

BOOK OF ABSTRACTS

2nd International Conference on Functional Biology

International House of Scientists “Frédéric Joliot-Curie”
16 – 18 September 2026
Varna, Bulgaria

SECOND INTERNATIONAL CONFERENCE ON FUNCTIONAL BIOLOGY

16 – 18 September 2026

International House of Scientists “Frédéric Joliot-Curie”, St. Constantine and Helena Resort, Varna, Bulgaria

FOREWORD

It is our great pleasure to welcome you to the **Second International Conference on Functional Biology (FunBio2)**, organised by the Institute of Plant Physiology and Genetics at the Bulgarian Academy of Sciences in collaboration with the Section “Plant Physiology and Biochemistry” of the Union of Scientists in Bulgaria.

Following the first edition of the conference, FunBio2 continues our ambition to create a stimulating forum where researchers from different fields, institutions and countries can meet, exchange ideas and establish new scientific connections. Functional biology is developing rapidly, increasingly bringing together knowledge from different levels of biological organisation - from genes and molecular networks to whole organisms, their interactions with the environment and their applications in agriculture and biotechnology.

The scientific programme reflects this breadth, covering **Plant Physiology and Biochemistry, Genetics and Epigenetics, Cell and Molecular Biology, Biotechnologies for Sustainable Development, Biodiversity and Genetic Resources, and Agrobiology and Food Science**. Through plenary and oral lectures, poster presentations and discussions, we hope to encourage exchange across these fields and to explore how fundamental biological knowledge can contribute to addressing current challenges related to environmental change, sustainable agriculture, biodiversity and food production.

A particular aim of FunBio2 is to bring together established scientists and the next generation of researchers. We hope that the conference will provide young scientists with opportunities to present their work, exchange ideas with experienced colleagues and develop collaborations that extend well beyond these three days in Varna.

We sincerely thank all invited speakers, participants, members of the Scientific and Organising Committees, and our supporting institutions and sponsors for making FunBio2 possible.

We wish you an inspiring and enjoyable conference, fruitful scientific discussions and a memorable time together on the Bulgarian Black Sea coast. We hope these three days will bring not only stimulating science, but also new ideas, new connections and lasting collaborations.

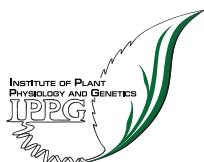
Welcome to Varna, and welcome to FunBio2!

ORGANIZING COMMITTEE

Prof. Dr. Valya Vassileva – Conference Chairperson – Institute of Plant Physiology and Genetics, BAS
Prof. Dr. Katya Georgieva – Institute of Plant Physiology and Genetics, BAS
Prof. Dr. Iskren Sergiev – Institute of Plant Physiology and Genetics, BAS
Assoc. Prof. Dr. Dessislava Todorova – Institute of Plant Physiology and Genetics, BAS
Assist. Prof. Dr. Ivan Iliev – Institute of Plant Physiology and Genetics, BAS
Assist. Prof. Dr. Elena Shopova – Institute of Plant Physiology and Genetics, BAS
Assist. Prof. Dr. Mariyana Georgieva – Institute of Plant Physiology and Genetics, BAS
Assist. Prof. Dr. Zlatina Gospodinova – Institute of Plant Physiology and Genetics, BAS
Assist. Prof. Tsvetina Nikolova – Institute of Plant Physiology and Genetics, BAS
Mrs. Neli Naydenova – Institute of Plant Physiology and Genetics, BAS

SCIENTIFIC COMMITTEE

Prof. Dr. Svetlana Misheva – Institute of Plant Physiology and Genetics, BAS
Assoc. Prof. Dr. Lyudmila Simova-Stoilova – Institute of Plant Physiology and Genetics, BAS
Assoc. Prof. Dr. Maria Geneva – Institute of Plant Physiology and Genetics, BAS
Assoc. Prof. Dr. Irina Vaseva – Institute of Plant Physiology and Genetics, BAS
Assoc. Prof. Dr. Georgi Bonchev – Institute of Plant Physiology and Genetics, BAS
Assoc. Prof. Dr. Ina Aneva – Institute of Biodiversity and Ecosystem Research, BAS
Assoc. Prof. Dr. Miroslava Zhiponova – Sofia University “St. Kl. Ohridski”
Prof. Dr. Pilar S. Testillano – Margarita Salas Center for Biological Research (CIB-CSIC), Spain
Prof. Dr. Sigita Jurkonienė – Nature Research Centre, Vilnius, Lithuania
Prof. Dr. Yalçın Kaya – Trakya University, Edirne, Türkiye
Assoc. Prof. Dr. Federico Martinelli – University of Florence, Italy



CONFERENCE TOPICS

PLANT PHYSIOLOGY AND BIOCHEMISTRY

Physiological and biochemical mechanisms in plants under normal and stress conditions

GENETICS AND EPIGENETICS

From gene regulation to heritable mechanisms driving adaptation and evolution of organisms

CELL AND MOLECULAR BIOLOGY

Cellular structures and molecular networks involved in the regulation of growth, development and stress responses

BIOTECHNOLOGIES FOR SUSTAINABLE DEVELOPMENT

Biotechnological tools and applications for environmental protection, human health and sustainable agriculture

BIODIVERSITY AND GENETIC RESOURCES

Conservation and use of biological diversity to maintain ecological balance and food security

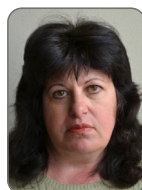
AGROBIOLOGY AND FOOD SCIENCE

Biological principles of agricultural systems and their links to food quality and safety

OUR KEY SPEAKERS



Valya Vassileva
Institute of Plant Physiology and
Genetics, Bulgarian Academy of
Sciences (Bulgaria)



Katya Georgieva
Institute of Plant Physiology
and Genetics, Bulgarian
Academy of Sciences (Bulgaria)



Edina Turkosi
Centre for Agricultural
Research (Hungary)



Adam Solti
Eötvös Loránd University
(Hungary)



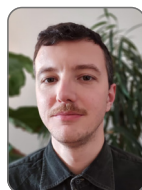
Faheem Shehzad Baloch
Mersin University (Turkey)



Tibor Janda
HUN-REN CAR, Agricultural
Institute (Hungary)



Ramona Duminicioiu
Eco Ruralis Association
(Romania)



Massimiliano Corso
INRAE-IJPB (France)



Philippe Gallusci
University of Bordeaux (France)



Yalçın Kaya
Trakya University (Turkey)

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Биомед фючар ЕООД

CONFERENCE PROGRAM

16 September 2026 (Wednesday)			
08:00 – 09:30 Registration & Wake-up Coffee			
09:30 – 10:00	Welcome	Valya Vassileva	Greetings and welcome from the Institute of Plant Physiology and Genetics – BAS.
10:00 – 13:00 Morning Session			
10:00 – 10:40	PL-01	Philippe Gallusci	Epigenetic memories in crops: studies from tomato and grape.
10:40 – 11:05	OP-01	Ane Kortabarria Pérez	Live slow and die old: developmental changes and leaf physiology in the resurrection angiosperm <i>Ramonda myconi</i> .
11:05 – 11:30	OP-02	Jurga Jankauskienė	Mitigation of herbicide-induced stress in common wheat by a microbial biostimulant: physiological and biochemical evidence.
11:30 – 12:10	PL-02	Tibor Janda	The role of light during cold acclimation processes in plants.
12:10 – 12:35	OP-03	Gergana Mihailova	Relationship between drought tolerance and fungal pathogen resistance in crop and resurrection plants.
12:35 – 13:00	OP-04	Gabriella Szalai	How <i>Eruca</i> plants respond to moderate UV-B radiation?
13:00 – 14:00	Lunch Break		
14:00 – 19:00 Afternoon Session			
14:00 – 14:40	PL-03	Katya Georgieva	<i>Haberlea rhodopensis</i> – resurrection as a survival strategy.
14:40 – 15:05	OP-05	Tsvetina Nikolova	Boosting wheat tolerance: how priming with spermine regulates the antioxidant system during biotic stress caused by <i>Fusarium culmorum</i> .
15:05 – 15:30	OP-06	Vasilissa Manova	Effects of plant growth regulators in two wheat species: from abiotic stress responses to field performance.
15:30 – 16:00	Coffee Break		
16:00 – 16:40	PL-04	Faheem Sh. Baloch	Exploiting Fertile Crescent diversity through precision genomics for climate-resilient agriculture.
16:40 – 17:05	OP-07	Tautvydas Žalnierius	<i>Heracleum sosnowskyi</i> control approaches based on gibberellin metabolism modulation.
17:05 – 17:30	OP-08	Alexander G. Ivanov	Restricted capacity for state transitions in the antarctic ecotype of <i>Colobanthus quitensis</i> implies differential strategies to cold acclimation of Antarctic and Andes ecotypes.
17:30 – 19:00	Poster Session		
19:30 – 23:00	Welcome Cocktail		



