with alpine plants and suggested a possible use of bacterium-based inoculants to improve cold tolerance in agronomically important crops.

35 - Distinct roles of AtCuAOβ and RBOHD in wound-induced local and systemic leaf-to-leaf and root-to-leaf stomatal closure in Arabidopsis

Ilaria Fraudentali (1) - Chiara Pedalino (2) - Riccardo D'Incà (1) - Paraskevi Tavladoraki (2) - Riccardo Angelini (2) - Alessandra Cona (1)

Roma Tre University, Department of Science, Roma, Italy (1) - Roma Tre University, Department of Science, Rome, Italy (2)

In plants, abiotic stress-induced signaling involves H2O2 production that mediates a wide range of physiological responses among which stomatal closure. Recently, it has been reported that amine oxidases contributes to H2O2 production in guard cells through polyamine catabolism, together with NADPH oxidases, the best-known sources of apoplastic ROS in these cells. The Arabidopsis copper-containing amine oxidase β (AtCuAOβ) was reported to be involved in stomatal closure induced by the wound-signal MeJA, as well as to mediate early differentiation of root protoxylem induced by leaf wounding, which suggests a role of this enzyme in the whole-plant coordination of water supply and loss through events of stress-induced phenotypic plasticity. Moreover, also isoform D of the respiratory burst oxidase homolog (RBOH) has been shown to be involved in stress-mediated modulation of stomatal closure. Here, the specific role of AtCuAOβ and RBOHD in local and systemic perception of leaf and root wounding that triggers stomatal closure was investigated at both injured and distal sites. Data evidenced that AtCuAOβ-derived H2O2 production mediates both local and systemic (leaf-to-leaf and root-to-leaf) stomatal closure, while RBOHD-driven ROS production contributes exclusively to systemic stomatal closure. Furthermore, data allow to hypothesize that RBOHD may act downstream of and cooperate with AtCuAOβ in inducing the oxidative burst that leads to systemic wound-triggered stomatal closure.

38 - Turgor-sensitive responses link cell wall integrity to a signalling module promoting apical hook formation in Arabidopsis thaliana

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Upon germination in the dark, dicotyledonous seedlings form an apical hook to protect young meristems from mechanical damage during soil emergence. Hook formation is a developmental process that results from differential growth on both sides of the hypocotyl apex and that is tightly controlled by environmental cues and hormones, among which auxin and gibberellins (GAs) are the main contributors. Cell wall composition is involved in the regulation of plant morphogenesis in general, but whether and how feedbacks from the cell wall contribute to apical hook development is still unclear. Here we show that dark-grown seedlings of Arabidopsis thaliana defective in pectin or cellulose biosynthesis, two main polysaccharidic components of the cell wall, displays severe defects in apical hook development, accompanied by loss of asymmetric auxin maxima and differential cell expansion. Moreover, our results suggest that disruption of hook formation in seedlings with altered cell wall are at least partially mediated by: i) the repression of a positive signalling module composed by HOOKLESS1 (HLS1) and PHYTOCHROME-INTERACTING FACTORs (PIFs), ii) the stabilization of the negative regulator of GAs signalling REPRESSOR OF ga1-3 (RGA) which is caused by iii) reduction of GAs levels. Accordingly exogenous GAs or HLS1 overexpression partially restored hook development in seedlings with defective cell wall. Notably, relieving cell wall stress throughout the reduction of turgor pressure also restored hook development and WT-like levels of the signalling elements that regulate it. Thus, we propose that turgor-dependent signals link cell wall integrity to a signalling module that controls differential cell elongation during hook development.

59 - The Lotus japonicus NPF4.6 is a KNO3 and ABA transporter involved in the lateral root elongation process

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Nitrate is an essential element for plant growth, both as a primary nutrient and as an important signal for plant development. In plants, the cross-talk between abscisic acid (ABA) and external nitrate has been recognized as a crucial mechanism for tuning root elongation in order to satisfy plant nutritional demands.

We present the functional characterization of the Lotus japonicus gene NRT4.6. Root expression analyses indicated that LjNPF4.6 is a nitrate and ABA responsive gene. Biochemical characterizations carried out in Xenopus laevis oocytes injected with LjNPF4.6 cRNA confirmed the involvement of LjNPF4.6 in nitrate- and ABA-related pathways.

The spatial profile of prom-LjNPF4.6-gusA expression in transgenic hairy roots grown on 12mM KNO3 was confined to the epidermal cell layer, root hairs and vascular bundle structures. In the absence of nitrate, GUS activity was mainly detected in the epidermal zone, whereas the addition of ABA induced the expression in the vascular bundles. Furthermore, the phenotypic characterization of three independent knock out insertion mutants revealed a nitrate-dependent defect in the root elongation process and an increased tolerance to the inhibitory effect induced by external ABA, on lateral root elongation. We will discuss the possible involvement of LjNPF4.6 in the nitrate/ABA cross talk controlling the root elongation process.

107 - Compartment-specific Ca2+ imaging in the green alga Chlamydomonas reinhardtii reveals high light-induced chloroplast Ca2+ signatures

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To investigate the role of intracellular Ca2+ signaling in the perception and response mechanisms to light in unicellular microalgae, the genetically encoded ratiometric Ca2+ indicator Yellow Cameleon (YC3.6) was expressed in the model organism for green algae Chlamydomonas reinhardtii, targeted to cytosol, chloroplast, and mitochondria. Through in vivo single-cell confocal microscopy imaging, light-induced Ca2+ signaling was investigated in different conditions and different genotypes, including the photoreceptors mutants phot and acry. A genetically encoded H2O2 sensor was also adopted to investigate the possible role of H2O2 formation in light dependent Ca2+ signaling. Light dependent Ca2+ response was observed in Chlamydomonas reinhardtii cells only in the chloroplast as an organelle-autonomous response, influenced by light intensity and photosynthetic electron transport. The absence of blue and red-light photoreceptor aCRY strongly reduced the light dependent chloroplast Ca2+ response, while the absence of the blue photoreceptor PHOT had no significant effects. A correlation between high light-induced chloroplast H2O2 gradients and Ca2+ transients was drawn, supported by H2O2 induced chloroplast Ca2+ transients in the dark. In conclusion, different triggers are involved in the light induced chloroplast Ca2+ signaling as saturation of the photosynthetic electron transport, H2O2 formation and aCRY-dependent light perception.

78 - Unveiling the mitochondrial unfolded protein response (UPRmt) in Arabidopsis thaliana

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Mitochondria are key organelle involved in many different cellular processes, from the production of energy to stress sensing and response. However, to preserve proper organelle function, is necessary to maintain mitochondrial protein homeostasis. This is usually assured by the action of the mitochondrial protein quality control (mtPQC), a complex network of chaperones and proteases. However, when there is an accumulation of misfolded/unfolded proteins that goes beyond its capability the cell mounts the mitochondrial unfolded protein response (UPRmt). UPRmt is a mitochondrial stress response by which mitochondria promote the expression of nuclear-encoded genes to restore organelle function.

In this work we investigate proteotoxic stress and UPRmt in Arabidopsis thaliana. Mutant plant lines showing a constitutively active UPRmt have been identified and characterized. Mitochondrial morphology and dynamics, Ca2+ signaling and stress marker gene expression profile were determined.

54 - Study of three putative Plastid Peptide Transporters mediating chloroplast-to-nucleus signalling in response to folding stress in Arabidopsis thaliana chloroplasts.

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Chloroplasts are DNA-containing organelles that originated from the endosymbiosis with cyanobacteria ancestors. The physical separation of nuclear and plastid genomes drove the evolution of signalling pathways that ensure coordinated nuclear and plastid gene expression, throughout development and in response to the environment. Albeit the signalling molecules behind the chloroplast-to-nucleus communication are still not fully understood, pieces of evidence suggest that peptides derived from proteolysis could act as signalling molecules. Studies conducted in nematode and yeast have revealed that mitochondrial ABC transporters mediate the extrusion of peptides from mitochondria to activate the mtUPR in response to heat. In this context, we identified three putative Plastid-located Peptide Transporters (PPTs) in the Arabidopsis genome. Lack of PPT proteins reduces the peptide extrusion from chloroplasts upon heat shock, altering the regulation of CLPB3 plastid unfoldase and increasing heat sensitivity. The extruded peptides were analysed via mass spectrometry revealing that they mostly belonged to fragile Photosystem II proteins. In addition, PPTs are capable of complementing heat-sensitive phenotypes in yeast, corroborating their function as peptide extruders.

BIOTECHNOLOGICAL APPROACHES FOR CIRCULAR BIO–ECONOMY AND SUSTAINABILITY

124 - Re-Waste: a green, sustainable and circular strategy for crop resilience

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Huge amounts of organic waste are generated by agriculture and food industries, whereas, in the EU, an estimated 20% of the total food produced is lost or wasted. Food waste is rich in biomolecules, able to cause changes in crop important physiological processes. The purpose of the current study was to exploit waste by-product to develop a sustainable strategy for crop plant growth and resilience to (a)biotic stresses. We, thus, conducted agronomic trials on two model species for agriculture (wheat and tomato) by using agro-food waste, that was compacted and dehydrated, and the obtained solid dry residue (SDR) was directly added to the growth substrate. These SDRs represent a potential reservoir of bioactive products, including oligosaccharins, such as chitooligosaccharides and oligogalacturonides, both potential alternative to traditional agrochemicals. We propose here a novel, safe and sustainable strategy based on a model that promotes circular economy by recycling wet-organic waste, replacing chemical fertilizers, reducing pollution and high management costs.

100 - Enhancing Biomass Productivity and Mitigating Photoinhibition under High Light through Biotechnological Engineering of Astaxanthin Accumulation in Chlamydomonas reinhardtii

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Astaxanthin, a valuable ketocarotenoid with strong antioxidative activity, naturally accumulates in selected microorganisms under environmental stress. Green microalgae are photosynthetic, unicellular organisms cultivated in artificial systems to produce, with a reduced carbon footprint, biomass and industrially relevant bioproducts. While light is required for photosynthesis, fueling carbon fixation processes, application of high irradiance causes photoinhibition and limits biomass productivity. We applied a biotechnological approach of engineering astaxanthin accumulation in model species Chlamydomonas reinhardtii. The constitutive accumulation of this pigment conferred high light tolerance, reduced photoinhibition and improved biomass productivity at high irradiances. In competitive co-cultivation experiments, astaxanthin-rich Chlamydomonas reinhardtii outcompeted its corresponding parental background strain and even the fast-growing green alga Chlorella vulgaris. Engineering pigment composition could offer a powerful strategy to improve biomass productivity in customized photobioreactor setups and outperform growth of other competing microalgal strains improving the sustainability of the process.

72 - Improving chromium removal from wastewater: the use of natural biostimulants towards circular economy

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Wastewater contamination by chromium (Cr) is widely diffused in anthropic areas due to its use in industrial processes. The removal of elements from contaminated waters through plant-based solution is the only environment-friendly technology possible, due to the huge volume of fluids to be cleaned each year. Here, we applied for the first time one of the most promising biostimulants, wood distillate (WD), to the environmental issue of wastewater treatment through constructed wetlands. The use of WD in phytotechnologies represents a valuable example of circular economy, with the effective reuse of a by-product of the energy valorisation of waste plant biomass. We administered WD to the macrophyte Spirodela polyrhiza while floating on Cr contaminated waters in order to assess its effects on plant growth and physiology and to evaluate if the possible biostimulation could improve the Cr removal from the solution. Among a series of WD concentrations (0–1000 µL/L), 250 µL/L was identified as the one able to increase the growth of S. polyrhiza over a 7-day trial on N-medium (leaf area, fresh and dry weight, chlorophyll fluorescence and content were the evaluated parameters). Administering this WD concentration in presence of Cr (2.5 and 5 mg L-1) not only the plant tolerance was increased, but also the Cr uptake capacity, thus leading to an increased removal efficiency. Therefore, WD deserves to be studied as biostimulant of cultivations even when devoted to phytoremediation practises.

108 - Space missions as a circular economy: plant production for a sustainable future

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Both in space and on Earth, there is a need to find new sustainable ways to use the limited available resources and guarantee food security. Applying the principles of circular economy (reduce, reuse, recycle, and recover) to create closed-loop life support systems in space outposts is already the most economically viable option for space exploration. Independence from resupply from Earth will become even more stringent as missions became longer and more challenging, like to the Moon and Mars. For this reason, in-situ resource utilization (ISRU) technologies are also being designed to extract resources from extraterrestrial bodies and convert them into usable materials, such as oxygen, water, and substrate for food production.

Central to a closed systems for future space missions are photosynthetic organisms, like plants, microalgae, and cyanobacteria, which are capable of regenerating oxygen, purify water, recycle waste and utilize in-situ resources, while contributing to food production and human wellbeing. For these reasons, the Italian Space Agency is fostering multiple projects, from plant physiology to technological demonstrators, for the study of plant production in the context of closed-looped systems and in-situ resource utilization.

63 - A sustainable biotech system to extract bioactive compounds from vegetable waste and by-products

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By following the circular economy principles, valorization of natural bioactive compounds from agro-industrial waste is a mandatory commitment for a sustainable future and society, which require more food, less waste and healthy people. Vegetable waste products still represent an abundant, renewable and cheap sources of high added value molecules to use as ingredients in food or plant biostimulants market. According to the sustainability principles, one of the main goals of our research is to investigate alternative solutions to the conventional extraction systems to minimize the use of toxic organic solvents and reduce the energy cost or use of expensive equipment, however ensuring high extraction yield. In our work, the raw culture medium used to grow a new, genetically modified Bacillus subtilis strain, showing increased secretion of pivotal enzymes for the digestion of the plant cell wall, was used to assist in the extraction of bioactives from cauliflower (Brassica oleracea) manufacturing by-products, using isothermal pressurization cycles. Performing this extraction system on cauliflower by-product, we observed how the amount of recovered polyphenols and isothiocyanates correlates with the amount of applied hydrolytic enzymes, demonstrating that the pre-treatment with non-purified culture broth effectively promotes the release of bioactives from the vegetable matrix. Moreover, testing the extract on some pathogenic fungal strains responsible of infections in rice plant, it was observed a relevant antifungal property of the cauliflower extract, inhibiting the fungal growth. Therefore, this approach is a valid and sustainable procedure for the recovery of bioactive compounds from food waste.

90 - Investigating the potential of microalgae-microbe interactions for efficient extraction of algal metabolites

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ABSTRACT

The extraction of intracellular metabolites from microalgae is limited by their rigid and thick cell walls, and developing low-cost and efficient extraction methods would greatly improve their utilization in different industrial applications, also in line with the needs of sustainable development.

Similarly to the use of microbial cellulases for the bioconversion of lignocellulosic wastes in bioethanol production, specific algalytic formulations can be exploited for the bioprocessing of microalgae in biodiesel production. However, microalgae-microbe interactions have not been well characterized so far. By using an algal trap, an algal-eating saprotroph was captured and identified as a novel Penicillium sumatraense isolate, which assimilated Chlorella vulgaris by secreting an enzymatic arsenal mainly composed of proteases, β-glycosidases and 1,3-β-glucanases. The treatment of whole C. vulgaris cells with the filtrate from P. sumatraense increased the release of triacylglycerols from algal cells up to 50%, highlighting the potential of algal-eating saprotrophs in the bioprocessing of microalgae [1]. To investigate the molecular mechanisms of 1,3-β-glucan metabolism in microalgae-microbe interaction, the exo-1,3-β-glucanase G9376 and 1,3-β-transglucanase G7048 from P. sumatraense were heterologously expressed and characterized. Their biochemical characterization revealed that G7048 possesses an antagonizing activity against the degrading action of G9376, thereby suggesting how P. sumatraense can feed on exogenous 1,3-β-glucan of algal origin without incurring in the autohydrolysis of 1,3-β-glucan present in its own cell wall [2].

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PLANT ADAPTATION TO ENVIRONMENTAL CONDITIONS

19 - The geomagnetic field (GMF) modulates Arabidopsis thaliana ROS metabolomics and transcriptomics

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The GMF is a natural component of Earth biosphere. GMF reduction to near-null values (NNMF) induces gene expression modulation that generates biomolecular, morphological, and developmental changes. Here, we show the effect of NNMF on gene expression and reactive oxygen species (ROS) production in time course experiments on Arabidopsis thaliana. Plants exposed to NNMF in a triaxial Helmholtz coils system were sampled from 10min to 96h to evaluate differentially expressed genes (DEGs) of oxidative stress (OS) responses by full transcriptomics. A total of 194 DEGs involved in oxidative reactions were selected, many of which showed a fold change ≥±2 in at least one timing point. Heatmap clustering showed DEGs both between roots/shoots and among the different time points. In 24-96h developing stages, H2O2 and polyphenols were also analyzed from roots and shoots. NNMF induced a lower H2O2 than GMF, in agreement with the expression of ROS-related genes. 44 polyphenols were identified, which content progressively decreased during NNMF exposition time. Our results indicate that the GMF induces a basic OS which is characterized by the downregulation of genes coding for scavenging enzymes and the upregulation of genes that code for enzymes for ROS production. We hypothesize that this condition generates OS condition that plants evolved in a GMF environment. MF variations induce in plants changes in the redox status, indicating a functional role of plant magnetoperception in response to stress.

32 - Disturbance of nitrogen-fixing symbiotic interactions by PET micro/nanoplastics: the case of Azolla filiculoides Lam. and Anabaena azollae

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The effect of micro/nanoplastics (MNPs) of polyethylene terephthalate (PET) was tested on the Azolla-Anabaena symbiosis, an association between the aquatic fern Azolla filiculoides Lam. and the nitrogen-fixing cyanobacterium Anabaena azollae [1]. MNPs (size ~ 200-300 nm) were produced as water dispersions from PET bottles through reiterated cycles of homogenization, used to prepare IRRI2-medium [2] at two environmentally realistic concentrations (around 0.05 gL-1 and 0.1 gL-1 of MNPs) and administered for ten days to Azolla-Anabaena symbiotic system.

The presence of MNPs did not significantly affect the fern growth, except a reduction in root length in plants grown with the highest PET concentration. Chlorophyll content was markedly reduced in PET-treated plants, but no impairment of photosynthesis was revealed by chlorophyll fluorescence parameters. Treated plants showed a decrease in the concentration of some elements (i.e. Ca, Mg, Mn) and an interesting reduction in the level of Co, a metal essential for the growth of A. filiculoides in absence of combined nitrogen [3]. Microscopy observations showed an accumulation of MNPs at the root level and an alteration in the morphology of the heterocysts.

Overall, results demonstrated that plastic particles can negatively impact on the symbiotic system Azolla-Anabaena.

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117 - NEW INSIGHTS INTO DURUM WHEAT SALT STRESS TOLERANCE

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Soil salinization affects seed germination and plant development by inducing osmotic stress and ion imbalance, leading to severe crop yield losses. Understanding genotypic responses to salt stress can enhance crop productivity stability. Here, roughly 180 recombinant inbred lines (RILF7:F8), obtained from crossing the modern durum wheat cultivar “Primadur” (Blondur//2587-8-6/Leeds) with the Ethiopian purple wheat “T1303” (PI352395), was genotyped with Illumina 25K Infinium SNP array and phenotyped for grain color. Exposure to high salt concentration was evaluated in 18 genotypes selected from the tails of the RIL. Germination efficiency, shoot and root length revealed different salt susceptibility, allowing us to select six genotypes with contrasting salt stress responses. In control conditions, pigmented genotypes had lower germination efficiency and higher antioxidant capacity, while under stress conditions yellow genotypes displayed increased antioxidant capacity and germination efficiency compared to the pigmented ones. Redox metabolism, including quantification of key metabolites, such as total phenols, chlorophyll A/B, carotenoids, ROS accumulation in roots and the oxidative stress experienced by seedlings were assessed to investigate antioxidant capability and germination efficiency relationship. Finally, molecular genotyping and identification of microsatellites related to salt tolerance in wheat kernels were performed to provide useful screening and selection tools.