CONFERENCE PROGRAM

17 September 2026 (Thursday)			
08:30 – 13:00 Morning Session			
08:30 – 09:00	PhD Students Workshop		
09:00 – 09:40	PL-05	Valya Vassileva	From model plants to legumes - epigenetic regulation of stress responses and adaptation.
09:40 – 10:05	OP-09	Bekim Gashi	Networking and opportunities for sustainable crop epitranscriptomics.
10:05 – 10:30	OP-10	Volodymyr Strashnyuk	Thermal adaptation and seasonal variation in endoreduplication and polyteny in the fruit fly <i>Drosophila melanogaster</i> (Diptera: Drosophilidae).
10:30 – 11:00	Coffee Break		
11:00 – 11:40	PL-06	Ádám Solti	The contribution of the nitric oxide signaling and the autophagy processes to remove iron from the chloroplasts during developmental senescence.
11:40 – 12:05	OP-11	Irina I. Vaseva	Integration of ethylene signaling in drought stress responses.
12:05 – 12:30	OP-12	Georgi Antov	Anticancer potential of <i>Levisticum officinale</i> in <i>in vitro</i> cell culture model systems.
12:30 – 13:00	Round Table Discussion I		
13:00 – 14:00	Lunch Break		
14:00 – 19:00 Afternoon Session			
14:00 – 14:40	PL-07	Massimiliano Corso	Remodeling seed specialized metabolism for stress adaptation and plant-microbe interactions.
14:40 – 15:05	OP-13	Liudmyla Kozeko	Cellular response in rapeseed to exogenous amino acids under drought.
15:05 – 15:30	OP-14	Michel Francis	Nitric oxide-associated regulation of plastidial iron homeostasis.
15:30 – 16:00	Coffee Break		
16:00 – 16:25	OP-15	Shazie Yusein-Myashkova	Insights into the mode of action of novel naphthalimide derivatives as selective anticancer agents.
16:25 – 16:50	OP-16	Amit Roy	Integrated metabolomic and dual-seq analysis of Norway spruce-fungal pathogen interactions.
16:50 – 17:30	PL-08	Ramona Duminicioiu	Agroecology and food systems in Romania: small-scale farmers' perspectives on the current crisis.
17:30 – 19:00	Poster Session		
19:30 – 23:00	Social Dinner		



CONFERENCE PROGRAM

18 September 2026 (Friday)			
08:30 – 13:20 Morning Session			
08:30 – 09:10	PL-09	Edina Türkösi	From wild gene pools to resilient crops: <i>Thinopyrum</i> as a source of drought tolerance in wheat.
09:10 – 09:35	OP-17	Georgi Bonchev	Revealing the hidden genetic diversity in natural plant germplasm collections through DNA barcodes – case studies in breeding research and conservation of endemic and threatened species.
09:35 – 10:00	OP-18	Jana Kvíderová	Biophysical markers in large-scale algal cultivation.
10:00 – 10:30	Round Table Discussion II		
10:30 – 11:00	Coffee Break		
11:00 – 11:40	PL-10	Yalçın Kaya	Herbicide resistance in crops: evaluating integrated weed management.
11:40 – 12:05	OP-19	Margarita Popova	Differential drought responses of Bulgarian sweet, super-sweet and forage maize hybrids under field conditions.
12:05 – 12:30	OP-20	Amrita Chakraborty	Friends or foes? Insect-microbe interactions in fruit agroecosystems.
12:30 – 13:10	General Discussion and Future Perspectives		
13:10 – 13:20	Closing Ceremony		
13:20 – 14:20	Lunch Break		
14:30 – 19:00	Excursion		



LIST OF PRESENTATIONS

PLENARY LECTURES PL – 01–10

- PL-01 Philippe Gallusci** – EPIGENETIC MEMORIES IN CROPS: STUDIES FROM TOMATO AND GRAPE.
- PL-02 Tibor Janda** – THE ROLE OF LIGHT DURING COLD ACCLIMATION PROCESSES IN PLANTS.
- PL-03 Katya Georgieva** – *HABERLEA RHODOPENSIS* – RESURRECTION AS A SURVIVAL STRATEGY.
- PL-04 Faheem Shehzad Baloch, Parnaz Mortazavi, Muhammad Azhar Nadeem, Ahmad Alsaleh, Cemal Kurt, Amjad Ali** – EXPLOITING FERTILE CRESCENT DIVERSITY THROUGH PRECISION GENOMICS FOR CLIMATE-RESILIENT AGRICULTURE.
- PL-05 Valya Vassileva, Dimitar Todorov, Mariyana Georgieva** – FROM MODEL PLANTS TO LEGUMES – EPIGENETIC REGULATION OF STRESS RESPONSES AND ADAPTATION.
- PL-06 Lili Petrovicz, Veronika Kovács, Michel Francis, Jagruti Bodele, Krisztina Kovács, Katalin Solymosi, Paula Pongrac, Katarina Vogel-Mikuš, Valentina Bonanni, Milan Žižić, Alexandra Gianoncelli, Ádám Solti** – THE CONTRIBUTION OF THE NITRIC OXIDE SIGNALING AND THE AUTOPHAGY PROCESSES TO REMOVE IRON FROM THE CHLOROPLASTS DURING DEVELOPMENTAL SENESCENCE.
- PL-07 Léa Barreda, Stéphanie Boutet, Chandrodhay Saccaram, Emel Sezan, Céline Brosse, Mélissa Sancharme, Audrey Le-Cabec, Ophélie Merzeaud, Anne-Solenn Valadon, Jean-Luc Cacas, François Perreau, Loïc Rajjou, Loïc Lepiniec, Massimiliano Corso** – REMODELING SEED SPECIALIZED METABOLISM FOR STRESS ADAPTATION AND PLANT-MICROBE INTERACTIONS.
- PL-08 Ramona Duminicioiu** – AGROECOLOGY AND FOOD SYSTEMS IN ROMANIA: SMALL-SCALE FARMERS' PERSPECTIVES ON THE CURRENT CRISIS.
- PL-09 Edina Türkösi, Klaudia Kruppa, Éva Darkó, Balázs Varga, Márton György, Zsolt Gulyás, Kristóf Jobbágy, Kateřina Holušová, Eszter Gaál, Balázs Kalapos, Mónika Cséplő, András Farkas, László Ivanizs, Péter Mikó, Andrea Gulyás, Norbert Hidvégi, Péter Kovács, András Cseh, Márta Molnár-Láng, Jan Bartoš, Éva Szakács, István Molnár** – FROM WILD GENE POOLS TO RESILIENT CROPS: *THINOPYRUM* AS A SOURCE OF DROUGHT TOLERANCE IN WHEAT.
- PL-10 Yalçın Kaya** – HERBICIDE RESISTANCE IN CROPS: EVALUATING INTEGRATED WEED MANAGEMENT.