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71 - Exploring durum wheat germplasm to minimize drought impact on the nutritional status of plants

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Climate change is an escalating global crisis that poses severe threats to our environment and food systems. One of the critical consequences of climate change is an increase in extreme weather events, including droughts. Droughts have far-reaching implications for agriculture, particularly for crops like durum wheat, a staple cereal crop used to produce semolina for pasta and couscous. Durum wheat (DW), with its specific requirements for moisture, is particularly vulnerable to prolonged dry spells. The impact of climate change on droughts is not limited to reduced rainfall alone. Indeed, drought may cause nutrient deficiencies even in fertilized fields. Therefore, this work aims to identify resilient durum wheat genotypes able to cope with suboptimal water, and with unbalanced nutrient availability deriving from drought. Starting from a preliminary phenotypic and genotypic characterization of a DW genotype population in this work, we characterize the impact of drought of different durum wheat genotypes (SVEMS16, BULEL, SVEMS1, CRESO, SVEVO, SVEVO 1B/1R, ETRUSCO, Sen. Cappelli) on mineral nutrients profile and nutrient uptake mechanism, including root exudation. The results highlight variability among genotypes, with some genotypes displaying higher ability to face drought maintaining nutrient homeostasis. In order to further investigate the observed variability, the Genotyping-by-Sequencing (GBS) analysis on DW genotypes has been performed revealing DNA polymorphisms directly related to the phenotypic differences observed. Investigating DW biodiversity to minimize the drought impact on the nutritional status of plants will allow to underpin improvement of crop management strategies for future agricultural use.

23 - Dissection of heat and drought tolerance traits in a Solanum pennellii introgression tomato line.

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Mediterranean agriculture faces major challenges associated to climate change and sustainability. In particular, two major concerns are the increased daily temperatures and water shortage. Previously a tomato line carrying a small region of the Solanum pennellii wild genome (IL12-4SL) was selected as potentially heat-tolerant. This study aimed to confirm and analyse this tolerance in the same line grown under prolonged high temperatures and water stress. Most of the morphological, biochemical and physiological traits were affected by single and combined stresses, but IL12-4-SL tolerated abiotic stresses better than the cultivated line M82. Moreover, under stress IL12-4-SL produced more flowers than M82, also characterized by higher pollen viability. When exposed to heat and drought stress, IL12-4-SL was more capable than M82 of photochemically dissipating excess absorbed light. Probably, the ability of this line to better cope with abiotic stresses was linked to its higher ascorbic acid accumulation, likely due to activation of the alternative D-galacturonate pathway in leaves. Results obtained in this study demonstrated that IL12-4-SL is a promising line to be used in breeding programs, considering possible future climate scenarios characterized by frequent and long-lasting heatwaves and low rainfall.

103 - Plant responses to complex environmental stresses: investigation of molecular crosstalk between low oxygen and iron deficiency response.

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Plants are sessile organisms, and as such they have developed systems to perceive and integrate stimuli provided by the environment. One of the most challenging conditions that plants are forced to cope with is represented by heavy rainfalls. These meteorological phenomena culminate in waterlogging, determining a decline in oxygen (O2) availability in the rhizosphere, and preventing proper nutrient uptake. O2 is an essential substrate for many biochemical reactions and a co-substrate for many enzymatic activities. The main oxygen sensors in Arabidopsis thaliana are represented by plant cysteine oxidase (PCO) enzymes, which oxidize ethylene-response factor VII (ERF-VII) members and target them for degradation, depending on oxygen availability. During waterlogging, PCO reaction is inhibited due to the lack of oxygen; therefore, ERF-VII members are stabilized and able to induce the expression of anaerobic genes. PCOs belong to dioxygenases superfamily, and as such they require a form of nonheme iron (Fe2+) as cofactor. Being O2 and Fe2+ required for their activity, they might represent a convergence point between low oxygen and nutritional stimuli. To shed light on the integration of these signals, we examined the response of erfVII mutants on iron-deficiency medium through both phenotypic and gene expression analyses. We also evaluated the plant tolerance to submergence in presence of iron-deplete soil. Furthermore, we investigated ERF-VII stability according to iron availability by means of ERF-VII-luciferase reporter lines in A. thaliana. Our preliminary results will allow us to expand the view of the effect of nutrient imbalances on flooding responses in plants.

105 - Phosphorylation-mediated regulation of alternative splicing in plant heat stress response with a focus on the role of cAMP

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Alternative splicing (AS) is nowadays considered as a key post-transcriptional regulatory mechanism for plant survival to high temperatures; however, little is known about the underlying mechanisms that regulate this process. We have recently demonstrated that several proteins involved in AS were affected by 3′,5′-cyclic adenosine monophosphate (cAMP). Here, we present a large-scale phosphoproteomic study to assess the phosphorylation and dephosphorylation events that regulate AS during heat stress (HS). Moreover, to better understand the role of cAMP in the HS response, we investigated the phosphoproteome changes in tobacco BY-2 cells overexpressing a genetic tool that reduces intracellular cAMP levels. The results show that, firstly, the phosphorylation status of several RNA-binding proteins is responsive to HS and the heat-dependent phosphorylation targets are conserved among plant species. Secondly, the cAMP depletion affects the HS response through the regulation of splicing factors and spliceosome components revealing a new cAMP-dependent signal transduction mechanism. Thirdly, changes in phosphoregulations depend on the activity of heat- and cAMP-dependent kinases, notably mitogen-activated protein kinases and serine/threonine kinases. Finally, the most highly connected and conserved proteins were investigated in order to evaluate their roles in the signal transduction cascade. Taken together, our findings provide new insights into the molecular mechanism by which HS affects phosphorylation levels of proteins involved in AS in plants and show that cAMP plays a key systemic role inducing remarkable cAMP-dependent phosphorylation changes during HS response.

70 - Constitutive sodium compartmentalization patterns differentially affect biomass production of two salt-treated edible halophytes

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ABSTRACT

Food supply reliability in the face of increasing salinity of agricultural soils may be improved by including edible halophytes among cultivated crops, especially those which possess constitutive tolerance mechanisms that lead to stable or increased productivity under high salinity. Here we hypothesized that constitutive salt tolerance mechanisms that isolate sodium (Na+) away from photosynthetic tissues, such as bladder cells, are more carbon-use efficient than mesophyll vacuole sequestration of Na+. Five replicates each of Salsola soda and Tetragonia tetragoiniodes were grown hydroponically with 0, 100 and 200 mM NaCl for 28 days. The increased accumulation of Na+ in the leaf epidermal bladder cells of T. tetragoiniodes relative to increased Na+ concentrations in S. soda mesophyll vacuoles was associated with increased carbon assimilation efficiency of available CO2 and more stable PSII activity and composition (quantum yield, NPQ and chlorophyll pigments). 100 mM and 200 mM NaCl resulted in increased biomass relative to control plants in T. tetragoiniodes despite less favorable K+/Na+ ratios in leaf tissues than S. soda, whose biomass did not differ among the treatments. This is supported by greater root efflux rate of K+ in response to NaCl treatment in T. tetragoiniodes. By sequestering excess Na+ in bladder cells, T. tetragoiniodes maximizes its carbon use efficiency by buffering photosynthetic tissues, especially at moderate salt levels (100 mM NaCl). By evaluating and selecting for highly efficient Na+ sequestration mechanisms, there is an increased likelihood of meeting the difficult challenges presented by climate change, such as increased temperature, drought, and soil salinity.

17 - Physcomitrium patens flavodiiron proteins for the adaptation to changing environmental conditions

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Plant development and metabolism depend on the ability to perform efficient photosynthesis. Two photosystems (PS) use sunlight to fuel an electron transport that produces NADPH and ATP necessary for carbon fixation. In response to changing light intensity, photoprotective mechanisms prevent an imbalance in the electron transport chain and the consequent damage to cell components. In all photosynthetic organisms of the green lineage but angiosperms, upon sudden illumination, flavodiiron proteins (FLV) act as safety valve for electrons via O2 photoreduction downstream PSI. Despite their important role, there is a lack of information about FLV structure and biological mechanism. In this work, we characterized the FLVA and FLVB isoforms of the moss Physcomitrium patens to decipher the role of FLV during land colonization and adaption to different environments. WT plants were acclimated to control (CL), high (HL) and fluctuating light (FL) conditions for physiological, biochemical and spectroscopic analysis. We observed higher accumulation of FLV in HL and FL acclimated WT P. patens than CL plants, suggesting FLV have an enhanced role under those conditions. We also generated lines expressing a 6xHis-tagged version of FLVB that allowed the purification of hetero-tetramers of FLVB and FLVA, stably bound together, indicating a strong association between the two proteins. The purified heterocomplex was functional and provided a solid basis for starting structural and interactome studies.

30 - Evidence from an Arabidopsis thaliana p5cdh mutant strengthening the occurrence of a proline-P5C cycle in plants.

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Since many plants accumulate proline under hyperosmotic stress conditions, the use of proline to fuel cell metabolism and as a source of organic nitrogen and carbon skeletons may play a main and still underestimated role to resume growth during the subsequent recovery. The mitochondrial degradation of proline to glutamate involves two oxidative steps catalysed in sequence by proline dehydrogenase (ProDH) and δ1-pyrroline-5-carboxylate (P5C) dehydrogenase (P5CDH). The former is believed to feed electrons directly to the respiratory chain, whereas the latter uses NAD+ as the electron acceptor. The occurrence of a shortcut in which the P5C released by ProDH is not further oxidized, but is reduced back to proline in the cytosol by P5C reductase (P5CR) has been hypothesized. Such apparently futile proline-P5C cycle may provide the cell with a mechanism for transferring reducing equivalents from the cytosol to the mitochondrion, and to fuel the respiratory chain. Moreover, ProDH activity may alternatively lead to ROS production, which can trigger in turn the apoptotic process, or increased ATP synthesis for protective autophagy. Although the role of the proline-P5C cycle is now well established in mammals, its occurrence in plants is still a matter of debate due to the physical separation of P5CR and ProDH. Here we report on the metabolic fate of exogenously supplied proline in wild-type and p5cdh cultured cells of A. thaliana that supports the functioning of a proline-P5C cycle.

69 - Novel function of isoprene in root physiology and salt stress tolerance

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Isoprene is the major volatile compound emitted by plants. In leaves, isoprene synthase (IspS) catalyzes isoprene formation from DMADP from the MEP pathway. There is much scientific evidence on the activities of in situ isoprene emission in leaves, but less is known in roots. Reports suggest that root tissues can also emit isoprene. However, the effect of isoprene on root physiology and root responses to abiotic stresses is currently unknown. We transformed Arabidopsis thaliana with Agrobacterium tumefaciens to generate Arabidopsis plants overexpressing IspS from Eucalyptus globulus (isoprene emitter, IE) or carrying an empty vector (non-emitter, EV). We explored the possible effects of isoprene on roots through (a) phenotype, (b) transcriptomic (RNA-seq) and (c) metabolic changes in transgenic Arabidopsis roots exposed to salt stress. We confirmed that root tissues can emit isoprene although at lower rate than leaves. MEP pathway metabolites were significantly enhanced in IE roots compared to EV. Our study shows that root isoprene enhanced root growth and biomass. RNA-seq data revealed an upregulation of several genes that encode metabolites crucial for salt tolerance and hormone signaling. Metabolomic data indicated that root isoprene affected amino acids, TCA cycle metabolites and hormone levels in roots. Our results highlight that the capacity to emit isoprene by roots affects root physiology via alteration of transcriptome and metabolome, suggesting a novel function of isoprene in the belowground.

8 - Increasing phosphoribulokinase affinity to improve CO2 flux: testing the approach and first results

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The rising number of extreme climate events challenges plant survival and thus final crop yield. Nevertheless, plant productivity is tested by our increasing food demand. Many approaches attempted to boost plant productivity by improving carbon fixation. Our research is focused on the Calvin-Benson cycle with the final aim of increasing the flux of carbon by improving phosphoribulokinase (PRK), which catalyzes the production of ribulose-1,5-bisphosphate, the substrate of Rubisco. Relevant information is available on PRK from the green algae Chlamydomonas reinhardtii (CrPRK), allowing us to test our approach. The experimentally determined KM of CrPRK for its substrate ribulose-5-phosphate (Ru5P) is about twice as much as the concentration of Ru5P in Chlamydomonas chloroplast. In vivo, the presence of inhibitors and reaction products diminishes the affinity, with calculated KM values even greater than experimental ones. These observations give scope to an engineering work based on a rational design that aimed at enhancing the affinity of CrPRK for its substrate. Using the computational approach MM-GBSA we simulated the Ru5P trajectory from the enzyme outer shell to the catalytic site, finding some residues of interest. After mutagenesis, the catalytic parameters were tested. The first round of mutants highlighted the reliability of the in silico analysis and provided a first promising mutant to improve with further cycles of in silico/in vitro analysis.

74 - Krebs cycle substrates starvation explains the decrease of O2 consumption induced by hexanoic acid in pea roots

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Plants exposed to waterlogging face anoxia and chemical toxicity caused by phytotoxins produced by microorganisms in the soil. One such toxin is hexanoic acid (HxA) that, at certain levels, causes a strong decline in root O_2 consumption at low pH values. However, the mechanism behind this response is still unknown. We treated pea (Pisum sativum L.) roots with 20 mM HxA acid at either pH 5.0 or 6.0 for 1 h and measured the leakage of metal cations (K^+, Na^+, Ca^{2+} and Mg^{2+}), organic acids (malate and citrate) and soluble non-structural carbohydrates. After treatment, mitochondria were isolated to assess their functionality, evaluated as electrical potential formation and O_2 consumption. HxA treatments induced a depletion of cations (mainly K^+), malate and citrate, but only the leakage of these organic acids was increased at pH 5.0 and correlated with the inhibition of O_2 consumption. Sugars were also released from roots, but without affecting their internal pool or evidencing starch depletion. Mitochondria isolated from treated roots were almost unaffected, showing a slight, but significant, reduction of O_2 consumption when roots were treated at pH 5.0. On the basis of these results, we propose a model in which HxA, in its undissociated form at acidic pH, enters the root cells and induces the efflux of malate and citrate, which in turn leads to starvation of mitochondrial respiratory substrates of Krebs cycle and a consequent decline in O_2 consumption.

104 - Effects of light quality on carbon assimilation/emission and growth of lettuce: use of an upgraded prototype-platform to unravel mechanisms of photosynthesis/respiration control.

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Responses of plant performances to light quality are focus of research to optimize plant indoor cultivation with artificial light. In a comparative study of lettuce grown with different light spectra, photosynthesis, root and shoot respiration, and growth traits through chlorophyll fluorimetry essays, biochemical and molecular characterization of photosystems, and plant-to-atmosphere gas exchange assessment have been measured. To answer physiological questions at the whole-plant level we tested two phenotyping platforms, one based on 3D and multispectral imaging technology and a prototype based on whole plant gas exchange analyses, capable of differentiating root from canopy gas exchange. Red and blue (RB) light maximized photosynthetic activity, but this advantage did not lead to greater biomass accumulation, which was greater in plants under red, green and blue (RGB) and further under full spectrum (FS) light. Plants subjected to the RB light regime invest less carbon in leaf blade extension and incorporate less carbon than what has been assimilated. RB leaves have a smaller PSII antenna size and the cyclic electron transport around PSI is higher, implying a lower NADPH / ATP ratio. Exposure to RB light affects plant overall metabolic biosynthetic pathways at the expense of growth, while RGB and especially FS light, by not triggering this acclimation, are more suited to constant low light growing conditions, as those usually provided by indoor growing technologies.

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118 - Physiological response of Lens culinaris Medik. to cerium and neodymium chlorides exposure

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Rare earth elements (REEs) are a group of 15 elements, the lanthanides, Yttrium, and Scandium, with similar chemical and physical properties. Their use for many advanced technological applications remarkably increased in the last decades, and it was associated with an intensive extraction of such elements from their ores. Consequently, increasing amounts of either REE-containing by-products, deriving from the extraction process, and REE-containing wastes, deriving from the disposal of REE-containing devices, are reaching the environmental systems both at the local and global levels, as never in the past. The effects of cerium chloride (CeCl3) and neodymium chloride (NdCl3) exposure on Lens culinaris Medik. were investigated. Both morphological and physiological responses were evaluated after germinating for two days in Petri Dish and after treatment at 10 and 100 µM for three days in a hydroponic system. Parameters measured were chlorophyll content, photosynthetic activity using JUNIOR-PAM, presence of reactive oxygen species (ROS) such as singlet O2 and H2O2. Preliminary results show an effect on the yield (Y) of the PSII; since Y(II) is a functional parameter, the determination of a more pronounced response in this may hint at the mode of action of contaminants. High concentrations induce an increase in ROS production too. This study aims to understand better the toxic effects of these two REEs and the relation between responses induced by the plant.

96 - Investigating holm oak forest dieback through a multiscale approach: physiological measurements and visual assessment data with remote sensing

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Keywords: drought, holm oak dieback, photosynthesis, remote sensing, tree-ring δ13C

Drought and heat waves profoundly affect the functioning of Mediterranean forests. As Quercus ilex dieback has been observed in many Mediterranean stands, it is crucial to develop effective tools for studying this phenomenon across multiple scales. Our study was conducted over three years (2019-2021) in declining (D) and non-declining (ND) Q. ilex stands in southern Tuscany, assessing physiological traits such as gas exchange and water relations. Dendrochronological and tree-ring δ13C analyses were combined to investigate the effects of previous droughts on tree growth and WUE. Finally, the relationships between physiological indicators and satellite-derived Normalized Difference Vegetation and Water Indices (NDVI and NDWI) were analyzed. Seasonality had a strong effect on physiological traits, with the main stress occurring during the summer of 2020, as evidenced by the lowest gas exchange values. Furthermore, plants in the ND stand exhibited significantly higher photosynthetic performances than those in the D stand across multiple seasons. The δ13C value at ND significantly exceeded that observed at site D over the last 20 years, revealing more conservative water use. While the high spatial resolution of Sentinel 2 helped identify the overall Q. ilex decline, these indices were inadequate for analyzing forest stands with patchy dieback. Our study emphasizes the significance of a multiscale approach that combines in situ physiological measures and satellite data to monitor Q. ilex decline.

114 - Does slow and fast wilting rates affect the recovery from embolism in poplar seedlings? New insights from micro-CT analysis.

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Severe water stress constrains, or even halts, water transport in the xylem due to embolism formation. Poplar trees respond to drought-induced embolisms by accumulating sugars in both xylem apoplast and stem tissues, thus providing energy and facilitating osmotic adjustment processes. We focused on how the rate of drought progression modulates energy reserves (sugar accumulation) and affects the recovery process. P. nigra seedlings were exposed to either fast-developing drought (by ceasing irrigation) or slow-developing drought (by progressively reducing the amount of water provided to the plants). Once similar level of water stress was reached, the plants were re-watered and 24 hours after re-irrigation the recovery rate was determined thorough x-ray micro-CT analyses. Plants rewatered after a slowly induced stress, recovered approximately 25% of embolized vessels, whereas in fast-stressed plants, over 50% of vessels restored their hydraulic functionality. The reduced growth and lower accumulation of soluble sugars in slowly stressed stems, combined with the measurements of embolism, indicate that slow developing drought reduced the hydraulic ability to recover compared to fast developing stressed plants. This suggests that embolism removal requires the accumulation of sugars. Image analyses indicated that embolism formation and its removal are spatially coordinated process, with embolism formation occurring from the inside out and recovery from the outside in, underlining the importance of xylem proximity to phloem (carbohydrates source). To better understand the effects of stress progression rate, we are now focusing on expression levels of genes related to carbohydrates reserves mobilization and transport.

92 - New tools to study in vivo the dynamics of Ca2+ in plant mitochondria

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How do plant cells generate specific Ca2+ signatures? Which are the mechanisms for their tight regulation? To help answering these questions, we have planned to generate new tools to study in vivo the dynamics of Ca2+ in different organelles with the aim of better defining their relationships with cytosolic Ca2+ homeostasis. Our strategy is based on the use of new red-shifted genetically encoded intensiometric Ca2+ indicators (GECIs) targeted to different subcellular compartments (mitochondria and ER) to be used in combination with the green-shifted GCaMP3 Ca2+ indicator localised in the cytosol.

Here we report the expression in plant cells of the jRCaMP1b targeted to mitochondria.

Specifically, Arabidopsis thaliana plants expressing the mitochondrial localized jRCaMP1b have been obtained and tested to verify their efficiency to report mitochondrial Ca2+ dynamics. For this aim, side by side experiments with the Arabidopsis plants expressing the Cameleon 4mt-YC3.6 have been carried out. In order to study how plant mitochondria accumulate Ca2+ in response to a stimulus-induced Ca2+ increase in the cytosol, the Mito-jRCaMP1b plants were crossed with plants expressing the green-shifted cytosolic-localized Ca2+ indicator GCaMP3. Moreover, the simultaneous expression in Arabidopsis mitochondria of the jRCaMP1b together with the pH sensor cpYFP allowed us to monitor mitochondrial Ca2+ and pH dynamics at single cell level. Analyses of mitochondrial Ca2+ dynamics in mature plants will be also presented.

102 - Exploring different phenotypic and molecular approaches to unravel early events in olive–Xylella interactions

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Xylella fastidiosa is a xylem-limited plant-pathogenic bacterium causing severe diseases in several crops. Infections in olives cause the olive quick decline syndrome, a disease that compromises the survival of susceptible trees. This represents one of the most challenging pathosystems to study, because infections have a long incubation period (i.e. > 1 year before any visual alteration induced by the bacterium can be detected) and bacterial virulence factors/host response mechanisms are largely unknown. Even so, it is extensively documented that the formation of cell aggregates in the vessels and the degradation of the cell wall cause the hydraulic collapse of the xylem network. Such phenomena are more prominent in the susceptible hosts, while resistant/tolerant genotypes may remain symptomless or show mild symptoms. We aimed to explore the use of different physiological traits, that combined with molecular markers, may identify biomarkers suitable for the early prediction of the host response. Phenotypic measurements likewise stomatal conductance, stem water potential, leaf and canopy temperature, electrical signal changes in the sap flux monitored by electrical impedance spectroscopy and massive transcriptomic data have been generated from plants under different infection conditions. Efforts are directed to integrate transcriptomic and physiological data, in a multiomic approach, for rapid and accurate predictions of resistant vs susceptible phenotype.

POSTER SESSION

11 - Plant waste-derived biostimulants as a sustainable approach to improve soybean germination under osmotic stress

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Plant-derived biostimulants are natural substances that can have an impact on sustainable agriculture and bio-economy by promoting plant growth and stress resilience while reducing the input of synthetic fertilizers. Such sustainable agriculture practices aim to minimize the negative impact on the environment while ensuring long-term food production and economic stability. The economic value of agricultural residues and plant by-products can be harnessed through plant waste recycling and precision farming technologies. Related to the latter, seed priming, defined as technologies designed to improve germination performance, can contribute to building up dynamic and sustainable agriculture practices. Therefore in this work, plant waste has been used to produce biostimulants that are applied as seed priming agents to boost seed quality. Red chicory and canola waste-derived extracts were used to treat three soybean varieties (OL 996, NAV 270, EM PURA), along with hydropriming and unprimed controls. The germination efficiency was monitored both under physiological and osmotic stress conditions induced by polyethylene glycol (PEG). The germination performance was monitored for three days and multiple parameters (e.g., percentage, speed, synonymity, uniformity, and radicle growth) were measured. The results evidence that the seed plant waste-derived biostimulant application improved germinability especially under PEG treatment, thus rescuing the germination under stress.

36 - Polyamines as seed priming agents.

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The ongoing climate changes expose plants to extreme environmental conditions, subjecting them to atypical abiotic stresses responsible for yield losses. The increasing needs to integrate environmental protection and food security involves implementation of sustainable agricultural practices able to enhance crop productivity. Plant Bio-stimulants (BSs) are promising eco-friendly tools to reduce the dependency on synthetic fertilizers. Among BSs, Polyamines (PAs) are biogenic ubiquitous amines involved in growth regulation and defense responses. Here it has been investigated the effect of Arabidopsis seed priming with exogenous PAs, among which putrescine, spermidine and spermine, on germination parameters and growth rate, in optimal conditions and under abiotic stress. The time-course analysis of germination parameters revealed an increase of both germination rate and index of PA-primed seeds over the first 3 days from sowing with optimal concentration of each PA, depending on respective chain length. Analysis of phenotypic traits such as root length of seedlings from PA-primed seeds showed a higher root growth rate than seedlings from untreated control seeds with respect to time zero, under both optimal conditions and salt stress conditions. Overall, results suggest that seed priming with exogenous PAs can improve germination rate and plant fitness, denoting a promising tool to increase yield and ensure productivity under both optimal and stress conditions.

49 - Valorization of wastewater from hydroponics using microalgae

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Vertical farms (VFs) are systems that are developed in height and continuously produce along the year. The most used cultivation technique in VFs is hydroponics, which provides the growth of plants in a liquid solution rich in salt nutrients. At the end of the productive cycle, hydroponic solutions still contain nutrient salts, thus they cannot be directly released into the environment.

Microalgae represent a reliable and “green" solution for the treatment and valorization of such spent solutions. In fact, microalgae can easily consume the residual nutrients, generating biomass that can be used as a biofertilizer or a biostimulant, following principles of circular economy.

We showed the ability of the fast-growing and robust eukaryotic microalga Chlorella vulgaris of using valuable resources derived from an industrial hydroponic cultivation. In particular, we reduced more than 80% and 70% the contents of inorganic phosphate and nitrogen, respectively, and more than 2 g/L of microalgal biomass was generated exploiting such wastewater. It is noteworthy that plant root exudates, possibly, affected microalgal growth in spent solutions.

In addition, a preliminary work has been conducted exploiting the hydroponic solution for the cultivation of genetically engineered microalgal strains, which accumulated non-native astaxanthin, aiming at generating microalgal biomass of high-added value. This will facilitate the achievement of the economical sustainability of the entire process.

50 - Effects of a Coffee Waste Solution (CWS) treatment on strawberry plants

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Coffee represents the 3rd most popular beverage in the world, with approximately 500 billion cups consumed worldwide annually. Due to the large amount of coffee produced and consumed, a large amount of coffee waste is made. In order to propose some reuse of these waste, the aim of this research was to evaluate the suitability of a coffee waste solution (patent pending) as a possible plants' biostimulant. Strawberry (Fragaria vesca) cultivation is of great importance worldwide, in fact the annual world production has reached over 3.6 million metric tons. We evaluated the effects of a 3-month-long growth supplementing a solution produced from wastes of coffee roasting (CWS) on the physiology and morphology of strawberries. During the three months of treatment, plants watered supplementing the CWS showed an increased number of leaves with respect to plants irrigated with water only (controls). Gas exchange measurements indicated a lower stomatal conductance of CWS-treated plants with respect to controls showing similar photosynthesis. As a consequence, CSW-treated plants were characterized by higher instantaneous water use efficiency, given by the ratio between photosynthesis and stomatal conductance. At the fruit level, irrigation supplementing CWS induced a rise of total proteins and antioxidant molecules in comparison to controls, leading to a substantial changes of fruit quality and to a modification of fruiting time. We conclude that the use of CWS improves physiological traits of strawberries and may be considered as a suitable biostimulant. Further studies focusing on long-term effects on plants and soils are needed to recommend CWS as a suitable analogous of chemical fertilizers for sustainable farming practices.

75 - Effects of drought stress and endogenous strigolactones on biosynthesis of glycoalkaloids in tomato (Solanum lycopersicum L.)

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Tomato (Solanum lycopersicum L.) produces a range of secondary metabolites in both fruits and vegetative organs. Among these compounds, steroidal glycoalkaloids (SGAs) are of great scientific and applicative interest, both for their role in plant defence against pathogens and for their useful properties for human health. The SGA biosynthetic pathway has been recently described as part of the GLYCOALKALOID METABOLISM (GAME) pathway from cholesterol; however, its regulation is still little understood. Obtaining information on the roles of exogenous and endogenous regulatory factors would allow steering SGAs concentrations for industrial application. Although SGA concentrations are genetically determined, it has been shown that biotic stress influences their production, while still little is known about the effect of abiotic stress. Strigolactones (SLs), a class of carotenoid-derived hormones, are involved in the tomato response to different types of abiotic stresses, while their role in secondary metabolism mostly remains to be elucidated. In this study, the role played by stress and endogenous SLs on the biosynthesis of SGAs was analysed by imposing drought stress onto WT and SL-depleted (SL-) tomato plants of the cultivar M82. Physiological parameters such as stomatal conductance, net assimilation rate, and stem water potential were assessed to analyse the progression of water stress. Leaf samples were collected at different stress levels and used for RNA extraction. Expression analysis by qRT-PCR of stress and SL signal markers, and of SGA biosynthetic genes is underway. The levels of mature miR1916, a putative regulator of SGAs biosynthesis, and of his putative target strictosidine synthase (STR2) will be checked as well.

82 - Mitigation effects of a biostimulant based on seaweed and yeast extracts on tomato (Solanum lycopersicum L.) grown under drought stress conditions

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The lack of water represents one of the main problems of the modern agriculture. This abiotic stress causes heavy crop losses in the face of an increasing food demand due to the growth of the world population. Therefore, the development of new technologies to be applied in agricultural field is necessary to improve crop productivity in drought conditions. One possible solution is the use of plant biostimulants, products able to enhance plant physiological processes including tolerance to abiotic stress.

In this work, the effects of ERANTHIS®, a biostimulant based on seaweed (Ascophyllum nodosum and Laminaria digitata) and yeast extracts were tested on tomato (Solanum lycopersicum cv. Micro-Tom) grown under drought stress. H2O2, osmolyte (proline and glycine betaine), non-protein thiol content and antioxidant enzyme (SOD, CAT, POX and GST) activity were measured in leaves along with the level of transcripts (RNA-Seq analysis). On fruits, qPCR analyses were performed to evaluate the expression of genes related to ripening, including those involved in lycopene biosynthesis. To correlate gene expression to metabolite content, the spectrophotometric quantification of lycopene was also performed.

In general, leaves of stressed-treated plants showed lower H2O2, osmolyte, non-protein thiol, and antioxidant enzyme activity levels compared to untreated plants. These data correlated to RNA-Seq analysis. The lycopene content in fruits from treated-plants was higher than in fruits of untreated plants, in agreement with gene expression analyses, in which an up-regulation of lycopene biosynthetic genes was observed.

In conclusion, ERANTHIS® can mitigate the effects of drought stress in tomato by improving plant yield and fruit quality

88 - Callus and cell suspension cultures from Coffea arabica for the production of valuable secondary metabolites

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Secondary metabolites (SMs) from coffee (Coffea spp.) plant have potential applications in medical and cosmetic field. Large-scale production of high-value SMs could be achieved by plant cell tissue culture, mainly as cell suspension cultures (CSCs) as widely demonstrated for many medicinal and aromatic plants. Nevertheless, for C. arabica a well-defined protocol for the induction of non-embryogenic and friable callus is still missing. We incubated leaf disc explants from C. arabica cv. Bourbon Red and Castillo on a full-strength MS medium in the presence of different concentrations and types of cytokinins (CK) and auxins (AUX). The percentage of callusing and the mean fresh weight of explant+callus (mFWe+c) were measured for each treatment. The callus texture was also considered. CSCs were stabilized into fine suspensions, monitoring the growth as fresh and dry weight increase every 3-4 days. The highest value of mFWe+c and friable calli was obtained in the presence of 1 mg l−1 of CK and AUX for both cultivars. In CK-free media, the mFWe+c measured were significantly different from the others, reaching the lowest values. Significant differences in mFWe+c detected in treatments with different CK, being Kinetin the best factor able to maximize weight and friable consistency. Significant differences were also observed between cultivars and type of AUX. The presence of SMs, such as caffeine, chlorogenic acids, trigonelline, theobromine, kahweol and cafestol was detected.

97 - TUNING CHLAMYDOMONAS METABOLISM TOWARD THE PRODUCTION OF HIGH VALUE COMPOUNDS

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Microalgae have the potential to ensure food, energy and raw material for a growing population. Nevertheless, microalgae cultivation nowadays is expensive their effective use on industrial scale is conditioned by costs break down. On of the most prospective strategies is the combination of the sole biomass production with production of high value compounds. Currently, Chlamydomonas reinhardtii is considered one of the most promising microalgae for that purpose. A wide range of high-value products, ranging from therapeutic proteins to pigments and fragrances, can be produced in Chlamydomonas through genetic engineering and here we report two of them.

First, the monoterpene geraniol is one of the most requested fragrances, with production exceeding 1000 tons/year. Beyond its flavor, industrial interest in geraniol comes from its many well-known properties that make it useful as a repellent, anti-inflammatory, anticancer. By overexpressing three key enzymes, the MEP pathway was re-directed to geraniol production, providing a green alternative to petroleum-based synthesis.

Secondly, Chlamydomonas reinhardtii, approved by the FDA for human consumption, was used to produce high-value proteins. In particular, the synthetic chimeric protein zeolin, obtained by merging γ-zein and phaseolin proteins, the major storage proteins for maize (Zea mays) and bean (Phaseolus vulgaris) was successfully accumulated. Seed storage proteins have unbalanced amino acid content which requires a complementation in dietary: the chimeric recombinant zeolin overcome this constrains providing an amino acid storage strategy with a balanced profile.

99 - Chlamydomonas reinhardtii as biofactory of dsRNA molecule for exogenous applications to increase sustainability and safety in fruit and vegetable supply chain

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RNA interference (RNAi) is an innovative and sustainable way to improve the quality of plant production and reduce pesticide use. The spray-Induced Gene Silencing (SIGS) approach represents a “green” alternative to chemical pesticides, in which dsRNAs are sprayed onto plant tissues. However, the high cost of dsRNA production has limited its commercial application. We propose, Chlamydomonas reinhardtii, as dsRNA biofactory. This green alga is GRAS, has already been used as a cell factory and genetic engineering tools are available for its transformation. We optimized an expression vector for dsRNA production and transformed 5 microalgae strains: 2 Dicer3-like (DCL3) mutants, the relative WT and the UVM4 strain, previously selected for its high protein expression level. The DCL3 mutants may allow reduced dsRNA processing. To evaluate the application of dsRNA produced by microalgae for silencing plant genes, we expressed a dsRNA against the tomato MADS-box genes AGL6 involved in the ovary development. At the same time, we expressed a dsRNA against the causative agent of Sharka PPV, a virus devastating to the prunus genus. The application of whole or partially degraded algae expressing dsRNA against a pathogen will offer an environmentally friendly alternative to chemical pesticides.

110 - Olive patè as a source of bioactive molecules to elicit defense against microbial pathogens in A. thaliana: from olive mill waste byproducts to new natural protectant against pathogens.

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Olive oil is a core component of the Mediterranean diet known for its nutritional properties and health benefits. Multi-phase decanter technology is a modern two-phase system producing, in addition to extra virgin oil, a solid waste called 'Pâté' consisting of a wet fraction composed by pulp, skin, and vegetative water. Pâté is enriched of bioactive molecules of great interest as bioprotectants against microbial pathogens. By exploiting the tangential-flow membrane filtration (TFMF) eco-friendly technology, the vegetation water, recovered from Pâté, was sequentially fractionated through a combination of microfiltration (MF), ultrafiltration (UF) and nanofiltration (NF). NMR spectroscopy indicated the presence, in specific TFMF fractions, of bioactive phenols such as tyrosol, hydroxytyrosol and oleuropein with a known antimicrobial activity. High Performance Anion Exchange Chromatography with Pulsed Amperometric Detection (HPAEC-PAD) profile showed the enrichment, of pectin-derived oligogalacturonides (OGs) a class of elicitors of defense responses against microbial pathogens especially in UF concentrated fraction (CUF). CUF was able to activate defense responses including the transient accumulation of cytosolic calcium and inductions of defense genes in A. thaliana plants. CUF enriched in bioactive molecules, also induced resistance of A. thaliana against Botrytis cinerea and Pectobacteriun carotovorum. This finding highlights the potential valorization of olive mill by-products to be exploited as sustainable plant protectants against microbial pathogens in alternative to chemical pesticides consistently with 'Green Deal' goals to halve the use of chemical pesticides by 2030.

112 - INDOOR PHYTOREMEDIATION OF FORMALDEHYDE BY TILLANDSIA PLANTS

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Tillandsia plants fall into the Bromeliad family, also known as air plants, can grow without soil and obtain nutrients and moisture from the air and rainwater. This characteristic makes Tillandsia suitable for phytoremediation of air pollutants, including formaldehyde (FA). FA, which can have detrimental effects on human health, is released from various sources in indoor environments. The main goal of this study was to test and compare the ability of two Tillandsia species (T. velutina and T. ionantha) to remove FA. Firstly, FA degradation capability of leaf extracts was tested in vitro and showed a higher efficiency of T. ionantha than T. velutina in the removal of this toxic compound. However, the experiments of FA removal in vivo, conducted in a glovebox equipped with a sensor probe for high temporal resolution monitoring of FA demonstrated that both the Tillandsia species were able to remove FA with a high efficiency. FA, probably due to a fast degradation and reutilization, was not accumulated in the leaves. Accordingly, after FA treatment the glutathione content, which is the substrate of formaldehyde dehydrogenase, significantly decreased in both the species; moreover, an increase in the content of soluble proteins was observed. In line to the higher FA degradation capability of T. ionantha, the analysis of photosynthetic pigments after FA treatment, indicated that this Tillandsia species better tolerates FA pollution.

116 - SUSTAINABLE STRATEGIES TO PRODUCE PLANT DEFENSINS WITH ANTIVIRAL PROPRIETIES

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The identification of plant metabolites with antiviral action can be useful for the production of phytotherapics for the treatment of human viral diseases. Plant defensins are promising antimicrobial peptides involved in plant defense responses against a wide range of environmental injuries. The expression of four defensin genes from Nicotiana tabacum (FST, S13, THIO, J1) has been stimulated by subjecting cultured Tobacco Bright Yellow 2 (TBY-2) cells to different elicitation treatments (JA, SA, ABA, NaCl, H2O2, heat stress). Unfortunately, gene expression analysis showed that tobacco endogenous defensins' genes were not expressed in TBY-2 cells, neither in the elicited cultures nor in those grown under basal conditions. These results can be due to the difficulties of undifferentiated cells to express genes involved in biosynthetic pathways of defense metabolites. To get around the problem, the use of hairy roots' cultures obtained from Petunia hybrida cv. Mitchell W115 subjected to Rhizobium rhizogenes infection has been chosen as alternative strategy to produce putative antiviral defensins. Based on the reported antimicrobial activity, the heterologous genes selected for transformation in P. hybrida among those encoding for peptides with antiviral activity, we started with a defensin from Arabidopsis thaliana (AT3G05727), the sequence of which was engineered to optimize the purification steps, and the KALATAB1 cyclotide precursor from Oldenlandia affinis.

123 - Monochromatic LED lighting affects the antioxidant machinery in lentil (Lens culinaris Medik.) seedlings

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In the last decades, light-emitting diode (LED) technology has gained more and more attention, and its applicability increased due to its advantages over conventional light sources, such as fluorescent, high-pressure mercury, high-pressure sodium, and metal-halide lamps. LED lighting allows to use single wavelengths of different regions of the light spectrum and adjust the light intensity. Therefore, this innovative system represents a great tool for studying plants' light responses, especially in the horticulture field. Light is known to be one of the key environmental factors influencing plants’ morphology and physiology. However, given its complex nature, the effects of light and its regulatory mechanisms are still not fully explained. Our study aimed at investigating the effects of continuous LED light treatment on the antioxidant systems during the early stages of Lens culinaris seedlings. Lentil seeds were germinated and grown in darkness and under blue and red monochromatic LED light, at the intensities of 100, 300 and 500 µmol m-2 s-1. Antioxidant and redox metabolism, including ROS level, ascorbate and glutathione content, antioxidant enzyme activities, and thiol-disulfide status of proteins, were assessed. This study highlighted the role of LED quality and quantity in affecting cell redox status and optimising lighting strategies in indoor cultivation.