ORAL PRESENTATIONS OP – 01–20

- OP–01 **Ane Kortabarría Pérez**, Enara Alday Echechipia, Irati Arzac Garmendia, Antonio Hernández Hernández, Usue Pérez López, José Ignacio García Plazaola, Beatriz Fernández Marín – LIVE SLOW AND DIE OLD: DEVELOPMENTAL CHANGES AND LEAF PHYSIOLOGY IN THE RESURRECTION ANGIOSPERM *RAMONDA MYCONI*.
- OP–02 **Jurga Jankauskienė**, Greta Šaltytė, Rima Mockevičiūtė, Elżbieta Jankovska-Bortkevič, Sigita Jurkonienė, Zornitsa Katerova, Iskren Sergiev, Desislava Todorova – MITIGATION OF HERBICIDE-INDUCED STRESS IN COMMON WHEAT BY A MICROBIAL BIOSTIMULANT: PHYSIOLOGICAL AND BIOCHEMICAL EVIDENCE.
- OP–03 **Gergana Mihailova**, Katya Georgieva, Eva Popova, Vasilissa Manova, Zornitsa Stoyanova, Stilian Simeonova, Francesca Rapparini, Alessio Tei, Fabio Osti – RELATIONSHIP BETWEEN DROUGHT TOLERANCE AND FUNGAL PATHOGEN RESISTANCE IN CROP AND RESURRECTION PLANTS.
- OP–04 **Gabriella Szalai**, Magda Pál, Mihály Dernovics, Kamirán Á. Hamow, Kinga Benczúr, Tibor Janda – HOW ERUCA PLANTS RESPOND TO MODERATE UV-B RADIATION?
- OP–05 **Tsvetina Nikolova**, Elena Shopova, Zornitsa Katerova, Lilyana Brankova, Iskren Sergiev, Ivo Yanashkov, Desislava Todorova – BOOSTING WHEAT TOLERANCE: HOW PRIMING WITH SPERMINE REGULATES THE ANTIOXIDANT SYSTEM DURING BIOTIC STRESS CAUSED BY *FUSARIUM CULMORUM*.
- OP–06 **Vasilissa Manova**, Yana Koycheva, Radoslav Chipilski, Zoia Tsekova, Iskren Sergiev, Konstantina Kocheva, Elisaveta Kirova, Irina Moskova – EFFECTS OF PLANT GROWTH REGULATORS IN TWO WHEAT SPECIES: FROM ABIOTIC STRESS RESPONSES TO FIELD PERFORMANCE.
- OP–07 **Tautvydas Žalnierius**, Dominykas Laibakojis, Sigita Jurkonienė, Saulė Rapalytė – *HERACLEUM SOSNOWSKYI* CONTROL APPROACHES BASED ON GIBBERELLIN METABOLISM MODULATION.
- OP–08 **León A. Bravo**, Beth Szyszka-Mroz, Dimitre A. Ivanov, **Alexander G. Ivanov**, Norman P. A. Hüner – RESTRICTED CAPACITY FOR STATE TRANSITIONS IN THE ANTARCTIC ECOTYPE OF *COLOBANTHUS QUITENSIS* IMPLIES DIFFERENTIAL STRATEGIES TO COLD ACCLIMATION OF ANTARCTIC AND ANDES ECOTYPES.
- OP–09 **Bekim Gashi**, Valbona Sota, Jesus Pascual Vazquez – NETWORKING AND OPPORTUNITIES FOR SUSTAINABLE CROP EPITRANSCRIPTOMICS.
- OP–10 **Volodymyr Strashnyuk**, Yelyzaveta Koptevtsova, Evgenij Vakulenko – THERMAL ADAPTATION AND SEASONAL VARIATION IN ENDOREDUPPLICATION AND POLYTENY IN THE FRUIT FLY *DROSOPHILA MELANOGASTER* (DIPTERA: DROSOPHILIDAE).
- OP–11 **Irina I. Vaseva**, Simona Galabova, Dominique Van Der Straeten – INTEGRATION OF ETHYLENE SIGNALING IN DROUGHT STRESS RESPONSES.
- OP–12 **Georgi Antov**, Zlatina Gospodinova, Gergana Mihailova, Natalia Krasteva, Trayana Kamenska, Svetozar Stoichev, Danail Pavlov, Antoaneta Georgieva, Stefka Valcheva-Kuzmanova – ANTICANCER POTENTIAL OF *LEVISTICUM OFFICINALE* IN *IN VITRO* CELL CULTURE MODEL SYSTEMS.
- OP–13 **Liudmyla Kozeko**, Dmytro Klimchuk, Yuri Akimov, Yulia Ovcharenko, Virgilija Gavelienė, Tautvydas Žalnierius, Sigita Jurkonienė – CELLULAR RESPONSE IN RAPESEED TO EXOGENOUS AMINO ACIDS UNDER DROUGHT.
- OP–14 **Michel Francis**, Maria Gracheva, Helga Zelenyánszki, Katalin Solymosi, Ádám Solti – NITRIC OXIDE-ASSOCIATED REGULATION OF PLASTIDIAL IRON HOMEOSTASIS.

- OP–15** Zlatina Vlahova, Lazar Lazarov, Maria Petrova, Shazie Yusein-Myashkova, Jordana Todorova, Maria Schröder, Monika Mutovska, Stanimir Stoyanov, Yulian Zaganyarski, Iva Ugrinova – INSIGHTS INTO THE MODE OF ACTION OF NOVEL NAPHTHALIMIDE DERIVATIVES AS SELECTIVE ANTICANCER AGENTS.
- OP–16** Amit Roy, Sellamuthu Gothandapani, Amrita Chakraborty – INTEGRATED METABOLOMIC AND DUAL-SEQ ANALYSIS OF NORWAY SPRUCE–FUNGAL PATHOGEN INTERACTIONS.
- OP–17** Georgi Bonchev, Irina Boycheva, Vasilissa Manova, Stiliana Simeonova, Miroslava Zhiponova, Yordan Georgiev, Iva Doycheva, Stoyan Stoyanov, Maria Rosa Marano, Lucila Garcia, Carolina Galli, Miroslav Svec, Michal Hrabovski, Dominika Gahurova – REVEALING THE HIDDEN GENETIC DIVERSITY IN NATURAL PLANT GERMPLASM COLLECTIONS THROUGH DNA BARCODES – CASE STUDIES IN BREEDING RESEARCH AND CONSERVATION OF ENDEMIC AND THREATENED SPECIES.
- OP–18** Jana Kvíderová, Jaromír Lukavský, Josef Elster – BIOPHYSICAL MARKERS IN LARGE-SCALE ALGAL CULTIVATION.
- OP–19** Margarita Popova, Momchil Paunov, Biserka Hinkina, Nataliya Petrovska, Atanas Atanassov, Miroslava Zhiponova – DIFFERENTIAL DROUGHT RESPONSES OF BULGARIAN SWEET, SUPER-SWEET AND FORAGE MAIZE HYBRIDS UNDER FIELD CONDITIONS.
- OP–20** Amrita Chakraborty, Paul G. Becher – FRIENDS OR FOES? INSECT–MICROBE INTERACTIONS IN FRUIT AGROECOSYSTEMS.

POSTER PRESENTATIONS PP – 01–39

- PP–01 Dominykas Laibakojis, Simonas Balčiūnas, Saulė Rapalytė, Jurga Būdienė, Tautvydas Žalnierius, Sigita Jurkonienė – UHPLC–MS/MS METHOD DEVELOPMENT FOR ABSOLUTE QUANTIFICATION OF GIBBERELLINS AND INDOLE-3-ACETIC ACID IN REPRODUCTIVE TISSUES OF *HERACLEUM SOSNOWSKYI*.
- PP–02 Elena Shopova, Liliana Brankova, Zornitsa Katerova, Lyudmila Dimitrova, Ivo Dimitrov, Dessislava Todorova – EFFECT OF UV-C SEED PRIMING ON THE PHENOLIC CONTENT AND ANTIOXIDANT CAPACITY OF DIFFERENT KALE CULTIVARS.
- PP–03 Greta Šaltytė, Jurga Jankauskienė, Rima Mockevičiūtė – ETHYLENE EMISSION IN WINTER WHEAT (*TRITICUM AESTIVUM* L.) UNDER SHORT-TERM COLD STRESS: INFLUENCE OF HERBICIDE AND MICROBIAL BIOSTIMULANT TREATMENTS.
- PP–04 Liliana Brankova, Milen Bogdanov, Elena Shopova, Savina Stoyanova, Lyudmila Dimitrova, Zornitsa Katerova, Dessislava Todorova, Tsvetina Nikolova, Denitsa Petkova – UV-C SEED PRIMING AS A TOOL FOR MODULATING THE GLUCOSINOLATE PROFILE OF KALE MICROGREENS.
- PP–05 Dilyana Doneva, Violeta Peeva, Svetlana Misheva, Katelina Prokopova, Gabriella Szalai, Magda Pal, Aleksander Angelov, Vladimir Aleksandrov – COMPREHENSIVE STUDY OF THREE WHEAT CULTIVARS SUBJECTED TO DROUGHT.
- PP–06 Brigita Gylytė, Rolandas Karitonas, Sigita Jurkonienė, Vaidevutis Šveikauskas, Vira Khoma, Jūratė Vaičiūnienė, Vitalija Mickevičiūtė, Oksana Stoliar, Levonas Manusadžianas – CHARACEAN *NITELLOPSIS OBTUSA* CELL RESPONSES TO YTTRIUM AND SELECTED LANTHANIDES.
- PP–07 Hristo Nikolov, Radoslav Chipilski, Zoia Tsekova, Irina Moskova – EVALUATION OF THE EFFECT OF TWO PLANT GROWTH REGULATORS – 4PU-30 AND MEIA, ON EINKORN (*TRITICUM MONOCOCCUM* L.) AND WINTER WHEAT (*TRITICUM AESTIVUM* L.) IN FIELD CONDITIONS, USING DIFFERENT APPROACHES.
- PP–08 Katya Georgieva, Zornitsa Stoyanova, Eva Popova, Vasilissa Manova, Rossitza Rodeva, Bistra Mihailova, Gergana Mihailova – RESPONSE OF THE CROP PLANT *SOLANUM LYCOPERSICUM* AND THE RESURRECTION PLANT *HABERLEA RHODOPENSIS* TO INFECTION CAUSED BY PHYTOPATHOGENIC FUNGI.
- PP–09 Nadya Agayn-Stoyanova, Juliana Ivanova, Veronika Deleva – COMBINED EFFECT OF TEMPERATURE AND LIGHT ON THE ANTIOXIDANT ACTIVITY OF THE NEWLY ISOLATED STRAIN *SCENEDESMUS SP. JV-25*.
- PP–10 Rolandas Karitonas, Eimantas Antanas Lapukas – TOXICITY OF BINARY MIXTURES OF SULFAMETHOXAZOLE AND PRASEODYMIUM TO *LEMNA MINOR*. PHYSIOLOGICAL AND OXIDATIVE STRESS MARKERS AND PEROXIDASE INDUCTIONS.
- PP–11 Dessislava Todorova, Liliana Brankova, Elena Shopova, Zornitsa Katerova, Tsvetina Nikolova, Ljudmila Dimitrova, Sigita Jurkonienė, Jurga Jankauskienė, Iskren Sergiev – BIOCHEMICAL RESPONSES OF WINTER WHEAT TO APPLICATION OF MICROBIAL BIOSTIMULANT AND SELECTIVE HERBICIDE.
- PP–12 Iskren Sergiev, Elena Shopova, Liliana Brankova, Zornitsa Katerova, Tsvetina Nikolova, Ljudmila Dimitrova, Jurga Jankauskiene, Sigita Jurkoniene, Dessislava Todorova – MODULATION OF WHEAT PHYSIOLOGICAL RESPONSES TO HERBICIDE BY APPLICATION OF MICROBIAL BIOSTIMULANT.
- PP–13 Simona Galabova, Irina I. Vaseva, Dominique Van Der Straeten – TISSUE-SPECIFIC MANIPULATION OF ETHYLENE SIGNALING IN *ARABIDOPSIS THALIANA* AFFECTS THE TRANSCRIPTIONAL DYNAMICS OF STRESS-INDUCIBLE GENES UNDER DROUGHT AND RECOVERY.