16 - Chromatin and transcriptome dynamics in the rehydration-dehydration cycle of primed and overprimed Medicago truncatula seeds

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Improving the quality of seeds has become crucial to meet the needs of high-standard agricultural markets. Seed priming represents a valid approach to ensure high yields in orthodox seeds, since it predisposes seeds for germination, enhancing germination performance as well as the resilience to various environmental stresses. Priming protocols mimick the first stages of germination, by exposing seeds to a controlled imbibition phase immediately followed by a dehydration step. The capacity to withstand dehydration is termed as “desiccation tolerance” and it depends on several molecular processes which are only partially identified. For this reason, seed priming often result in a hetereogeneous response. Despite a growing supporting literature, an exhaustive molecular landscape concerning seed priming is still missing. We therefore investigated these aspects by combining the results of two high-throughput analysis, RNA-Seq and ATAC-Seq, in the well characterized priming versus overpriming experimental system (Pagano et al., 2023. https://doi.org/10.1111/pce.14295). In this work, we higlight the most prominent trascriptome and chromatin accessibility dynamics emerging from the dehydration-rehydration cycle of Medicago truncatula seeds. These preliminary results will be used for better adjustment of pre-sowing treatments as well as identifying new hallmarks of seed priming. Additionally, results could serve as novel layers of understanding germination physiology in Legumes

21 - A potential role of two distinct cytochromes b561 in intracellular ascorbate homeostasis and ROS-mediated signaling in A. thaliana

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In plants, ascorbate (ASC) plays numerous physiological roles: it is one of the most important antioxidant metabolites, but it also acts as a substrate for several enzymes, among which the transmembrane proteins belonging to the cytochromes b561 (CYB561s) family. ASC needs to be kept in a reduced state to perform its biological functions, but the enzymes that regenerate ASC from its oxidized forms seem to be absent in some organelles (e.g., vacuoles). CYB561s catalyze a transmembrane electron transfer from ASC to monodehydroascorbate, possibly allowing ASC regeneration in those compartments that lack other ASC regeneration systems.

The genome of A. thaliana contains 4 CYB561s-encoding genes (CYB561-A, -B, -C and -D). We recently demonstrated that the vacuolar CYB561-A catalyzes a bidirectional electron transfer in vivo. We are now investigating the subcellular localization of the other three paralogs and we are studying the physiological role of CYB561-A and -B by performing molecular phenotyping of T-DNA mutant plants. All mutants show a decreased ROS content and an altered activity of several ASC-related enzymes (ASC peroxidases and dehydroascorbate reductases), although no differences in the ASC content were measured. CYB561-A mutants also show a delay in flowering onset and a greater accumulation of anthocyanins when exposed to high light. Thus, our results suggest that CYB561s may be involved in the ASC homeostasis and ROS-mediated signaling.

37 - Role of Arabidopsis ACA8 and ACA10 in stomatal modulation upon root wounding or MeJA treatment

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In Arabidopsis, stress-driven stomatal closure is regulated by oscillations of the cytosolic Ca2+ concentration occurring in guard cells, which vary in frequency, duration and amplitude depending on the specific stress perceived. Auto-inhibited Ca2+-ATPases 8 and 10 (ACA8/10) are two plasma membrane proteins localized in guard cells, which are key regulators of Ca2+ homeostasis in plant cells. In this study, it was investigated the role of ACA8 and ACA10 in stomatal closure induced by root wounding and by the wound-signalling phytohormone methyl jasmonate (MeJA), using loss- and gain-of-function genetic approaches. In wild-type seedlings, root wounding triggers a consistent stomatal closure starting already after 5', while MeJA treatment triggers a delayed stomatal closure compared to root wounding, starting only after 30'. In contrast, aca8 and aca10 insertional mutants, as well as ACA8 overexpressing seedlings, are completely unresponsive to both stimuli here considered. Moreover, RT-qPCR analyses show an induction of ACA8 and ACA10 expression upon root wounding, stronger for ACA10 than for ACA8. Overall, results obtained showed that these two pumps are indeed necessary for the modulation of stomatal movement in response to both root wounding and MeJA treatment, probably due to their role in the correct regulation of cytosolic Ca2+ homeostasis.

44 - Integration of transcriptomics and metabolomics data provides a glimpse of early steps of orchid mycorrhiza establishment

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Terrestrial orchids rely on their associated mycorrhizal fungal symbionts for seed germination and early plant development. However, the molecular mechanisms involved in the early stages of the interaction remain poorly understood. To address this gap, a time-course experiment using a mycorrhizal model system, i.e., the Mediterranean orchid Serapias vomeracea and its fungal symbiont Tulasnella sp., was performed, resembling early events in orchids-fungus interaction. The experiment aimed to investigate the fungal gene expression at different time points during the establishment of the orchid mycorrhizal symbiosis, to reveal the signaling pathways involved in the interaction. By combining RNA sequencing and untargeted metabolomics approaches, we compared the differential accumulation of transcripts and metabolites in different stages of the mycelium growth in the presence of orchid seeds before (T1, T2) and after protocorm development (MYC, SYMB), comparing them with the free-living mycelium (FLM). First analysis revealed 38, 8, 302 and 8561 differentially expressed genes (DEGs) in stages T1 and T2, MYC and SYMB, respectively, that were functionally characterized through in silico analysis. Additionally, metabolomic analyses showed significant changes of lipids, amino acids and peptide-like compounds in each condition. Integration of both omics approaches will provide useful data on the fungal changes in early events of symbiosis.

48 - Study of the transcriptional regulation controlling plant response to the Red and Far Red ratio.

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Plants live in a changing environment, and they have to constantly adapt their growth to that. Light is one of the most important environmental stimuli that affects all plant life-cycle and provides information about the surrounding. For example, the decrease in the ratio between the Red and Far Red (R/FR) components of sunlight is a signal of the proximity of other plants, which could compete for light and nutrients. This signal triggers a series of elongation responses in sun-loving species, such as Arabidopsis thaliana, that serve to avoid the shaded environment created by dense vegetation. The low R/FR-mediated elongation responses are achieved by the remodeling of gene expression, which is induced by the transcription factors Phytochrome Interacting Factors (PIF). In this work, we deeply characterized the transcriptional response showing that genes induced by low R/FR have an accessible chromatin conformation on their promoters that ensures a rapid but transient upregulation that depends mainly on PIF levels. Furthermore, we found that HFR1 (long Hypocotyl in Far Red 1), a negative regulator of hypocotyl elongation, is the only PIF target that shows a rapid but persistent upregulation in low R/FR. This persistent upregulation is achieved by the opening of the chromatin at the transcriptional starting site, which does not depend on PIF activity. The discovery of this regulation is particularly interesting considering that HFR1 is a negative regulator of PIFs activity and levels, suggesting that Arabidopsis evolved a complex gas-and-brake mechanism that allows a prompt but modulable response to light fluctuations.

58 - New insights into the molecular mechanism regulating primary root development in rice (Oryza sativa, L.)

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The root is a fundamental organ for plant nutrition, anchoring and perception of soil-born stresses. The root structure is the result of genetic and environmental factors, and it is dynamic, so that its development is modulated during the plant growth in response of environmental conditions (plasticity). This ability is related to the activity of the root apical meristem (RAM) which has been shown to be regulated by hormones and ROS in Arabidopsis. In a translational work, we found out similarities between rice and Arabidopsis root development. First, we imaged the rice seminal root and revealed its structure at the cellular level. We observed that the transition zone (TZ) positioning, and thus the meristem size, was influenced by cytokinins, ROS and high salinity. We also found similarities in the distribution of H2O2 and O-2. Finally, we searched for genes homologous to that of Arabidopsis and involved in determining the meristem size. We found candidate genes belonging to the families of PINs, RRs, GH3s that showed different expression profiles in response to cytokinins and H2O2. Our results showed that similar mechanisms regulated the RAM size in rice and Arabidopsis laying the base for the understanding of rice root plasticity under high salinity conditions, a major threat to rice productivity worldwide.

References: Formentin et al., Front Plant Sci 2018; Formentin et al., Plants 2018; Kajala et al., Cell 2021; Reynoso et al, Dev. Cell 2022

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62 - The miR319-LA-SFT module is needed for strigolactones to promote flowering in tomato

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Strigolactones (SL) are phytohormones with functions in the interaction with (micro)organisms in the rhizosphere and in stress responses. Also, while their effects on vegetative development are well studied, not so is their role in reproduction. We investigated the effects of genetic and chemical modification of SL levels on flowering in tomato (Solanum lycopersicum L.), and the molecular mechanisms underlying such effects. Results showed that SL levels in the shoot correlate with the speed of flower development, with the number of flowers, and with the transcripts of the florigen-encoding SINGLE FLOWER TRUSS (SFT). Genome-wide and targeted transcript quantifications in the leaves, coupled to metabolite analyses, demonstrated for the first time that SL promote tomato flowering by activating the microRNA319-LA (LANCEOLATE) module in leaves, which in turn decreases the content of bioactive gibberellins and increases SFT transcription. Several other floral markers and morpho-anatomical features of developmental progression are induced in the apical meristems upon treatment with SL, which seem to affect flower development more markedly than floral transition. Nonetheless, the induction of SFT and the promotion of flower development by exogenous SL can occur both before and after floral transition, and are blocked in plants expressing a miR319-resistant version of LA. Our study represents the first positioning of SL in the flowering regulation network in any plant species.

66 - Deciphering the biochemical properties of aldo-keto reductases from Arabidopsis thaliana: a new class of enzyme with a central role in GSNO homeostasis

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Recently, human aldo-keto reductase (AKR) 1A1 has been identified as being part of a new class of enzymes involved in S-nitrosoglutathione (GSNO) catabolism by a mechanism resembling that of S-nitrosoglutathione reductase (GSNOR), a ubiquitous NADH-dependent enzyme, but using NADPH as a source of reducing equivalent. Scoring the Arabidopsis thaliana genome of orthologs for human AKR1A1, we found four genes encoding AKRs that exhibit the greatest sequence similarity with the human enzyme. Arabidopsis AKR4Cs were then expressed in bacterial cells and purified to homogeinity. Their kinetic properties were determined starting from the Michaelis-Menten constants (Km) and turnover numbers (kcat) for both GSNO and NADPH. We have also tested their ability to use alternative substrates such as nitrosylated form of truncated glutathione (i.e., nitrosylated γ-Glu-Cys and Cys-Gly) and investigated the redox sensitivities of each enzyme to oxidative molecules like hydrogen peroxide, oxidized glutathione (GSSG), and GSNO. The results obtained revealed that AtAKR4C8 seems to be the most catalytically efficient isoform with an affinity toward GSNO similar to that measured for GSNOR (~30 μM). In addition, it is the only paralog that could be inactivated by oxidant treatments, indicating that it contains one or more redox sensitive cysteine thiols. Finally, these results suggest the potential role of AKR4Cs in controlling GSNO homeostasis in plants.

81 - The Arabidopsis NPK1-related protein kinases regulate responses triggered by the endoplasmic reticulum stress

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The endoplasmic reticulum (ER) is involved in the biosynthesis and folding of secreted and plasma membrane proteins among which there are elements involved in cell wall (CW) deposition. In presence of adverse conditions, unfolded/misfolded proteins accumulate in the ER leading to a condition known as ER stress which is mitigated by the activation of unfolded protein response (UPR). In Arabidopsis, treatment with ER stress inducer tunicamycin (Tm) or with the cellulose biosynthesis inhibitor isoxaben (ISX), causes cellulose reduction triggering the CW damage syndrome, ER stress and the concomitant activation of UPR pathways. The MAP triple kinases (MAP3K) named ARABIDOPSIS NPK1-RELATED PROTEIN KINASES (ANP1-3) are involved in this process since their loss phenocopies ISX and Tm responses and their overexpression confer resistance to prolonged ER stress. ANPs are critical components of intracellular transduction in biotic stress responses and localized in different subcellular compartments in response to CW-derived signals. The aim of this study is providing knowledge of the role of ANPs on ER stress response and cell wall deposition and how different organelles contribute to the perception and adaptation to environmental clues.

84 - New insights on oxygen-dependent proteostasis in plants from a synthetic strategy

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Plants use a complex set of molecular pathways to attune their physiology and development to the oxygen available within tissues. Direct O2 perception relies on plant cysteine oxidase (PCO) enzymes, whose kinetic properties qualify them as genuine O2 sensors under physiological conditions. PCOs use molecular oxygen to oxidize N-terminal Cys residues from substrate proteins and channel them to the Cys/Arg N-degron pathway of proteasomal degradation (Cys-NDP). The identification of PCO substrates is therefore crucial to gain full understanding of the role of oxygen as a signal in plant endogenous processes. The only PCO substrates characterized since the discovery of the pathway are a handful of Met-Cys-starting (MC-) proteins. To create a simplified in vivo platform for the investigation of PCO targets, we reconstituted the plant Cys-NDP by introducing a PCO enzyme in Saccharomyces cerevisiae, where cysteine oxidases are absent. Incorporation of MC- protein sequences in suitable reporter constructs helped us evaluate PCO activity with a simple luciferase assay in this synthetic strain. We assayed a broad array of previously unexplored MC-proteins from the Arabidopsis proteome and successfully recovered a set of novel Cys-NDP targets, currently under validation in Arabidopsis. The new substrates, encompassing enzymes and transcription factors, potentially expand by far the information on the extent of O2-dependent regulation in plant development and stress responses.

93 - EXOGENOUS STRIGOLACTONES PROMOTE TOMATO BERRY RIPENING.

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Strigolactones (SLs) are known to play a role in several plant developmental processes, also through the interaction with other phytohormones. Nevertheless, there is a huge lack of information regarding their role during fruit ripening. It is known that in grapevine berries, SLs interact with abscisic acid-mediated accumulation of anthocyanins (Ferrero et al., 2018). Ethylene is the plant hormone responsible for ripening onset in climacteric fruits. In Arabidopsis, Ueda et al. (2015) demonstrated how SLs enhance ethylene action in leaf senescence processes.

In order to investigate changes in metabolic patterns of tomato fruits in post-harvest phase and to evaluate putative relations with ripening processes, we performed exogenous application of rac-GR24 (5DS), a biosynthetic analogue of SLs, on tomato berries of the Butalina and Micro-Tom varieties, both collected at the 'mature green' stage. The treatment was effective in promoting a faster change of Butalina berry skin color from green to red. Color changes in MicroTom were less visible, but lycopene quantification and induction of genes related to lycopene biosynthesis (SlPDS and SlPSY1) confirmed the activating effect on ripening. Treated berries displayed a final decrease in total soluble sugar content. ABA content and SlNCED1 transcription were negatively affected by the treatment, whilst ABA sensitivity was probably increased. ABA catabolism-related genes, SlCYP707A1 and SlUGT75C1, were indeed activated by rac-GR24 (5DS). Moreover, genes encoding for enzymes directly involved in ethylene production, such as SlACO1 and SlACS2, were strongly up regulated.

These results suggest that SLs may be involved in the maturation processes of tomato fruits.

106 - BON1: a Ca2+-binding protein at the crosstalk between plant development and plant response to environmental stimuli

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Arabidopsis BON1 is a copine with two domains that bind Ca2+ and a vWA domain responsible for interaction with targets. BON1 is plasma membrane localised and is involved in the plant immunity. Our analyses on two independent bon1 homozygous mutants show a drastically reduced adult plant size and defects in phyllotaxis that start to be visible 3 weeks after germination and has gene-dosage dependence revealing that BON1 has also a role in plant development and in particular during the progression from vegetative to reproductive growth. The vWA domain of BON1 interact with the autoinhibitory domain of 2 plasma membrane Ca2+-ATPases, ACA8 and ACA10, to possibly regulate their activity. The BON1-Ca2+-ATPase interaction caught our attention, and we examined the effects of BON1 on ACA8. Results show that the ACA8 activity is significantly stimulated by BON1 in a concentration-dependent manner showing that BON1 activates the pump. To investigate if the developmental defects of bon1 can be ascribed to its regulative effect on Ca2+-ATPases, hence, on cytosolic Ca2+ homeostasis, bon1 and aca8aca10 mutants expressing the biosensor NES-YC3.6 have been analysed for both the resting cytosolic Ca2+ concentrations and for Ca2+ transients induced in response to external stimuli. Analysis in the shoot apical meristem, where the two partner proteins are co-expressed, reveal that Ca2+ dynamics are altered. To study in planta the dynamics of BON1-ACA8 interaction, FRET-FLIM analyses are ongoing.

119 - mtDNA repair machinery is central in salt stress response

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The importance of organelles as central coordinators of plant responses to internal/external stimuli has become increasingly important. Mitochondria play a role as stress sensors of environmental stimuli, being a component of a complex communication network involving organelles and the nucleus. WHIRLY2, one of the most abundant proteins involved in mtDNA repair, has been shown to be crucial in mitochondria morphology and functionality.

Our analyses show that in the presence of salt, WHIRLY2 is crucial for protecting mtDNA from oxidative damage and regulating its repair. The destruction of nucleoid integrity, due to the absence of WHIRLY2, impairs the proper redox-dependent response. Loss-of-function why2 mutants are indeed highly sensitive to salt stress, mainly due to their inability in orchestrating a proper response against the stress.

126 - GLR3.7 as negative regulator of GLR3.3 mediated stress responses

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During evolution, plants have developed sophisticated mechanisms to perceive and respond to the environment. Adaptability and responsivity to biotic and abiotic stresses is a key mechanism that involves multiple factors: second messengers, phytohormones, metabolites and a whole variety of molecules involved into the perception, distribution, and transduction of stress signals. Recent works showed that wounding-driven long-distance propagation of Ca2+ waves depend on the activity of the cation permeable channel Glutamate Like Receptor 3.3 (GLR3.3), but further information on how this key factor is regulated is still scarce.

GLRs are homologous to the animals' ionotropic glutamate receptor channels (iGluRs), which can form both homo and hetero-tetramer. This allow us to hypothesize that different assemblies of plant GLRs as well can bring to the formation of channels with different properties.

Here we present multiple lines of evidence supporting the molecular and functional interdependency between GLR3.3 and another relatively uncharacterized GLR isoform, GLR3.7.

Colocalization and close proximity of GLR3.3 and GLR3.7, both at tissue and cellular level, has been demonstrated by GUS reporter assay, complemented by a FLIM analysis.

Subsequently, in vivo Ca2+ imaging and qReal-Time PCR experiments assessed the impact of GLR3.7 activity on GLR3.3-dependent responses, such as burning stress and amino acid administration.

Taken together, our results suggest that GLR3.7 can regulate the activity of GLR3.3 by possibly forming, in planta, a heteromeric channel.

28 - How much disordered is the conditionally disordered protein CP12 in its different redox states?

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CP12 is a conditionally disordered protein (IDP) known as a light-dependent redox switch containing two pairs of regulatory cysteines located at either terminus of the protein. By serving as a scaffold in the formation of a multiprotein complex, CP12 regulates the enzymatic activity of two non-consecutive and energy-consuming enzymes of the Calvin-Benson cycle. Until now, structural studies of unstructured proteins were hardly feasible. However, thanks to the implementation of the small angle X-ray scattering (SAXS) approach it is now possible to shed light on this complex topic.

SAXS analyses of Arabidopsis thaliana CP12 (AtCP12) in its different redox states confirm the highly disordered nature of the protein in solution, showing that the reduced form appears more expanded than the oxidized one. In both redox states, AtCP12 behaves like a highly flexible biomolecule, as can be deduced from Kratky plot profiles of AtCP12 compared with folded and unfolded reference proteins. However, the oxidized AtCP12 appears more compact than the reduced form. Using the EOM approach, it was possible to describe oxidized AtCP12 as a mixture of conformers in which highly disordered molecules (in spite of the two disulfide bridges) account for 75% of the total, while molecules that are partially structured at the N-teminus account for the remaining 25%. These results rule out the existence of significant amounts of structured and compact conformations of free AtCP12 in solution.

39 - Uncovering the functional properties of ribulose-5-phosphate 3-epimerase (RPE) from the green microalga Chlamydomonas reinhardtii

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Photosynthetic carbon fixation is realized by eleven enzymes in the so-called Calvin-Benson-Bassham cycle (CBBC), where essential epimerization of xylulose-5-phosphate (Xu5P) into ribulose-5-phosphate (Ru5P) is catalyzed by the ribulose-5-phosphate 3-epimerase (RPE). In this study, we aimed at investigating the structural and functional characteristics of RPE from the model microalga Chlamydomonas reinhardtii (CrRPE), which is demonstrated to be an amphibolic enzyme participating both in the CBBC and in the oxidative pentose phosphate pathway.

The crystal structure of CrRPE has been solved at a resolution of 1.96 Å. A sequence comparison with homologous epimerases from plant and non-plant sources outlined a high conservation of the active center, suggesting a possibly shared catalytic mechanism. Reliable multi-step enzymatic assays were set up and optimized to determine the kinetic parameters related to the conversion activities of Ru5P to Xu5P and vice-versa, which allowed to deduce RPE catalytic functioning in the context of physiological conditions. Finally, we investigated the effects of redox-based modifications on protein catalysis, since in the last decades several proteomic studies identified plant RPEs as putative targets of various oxidative modifications.

In conclusion, this work aims at expanding the basal knowledge on the functioning and regulation of the CBBC, a key step in laying the basis for future improvements of carbon fixation and primary production.

41 - Differential stomatal density and size responses to drought in three Cannabis sativa genotypes: effect on stomatal conductance and physiological stomatal behaviour

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The identification of plant genotypes with effective physiological and morphological stomatal control is a major component of phenotyping efforts to select more productive climate resilient varieties. We examined the physiological and stomatal responses to water deficit of three varieties of Cannabis sativa with contrasting leaf areas (LA). Under full irrigation, all three varieties showed identical stomatal size (SS) and density (SD) values. However, under water deficit, the variety with the largest LA (Fibror) showed the greatest increase in SD (83%) and reduction in SS (61%). The variety with the smallest LA (Earlina) exhibited a lower increase in SD (40%) and reduction in SS (35%). Interestingly, stomatal initiation increased equally in all three genotypes under water deficit, and the area of the epidermis allotted to gas exchange was unaffected by soil water availability. The speed of stomatal conductance (Gs) adjustment is thought to be faster in plants with a high density of small stomata. This was not the case in leaves of the C. sativa genotypes developed under both irrigation regimes. However, the relative extent of adjustment in Gs values (from maximum Gs in the light to stomatal closure induced by darkness) was related to size and density. This may suggest that the interaction between stomatal morphology and physiology is more complex than previously considered and likely involves variation in guard cell signalling and biochemical properties.

PLANT IMMUNITY

3 - Protein precursors for effective plant immunity: the case of pectin methylesterases

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ABSTRACT

Plants exploit an immunity-related pectin methylesterase (IT-PME) activity against microbes (Del Corpo et al., 2020). This process could induce the formation of "egg box" structures, resulting in stiffening of the cell wall. IT-PME activity may also promote the production/sensing of damage-associated molecular patterns such as oligogalacturonides and methanol, promoting defensive priming in plants. PME activity can also favour the binding of pathogen recognition receptors such as wall-associated kinases (WAKs), RESISTANCE TO FUSARIUM OXYSPORUM 1 (RFO1) and FERONIA to de-methylesterified pectins. A precise on/off mechanisms regulating PME activity seem necessary for an efficient defense response without compromising growth (Lionetti et al., 2017; Coculo et al., 2022). We revealed that Arabidopsis PMEs involved in plant defense are pro-proteins. We identified two subtilases (SBT), SBT3.3 and SBT3.5, as modulators of IT-PME activity in Arabidopsis immunity to Botrytis. SBT3.3 follows an unconventional secretion pathway to reach the CW where they can activate Pro-PMEs secreted through a conventional protein secretion. The Pro region shares structural similarities with functionally characterized PME inhibitors, mediators of maintenance of CW integrity. Our research reveals the activation of PRO-PMEs as a resistance factor against pathogens. Modification, perception, and signal transduction of pectin methylesterification in plant immunity will be discussed.

Coculo, D.et al. (2022) bioRxiv 2022.07.28.501549

Del Corpo, D. et al (2020) Mol. Plant Pathol., 21, 1620–1633.

Lionetti, V. et al. (2017) Plant Physiol., 173, 1844–1863.

5 - Priming of special metabolism in Arabidopsis thaliana mediates elicitor-induced resistance to fungal infection.

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Biotic elicitors trigger in plants a robust but transient expression of defense-related genes, followed by a reduced susceptibility to pathogen infection. To investigate the molecular basis of elicitor-induced resistance, we analzyed the transcriptome of Arabidopsis leaves treated with the bacterial elicitor flg22 and inoculated, after 24 h, with the fungal pathogen Botrytis cinerea. In the absence of infection, defense-related genes were not significantly up-regulated 24 h after elicitation, though a transient repression of growth-related genes could be observed. In contrast, flg22-treated plants showed, upon infection, increased expression of genes involved in special metabolism. Genetic analysis confirmed that indolic compounds and aliphatic glucosinolates contribute to flg22-induced resistance. Moreover, elicitation did not affect seed production, suggesting that priming of special metabolism allows increased resistance to pathogens with minimal impact on fitness.

46 - (±)-3-Deoxyradicinin Induces Stomata Opening and Chloroplast Oxidative Stress in Tomato (Solanum lycopersicum L.)

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Radicinin is a phytotoxic dihydropyranopyran-4,5-dione isolated from the culture filtrates of Cochliobolus australiensis, a phytopathogenic fungus of the invasive weed buffelgrass (Cenchrus ciliaris). Radicinin proved to have interesting potential as a natural herbicide. Being interested in elucidating the mechanism of action and considering radicinin is produced in small quantities by C. australiensis, we opted to use (±)-3-deoxyradicinin, a synthetic analogue of radicinin that is available in larger quantities and shows radicinin-like phytotoxic activities. To obtain information about subcellular targets and mechanism(s) of action of the toxin, the study was carried out by using tomato (Solanum lycopersicum L.), which, apart from its economic relevance, has become a model plant species for physiological and molecular studies. Results of biochemical assays showed that (±)-3-deoxyradicinin administration to leaves induced chlorosis, ion leakage, hydrogen peroxide production, and membrane lipid peroxidation. Remarkably, the compound determined the uncontrolled opening of stomata, which, in turn, resulted in plant wilting. Confocal microscopy analysis of protoplasts treated with (±)-3-deoxyradicinin ascertained that the toxin targeted chloroplasts, eliciting an overproduction of reactive singlet oxygen species. This oxidative stress status was related by qRT-PCR experiments to the activation of transcription of genes of a chloroplast-specific pathway of programmed cell death.

47 - A biostimulant based on a brown alga (Ecklonia maxima) and yeast extract confers resistance toward Pseudomonas syringae pv. tomato DC3000

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Biotic stress is one of the leading causes of the loss of crop productivity. The need to reduce the use of chemical agents in agriculture requires to develop sustainable alternatives. One of the most promising solutions to address this important challenge involves the use of biostimulants, commonly based on seaweed extracts, complex organic materials, hormone-like compounds, amino acids, and humic acids. In this work, a first screening of different plant biostimulants such as triacontanol, chitosan, fulvic acids, and a seaweed/yeast combination extract was performed on tomato seedlings, to evaluate their possible antimicrobial role against the infection with the bacterium Pseudomonas syringae pv. tomato DC3000 (Pst). The results underlined the promising bioprotectant activity of the Ecklonia maxima-based biostimulant. Therefore, its role in mature tomato plants was deeply investigated. After the infection, the biostimulated plants showed a less affected phenotype and significantly reduced Pst replication in leaves. Consistently, callose deposition was promoted in biostimulated plants. Furthermore, ion leakage and oxidative damage induced by the biotic stress were mitigated, while the hormone-mediated response to stress and the expression of biotic stress-related genes were increased. In conclusion, our study demonstrated that biostimulation confers resistance to Pst and proposes a low-cost and eco-friendly strategy as a useful tool to increase plant resistance to pathogens.

57 - Systemin is not alone: its precursor provides new peptides with still unexplored functions

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In the history of tomato defense responses, the focus has often been the systemin peptide (Sys) as a key component of plant immunity, activating multiple responses to a wide range of stressors. However, the mechanisms linked to such a broad protection capacity are also possibly linked to the intrinsic disorder of its precursor protein, known as Prosystemin (ProSys). Since recent findings suggest that ProSys has its own biological functions besides Sys sequence, we assumed that this precursor provides other peptide motifs with a role in defense-related pathways. Candidate peptides were identified in silico and synthetically produced to investigate their performance in protecting tomato plants from different biotic stressors. Our results reveal that ProSys harbours several repeats motifs that, when exogenously supplied in small doses, are able to trigger plant immune responses against pathogens and pest insects. An intriguing result comes from mass spectrometry analysis performed on plants overexpressing ProSys, validating the existence in vivo of three peptide motifs. These experimental data shed new light on ProSys unreported functions and provide a new chapter to be explored in the study of plant peptidome during stress events.

60 - Beyond Systemin: An in-deep screening of molecular mechanisms underpinning tomato Prosystemin

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As known plants constantly counteract several stresses through a very sophisticated immunity system, based on the recognition of the enemy's molecular patterns, called in general elicitors, that act as danger signals that trigger plant defenses. Among them tomato Systemin (Sys) promotes long-distance defense responses by amplifying the jasmonic acid pathway, leading to the production of defense compounds. Sys is proteolytically released from the C-terminal region of its precursor, Prosystemin (ProSys). Interestingly, ProSys is an intrinsically disordered protein and this suggest that it may interact with different molecular partners belonging to diverse biological pathways. In addition, we demonstrated that the exogenous application of ProSys devoid of Sys peptide (here referred like ProSys(1-178)) on tomato leaves, is able to confer protection against different stress agents, highlighting different functions of ProSys(1-178). To deeply understand the role of ProSys(1-178) in tomato defenses we designed ad hoc experiments in which we compared the impact on tomato plants of ProSys(1-178) and Sys peptide, when exogenously delivered. Preliminary studies, conducted through the expression analysis of genes related to different biosynthetic pathway, suggested that ProSys(1-178) may be involved in different phytohormone pathways. Our previous works and this preliminary findings highlight the multifaceted roles of ProSys(1-178) and open up new opportunities of research on it functional mechanism.

80 - Phyllosphere microbiota composition of tomato plants treated with Prosystemin-derived peptide

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The employment of growth or defense inducers, commonly used in agriculture to maintain crop health, may impact on both structure and behaviour of plant microbial population. In this context, plant phyllosphere is a flexible and challenging habitat involved in a dynamic microbial selection. Recent findings suggest that the application of protein hydrolysates and plant hormones can shape plant immunity responses, positively impacting the leaf microbiota. It is still unclear what molecular processes are responsible for the selection or the recruitment of useful bacteria, but many efforts have to be made. In this direction, we explored the impact of an elicitor peptide derived from Prosytemin protein, notably involved in tomato defense responses against biotic stress, on the epiphytic community of tomato leaves. 16S rRNA communities of tomato (cv. San Marzano Nano) phyllosphere of peptide and mock treated plants were profiled. Our results show that leaves microbiota undergoes a shift upon peptide treatment compared to control plants, with main differences in bacterial abundance. For example, the genera Photobacterium and Psychrobacter, associated with growth-promoting or antagonistic traits, increased in peptide-treated samples. This exploratory analysis strengthens the importance of using tools inspired by nature like plant-derived peptides to ensure plant health at multiple levels, including the promotion of beneficial plant-microbe interactions.

95 - The homeostasis of oligogalacturonides in the growth-defence trade off

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Plant immunity can be activated by endogenous elicitors referred to as damage-associated molecular patterns (DAMPs). Typical DAMPs are the oligogalacturonides (OGs), fragments released from cell wall pectin by the action of endopolygalacturonases (PGs). The accumulation of OGs in the apoplast is favored by plant-encoded polygalacturonate-inhibiting proteins (PGIPs) that inhibit microbial PGs. Expression of an inducible PGIP-PG chimera named "OG-machine" in transgenic Arabidopsis plants leads to the release OGs that confer resistance against pathogens. However, over-accumulation of OGs in these plants also results in growth inhibition (growth–defense trade-off) and hyper-immunity. A homeostatic control preventing the potential deleterious effects of OGs is played by at least four specific oxidases, named OGOXs, belonging to the Berberine Bridge Enzyme-like protein family. Through a reverse genetic approach, we show that OGOXs play a crucial role in the homeostasis of OG levels in the apoplast and in dampening OG elicitor activity. Furthermore, by expressing the OG machine in genetic backgrounds defective in key elements of plant immunity such as EDS1, RBOHD and JAR1, we show that the effects on growth and defense can be uncoupled. Our results suggest that a tuned orchestration of OG elicitor activity is crucial for maintaining the growth-defence trade-off in Arabidopsis.

NOVEL TECHNOLOGY FOR MULTISCALE LEVEL OF INVESTIGATION IN PLANT PHYSIOLOGY

20 - Mining the seagrass microbiota: the endophyte community of Posidonia oceanica seeds

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Several species of endophytic bacteria, fungi and actinomycetes colonize the inner plant tissues, where they establish mutualistic interactions with beneficial effects for their hosts. Most of the information available is referred to terrestrial plants, while less is known for marine plants. Adult Posidonia oceanica hosts a complex endophytic microbial community, but we do not know what endophytes are already present in seeds. This work investigated the composition of P. oceanica seed microbiota by employing a double strategy: compiling the whole inventory by metagenomic analysis of the bacterial and fungal community, and isolating their culturable fraction. A total of 42 bacterial strains were isolated and identified, including 18 species of halophilic or salt-tolerating species. Some species are known for their putative beneficial effects for the host plant and could be exploited in conservation/restoration programs to improve the seed storage and seedling development.

26 - Exploring new frontiers in botanical experiments: harnessing the potential of 3D printing for innovative research setups

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3D printing emerged in the early 1980s and experienced rapid growth from 2006 onwards, becoming widely accessible to the public while simultaneously lowering costs and increasing user-friendliness. This technology enables the conversion of digital models into physical objects, layer by layer, utilizing selective laser sintering (SLS) or fused deposition modeling (FDM). Notably, 3D printing offers numerous advantages, including rapid and cost-effective object prototyping, as well as the ability to fabricate shapes and solutions unattainable with other techniques. Furthermore, it provides flexibility in material selection, allowing, for example, for variations in elasticity and degradability.

As a case study, an innovative split-plot experimental design was developed to examine the interaction of plant roots with different environments. The pot design incorporates a specialized partition barrier, that create two distinct sectors: a Confined sector (CS) and a Free sector (FS). This configuration permits root passage between the sectors exclusively in one direction (from FS to CS). By leveraging the shape of the barrier and the root growth behaviors, such as gravitropism and response to obstacles, the roots are guided towards or away from the passage holes in the barrier, depending on its orientation. Additionally, the pot system has the capability to maintain separate watering for each sector, ensuring controlled hydration conditions or the utilization of different solutions.

51 - Enhancing secondary metabolites in ready-to-eat salads: Postpackaging UVB treatments to improve leaf quality of Lactuca sativa and Diplotaxis tenuifolia

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Ready-To-Eat (RTE) salads can promote the intake of secondary metabolites due to their easy handling and minimal processing. However, these compounds in the leaves of RTE species are usually low due to limited solar UV exposure during cultivation. To address this, we treated packaged fresh-cut lettuce and wild rocket leaves by UVB lamps during storage at 5 °C, 80% RH. Samples were kept under a 20 µmol m-2 s-1 white light 12h/day for 6 days. Half of them were additionally treated for 3 days, 9h/day, with UVB lamps at 3.2 and 0.9 µmol m-2s-1 of UVB and UVA, respectively. Photosynthetic parameters, epidermal phenolics (Ephen) and chlorophyll indices were monitored daily with fluorescence sensors during the 6-day storage. HPLC analysis of phenolics, carotenoids, chlorophylls and glucosinolates, as well as measurements of antioxidant activity and fresh weight loss were performed.

The UVB treatment increased Ephen without affecting chlorophyll, carotenoids, or photosynthesis. EPhen changes occurred~15 hours after the first UVB application, with wild rocket responding faster than lettuce. Treated samples had higher (about 50-60%) flavonoid contents (mainly quercetin derivatives) compared to controls. We proved that RTE salads can be treated with UVB radiation while packaged, resulting in increased phenolic content. These results contribute to the optimization of UVB treatments to enhance postharvest leaf quality toward promising food industry applications.

Keywords: Flavonols– Non-destructive optical sensors – UVB radiation – postharvest quality – ready-to-eat

64 - Naked Roots: unveiling the hidden dialogue between root and shoot

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Permanently restricted to their site of germination, plants evolved mechanisms to react to ever-changing environmental conditions and harmful stresses. Information about environmental stimuli that are locally perceived is often transmitted systemically by chemical and electrical signals, triggering responses in unharmed tissues and/or directing the coordination of developmental programs of different plant organs. In the last decade, the usage of fluorescence microscopy techniques for the measurement of second messengers in the entire plant (e.g Ca2+, reactive oxygen species, pH), coupled with the quantification of electrical signals, provided a detailed picture of both events and molecular players taking place in the aerial part of the plant. A large part of the data comes from studies in Arabidopsis subjected to leaf wounding, in which the activity of plant Glutamate Receptor-Like (GLRs) promotes (i) the transmission of leaf-to-leaf electrical signals and Ca2+ waves propagation, (ii) the increase in the concentration of the defense hormone jasmonate and (iii) the increase in the expression of jasmonate responsive genes in unwounded leaves. Unfortunately, technical limitations hamper the assessment of events orchestrating the communication between the root system and the shoot, which is fundamental for optimizing resource allocation and water balance. Here, by developing a rhizotron-based system to gain direct optical access to both root and shoot in Arabidopsis, we describe a novel shoot-to-root Ca2+- and pH-wave propagation upon the mechanical damage of the leaf. Moreover, we show that rehydration of plants under drought stress induces the rapid generation of sustained root-to-shoot Ca2+ waves.

120 - Does exposure to Rare Earth Elements cause adverse effects on the Brassicaceae Sinapis alba?

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Rare Earth Elements (REEs) have become essential in today's technology due to their range of properties. Anthropogenic activities have led to an increase in their environmental concentrations, and the potential effects on the environment are not yet well known.

In this study, effects on Sinapis alba were evaluated by the individual exposure to REEs Lanthanum (La) and Gadolinium (Gd) for 72h with concentrations ranging from 0.005 to 50 mg L-1. The exposure was performed under dark conditions at 24 ± 2 °C, and the solutions were adjusted to an initial pH of 6.3 (± 0.2) to avoid precipitation.

Preliminary results display differences in the effects between the two REEs on the growth, ascorbate (ASC), and dehydroascorbate (DHA) content. Under the exposure conditions, there was no effect on the germination. Exposure to La caused an increase in the plant length, in comparison to the control in four of the five concentrations. Conversely, all concentrations of Gd exposure caused a decrease in length in comparison to the control. La exposure caused also a reduction in the ASC content at lower concentrations, with the highest reduction observed at 0.05 mg L-1, followed by the highest increase at 0.5 mg L-1. For the Gd exposure, all concentrations caused a reduction in the ASC and DHA content, with the lowest concentration resulting in the highest reduction.

These data indicate the presence of metabolic perturbations induced by Gd and La, however, no clear pattern behavior can be yet established. Further endpoints will be studied including the protein content, enzymatic and non-enzymatic antioxidants, generic peroxidases, and catalase to better identify a link, or a lack of one, on the effects caused by the exposure.