- PP-14 **Maria Stasińska-Jakubas, Barbara Hawrylak-Nowak** – EFFECTS OF CHEMICALLY DIVERSE ELICITORS ON OXIDATIVE STATUS AND SPECIALIZED METABOLISM IN MEDICINAL PLANTS.
- PP-15 **Saulė Rapalytė, Dominykas Laibakojis, Tautvydas Žalnierius, Sigita Jurkonienė** – *HERACLEUM SOSNOWSKYI* FRUIT DEVELOPMENT DISRUPTION BY SPRAY-INDUCED GENE SILENCING OF GIBBERELLIN 2-OXIDASE.
- PP-16 **Violeta Peeva, Dilyana Doneva, Svetlana Misheva, Katelina Prokopova, Alexander Angelov, Vladimir Aleksandrov** – INSIGHT INTO PHOTOCHEMICAL AND PHYSIOLOGICAL TRAITS OF TWO WHEAT VARIETIES UNDER WATER DEFICIT.
- PP-17 **Zornitsa Katerova, Tsvetina Nikolova, Liliana Brankova, Elena Shopova, Iskren Sergiev, Ivo Yanashkov, Dessislava Todorova** – ALTERATIONS IN THE CONTENT OF PHENOLIC COMPOUNDS AND FLAVONOIDS IN WINTER WHEAT AFTER PHYTOPATHOGENIC FUNGUS INFECTION IS MODULATED BY SPERMINE SEED PRIMING.
- PP-18 **Zornitsa Stoyanova, Agnieszka Jamiołkowska, Weronika Kurska, Ewa Zalewska** – PATHOGENIC POTENTIAL OF *FUSARIUM OXYSPORUM* ISOLATES FROM TOMATO AND WHEAT ROOTS, ASSESSED ON DETACHED TOMATO FRUITS.
- PP-19 **Sigita Jurkonienė, Virgilija Gavelienė, Rima Mockevičiūtė, Elžbieta Jankovska-Bortkevič, Tautvydas Žalnierius, Vaidevutis Šveikauskas, Liudmyla Kozeko** – SCREENING OF BIOSTIMULANTS TO INCREASE CELLULOSE CONTENT IN FLAX FIBERS.
- PP-20 **Yildiz Dogan, Abdullah Unlu, Musa Kirisik, Kubra Yildiz, Mine Unlu, Levent Keskin, Muhammad Azhar Nadeem, Faheem Shehzad Baloch** – EVALUATION OF LOCAL MELON GENOTYPES FOR RESISTANCE TO APHID (*APHIS GOSSYPYII* GLOVER) AND ZUCCHINI YELLOW MOSAIC VIRUS (ZYMV) AND DEVELOPMENT OF A RESISTANT LINE.
- PP-21 **Iliyan Pochileev, Ivanka Dimova** – INTEGRATIVE miRNA-mRNA REGULATORY SIGNATURES IN BRAF-STRATIFIED METASTATIC MELANOMA.
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PLENARY LECTURES

PL-01

EPIGENETIC MEMORIES IN CROPS: STUDIES FROM TOMATO AND GRAPE

Philippe Gallusci

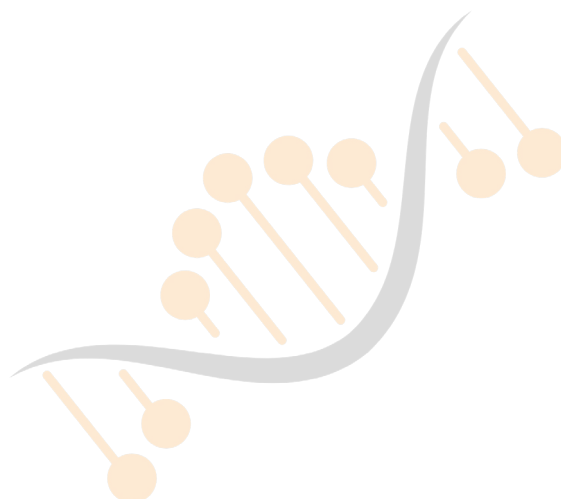
UMR EGFV, Bordeaux University, department of environmental sciences, INRAE, Bordeaux Science Agro, Villenave d'Ornon, France

Presenting author: philippe.gallusci@inrae.fr; philippe.gallusci@u-bordeaux.fr

Epigenetics encompasses the complement of genetic information carried in chromatin beyond the DNA sequence. Studies in *Arabidopsis*, tomato and other plants, have demonstrated the relevance of epigenetic mechanisms in the control of development, impact on agronomic traits and genome stability. However, the transmission of epigenetic information through generation is still a matter of debate. So far, there is no comprehensive view on an epigenetic inter/trans-generational memory, which may vary depending on the factors inducing the epigenetic variations, the type of variations as well as on reproduction mode.

To investigate the transgenerational memory of epigenetic marks, we have used a unique tomato EpiRIL population generated by crossing WT and *SIMET1* RNAi plants of the same genetic background. The population of EpiRILs consists of 115 lines that were propagated by single seed descent over 8 generations. Observable epigenetic and phenotypic differences are evident within the population. Methylome analysis of F4 and F7 plants of the EpiRIL population indicates methylation profiles in part corresponding to the WT or *SIMET1* RNAi lines and provide evidence of inheritance of DNA methylation imprints over several generations. This EpiRIL population will now provide a valuable epigenetic community resource to evaluate the contribution of epigenetic variations to traits of agronomic interest including chemo/phenotypes and responses to environmental stresses. In addition, we are currently developing works in grapevine to investigate the consequences of (a) biotic stresses on the plant DNA methylation landscapes, and analyze the epigenetic memory of such stresses over years, and through clonal propagation.

Keywords: Epigenetic memory; DNA methylation; transgenerational inheritance; EpiRILs; tomato; grapevine; environmental stress; clonal propagation.



PL-02**THE ROLE OF LIGHT DURING COLD ACCLIMATION PROCESSES IN PLANTS****Tibor Janda**

Department of Plant Physiology and Metabolomics, Agricultural Institute, HUN-REN Centre for Agricultural Research, Martonvásár, Hungary

Presenting author: janda.tibor@atk.hun-ren.hu

During growth at low temperatures, plants are able to adapt to a subsequent severe cold stress, one that would have been lethal without acclimation. Although the mechanisms of cold acclimation are well known in various plant species, many details, especially in the area of regulatory processes, have not yet been clarified. Not only temperature but also light plays a significant role in cold acclimation: although photoinhibition can occur at low temperatures, cold hardening is not sufficiently effective without adequate light intensity. The main aim of our recent studies was to better understand how the low temperature signal from the leaves may affect the stress responses in the roots, and how the light conditions modify certain stress acclimation processes in different crop plants.

In the 1st experiment, rice plants were exposed to moderate low temperatures with different light intensities, and in the case of some groups of plants, only the leaves received the cold, while the roots remained at control temperature. RNA sequencing focusing on the roots of plants grown under normal growth light conditions revealed that nitrogen metabolism and nitric oxide-related signalling, as well as the phenylpropanoid-related processes, were specifically affected. Real-time PCR results focusing on the COLD1 and polyamine oxidase genes, as well as metabolomics targeting hormonal changes and phenolic compounds also showed that not only cold exposure of the leaves, either alone or together with the roots, but also the light conditions may influence certain stress responses in the roots of rice plants (Gholizadeh et al. 2024.)

In the 2nd experiment, wheat plants were grown under white light or blue light conditions, then plants were exposed to low, acclimating temperatures. Measurements of the chlorophyll-*a* fluorescence parameters, targeted and untargeted HPLC analyses performed on the leaves and roots of plants, completed with gene expression analyses demonstrate that blue light may induce specific cold acclimation processes in wheat leaves, and the effects of light signal can also be detected in roots (Szalai et al. 2025).

In the 3rd experiment, young maize plants were cold acclimated at different light intensities. After that, not only its effect on cold stress responses, but also the responses to another stressor, high salinity, were analysed at metabolite and gene expression levels.

Keywords: Acclimation; cereals; light; low temperature; photoinhibition; shoot-root signalling.

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PL-03

HABERLEA RHODOPENSIS – RESURRECTION AS A SURVIVAL STRATEGY

Katya Georgieva¹, Gergana Mihailova¹, Liliana Gigova¹, Lyudmila Simova-Stoilova¹, Éva Sárvári², Áron Keresztes², Katalin Solymosi², Gianpaolo Bertazza³, Francesca Rapparini³, Ádám Solti²

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. G. Bonchev Str., Building 21, 1113 Sofia, Bulgaria

²Institute of Biology, ELTE Eötvös Loránd University, Pázmány Péter Sétány 1/C, H-1117 Budapest, Hungary

³Institute of BioEconomy, National Research Council of Italy (CNR-IBE), Via P. Gobetti 101, 40129 Bologna, Italy

Presenting author: georgieva.katya.m@gmail.com; katya@bio21.bas.bg

Haberlea rhodopensis Friv. (Gesneriaceae) is a perennial herbaceous, poikilohydric flowering plant that can survive desiccation to an air-dried state and fully recover upon rehydration. It is endemic to the Balkan Peninsula and a relict species of the Tertiary period, belonging to the group of homoiochlorophyllous resurrection plants that retain their chlorophyll content during dehydration. It inhabits mostly shady, northern, limestone or silicate rocks in mountain areas with relatively high humidity, but can be rarely found on rocks directly exposed to sunlight. Unlike most resurrection plants, *H. rhodopensis* is exposed to and can withstand freezing temperatures in winter. Similar to drought, freezing stress also causes plant dehydration, and they survive the harsh winter conditions in a desiccated state. Our studies were focused on the mechanisms of desiccation tolerance at optimal, high and low temperatures. The effect of light during desiccation was investigated by comparing the response of shade and sun plants. To elucidate the survival strategies of this unique species, we examined the protective responses during both desiccation and rehydration, as rapid water uptake represents an additional physiological challenge. Our results showed that membrane integrity is preserved during desiccation of *H. rhodopensis*. The enhanced leakage in air-dried leaves likely reflects structural modifications in the cell wall, plasma membrane, and vacuolar system. Drought stress induced a complete rearrangement of cellular architecture: small secondary vacuoles formed during moderate dehydration, progressively expanding and displacing the cytoplasm and organelles toward the cell interior. Chloroplasts decreased in size, became more rounded, and retained grana and stroma thylakoids containing dense luminal substance. A characteristic feature of resurrection plants is the reversible downregulation of photosynthetic activity. CO₂ assimilation was more sensitive to drought stress than photochemical activity, declining markedly under moderate water loss when PSII quantum efficiency showed only slight reduction. The decline in PSII activity was accompanied by enhanced PSI activity. Under severe water stress, reduced photochemical efficiency was coupled with increased non-photochemical quenching, preventing PSII overexcitation. Desiccation of *H. rhodopensis* to an air-dried state caused slight reductions in key photosynthetic proteins and minor changes in the relative amounts of chlorophyll-protein complexes. Protection against desiccation damage involved the accumulation of non-reducing sugars and other osmoprotectants to maintain hydration shells, synthesis of protective proteins (such as dehydrins, ELIPs, sHSPs) that stabilize membranes and macromolecular structures, and strong upregulation of antioxidant systems. Antioxidant activity peaked in the air-dry state, a characteristic feature of resurrection plants.

Keywords: Drought stress; rehydration; cellular integrity; photosynthesis; chlorophyll fluorescence; photosynthetic proteins; antioxidant activity; protective proteins.

PL-04

EXPLOITING FERTILE CRESCENT DIVERSITY THROUGH PRECISION GENOMICS FOR CLIMATE-RESILIENT AGRICULTURE

Faheem Shehzad Baloch¹, Parnaz Mortazavi², Muhammad Azhar Nadeem¹, Ahmad Alsaleh³, Cemal Kurt⁴, Amjad Ali²

¹Department of Biotechnology, Faculty of Science, Mersin University, Mersin, Türkiye

²Department of Plant Protection, Faculty of Agricultural Science and Technologies, Sivas university of Science and Technology, Sivas, Türkiye

³Department of Agriculture and Food Hemp Research Institute, Yozgat Bozok University, Türkiye

⁴Department of Field Crops, Faculty of Agriculture, University of Çukurova, Adana, Türkiye

Presenting author: balochfaheem13@gmail.com

Türkiye, located within the Fertile Crescent, represents one of the world's most significant centers of crop domestication and genetic diversity, particularly for cereals and legumes. The rich genetic resources originating from this region harbor novel alleles governing key agronomic, nutritional, and stress-resilience traits, offering immense potential for sustainable crop improvement.

Despite substantial yield gains achieved during the Green Revolution through dwarfing genes and advanced agronomic practices, modern agriculture faces escalating challenges, including climate change, reduced arable land, and increasing global food demand. Addressing these constraints requires innovative, genomics-driven breeding strategies.

In this study, we present an integrated genomics-assisted breeding framework leveraging high-throughput genotyping and phenotyping approaches. Our research encompasses extensive germplasm collections of both wild and cultivated species, including bread and durum wheat, chickpea, lentil, common bean, sesame, maize, peanut, soybean, and medicinal plants. These resources have been systematically characterized using genotyping-by-sequencing (GBS) and whole-genome resequencing, enabling high-resolution discovery of genomic variation.

We have identified and validated numerous QTLs and SNP markers associated with adaptive, physiological, and nutritional traits under diverse stress conditions. Ongoing work focuses on dissecting heat stress tolerance in an interspecific F₂ population derived from *Cicer arietinum* × *C. reticulatum*, targeting key physiological traits. Furthermore, climate-resilient and underutilized crops such as buckwheat, finger millet, and sorghum have been introduced into the Mediterranean agro-ecosystems of Türkiye to enhance system resilience.

Through ERANET and TÜBİTAK-funded projects, we have successfully applied GBS and resequencing approaches to identify loci linked to performance under nitrogen- and water-limited conditions. Complementary studies include marker-assisted breeding for aphid and Zucchini yellow mosaic virus resistance in melon, GWAS and KASP marker development in industrial hemp, validation of drought-associated SNPs in tomato, and integrative GWAS in buckwheat combining genomics with image-based phenotyping.

Collectively, these findings demonstrate the power of integrating genomics, phenomics, and marker-assisted selection to unlock crop genetic diversity and accelerate the development of climate-resilient cultivars, thereby contributing to sustainable agricultural systems.

Keywords: Genome diversity; High-throughput genotyping and phenotyping; QTL mapping; Germplasm characterization; Sustainable agriculture.

Acknowledgements: This work was supported by ERANET (Project No: 220N276), TÜBİTAK (Projects No: 121O313, 125O351) and COST Projects (225O388, 123O988).