122 - Enhancement of antioxidant capacity induced by LED Lights in microgreens of Cucurbita pepo L. var. sapling of sarzana and biological activity's evaluation in vitro

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Microgreens are newly sprouted, immature plants without roots that are harvested after the development of the cotyledon leaves, or seed leaves, usually between 10 and 14 days from seeding. This functionality is attributed to their high content of vitamins and minerals, as well as other bioactive compounds. It has been reported that many species of microgreens are more saturated with micronutrients than the adult versions of the same plants. For instance, microgreens have shown to be high in vitamins or their precursors, including carotenoids, ascorbic acid, chlorophyll, phenolic compounds, and glucosinolates. This study aimed to evaluate the effect of light-emitting diodes (LEDs) on the antioxidant nutrient content. Seeds were grown under monochromatic blue (470nm) and red (630nm) LED lights with an intensity of 100 ± 10 µmoL m−2 s−1 at 24 ± 1 °C for ten days and in dark conditions for the control. Analyses were conducted on both the aerial and root parts. The parameters measured were dry matter content, ascorbic acid content, glutathione content, chlorophylls and carotenoids, and polyphenols. The antioxidant capacity of polyphenolic extracts of the aerial part and root of zucchini microgreens was evaluated on Caco-2 cell line by cellular antioxidant activity (CAA) assays. Results showed that blue light had a positive effect on the increase of antioxidant compounds in aerial part of zucchini microgreens and, in addition, the efficacy of this blue light was also confirmed in vitro by CAA biological assay.

10 - Seed priming as a tool to improve germination in poplar seeds and preserve local biodiversity

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The need to recover degraded areas demands for new approaches to plan effective ecosystem restoration programs. These are often associated with nature-based solutions (NBS), and the development of novel tools to support and maximize ecosystem services. In a local context, Populus spp. (poplar) is used both for restoration and economically relevant purposes (e.g., plywood and other wood base panels). Currently, the natural poplar plantations are facing a crisis due to the reduction of land availability, and a decline in genetic variation. The latter may be also due to seed germinability issues which can have a negative impact on breeding programs. Seed priming, a pre-sowing treatment consisting of controlled rehydration-dehydration cycle resulting in enhanced seed germination, may represent a tool to overcome this bottleneck. However, poplar seeds are classified as recalcitrant since they have low dehydration tolerance, relatively high water content at maturity, and high metabolic activity at dehiscence. Therefore, there is an urgency to design specific priming protocols for poplar seeds. The current study proposes to look into easy-to-use and environmentally-friendly approaches to develop poplar seed priming techniques as tools to be included in the NBS catalog to support ecosystem restoration as part of the National Recovery and Resilience Plan (PNRR), Activity 5 “Conceptual framework and methodological tools of Nature Based Solution and Restoration Ecology”.

12 - Boosting technologies of orphan legumes towards resilient farming systems in the Greater Mediterranean Region: from bench to open field (BENEFIT-Med)

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The BENEFIT-Med project, funded by PRIMA (Partnership for Research and Innovation in the Mediterranean Area) programme in the EU framework for research and innovation, aims to develop an innovative and sustainable technology based on functional, highly resilient orphan legume accessions and 'on-farm' seed biopriming to boost seed vigor and seedling performance under adverse environments. Such novel farming solutions will be tested on trigonella (Trigonella foenum-graecum L.), grass pea (Lathyrus sativus L.), and forage pea (Pisum sativum var. arvense L.), regarded as an excellent source of beneficial nutrients. Using a participatory approach, local farmers will be trained on the best practices of 'on-farm' seed priming applied to orphan legumes. By reinforcing technical knowledge at individual, organizational, and community level, the capacity building of stakeholders will be promoted. An ex-ante evaluation integrated by Life Cycle Assessment (LCA), Life Cycle Costing (LCC), Social Life Cycle Assessment (S-LCA) will define the economic performance and the environmental/health/social impact of the new farming solutions. Crop suitability will be provided for the Mediterranean basin area to identify vulnerable regions under future climate conditions. The BENEFIT-Med consortium, coordinated by the University of Pavia, includes eleven partners from eight different countries.

13 - From bench to field: scaling-up seed priming technology on orphan legumes of the Mediterranean area

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Understanding and improving seed quality is crucial for globally established crops as well as for valorization of agrobiodiversity. One of the main objectives of the BENEFIT-Med project (https://www.benefit-med.eu/) is the optimization of seed priming protocols as sustainable approaches to improve germination performance in legume varieties adapted to dry environments. Six varieties from three Fabaceae species, Latyrus sativus, Pisum sativum and Trigonella foenum-graecum, have been selected as representative of commercially underutilized ‘orphan’ legumes of the Mediterranean area. Hydropriming and biopriming protocols have been developed and tested under in vitro and in-soil controlled conditions in order to establish suitable experimental systems to explore the molecular bases of seeds priming effectiveness in climate-ready crops.

15 - The importance of bedrock water for tree water relations under drought: experimental evidence and challenging questions

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Large portions of terrestrial habitats are dominated by shallow (< 50 cm) soils overlying compact bedrock. Experiments suggest that some rock types can store appreciable amounts of water, which can be released in a water potential interval compatible with physiological ranges of root water uptake. However, it is not clear how and how much of this water can be exploited by plants under drought, depending on rock characteristics (porosity, hydraulic conductivity), root hydraulics and rock-root interaction. Indeed, Fraxinus ornus trees facing summer drought maintained a better water status when growing on a more porous bedrock formation (breccia, with higher plant available water content, AWC) than on dolostone (less porous, lower AWC). Potted F. ornus saplings grown in soil-rock mixtures were subjected to well-watered and drought conditions and the hydraulic conductivities of the single components of the system, i.e. rocks (breccia and dolostone), soil, roots and whole plant were quantified. Data were implemented into 2D model simulations of the rock-root water exchange. Simulations indicated higher exploitation of water pools in the breccia than in the dolostone systems in the dry range (soil water potentials between -1.0 and -1.5 MPa). These results support the field evidence and suggest to consider bedrock available water for a better understanding of spatial variations in vegetation water status and dieback patterns under drought in rock-dominated landscapes.

18 - Antarctic psychrotolerant bacteria enhance tomato tolerance to cold stress and cause metabolic changes

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Plant-associated microorganisms can mitigate cold stress, promoting plant acclimation to low temperatures. However, scarce information is available on the molecular mechanisms underlying this process. This work aims to understand the mechanisms activated by psychrotolerant bacteria on tomato plants and identify metabolites involved in mitigating cold stress. Bacterial isolates from the Antarctic plant Colobanthus quitensis (Hafnia sp., Pseudomonas sp.) and a well-studied endophyte (Paraburkholderia phytofirmans PsJN) were selected for their ability to promote tomato shoot growth at 15°C. The isolate Hafnia sp. produced indolacetic acid in vitro at 25°C and at 4°C, when the medium was supplemented with tryptophan. To characterize the capacity of these isolates to affect plant metabolism, surface-disinfected seeds were inoculated with the respective bacterial strain, and mock-inoculated seeds were used as a control. Four-week-old plants were exposed to 4°C for 7 days in the dark and incubated at 25°C for zero, two, and four days to allow their recovery from the stress. Metabolites were determined with high-performance liquid chromatography-high resolution mass spectrometry (HPLC-HRMS). Bacterium-inoculated plants showed lower concentrations of polyphenols compared to mock-inoculated plants. In addition, bacterium-inoculated plants accumulated higher content of putative phenylalanine-containing dipeptides, suggesting that these compounds may be involved in cold tolerance. This study indicated that bacterial isolates were able to reprogram phenylalanine metabolism in cold-stressed tomato plants. A deeper understanding of the mechanisms promoted by beneficial bacteria in tomato plants will allow the development of sustainable strategies to protect plants against cold stress.

24 - Unravelling rice-native soil microorganism interactions involved in rice seedling establishment under flooding

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Flooding events caused by severe rains and poor soil drainage can interfere with plant germination and seedling establishment. Rice (Oriza sativa) is a cereal crops with unique germination strategies under flooding, which are the consequence of complex physiological and molecular mechanisms. In this context, a better understanding of the soil microbial biodiversity in agroecosystem is crucial. Soil microorganisms can contribute to enhance nutrient uptake and to lessen the impact of flooding stress, reducing costs and improving quality and quantity of the production. Our aim is to conduct a multiscale approach to assess the interaction between rice and the native soil microorganism in order to understand the plant molecular mechanisms involved. Rhizosphere and rhizoplane potential and active microbial community will be identified at different stages of plant development on rice plants germinated on paddy soil. This will shed light on possible microorganism harbouring plant growth promoting activities that contribute to rice seedling establishment under submergence.

27 - Unraveling the role of second messengers in salt tolerance in rice (Oryza sativa, L.)

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Unraveling the role of second messengers in salt tolerance in rice (Oryza sativa, L.)

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Soil salinization is one of the greatest threats to crop production and this problem is expected to increase in the coming years due to climate change. Rice, a staple food for more than half the world population, is the most salt-sensitive species among cereals. In a previous comparison between salt tolerant and sensitive rice varieties, Baldo (B) and Vialone Nano (VN), respectively, we found different salt-stress induced Ca2+ transients in the two roots and salt-stress induced early peak of H2O2 in B roots. How tolerant plants can respond to salt stress earlier than sensitive plants is still unknown. Since plasma membrane is involved in mediating Ca2+ and ROS signals, it is possible that B and VN have a different membrane protein composition. In this work, transcriptomic data from VN and B plants were analysed to identify DE membrane proteins that could be related to B tolerance. Among the genes involved in Ca2+ transport, OSCA2.2, MLO8, two MCU isoforms, and GLR3.1/4 were found in DEGs as well as two aquaporin genes putatively involved in H2O2 transport. Interestingly, CIPK9 is expressed at a higher level in B roots. Furthermore, we generated rice plants harbouring a genetically encoded probe for ROS. Preliminary analyses showed different responses to various stresses. Our results show that differences in the plasma membrane components are present. Whether these differences are responsible for the different Ca2+/ H2O2 dynamics will be the focus of further experiments.

29 - Functional properties and regulation of delta-1-pyrroline-5-carboxylate synthetase isoforms from Arabidopsis thaliana.

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Plant genetic improvement for increased stress tolerance is required to ensure higher crop productivity, also taking into account the threat represented by the ongoing climate change. Under water stress conditions, plants achieve osmotic compensation through the accumulation of the so-called compatible osmolytes, among which the amino acid proline is the most widespread. The beneficial effects of proline are believed to go well beyond counteracting the adverse effects of low water potential, and comprise regulation of the redox status of the cell and the NAD(P)H/NAD(P)+ ratio. The limiting step in proline synthesis is catalysed by δ1-pyrroline-5-carboxylate (P5C) synthetase (P5CS), which mediates the phosphorylation of glutamate, and the reduction of the product to glutamate-5-semialdehyde, which spontaneously cyclizes to P5C. At least two P5CS isozymes have been found in most angiosperms, one constitutively expressed to satisfy proline demand for protein synthesis, the other stress-induced. Despite the number of papers to investigate the regulation of P5CS at the transcriptional level, the properties of the enzyme have been subjected to limited study, and very little is known about post-translational regulatory mechanisms of the two isoforms. Here we report heterologous expression and purification of P5CS1 and P5CS2 from Arabidopsis thaliana. Their biochemical characterization showed different features that provide the basis for their specific role in plant cell metabolism.

31 - Strigolactones affect the transcriptional and transcriptomic drought stress memory in tomato

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The global climate crisis, characterised by increasing temperatures and changes in precipitation patterns, significantly impacts crop productivity. Plants are frequently exposed to repeated cycles of water stresses; responding differently to them, thus displaying “stress memory”, is an adaptive strategy to cope with fluctuating environmental conditions. For example, the so-called “after-effect” of drought is a feature of stress memory seen at the stomatal level: an incomplete recovery of conductance after drought, even when water potential has fully recovered. Besides the likely dependence on abscisic acid (ABA), the molecular and physiological mechanisms driving the after-effect are not clear yet. This study asked how the phytohormone strigolactones (SL) may contribute to this mechanism in tomato plants, as SLs are described to promote stomatal closure in both ABA-dependent and independent manners and are involved in the drought acclimation processes. We investigated their physiological and molecular contributions to the after-effect by comparing stomatal conductance and transcriptome of wt versus SL-biosynthetic mutants of tomato in repeated dehydration cycles. Interestingly, SL were indispensable for “trainability” of the transcriptome and stomatal response by repeated dehydration cycles. Candidate genes and mechanisms will be discussed.

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33 - The role of tomato B-BOX MicroProteins in reproductive development

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Microproteins (miPs) are single-domain proteins that act as post-translational regulators of protein complexes. Recently, two Arabidopsis miPs, miP1a and miP1b, have been shown to play a central role in the control of flowering time. These miPs belong to the BBX family, which is characterized by the presence of a B-BOX motif responsible for protein-protein interaction. Both miP1a and miP1b actively participate in the flowering process by forming a repressor complex with CONSTANS and TOPLESS. In Arabidopsis, overexpression of miP1a/b causes a delay in flowering linked to a reduction in the expression of the florigen FLOWERING LOCUS T gene. SlBBX16 and SlBBX17 are the homologs of miP1a/b in tomato, and to investigate their functional role we overexpressed them both in tomato (cv MicroTom) and Arabidopsis, focusing on phenotypic analyses during the reproductive development. Overexpression of SlBBX16 and SlBBX17 in Arabidopsis caused a slight delay in the transition from vegetative to flowering stage. In tomato, overexpression of SlBBX16 resulted in a delay in the first phases of fruit development, whereas SlBBX17-overexpressing plants displayed a prolonged flowering period and a reduced number of ripe fruits at harvest, indicating nonredundant roles for these two genes. Because there is no evidence in the literature for the presence in tomato of a flowering inhibitory complex similar to that in Arabidopsis, we are testing the interactions of SlBBX16/17 with key regulators of flowering by yeast-two-hybrid analysis.

34 - Effect of natural compounds with a potential role as priming agents on the mycorrhization efficiency of arbuscular mycorrhizal fungi

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Development of innovative agricultural strategies should enable sustainable and efficient use of natural resources, thereby reducing the use of external inputs in agriculture. The use of natural compounds and soil microbes as seed priming treatments can be an effective strategy to improve resilience and modulate responses to stresses. In the frame of the project Optimus Prime, two experiments were conducted to test whether seed priming agents could disfavour mycorrhization in Solanum lycopersicum (cv Moneymaker) plants: i) seeds from each treatment [chitosan+ melatonin (CHI+MEL); chitosan (CHI); salicylic acid (SA), chitosan + salicylic acid (CHI+SA), hydroprimed (HP) and untreated (NT)] were inoculated with spores of the arbuscular mycorrhizal fungi (AMF; Rhizophagus irregularis DAOM 197198) directly on the root system according to a magenta sandwich system; ii) the effect of the same priming treatments was evaluated directly in contact with AMF (R. irregularis) spores, then inoculating them directly on the tomato root. Roots were checked after two months for AMF colonization. Experiments were successful in demonstrating that the considered compounds should be used as seed priming agents without negative effect on mycorrhization. Thus, the data provided useful information for the developing of a seed priming system involving the coating of natural products in combination with AMF spores to improve plant tolerance and resilience upon abiotic and biotic stress conditions.

42 - A polyamine oxidase controls water-use efficiency in Solanum lycopersicum

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The polyamine thermospermine (T-Spm) is involved in several plant physiological processes. In Arabidopsis, polyamine oxidase 5 (AtPAO5) oxidises T-Spm and plays an important role in T-Spm homeostasis, plant development, xylem differentiation and abiotic stress tolerance. In tomato (Solanum lycopersicum), three AtPAO5 homologs were identified (SlPAO2, SlPAO3 and SlPAO4), and CRISPR/Cas9 mediated slpao3 mutants were obtained. The slpao3 mutants have altered T-Spm levels and exhibit changes in growth parameters, number and size of xylem elements, and expression levels of auxin-related genes compared to wild-type plants. The slpao3 mutants are also characterized by improved drought tolerance, presenting diminished xylem hydraulic conductivity and thus limited water loss, as well as reduced vulnerability to embolism. On the contrary, no significant difference was observed between slpao3 mutants and wild-type plants in leaf and root area, leaf dry matter content, stomatal index, density and size, and leaf water potential at turgor loss point, suggesting that the increased drought tolerance of slpao3 mutants likely does not involve mechanisms related to biomass partitioning and osmotic adjustments. Altogether, this study evidences conservation of the T-Spm-mediated regulatory mechanisms controlling plant growth and differentiation across different plant species and highlights a role of T-Spm in improving stress tolerance, while not constraining growth.

43 - Structural and biochemical characterization of Arabidopsis alcohol dehydrogenases reveals distinct functional properties but similar redox sensitivity

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ABSTRACT

Alcohol dehydrogenases (ADHs) are a group of zinc-binding enzymes belonging to the medium-length dehydrogenase/reductase (MDR) protein superfamily. In plants these enzymes fulfill important functions involving the reduction of toxic aldehydes to the corresponding alcohols as well as catalyzing the reverse reaction (i.e., alcohol oxidation; ADH1) and the reduction of nitrosoglutathione (GSNO; ADH2/GSNOR). In the current study, we investigated and compared the structural and biochemical properties of ADH1 and GSNOR from Arabidopsis thaliana. To this end, we expressed and purified ADH1 and GSNOR and determined two new ADH structures, NADH-ADH1 and apo-GSNOR, thus completing the structural landscape of Arabidopsis ADHs in both apo- and holo-forms. The structural comparison between Arabidopsis ADHs revealed a high sequence conservation (59% identity) and very similar native fold. In contrast, a striking dissimilarity was observed in the catalytic cavity supporting substrate specificity and accommodation. Consistently, ADH1 and GSNOR were strictly specific for their substrates (ethanol and GSNO, respectively), although both enzymes exhibited variable catalytic activity in the presence of long-chain alcohols. Given the high cysteines content (12 and 15 out of 379 residues for ADH1 and GSNOR, respectively), the number and position of accessible cysteine residues along with redox sensitivity were evaluated. Both enzymes showed a prominent and similar responsivity to thiol-oxidizing agents, indicating that redox modifications may constitute a mechanism for controlling enzyme activity under both optimal growth and stress conditions.

45 - Short and long-term effects of prescribed burning on xylem anatomy and hydraulics in Pinus sylvestris L.

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The current increase in large forest fires is an ecological and socio-economic threat, particularly in mountain regions. Prescribed burning (PB) is a widespread fuel treatment technique based on a planned application of fire to manage understory fuels in forests and mitigate the wildfire hazard. However, little it is known about PB's physiological impacts on European tree species. This study analyzed how the different PB intensities affected the functionality of tree hydraulics to establish the optimal prescriptions to reduce wildfire hazards, maximize surface fuel reduction and minimize tree damage. PB was carried out in May 2022 in a highly flammable montane forest stand located in Val Susa, Southwestern Italian Alps. The site is dominated by P. sylvestris and characterized by the presence of different tree diameter classes. The burning treatment, consisting of two levels of fire intensity, was quantified (fuel biomass consumption, rate of spread, fireline intensity) and monitored by thermal sensors placed on trees (at the stem base) to assess the residence time above threshold temperatures. One, six and twelve months after the PB, branches and wood cores were collected in both PB-treated and untreated sites. Our results showed that, although the fire dramatically reduced the amount of lying dead wood and litter (by 64 and 41%), the two levels of PB intensity did not affect considerably the xylem hydraulic properties of the vascular system and its anatomical features, revealing a high resistance of P. sylvestris to surface fire at the stem base. Overall, the present results contribute to understanding the impacts of fire treatment and its severity on tree survival and health status, thus enabling the correct management of PB prescriptions.