PL-05

FROM MODEL PLANTS TO LEGUMES - EPIGENETIC REGULATION OF STRESS RESPONSES AND ADAPTATION

Valya Vassileva, Dimitar Todorov, Mariyana Georgieva

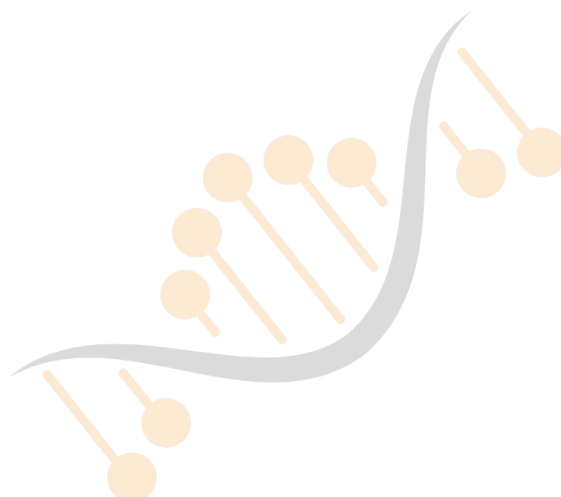
Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., Bldg. 21, Sofia, Bulgaria

Presenting author: valyavassileva@bio21.bas.bg

Epigenetic regulation helps plants respond to environmental stress through changes in gene expression that occur without alterations in the underlying DNA sequence. Research in *Arabidopsis thaliana* demonstrates that DNA methylation, chromatin remodelling and non-coding RNAs work together to control stress-related genes and mobile genetic elements. Some stress-induced chromatin modifications can persist beyond the initial stress event and contribute to priming and the establishment of stress memory. However, it remains unclear whether stress-induced epigenetic changes drive tolerance or merely accompany it, to what degree their establishment and persistence are shaped by genetic background, and how these processes operate in crops with larger, more repetitive genomes. We examine these questions in model plants and assess how far the emerging concepts translate to crop plants, including legumes, focusing on drought and heat stress responses. Studies in *Arabidopsis*, legumes and other crops indicate that stress-induced remodelling of DNA methylation and small RNA networks depends on genotype, organ and tissue type, and stress intensity, and is associated with differences in root system architecture, stress tolerance and recovery. We also consider the distinction between short-term, reversible epigenetic responses and more persistent modifications that potentially contribute to stress memory, and the challenges of the identification of truly heritable epigenetic variation. Finally, we discuss the opportunities and current limitations of epigenetic variation as a resource for crop improvement, including methylation-based biomarkers for the selection of stress-resilient germplasm, epigenetic priming strategies and emerging epigenome-editing tools for the enhancement of stress resilience.

Keywords: DNA methylation; non-coding RNAs; stress priming and memory; drought; heat stress; grain legumes.

Acknowledgements: This work was supported by the EU Horizon Europe PROSPER project (Grant Agreement No. 101181400).



PL-06

THE CONTRIBUTION OF THE NITRIC OXIDE SIGNALING AND THE AUTOPHAGY PROCESSES TO REMOVE IRON FROM THE CHLOROPLASTS DURING DEVELOPMENTAL SENESCENCE

Lili Petrovicz¹, Veronika Kovács¹, Michel Francis¹, Jagruti Bodele¹, Krisztina Kovács², Katalin Solymosi³, Paula Pongrac^{4,5}, Katarina Vogel-Mikuš⁴, Valentina Bonanni⁶, Milan Žižić^{6,a}, Alexandra Gianoncelli⁶, Ádám Solti¹

¹Department of Plant Physiology and Molecular Plant Biology, ELTE Eötvös Loránd University, Hungary

²Laboratory of Nuclear Chemistry, ELTE Eötvös Loránd University, Hungary

³Department of Plant Anatomy, ELTE Eötvös Loránd University, Hungary

⁴Department of Biology, University of Ljubljana, Slovenia

⁵Microanalytical Center, Jožef Stefan Institute, Slovenia

⁶Elettra-Sincrotrone Trieste S.C.P.A, Italy

^aCurrent affiliation: Department of Life Sciences, University of Belgrade, Serbia

Presenting author: adam.solti@ttk.elte.hu

Iron (Fe) allocation in the mesophyll cells primary targets chloroplasts to operate the photosynthetic electron transport chain, that Fe content is necessarily undergo to mobilization when Fe source-sink relations change, and when chloroplasts are directed towards degradations such as under developmental senescence. However, regulation of the chloroplast Fe homeostasis as well as the ways of the removal of Fe from the chloroplasts are hardly explored. Although nitric oxide (NO) is known to affect the Fe homeostasis of plants by affecting particularly the Fe uptake machinery of roots, evidence is missing that NO signal is also involved in the Fe status regulation of mesophyll cells. S-nitrosylation of glutathione (GSNO) has a pivotal role in the NO signalling that can trans-nitrosylate Cys residues of other proteins. The S-nitrosylation status is also regulated by the S-nitrosogluthatione reductase (GSNOR). Our expression analysis studies confirmed that the activation of the developmental senescence is also associated with the rising expression of *GSNOR*. In *Arabidopsis thaliana* Col-0 leaves the peak Fe content of chloroplasts is associated with early senescence that is followed by a gradual decrease in the Fe content associated with chloroplast ferritins. In macroautophagy defective lines, that process plays a key role in the removal of Fe from the chloroplasts, the ferritin apoprotein level remains low. Chloroplasts of the *GSNOR* knock-out *gsnor1.3* line overaccumulate Fe in the chloroplasts with a low Fe content in the leaves. Chloroplast Fe content remains high in developmental senescence, but in a high FER (apo)protein environment. In contrast, in *35S:GSNOR:FLAG* line overexpressing *GSNOR* the chloroplast Fe content remained close to that in Col-0. Our data indicates that NO signaling has a key role in the regulation of the chloroplast Fe status, and *GSNOR* is involved in the release of chloroplast Fe content upon developmental senescence.

Keywords: Chloroplast; iron; nitric oxide; S-nitrosylation.

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PL-07

**REMODELING SEED SPECIALIZED METABOLISM FOR STRESS ADAPTATION
AND PLANT-MICROBE INTERACTIONS**

Léa Barreda, Stéphanie Boutet, Chandrodhay Saccaram, Emel Sezan, Céline Brosse, Mélissa Sancharme, Audrey Le-Cabec, Ophélie Merzeaud, Anne-Solenn Valadon, Jean-Luc Cacas, François Perreau, Loïc Rajjou, Loïc Lepiniec, Massimiliano Corso

Université Paris-Saclay, INRAE, AgroParisTech, Institute Jean-Pierre Bourgin for Plant Sciences, 78000 Versailles, France

Presenting author: massimiliano.corso@inrae.fr

Specialized metabolites (SMs) play essential roles in seed quality by modulating developmental processes and enhancing tolerance to environmental stresses. In seeds, SM composition is highly dynamic and responsive to environmental conditions, contributing to protection against desiccation, heat stress, pathogens, and other environmental challenges.

Our research investigates how environmental stresses and nutrient availability reshape the seed specialized metabolome and how these metabolic changes contribute to stress adaptation. Using multi-omics approaches combined with functional genetics, we have shown that seeds of several Brassicaceae species (*Arabidopsis thaliana*, *Camelina sativa*, and *Brassica napus*) exhibit remarkable metabolic plasticity by adjusting their SM profiles in response to changing environmental conditions.

A major focus of our work has been the characterization of seed glucosinolate (GSL) diversity and its functional significance. Data from our groups have identified genes and enzymes involved in GSL side-chain elongation, substantially advancing our understanding of GSL diversity and its ecological function. We have demonstrated that environmental conditions strongly influence seed GSL accumulation and the production of their degradation products. Furthermore, we identified and characterized the gene network responsible for the biosynthesis of newly discovered acylated GSLs under both optimal and heat-stress conditions. These metabolic changes were associated with altered antioxidant capacity and reduced seed performance under stress. Further analyses on the role of glucosinolates in regulating seed-pathogen interactions are ongoing.

Our results reveal both conserved metabolic responses to environmental stress and genotype-specific regulatory mechanisms that shape seed specialized metabolism. These findings provide new insights into the adaptive significance of GSL diversity and the molecular mechanisms underlying seed quality, stress resilience, and plant defense.

Keywords: Specialized metabolites; glucosinolates; seed quality; heat stress; stress adaptation; plant-microbe interactions.

Acknowledgements: This work was supported by the French Government through the French National Research Agency (ANR) under the France 2030 Programme, “Grand Challenge: Biocontrol and Biostimulation for Agroecology” (MAMBO, ANR-25-GDBB-0004; SUPRASEED, ANR-25-GDBB-0003), and by the EU Horizon Europe PROSPER project (Grant Agreement No. 101181400). IJPB also acknowledges support from Saclay Plant Sciences-SPS (ANR-17-EUR-0007) and the Plant Observatory platforms PO-Chem and PO-Plants.



PL-08**AGROECOLOGY AND FOOD SYSTEMS IN ROMANIA: SMALL-SCALE FARMERS' PERSPECTIVES ON THE CURRENT CRISIS****Ramona Duminicioiu**

Eco Ruralis Association, Romania

Presenting author: ramona@ecoruralis.ro

Romania's agricultural system has been standing at a crossroads for decades, without fully mobilising two of its most important resources: the knowledge and potential offered by functional biology and the country's exceptionally large and diverse community of small-scale and peasant farmers. Almost one third (31.8%) of all farms in the European Union are located in Romania, the largest number of any EU member state. Yet the connection between scientific knowledge and this extraordinary farming community remains largely underexplored. Working with biological diversity, locally adapted seeds, complex agroecosystems and traditional and evolving knowledge, small scale farmers and peasants have a unique capacity to put biological and ecological processes into practice in food production and to advance agroecology at scale. This unrealised potential is part of a long-standing and largely neglected structural crisis, now intensified by the overlapping economic, social, climate and geopolitical crisis. Conflict, climate change, unstable and unequal markets, rising production costs, concentration of land and resources, demographic decline, increasingly concentrated food chains and weak democratic institutions and governance are undermining the viability of small scale farming and agroecology. Yet peasants and small-scale farmers continue to sustain local food systems, agricultural biodiversity, knowledge systems and rural economies, as well as forms of agricultural production that depend on diversity rather than uniformity. The current crisis is not only a crisis of agricultural production, but of the structures that determine who controls the resources, whose knowledge is recognised and which models of farming are supported. Small-scale farmers must be recognised not merely as beneficiaries of an agroecological transition, but as central actors capable of advancing and scaling it. Romania's agroecological transition cannot advance in isolation. It is inseparable from the wider Eastern European context, shaped by interconnected ecosystems, shared histories of agrarian reforms and similar social and economic realities, including persistent economic migration that is emptying rural areas and weakening the continuity of farming communities. These common conditions require more than fragmented national responses. The capacity of agroecology to expand depends on structural conditions, including access to land and seeds, fair markets and prices, generational renewal, recognition of farmers' knowledge, adequate public support and the effective implementation of human rights obligations, many of which require stronger regional cooperation and coordinated policy approaches. Eastern Europe therefore needs a common agroecological agenda, rooted in its own ecological, agrarian and social realities and built through sustained cooperation among farmers, researchers, civil society and public institutions.

Romania's and Eastern Europe's small-scale farms and peasant households constitute a living infrastructure of knowledge, biodiversity and food production. Connecting this capacity with functional biology and agroecological science, while strengthening regional cooperation and the rights and agency of small-scale farmers, offers a pathway for responding to both long-standing and emerging crises and for building more resilient, biodiverse, socially just and democratic food systems.

Keywords: Agroecology; small-scale farming; peasant agriculture; agrobiodiversity; food systems; farmers' knowledge; rural resilience; Eastern Europe.

PL-09

FROM WILD GENE POOLS TO RESILIENT CROPS: *THINOPYRUM* AS A SOURCE OF DROUGHT TOLERANCE IN WHEAT

Edina Türkösi¹, Klaudia Kruppa¹, Éva Darkó¹, Balázs Varga¹, Márton György¹, Zsolt Gulyás¹, Kristóf Jobbágy¹, Kateřina Holušová², Eszter Gaál¹, Balázs Kalapos¹, Mónika Cséplő¹, András Farkas¹, László Ivanizs¹, Péter Mikó¹, Andrea Gulyás¹, Norbert Hidvégi¹, Péter Kovács¹, András Cseh¹, Márta Molnár-Láng¹, Jan Bartoš², Éva Szakács¹, István Molnár¹

¹Centre for Agricultural Research, Hungarian Research Network (HUN-REN), Agricultural Institute, 2462 Martonvásár, Hungary

²Institute of Experimental Botany of the Czech Academy of Sciences, Centre of Plant Structural and Functional Genomics, Olomouc 779 00, Czech Republic

Presenting author: turkosi.edina@atk.hun-ren.hu

Bread wheat (*Triticum aestivum* L., $2n = 6x = 42$, BBAADD) is one of the world's major staple crops, but its productivity is increasingly threatened by drought and other climate-related stresses. In the context of climate change and decreasing availability of arable land, the development of high-yielding, stress-tolerant cultivars has become a major breeding objective. Wild relatives of wheat represent valuable sources of genetic diversity for crop improvement, and recent genomic advances have greatly enhanced the efficiency of alien introgression breeding. Species of the genus *Thinopyrum* (wheatgrasses) are important genetic resources for wheat improvement due to their adaptation to harsh environmental conditions and broad stress tolerance. Agropyron glael, an artificial hybrid of *Thinopyrum intermedium* and *Th. ponticum*, was introduced into the Martonvásár pre-breeding program to increase wheat genetic diversity through novel allelic variants.

Stable introgression lines carrying 42 or 44 chromosomes, including the addition (GLA7) and substitution (GLA8) lines, were evaluated for drought tolerance during flowering. Automated phenotyping of shoot traits, together with measurements of leaf relative water content and yield components, indicated improved drought tolerance in the GLA7 line compared with the parental wheat genotypes. The semidwarf GLA8 line, carrying the *Rht8* and *Rht2* dwarfing alleles, also displayed favorable physiological responses under drought conditions, including improved water status and maintenance of photosynthetic activity during stress. Changes in vegetation indices, activation of stress-responsive genes, and alterations in root architecture under drought conditions suggest adaptive mechanisms contributing to enhanced stress tolerance and reduced yield loss in these genotypes.

The enhanced drought tolerance observed in these introgression lines may be associated with the introgressed *Thinopyrum* chromatin. However, precise identification of the underlying quantitative trait loci (QTLs) will require further reduction and characterization of the alien chromosomal segments. Advanced genomic approaches and the development of molecular markers will facilitate the identification and selection of drought-tolerant genotypes for future wheat breeding programs. These results demonstrate the potential of *Thinopyrum*-derived introgressions to improve drought adaptation in wheat and provide a foundation for the identification of drought-associated loci for marker-assisted breeding.

Keywords: *Thinopyrum*; introgression breeding; drought tolerance.

Acknowledgements: The research leading to these results received funding from the Hungarian National Research, Development, and Innovation Office under Grant Agreement No FK145848 and the MTA Bolyai János Research Grant KK(BO/00013/25).

PL-10**HERBICIDE RESISTANCE IN CROPS: EVALUATING INTEGRATED WEED MANAGEMENT****Yalçın Kaya**

Trakya University, Engineering Faculty, Genetic and Bioengineering Department, Edirne, Türkiye

Presenting author: yalcinkaya22@gmail.com

Weeds are among the major biotic constraints to crop production and can substantially reduce yield and crop quality. Chemical herbicides have greatly improved the efficiency of weed control; however, their intensive and repeated use has imposed strong selection pressure on weed populations and led to the evolution of resistance to multiple herbicide sites of action, including ALS and ACCase inhibitors and glyphosate. The development of herbicide resistance is influenced by weed biology, gene flow, cropping intensity, monoculture and repeated reliance on herbicides with the same mode of action.

Herbicide-tolerant crop technologies have provided additional options for weed management. Glyphosate-tolerant crops such as Roundup Ready® soybean, maize, cotton and canola have simplified weed control and facilitated conservation tillage. Non-GM systems such as Clearfield®, based on imidazolinone-tolerant crops, have been widely adopted in sunflower, rice and other crops. Tribenuron-methyl-tolerant sunflower hybrids and newer technologies combining tolerance to different ALS-inhibiting herbicides provide further management flexibility. In rice, Clearfield® and Provisia™ systems offer alternative approaches to control red rice, weedy rice and other problematic grasses. However, reliance on any single herbicide-tolerance system may ultimately increase the risk of resistance evolution.

Sustainable weed control therefore requires integrated weed management (IWM) combining crop rotation and diversification, competitive cultivars and cover crops, optimized sowing rates and planting geometry, mechanical and physical weed control, harvest weed-seed control, and judicious rotation of herbicides with different modes of action. Site-specific weed management, decision-support systems, remote sensing and machine-vision-based spot spraying can further improve the efficiency of IWM. Successful implementation will also depend on addressing barriers related to farmer awareness, short-term economic costs, labour and equipment availability, and appropriate policy support.

Keywords: Herbicide resistance; integrated weed management; Clearfield system; A.I.R. technology; Provisia rice; sustainable agriculture; weed biology.