53 - Integrated Approaches to Increase Quality and Stress Resilience in Tomato Cultivars

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Drought stress and heavy metal contamination of soil pose challenges to agriculture and human health. Selecting plant genotypes with low heavy metal accumulation capacity in edible parts and more tolerant to water deficit is one of the goals to ensure food security and quality. Tomatoes are one of the most widely consumed vegetables being a source of antioxidants and mineral elements. In collaboration with ISI Sementi S.p.A, we compared four tomato lines (L1-L4), used by the Company in breeding programs, for their capacity to accumulate low levels of nickel (Ni) in shoots/fruits and to tolerate drought. Ni treatments (0.5 µM, 20 µM, and 85 µM) were performed under hydroponic conditions. Physiological and ICP-MS analyses were carried out on roots and shoots. Considering root elongation, L1 appeared less affected by Ni treatments than the other lines. Quantification of Ni in shoots revealed that L4 accumulated a higher level of the element than the other lines. The same analyses on fruits are ongoing using potted plants grown under greenhouse conditions. To test drought tolerance, in vitro germination assays were done in the presence of 4% and 8% PEG. L1 and L2 appeared insensitive to PEG, suggesting a potentially greater capacity to cope with water stress at the germination stage. An independent trial was conducted with potted plants on soil, imposing severe water stress. L1 was found to be the most sensitive, while L4 and L3 showed complete recovery after restoring watering. Comparative RNA-Seq analyses will be conducted to identify differentially expressed genes related to both Ni accumulation and water stress resilience.

55 - WATER AND NUTRITIONAL SAVINGS SHAPE NON-STRUCTURAL CARBOHYDRATES IN GRAPEVINE (Vitis vinifera L.) CUTTINGS.

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Global changes and sustainability challenge researchers in saving water and nutrients. The response of woody crops, which can be forced at facing more drought events during their life, is particularly important. Vitis vinifera can be an important model for its relevance in countries subjected to climate changes and its breeding, requiring cuttings plantation and strong pruning.

Drought leads to an impairment between growth and reserves which can be a key point in the survival of plantings. This work aims at understanding the role of non-structural carbohydrates (NSC) in: i) the maintenance of hydraulic function in cuttings subjected to water deficit and limited nitrogen: ii) identify the best regime of water and nitrogen to achieve a correct compromise between plant growth and NSC.

Cuttings of two different cultivars and three rootstocks of grapevine, were grown in pots under different water and N regimes. During the vegetative season, morphological and physiological traits were measured. In particular, NSC content analysis were performed in roots, rootstocks, shoots and canes on blooming and on cane ripening period.

During early vegetative phases, plants under water deficit shown a reduced growth and lower water potential respect well-watered plants. At the end of the vegetative season, NSC was affected also by the type of cultivar or rootstock. Nitrogen does not seem to have any effect on carbohydrates content.

Our results suggest that imposing a controlled water deficit to grapevine from budburst, can support plants to accumulate NSC, useful to help cutting survival and face incoming drought events.

56 - High density cultivation change responses to drought in one tomato genotype

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Today, one of the main challenges of agriculture is to implement sustainable farming systems. One of these systems could involve the increase of land-use efficiency through high-density (HD) cultivation under limited water availability. In this study one tomato genotype (cv. Money Maker) was cultivated under combined HD and drought. HD changed the light intensity and its quality. The photosynthetic active radiation and the ratio between red and far red were 13 folds and 3.8 folds higher, respectively, under low compared to high density. HD and drought had a different impact on plant growth parameters. Plants growing under HD showed the symptoms of shading, including increased height and leaf area and lower leaf mass per area. Four days after water withholding, HD plants showed a strong reduction of CO_2 assimilation and stomatal conductance compared to non-stressed plants, while in low-density plant this reduction was milder. An increase in H_2O_2, lipid peroxidation and proline accumulation were demonstrated in all the samples under drought. Results here obtained can help to better understand the physiological responses of plants under high density cultivation and drought, highlighting also the traits involved in the adaptation to abiotic stresses.

61 - Impact of chronic cadmium stress on tomato plant hydraulics and fruit quality

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Climate change adversely impacts not only the availability but also the quality of the irrigation water. Higher temperatures, drought and flood events increase the risk of low-quality water use, as they exacerbate many forms of water pollution. Cadmium (Cd) is a heavy metal not required for plant metabolism and toxic to many plant species.The aim of the present study was to investigate the poorly explored long-term effects of Cd-contaminated irrigation water on hydraulic traits of Solanum lycopersicum L. Cd treatment caused significant changes in the water relations, mineral content and wood anatomy of tomato plants. Different values of turgor loss point, osmotic potential at full turgor and bulk modulus of elasticity were recorded in leaf cells and in root cells of Cd-treated vs control (C) plants. Wider xylem vessels were recorded in stem, petiole and fruits peduncle of samples irrigated with Cd-polluted water vs C. This, in turn, may have favored the transport of Cd in leaves and fruits. A higher risk of drought-driven vulnerability to xylem embolism occurred in Cd-treated plants compared to C ones.

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65 - The maintenance of cell turgor in wood parenchyma is critical for xylem hydraulic safety: experimental evidence in <i>Populus nigra</i> (L.)

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Non-structural carbohydrates (NSCs) are involved in several plant functions such as growth, metabolism, transport, osmoregulation and defence, but recent evidence supports their putative role in maintaining plant hydraulic integrity under drought. Indeed, stem NSC-depleted Populus nigra (L.) saplings showed an increased vulnerability to xylem embolism formation, but a mechanistic explanation for this phenomenon is missing.

We hypothesised that low NSC content in wood parenchyma might impede osmoregulation under drought, leading to plasmolysis. Consequently, an earlier turgor loss could lead to formation of air gaps between xylem vessels and adjacent plasmolysed parenchyma cells that would serve as additional gas-entry points for xylem embolism build-up.

To test this hypothesis, plasmolysis was induced in xylem parenchyma cells by applying polyethylene glycol (PEG) solutions at known osmotic potentials on branch segments of adult trees, and inducing embolism with a pressure collar. The results showed that PEG-treated branches had an increased stem xylem vulnerability compared to controls.

These results highlight two key questions deserving further investigation: (1) what is the role of xylem vessel-associated cells and of conduit-parenchyma cell pit characteristics in embolism propagation in the xylem? (2) What is the role of NSC reserves in shaping plant hydraulic behaviour and plant survival under drought?

73 - The interplay between mannitol and sucrose drives hydraulic recovery in Fraxinus ornus saplings exposed to drought and stem shading.

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Plants exposed to drought undergo a re-allocation of non-structural carbohydrates (NSC), and in some cases sugar accumulate in stem and roots. Recently, bark was proposed as the main site for sugar accumulation, suggesting the contribution of stem photosynthesis in this process. In addition, some species accumulate large amounts of poly alcohols (i.e. mannitol). We investigated how the interplay between NSC and mannitol drives the response of Fraxinus ornus saplings to water shortage and recovery coupled to two light regimes: short and long stem shading. Percentage loss of hydraulic conductivity (PLC), starch, soluble NSC pool, glucose, fructose, sucrose and mannitol were measured. Drought induced a decrease of total NSC in bark, wood and roots. Glucose was constant in bark but not in roots highlighting a different engagement during recovery. Sucrose and PLC were directly correlated, indicating the pivotal role of sucrose in plant hydraulic responses under drought. Fructose concentration was extremely low, suggesting a rapid conversion to mannitol, which was indirectly related to starch. Finally, mannitol increased in all organs at the onset of drought and remained high during recovery. Our findings confirm the role of stem photosynthesis during drought and recovery. Mannitol and sucrose emerged as key metabolites, revealing that the role of NSC during abiotic stresses should be reconsidered evaluating single saccharides and other osmotic active compounds derived from NSC.

79 - Effects of $Cs_2AgBiBr_6$ Perovskite panels covering in-door greenhouses on plant growth and metabolite content in Artemisia annua L.

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By covering greenhouses with semi-transparent solar panels, sunlight can be exploited to generate electricity, allowing at the same time the healthy growth of plants. Perovskite-based halide solar cells appear to be the ideal candidate to achieve this goal, being also an environmentally friendly solution. Here, we report preliminary data concerning the influence of the lead-free $Cs_2AgBiBr_6$ perovskite, used as active layer for solar cells panels, covering the in-door green-house, on plant growth and metabolite contents of Artemisia annua L. The experiments were carried out “in-door” in a growth chamber with controlled conditions of light and temperature, using mini green-houses covered by transparent glass panels placed on the control green-houses (P-), and semi-transparent panels, constituted by glass covered by indium tin oxide (substrate) and $Cs_2AgBiBr_6$ perovskite layers (P+). A. annua plants were grown in pots placed inside P- and P+ mini green-houses. From well grown plants (about 90 days), the first leaves were picked up for chlorophyll analysis, while the remaining leaves and stems used for evaluating artemisinin contents. The average of total chlorophylls (a+b) in (P+) plants was more than 2-fold higher than control (P-) plants (1.68 vs 0.73 mg/g FW). Differently, preliminary results of artemisinin contents indicate that the Perovskite “screen” negatively affected metabolite accumulation.

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89 – Two is better than one: ultraviolet B radiations improve the salt-induced responses in the facultative halophyte Chenopodium quinoa

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In saline environments, in addition to salt stress, plants generally experience high levels of ultraviolet (UV) radiations. While much is known about responses to salinity and UV as individual stressors, there is a surprising deficiency in knowledge of how these two factors interact and their combined effects on plants. Here, we evaluated the responses of Chenopodium quinoa to combined salt (200 mM NaCl) and UV-B stress. Our data indicated that acclimation to salt involved an energy-saving strategy associated with limited Na translocation to shoots and reduced production of secondary metabolites. Compared to control plants, salt significantly reduced plant growth, leaf gas exchanges and altered K and water relations, while the UV-B treatment alone induced a shift in secondary metabolites towards the production of hydroxycinnamic acids, which absorb in the UV spectral region, with no relevant changes for all other measured parameters. In the combined UV-B and salt treatment, however, there was an altered K compartmentalization between the leaf mesophyll and the epidermal bladder cells. As a result, while no differences were seen for the biometric parameters, plants exposed to both UV-B and NaCl had improved water relations and leaf fluorescence parameters compared to those exposed only to salinity. Overall, the results demonstrated that UV-B alters ion and water relations in salt-treated plants, mitigating salt-induced damages in this facultative halophyte.

91 – Plant microbiomes in sustainable viticulture

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The intensification of agriculture has mostly been sustained by the increasing use of water resources at the expense of the environmental water balance. Furthermore, in recent decades climate change and the consequent expansion of drought have further compromised water availability, making current agricultural systems even more fragile both ecologically and economically. Therefore, there is an urgent need to improve the resilience of agrosystems by promoting more rational use of biological and natural resources. Microbiomes have emerged as important influencers of the physiology of many multicellular organisms. As part of the PRIMA programme, the present work aims to investigate the natural biodiversity of the endophytic microbiota of grapevine plants located in arid Mediterranean areas to define microbe consortia to be used in sustainable viticultural practices under adverse environmental conditions. More closely, the research aims to understand to which extent different Vitis vinifera varieties adaptation to water deficit is influenced by the microbiota, and whether such a beneficial association is transmissible to other varieties.

94 – Transgenerational stress memories in plants: forget or remember?

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Being sessile, plants endure and adapt to stresses using an armament of constitutive and stress-induced mechanisms. While many of these mechanisms are well-known, whether plants also possess memory of stress and whether stress resistance, once acquired is maintained at transgenerational level is more controversial. How plants are able to efficiently trade-off between the costs and benefits of remembering a past experience? How these memories are transmitted? Are there specific molecules, cellular components or key tissues that are specialized to memorize, store and use information? All these questions, and many more, remain largely unanswered. We present results from two studies dealing with transgenerational phenotypic and molecular responses to abiotic stresses. In the first study, Arabidopsis F1 plants from parents subjected to both middle and long chronic Cr stress showed increased Cr tolerance at the level of seed germination. RNA-seq analysis showed that F1 of plants exposed to Cr stress optimize their transcriptome activating mostly genes involved in Cr-Fe crosstalk such as several bHlH TF members. In the second study, two chickpea genotypes with contrasting tolerance to drought stress were identified and RNA-seq analysis. F1 plants are now grown in controlled environment under drought conditions, and phenotypic, transcriptomic and epigenomic (whole genome bisulfite sequencing) analyses are now carried out to investigate whether transgenerational effects occurs.

96 – Investigating holm oak forest dieback through a multiscale approach: physiological measurements and visual assessment data with remote sensing

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Keywords: drought, holm oak dieback, photosynthesis, remote sensing, tree-ring δ13C

Drought and heat waves profoundly affect the functioning of Mediterranean forests. As Quercus ilex dieback has been observed in many Mediterranean stands, it is crucial to develop effective tools for studying this phenomenon across multiple scales. Our study was conducted over three years (2019-2021) in declining (D) and non-declining (ND) Q. ilex stands in southern Tuscany, assessing physiological traits such as gas exchange and water relations. Dendrochronological and tree-ring δ13C analyses were combined to investigate the effects of previous droughts on tree growth and WUE. Finally, the relationships between physiological indicators and satellite-derived Normalized Difference Vegetation and Water Indices (NDVI and NDWI) were analyzed. Seasonality had a strong effect on physiological traits, with the main stress occurring during the summer of 2020, as evidenced by the lowest gas exchange values. Furthermore, plants in the ND stand exhibited significantly higher photosynthetic performances than those in the D stand across multiple seasons. The δ13C value at ND significantly exceeded that observed at site D over the last 20 years, revealing more conservative water use. While the high spatial resolution of Sentinel 2 helped identify the overall Q. ilex decline, these indices were inadequate for analyzing forest stands with patchy dieback. Our study emphasizes the significance of a multiscale approach that combines in situ physiological measures and satellite data to monitor Q. ilex decline.

98 – Impact of drought-induced holm oak dieback on understory species composition and terpene emissions in a Mediterranean maquis

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The occurrence of drought-induced holm oak dieback may alter the maquis species composition and volatile organic compounds (VOCs) emissions. Therefore, seasonal changes in holm oak canopy cover, herbaceous and shrub species richness and alpha biodiversity were investigated in two holm oak stands characterized by different degrees of crown dieback over four years (2019-2022) in Tuscany, while VOCs were monitored from 2019-2021. Holm oak canopy cover decreased by 50% in both stands, leading to increased radiation at the ground and a subsequent species richness increase in 2021. However, in 2022, the species richness was lower than the 2019 values in both stands, possibly because of the large amounts of dead wood on the ground. Although no changes in alpha biodiversity indices were observed, in 2021 a reduction in monoterpene emission was observed, thus reflecting the decline in holm oak canopy cover rather than the species richness modification. In conclusion, although the holm oak dieback did not have a significant impact on alpha biodiversity, it resulted in noticeable changes in the species composition of understory vegetation and terpene emissions. Our findings suggest that terpene emission in the Mediterranean maquis is more affected by holm oak dieback than species richness modification.

113 – Cyclic AMP involvement in the acquisition of basal thermotolerance in Arabidopsis thaliana

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Heat stress (HS), affecting growth and productivity, is one of the most harmful abiotic stresses for plants. To avoid damage and ensure the protection of cellular homeostasis from the effects of high temperatures, plants activate complex signaling networks leading to Heat Stress Response (HSR). Among signalling molecules, 3',5'-Cyclic Adenosine Monophosphate (cAMP) has been proved to have a key role in triggering plant HSR. It has been proposed that cAMP dampening increases HS susceptibility in tobacco BY-2 cells, as well as exogenous cAMP promotes the expression of heat shock proteins in Arabidopsis plants.

In this study, Arabidopsis plants overexpressing the 'cAMP-sponge', a genetic tool that reduces intracellular cAMP levels, were used to investigate the role of cAMP in HSR. For this aim, fifteen days-old wild type (wt) and transgenic (cAS) plants were exposed for 2 hours at 45°C and successively recovered for 3 hours at growth temperature (22°C). Phenotypic response, photosynthetic pigments, reactive oxygen species and antioxidants were analyzed to clarify the relationship between cAMP and redox homeostasis.

The compromised plant growth in the recovery phase indicates that cAMP lowering negatively affects basal thermotolerance.

115 – Durum wheat response under different phosphate fertilization regimes

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Durum wheat (Triticum durum Desf.) is one of the most important crops in the Mediterranean basin. Its cultivation presents a dominant position in Italian agriculture due to its adaptability and large consumption. A number of projects worldwide have been focusing on understanding the uptake, assimilation and utilization of phosphorus to improve the efficiency of phosphorus recovery in the grain. Whilst the physical processes of phosphorus remobilization have been studied in detail, the genetic control of these processes and their contribution to agronomic productivity are less well understood.

Following these guidelines, a multidisciplinary approach was applied to detect durum wheat response to different phosphorus fertilization regimes (Control, no fertilization; P120, phosphate fertilization; NPK, nitrogen, phosphate and potassium fertilization). Biometric data of two wheat cultivars (Ciccio, Svevo) was combined with phosphorus accumulation and metabolization data to get a proper picture of different genotypes and treatments. Furthermore, a proteomic approach based on two-dimensional electrophoresis coupled with mass spectrometry was applied to leaf samples collected in the stem elongation phase to deepen the physiological mechanism involved in plant response. Proteomic data were thus confirmed by qPCR analysis linked to the identified proteins and specific phosphorus transporters.

The results indicate consistent differences both between fertilization regimes and genotypes.

121 – REGULATORY MECHANISMS OF SULFUR METABOLISM INVOLVED IN SALT STRESS RESPONSE IN RICE

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Crop yield loss due to soil salinization is an increasing threat to agriculture worldwide. Salt stress drastically affects the growth, development, and grain productivity of rice (Oryza sativa), and the improvement of rice tolerance to salt stress is a desirable approach for meeting increasing food demand. A differential modulation of GSH metabolism was found in rice varieties characterized by a contrasting sensitivity to salt stress. GSH is considered the major organic sulfur compound in plants and its biosynthesis requires the activation of three key enzymes, ATPS and APR directly involved in the biosynthesis of cysteine and Y-ECS involved in the conversion of γ-glutamylcysteine into glutathione. Coherently, an impairment in S translocation and organication was observed in sensitive varieties of rice. The different gene expression of S metabolism regulatory enzymes was investigated at transcriptional and post transcriptional level throw DNA methylation profile and miRNA395 analysis respectively.

Modulation of DNA methylation pattern occurred after exposure to salt stress and characterized tolerant and sensitive rice varieties. The expression of miRNA395 and its targets genes under stress condition confirms the different response of sensitive and tolerant rice varieties to salt stress. Therefore, the regulation of sulfur and glutathione metabolism could be part of the redox network mechanisms in conferring salt tolerance in rice.

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125 – Plant cytosolic glucose-6P dehydrogenase isoforms display differential regulation under low temperatures and in relation to development: the role of alternative splicing

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The cytosolic isoform of glucose-6P dehydrogenase (Cy-G6PDH) contributes to 75% of the total activity in plants, playing specific roles in abiotic stress. In Arabidopsis, Cy-G6PDH is phosphorylated by SHAGGY-related kinase 11 (AtSK11). Due to AtSK11 activation upon low temperatures, it could be the possible trigger improving Cy-G6PDH under cold.

Arabidopsis plants defective in SK11 (KO-SK11) were compared with wild type (Col-1) grown under control (20°C) and cold stress (4°C). The lack of SK11 in mutants induced a cold-susceptible phenotype.

In KO-SK11 14 days seedlings (in-vitro), cy-G6PDH activity showed no difference compared to WT at 20°C and 4°C. Conversely, after 3 weeks in pot (full-expanded rosette) KO-SK11 showed a significant reduction in cy-G6PDH rate under both conditions.

Changes during development were investigated on the transcripts of the cy-G6PDH encoding genes (G6PD5; G6PD6) and their alternative spliced transcripts (G6PD5v1-5; G6PD6v1-2).

At 20°C, G6PD5 variants (G6PD5v3-4-5) were substantially absent in seedlings, while highly expressed in full-expanded rosette plants. G6PD5v1-2 were scarcely expressed in during developmental stages. Both G6PD6 variants did not change their expression during development at 20°C. Interestingly, G6PD5v4 and G6PD6v2 showed became prominent in response to cold in full-expanded rosette plants. We suggest that cy-G6PDH role(s) under cold-stress may be influenced by the alternative splicing processes, depending on development.

87 - Bacterial endophytes contribute to rice seedling establishment under submergence

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Rice can germinate and grow successfully under oxygen shortage. Previous studies showed that rice plants host a wide range of endophytic bacteria, capable of producing plant growth promoters which support plant development and survival under abiotic stress conditions. Currently, the role of endophytic bacteria under hypoxia is still poorly defined. To characterize the community composition of the microbiome in rice germination under submergence, a 16S gene profiling metagenomic analysis was performed of temperate japonica rice varieties, showing contrasting phenotypes in terms of coleoptile length when submerged. This analysis showed a distinct microbiota composition on the tolerant variety (long coleoptile) under submergence. Furthermore, culturable bacteria were isolated, identified and tested for plant growth-promoting activities. Selected bacteria were inoculated in seeds to evaluate their effect on rice under submergence, showing a response that is dependent of the rice genotype. Our findings suggest that endophytic bacteria can substantially contribute to rice seedling establishment under submergence and that diverse rice genotypes may benefit differently from bacteria inoculant.

Key words: coleoptile, endophytes, microbiota, Oryza sativa, submergence.

68 - Elicitor-Induced Transgenerational Priming of Defense Responses in Arabidopsis thaliana

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Plant diseases cause crop losses and compromise food safety due to fungal contamination and toxins. Crop protection is currently based on genetic resistances, which are not always available and can be overcome by pathogens, or on pesticides that can pose environmental and health issues. These problems might be overcome by novel strategies that aim at promoting the natural plant defense mechanisms. The plant immune system is based on the recognition of elicitors, molecules present in pathogenic microorganisms (Pathogen-Associated Molecular Patterns, PAMPs) or released by the host in response to attack (Damage-Associated Molecular Patterns, DAMPs). Foliar treatments with elicitors often lead to an increased resistance against microbial pathogens. For instance, Arabidopsis thaliana plants sprayed with flg22, a peptide derived from bacterial flagellin, show increased resistance to the fungal pathogen Botrytis cinerea if treated 24 h before inoculation. We show here that elicitor-induced resistance (EIR) lasts for at least one week and can be transmitted to the progeny of the treated plants. The involvement of known defense-related pathways in transgenerational EIR will be discussed. Notably, EIR does not affect plant growth and seed production, suggesting that transgenerational priming is a viable and cost-effective solution for crop protection.

6 - Post-translational modification of SPATULA by SECRENT AGENT and SPINDLY promotes organ symmetry transition at the gynoecium apex.

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Decoration of eukaryotic proteins by the monosaccharide posttranslational modification, O-linked N-acetylglucosamine (O-GlcNAc), is essential for cell survival in both plant and animal kingdoms, yet its precise functions are largely unknown.

We have investigated how two O-glycosyltransferase enzymes of Arabidopsis thaliana, SPINDLY (SPY) and SECRET AGENT (SEC) synergistically promote a rare bilateral-to-radial symmetry transition during patterning of the plant reproductive organ, the gynoecium.

SPY and SEC modify residues at the N-terminus of the bHLH transcription factor SPATULA (SPT) in vivo and in vitro by attaching O-Fucose and O-GlcNAc respectively, to promote style development. This post-translational regulation does not impact SPT homo- and hetero-dimerisation events with its partners INDEHISCENT (IND) and HECATE 1 (HEC1), although it enhances the affinity of SPT for the kinase PINOID (PID) gene locus to promote transcriptional repression. Altogether, our findings reveal a previously unrecognized mechanism for O-GlcNAc and O-Fucose post-translational decorations in controlling style development and offer the first molecular example of a synergistic role for SEC and SPY in plant post-embryonic organ patterning. Moreover, our study provides the best characterised molecular example of the biological function for these key signalling molecules in the control of a transcription factor, which is a key regulator of plant reproduction.

9 - Evidence of miRNA regulation on the DNA damage response during germination of the irradiated wheat seeds

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DNA damage response (DDR) is a complex network of cellular pathways that cooperate to sense and repair any lesions in DNA. It is one of the most important processes and is regulated by several mechanisms, one of which is MicroRNAs (miRNAs). MicroRNAs are small 18-24 nucleotide long single-stranded RNA molecules that regulate their target genes at the post-transcriptional level by cleavage or translation inhibition of the mRNA. Knowledge of miRNA regulation of the genes involved in the DDR is still scanty, hence novel experimental designs are required to enhance the understanding of their influence on the complex regulation of DDR. In this work, an initial bioinformatic analysis was performed to identify putative miRNAs that target the DDR sensors, signal transducers and effector genes in wheat. The selected miRNA-gene pairs were tested in an experimental system consisting of wheat seeds irradiated with 50 Gy and 300 Gy doses of gamma radiation. To evaluate the effect of the treatments on wheat germination phenotypic (germination behavior, seedling growth) and molecular (DNA damage, gene/miRNA expression profile) analysis has been carried out. The results showed that while the 50Gy dose seems to have a priming effect, the 300 Gy dose negatively impacted the seedling growth, also supported by enhanced accumulation of DNA damage. Importantly, the miRNA-gene expression analysis indicates a negative correlation between expression levels of the miRNA and their target genes.

2 - Spatiotemporal internal oxygen dynamics define cyclic hypoxia in plants

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ABSTRACT

Oxygen is essential for plant growth and development. An oxygen sensing mechanism deploys a class of transcription factors called ETHYLENE RESPONSE FACTORS type VII (ERFVIIs) that activate the transcription of hypoxia responsive genes (HRGs) to mitigate the decline in internal oxygen by promoting anaerobic metabolism. Hypoxia occurs due to limited oxygen availability following adverse environmental conditions, but the presence of hypoxic niches in plants was recently demonstrated. However, the existence and functional integration of spatiotemporal internal oxygen dynamics with plant development remains unknown. In this study, we observed a decrease in internal oxygen levels in young emerging leaves of Arabidopsis during the night. An intermittent fluctuation in oxygen availability is known as cyclic hypoxia in animal systems. Here we demonstrate that cyclic hypoxia exists in plants and generates an ERFVII-dependent mechanism that controls the rate of starch consumption and leaf growth based on internal oxygen availability.

52 - Effect of the trophic regime on the growth of microalgae

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Microalgae form a heterogeneous group of single cell organisms capable of colonizing different ecosystems. To date, only 30.000 different species are known out of an estimated number of about 800.000. This extraordinary biodiversity makes microalgae an excellent bioresource for a wide array of applications.

Microalgae are metabolically flexible. Their trophic habits can vary greatly, so that many microalgae species can adopt a photoautotrophic or heterotrophic metabolism but also combine both through a mixotrophic growth. Although the heterotrophic cultivation might increase the cost of an industrial plant, the ability to remove unwanted organic molecules ensures an added value to the cultivation. For this reason, several attempts to grow microalgae in wastewater of various anthropic origins have been investigated.

In this context, we have studied the growth of six different microalgae, including the filamentous cyanobacterium Arthrospira platensis. The microalgae were grown in both autotrophic and mixotrophic conditions. For some strains, mixotrophic conditions have been artificially imposed through the addition of organic molecules to the mineral medium. This artificial mixotrophy system allows to compare the biomass production and carbon partioning between the two trophic regimes (auto v.s. mixo). This approach prevents interferences deriving from the use of waste products, like dairy waste, that were used for other species. Experiments with the two approaches are ongoing.

77 - NUTRITIONAL AND NUTRACEUTICAL COMPOSITION OF ITALIAN WILD PEAR (PYRUS COMMUNIS VAR. ZINGARO) AT DIFFERENT MATURATION STAGES: A COMPARATIVE ANALYSIS OF PEEL AND PULP

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Keywords: ancient fruits, DPPH, FRAP, HPLC-DAD, NMR analysis, phenolic compounds

Ancient wild fruits are gaining attention due to their high nutritional value and low agronomic input. This study aimed to compare the nutritional and nutraceutical compositions of peels and pulps of an ancient Italian pear (P. communis var. Zingaro) at different maturation stages (S1-S6). The browning index, firmness, nutritional content (by NMR), phenolic composition (by HPLC-DAD), and antioxidant properties (DPPH and FRAP) were evaluated.

During maturation, peels changed from yellow (S1) to orange (S3) and finally brown (S6), while the pulp remained yellow until S4 and darkened at S5. Browning was attributed to phenolic oxidation and lower malic acid content. NMR revealed the presence of various compounds in the pulp: 10 organic acids, 11 amino acids, 5 sugars, and phytochemicals, such as vitamin B8. These compounds decreased after S4, reaching their lowest levels at S6. Total phenolic content (TPC) was higher in peels than in pulps, decreasing in both fruit parts throughout ripening. For both, the highest TPC was at the beginning of ripening: in S1-S2 for pulps and in S1-S3 for peels. Antioxidant assays confirmed a greater antioxidant capacity in S1-S2 (pulps) and S1-S3 (peels).

In conclusion, due to the highest TPC and antioxidant properties, the first three maturation stages are suggested as suitable for nutraceutical purposes. The ancient pear "Zingaro" showed to be promise as a source of nutrients and nutraceuticals with potential applications in food sectors.

67 - Better safe than sorry: the surprising drought tolerance of a wetland sedge (Cyperus alternifolius)

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Increased frequency and severity of drought events puts at risk crop productivity and survival. High leaf hydraulic efficiency assures high productivity, but typically comes at the cost of reduced hydraulic safety. Few studies have investigated leaf hydraulics of monocots, especially those inhabiting wetlands which can experience frequent water availability fluctuations. We studied leaf hydraulics in Cyperus alternifolius (Ca), an emergent aquatic plant. We focused on key indicators of leaf hydraulic efficiency (leaf hydraulic conductance, KL) and drought tolerance (water potential at turgor loss point, ΨTLP; vulnerability to xylem embolism, P50). Ca has high KL values (16.9±1.3 mmol.m-1.s-1.MPa-1), similar to some of the most productive crops, and apparently operates at water potential (ΨL) close to ΨTLP (-1.8±0.1 MPa) but far away from ΨL inducing embolism events in major veins (-3.5±0.1 MPa). P50 (-4.0 MPa) is similar to values observed in some xerophytes and substantially more negative than midday ΨL. Our results indicate that Ca has high hydraulic efficiency coupled to a remarkable resistance to xylem embolism, making Ca a good model to identify anatomical, physiological and molecular traits allowing to reconcile high hydraulic efficiency with high hydraulic safety, unlike most other plants where a trade-off between these opposite functions has been demonstrated. We speculate that Ca represents an ideotype for the development of productive and climate resilient crops.

1 - Digging up through Artificial intelligence how two wheat cultivars response to Polyethylene Glycol-Simulated Drought Stress by analyzing root morpho-anatomical traits

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Drought stress is one of the major environmental stresses limiting plant growth and crop productivity, including grain crops. We aim to accomplished a comparative study among Saragolla, an old wheat cultivar and Svevo modern one, under PEG—6000 treatment. The root system is the first organ which can perceive water deficiency and adapt their architecture accordingly. Therefore, roots were selected as materials to explore the anatomical mechanisms changes in the main growth using SEM (scanning electron microscope) images and Artificial Intelligence. The great root properties under drought stress found in Saragolla old wheat were characterized by a larger cross-sectional area root, a greater stele area and an increase of the xylem lumen size vessels. Artificial Intelligence algorithms underscored a clear-cut differentiation between the two cultivars root properties, emphasizing the fact that Saragolla wheat tended to exhibit a better plasticity under osmotic stress in order to maintain the water potential, and to maximize the root-to shoot ratio. Smart Root software was used to evaluate the changes in the morphology of primary and lateral root, confirming that the Saragolla old cultivar maintained a stable root ultrastructure. Overall, we concluded that the Saragolla wheat had a better drought resistance mechanism, exhibiting higher water retention capability and root to shoot ratio, possessing a higher phenotype plasticity that as a trait may have been reduced during selection of modern elite varieties cultivated under more stable condition.

14 - Physiological responses to salt stress in wild and domesticated rice

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Soil salinization is a major abiotic constraint affecting global food production. Domesticated rice (Oryza sativa L.) originated from the wild ancestor Oryza rufipogon Griff. and today is the most consumed food crop. Among the cereals, rice is the most sensitive to salinity stress. Its ancestor seems to possess superior salinity tolerance.

Here, we examined the physiological responses to salinity stress (7 days exposure to 80 mM NaCl) at the early seedling stage in O. rufipogon compared to a salt-susceptible (Vialone nano) and a salt-tolerant (Baldo) variety of O. sativa largely cultivated in Italy.

Evaluating rice for salinity tolerance based on phenotypic traits indicated O. rufipogon as the most tolerant, with 80% of plants survival with none to moderate injury symptoms, followed by Baldo (65%) and Vialone nano (55%). The survived plants of each genotype were assessed for biomass production, morphological traits, Na, K, C and N tissue content (roots and leaves), photosynthetic apparatus integrity and efficiency (JIP test), pigment content, leaf anatomy and stomatal characteristics revealing different strategies for salt tolerance. O. rufipogon, despite the highest reduction in biomass and shoot-root elongation, showed a superior ability of maintaining a lower Na^+/K^+ ratio in both roots and leaves. Baldo showed the highest leaf stomatal density and was the less affected from a photosynthetic point of view. Vialone nano displayed the most severe stress effects at leaf level.

40 - Exploiting somaclonal variability to increase drought stress tolerance in grapevine

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Vitis vinifera is one of the most sensitive species to climate-change induced stresses, such as drought. Whether on one side it is crucial to understand how grape cultivars physiologically perceive and react to adverse climate conditions, on the other side it is necessary to improve existing breeding platforms using sustainable techniques allowing the selection of resilient genotypes. Somatic embryogenesis (SE) (i.e. the initiation of embryos from somatic tissues) represents a powerful green biotechnological tool for genetic improvement purposes. SE can spontaneously generate new genetic variability, called somaclonal variation, which results from genetic mutations, changes in epigenetic marks altering transposon activity and/or gene expression, or phenotypic alterations.

This study was tailored to demonstrate whether vines in vitro regenerated through SE, namely somaclones, can endure water stress conditions better than the mother plant. A severe water deficit treatment was thereby imposed on a population of different somaclone lines of Vitis vinifera ‘Nebbiolo’ and on the corresponding mother plant line. During the trial, the main ecophysiological parameters (e.g. gas exchanges, relative water content, stem water potential) were monitored. Changes in biometric and anatomical traits were also inspected. Collected data allowed to identify those lines with improved tolerance to water stress. To deepen the mechanisms underlying the observed tolerance, biochemical and molecular analyses are ongoing to profile changes in the content of defense secondary metabolites and hormones and in the transcription of stress-responsive genes. In parallel, the genomes of the best and worst performing lines have also been sequenced.

22 - Biodiversity of photosynthetic response in various bryophyte accessions

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Non-photochemical quenching (NPQ) helps plants to prevent overreduction of the photosynthetic apparatus and consequent photodamage. It is known that NPQ depends on the activity of PSBS and LHCSR in the moss Physcomitrium patens, but it is largely unknown how NPQ is regulated in several bryophytes. We therefore characterized the variability of NPQ in different bryophytes to unravel their ability of acclimation/adaptation to high light intensities. Each species was grown in two conditions: in mild light (50 µmol photons m-2 s-1) for 6 weeks, then moved for 7 days in saturating light condition (350 µmol photons m-2 s-1), and only in mild light condition. NPQ activity was measured with a Dual-PAM by exposing the gametophores to 8 minutes of saturating actinic light (540 µmol photons m-2 s-1) and then NPQ recovery was followed for 10 minutes in the dark. In control condition the species showed very different NPQ behaviours, indicating different adaptive response. The exposure to saturating light acclimation induced a substantial change of NPQ induction profile in most of the accessions. Specifically, NPQ kinetics of saturating light acclimated gametophore arose rapidly in the first second but decreased in the first minute, generating a pick at the onset of light exposure, probably linked to the activation of the Calvin cycle. Data suggested that bryophytes can remodel the photosynthetic apparatus in order to cope with higher light radiance.

83 - Efficient utilization of monosaccharides from agri-food by-products supports Chlorella vulgaris biomass production.

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Microalgae are a sustainable source for biofuel production, feed, and high-value chemicals. Several strains can grow hetero- or mixotrophically using various organic substrates. However, costly organic carbon sources like acetate and glucose hinder scalability and economic sustainability. The utilization of sugars derived from agri-food waste biomass as organic carbon sources could enhance the sustainability of mixotrophic microalgae cultivation while simultaneously valorizing underutilized by-products. This study aimed at investigating biomass production and sugar utilization of C. vulgaris during cultivation using hydrolysates from two different types of agri-food waste: barley straw (BS) and citrus processing waste (CPW). CPW hydrolysate supported higher biomass production, compared to BS digestates, likely due to the presence of significant amounts of galactose alongside glucose. Notably, these two monosaccharides are rapidly consumed when C. vulgaris was grown in the light in the presence of CPW hydrolysate. In contrast, algal growth stopped before complete sugar utilization when pure monosaccharides were provided as the sole source of organic carbon. The efficient use of monosaccharides in the digestates is possibly dependent on the availability of different nitrogen sources, that affect extracellular pH during algal growth. Overall, our findings suggest that additional factors, besides the abundance of sugars that can be metabolized by the alga, increases C. vulgaris production in the presence of agri-food waste digestates.

NOVEL TECHNOLOGY FOR MULTISCALE LEVEL OF INVESTIGATION IN PLANT PHYSIOLOGY

86 - New state-of-the-art imaging tools to study how crops adapt to environmental changes: Lycopersicum esculentum key study

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Being sessile organisms, plants are subjected to environmental challenges during their entire life. Both abiotic and biotic stresses, such as water deficiency, salt stresses and pathogen attack, could significantly influence plant growth as well as crop yield.

Therefore, understanding plant response to external challenges is of vital importance.

In the last decades, molecular signalling from the stimulus perception to plant response was largely investigated. However, most of the available knowledges on plant signalling derive from studies performed on Arabidopsis thaliana model plant.

To investigate how crops deal with environmental stimuli, we decided to focus our attention on Lycopersicum esculentum. Tomato represents a promising model plant since it is a worldwide economically important crop and its growth as well as its productivity are sensitive to inappropriate external conditions.

To better understand crop signalling we aimed at combining the development of a battery of tomato lines expressing genetically encoded biosensors with tailored in-vivo imaging approaches.

So far, we set up an efficient transformation protocol by employing an ad hoc vector. We succeeded in obtaining MicroTom lines expressing Ca^{2+} and pH biosensors which will allow us to study their dynamics in response to external stimuli or during plant development. Preliminary experiments carried out with the two sensor lines will be presented.

109 - Methodological pipeline for monitoring post-harvest quality of leafy vegetables

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Plants are primary source of nutrients for humans. However, the nutritional value of vegetables tends to decrease once organ and tissue sinks are detached from the plant. Minimal processing of leafy vegetables involves cutting and washing before packaging and storage. These processing procedures result in stressful conditions and post-harvest disorders senescence-related can also occur. The aim of this work is to define a methodological pipeline to evaluate the “quality” changes of fresh cut leafy vegetables over their shelf life. At this purpose, intra-species variability has been investigated taking into account two cultivars of Lactuca sativa (var. longifolia and capitata), showing different susceptibility to browning. Since browning mainly depends on phenol oxidation, redox parameters as well as the activity of the enzymes involved in phenol biosynthesis and oxidation have been monitored over storage time. At the same time, the metabolic changes of the lettuce leaves have been estimated as response patterns to electrochemical sensors. Electrochemical patterns were predictive of browning-related biological features in a cultivar-dependent manner. The integration of the results obtained by this multivariate methodological approach allowed the identification of the most appropriate quality markers in lettuce leaves from different varieties. This methodological pipeline is proposed for the identification and subsequent monitoring of the post-harvest quality of leafy vegetables.

Authors index

A

B

C

L

M

N

Sommario