ORAL PRESENTATIONS

OP-01

LIVE SLOW AND DIE OLD: DEVELOPMENTAL CHANGES AND LEAF PHYSIOLOGY IN THE RESURRECTION ANGIOSPERM *RAMONDA MYCONI*

Ane Kortabarria Pérez, Enara Alday Echechipia, Irati Arzac Garmendia, Antonio Hernández Hernández, Usue Pérez López, José Ignacio García Plazaola, Beatriz Fernández Marín

Vegetal Biology and Ecology Dpt., UPV/EHU, 48940 Leioa, Bizkaia, Spain

Presenting author: ane.kortabarria@ehu.eus

The paleotropical resurrection plant, *Ramonda myconi* (L.) Rchb., endemic to the Iberian Peninsula, has the unique ability to survive desiccation and freezing. Despite its long lifespan, the specific duration of *R. myconi* leaves and the mechanisms they use to withstand desiccation multiple times throughout their lifetime remain uncertain. Considering that the plant's survival depends on the physiological performance of its leaves, we hypothesize that leaf ageing is a significant factor in this species survival. To address this, we characterised, across an annual cycle, leaf and plant growth, desiccation and freezing tolerance, photosynthesis, pigments, and antioxidants in three age classes of leaves (young, adult and senescent) of *R. myconi*. Based on preliminary findings, we estimate that the lifespan of an *R. myconi* leaf is at least two years. The leaves of different ages of *Ramonda myconi*, being a double-tolerant species, differ in their physiological responses to desiccation induced by drought and freezing. In short, our data suggest that freezing tolerance depends mostly on the acclimation process, while desiccation tolerance is constitutive. Notably, senescent leaves, due to their longer lifespan, have a higher tolerance. Our results show that *R. myconi* behaves like a typical shade-tolerant leaf in terms of its photosynthetic parameters and maintains a low assimilation rate throughout the year; nevertheless, as the leaves get older, photosynthetic capacity decreases, and α -tocopherol content on a chlorophyll basis increases. Consequently, senescent leaves are more tolerant to environmental stresses. Overall, the long lifespan of the leaves and the physiological differences between leaf classes allow the plant to grow successfully in a hostile environment while coping with both desiccation and freezing conditions.

Keywords: Desiccation; freezing; leaf ageing; photosynthesis; *Ramonda myconi*.



OP-02

MITIGATION OF HERBICIDE-INDUCED STRESS IN COMMON WHEAT BY A MICROBIAL BIOSTIMULANT: PHYSIOLOGICAL AND BIOCHEMICAL EVIDENCE

Jurga Jankauskienė¹, Greta Šaltytė¹, Rima Mockevičiūtė¹, Elžbieta Jankovska-Bortkevič¹, Sigita Jurkonienė¹, Zornitsa Katerova², Iskren Sergiev², Dessislava Todorova²

¹Laboratory of Plant Physiology, State Scientific Research Institute Nature Research Centre, Akademijos Str. 2, 08412, Vilnius, Lithuania

²Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. G. Bonchev Str., Bl. 21, 1113, Sofia, Bulgaria

Presenting author: jurga.jankauskiene@gamtc.lt

The use of herbicides in modern agriculture plays an important role in effective weed management; however, these agrochemicals may adversely affect plant physiological processes. Therefore, this study aimed to investigate the impact of a herbicide containing metsulfuron-methyl (68 g kg⁻¹) and thifensulfuron-methyl (682 g kg⁻¹) on selected physiological and biochemical parameters of common wheat, as well as to evaluate the potential of the environmentally friendly microbial biostimulant “ProbioHumus” in alleviating herbicide-induced stress.

To assess plant responses, biometric parameters, ethylene production, photosynthetic pigments content (chlorophyll *a*, chlorophyll *b*, carotenoids, and chlorophylls *a/b* ratio), oxidative stress indicators (malondialdehyde – MDA and hydrogen peroxide), osmolyte accumulation (proline), and components of the non-enzymatic antioxidant defense system (total phenolic compounds and antioxidant capacity) were analyzed.

The results demonstrated that herbicide application caused a 1.3-fold increase in ethylene content, indicating the activation of stress-response mechanisms even when the herbicide was applied at the manufacturer’s recommended rate. The combined application of the herbicide and the microbial biostimulant reduced ethylene levels by 35,74% compared with herbicide-treated plants. Herbicide treatment also resulted in a statistically significant decrease in chlorophylls and carotenoid content, whereas the addition of the microbial biostimulant partially mitigated these negative effects.

A substantial 2-fold increase in the oxidative stress markers MDA and hydrogen peroxide was observed following herbicide application, with MDA levels increasing 2.04-fold and hydrogen peroxide levels rising 2.07-fold relative to the control. In contrast, plants receiving the combined treatment exhibited only a 60% increase in MDA content, while hydrogen peroxide concentrations remained elevated compared with those of the control plants. Proline accumulation was also enhanced under herbicide stress; however, lower proline levels were detected in common wheat plants treated with both the herbicide and “ProbioHumus”.

The analysis of total phenolic compounds and antioxidant capacity indicated that the non-enzymatic antioxidant defense system was activated by both herbicide and microbial biostimulant treatment. Overall, the findings suggest that the microbial biostimulant “ProbioHumus” can alleviate herbicide-induced damage, maintain physiological stability, and enhance stress tolerance in common wheat. These results highlight its potential contribution to sustainable agricultural practices and support efforts toward reducing reliance on chemical herbicides.

Keywords: Common wheat; herbicide stress; microbial biostimulant; oxidative stress; antioxidant defense system.

Acknowledgements: This work was supported by the Bulgaria-Lithuania joint research project (ID: IC-LT/02/2025–2027).

OP-03

RELATIONSHIP BETWEEN DROUGHT TOLERANCE AND FUNGAL PATHOGEN RESISTANCE IN CROP AND RESURRECTION PLANTS

Gergana Mihailova¹, Katya Georgieva¹, Eva Popova¹, Vasilissa Manova¹, Zornitsa Stoyanova¹, Stiliana Simeonova¹, Francesca Rapparini², Alessio Tei², Fabio Osti²

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. G. Bonchev Str., Bl. 21, 1113 Sofia, Bulgaria

²Institute of BioEconomy, National Research Council of Italy (CNR-IBE), Via P. Gobetti 101, 40129 Bologna, Italy

Presenting author: mihailova.gergana.k@gmail.com; gmihailova@bio21.bas.bg

Drought and plant pathogenic fungi cause substantial yield losses in many economically important crops. Although plant responses to individual biotic and abiotic stresses have been widely studied, the mechanisms underlying exposure to both stress types simultaneously remain poorly understood. Tomato (*Solanum lycopersicum* L.) is a crop plant with significant importance in the human diet, and its production could be affected by pathogens. Resurrection plants are a unique plant group that can survive extreme dehydration of their vegetative tissues and completely recover upon rehydration. While their tolerance to drought is well studied, natural fungal infections in these species have not yet been reported.

This study aims to reveal how the resurrection plant *Haberlea rhodopensis* and the crop plant *Solanum lycopersicum* cope with both abiotic (drought) and biotic (pathogenic fungi) stress simultaneously and whether there is a cross-tolerance between drought resistance and resistance to biotic stress. The specific objectives are 1) to identify differences in fungal susceptibility between resurrection plants and crop plants; 2) to reveal how the dehydration and rehydration influence plant susceptibility to phytopathogens; 3) to characterize any inherent antifungal defenses or vulnerabilities in *H. rhodopensis* and *S. lycopersicum* through investigations at phenotypic, biophysical, biochemical, and molecular levels. The susceptibility to pathogenic fungi of *H. rhodopensis* and *S. lycopersicum* will be compared under optimal conditions (fully hydrated state), during moderate drought stress, as well as after plant rehydration. The functional state of the photosynthetic apparatus will be assessed by measuring the stomatal conductance, quantum efficiency of photosystem I (PSI) and PSII and the alterations in the content of key photosynthetic proteins. The role of soluble sugars, stress-induced proteins, and antioxidant activity during the simultaneous action of drought and pathogenic fungi will be examined. The transcriptional activity of stress-related genes, such as those involved in the antioxidant defense response, photosynthesis, plant immunity, genome integrity maintenance and DNA damage signaling will be analyzed. Additionally, the hormonal status and temporal shifts in the type and quantity of emitted biogenic volatile organic compounds in response to drought and fungal pathogens will be investigated. The comparison between resurrection and crop plant could provide valuable information for improving breeding of crop plants, resistant to the simultaneous action of multiple stresses.

Keywords: *Solanum lycopersicum* L.; *Haberlea rhodopensis*; fungal infection; drought stress; cross resistance.

Acknowledgements: The project was funded by the Bulgarian National Science Fund, grant number КП-06-H96/3.

OP-04**HOW *ERUCA* PLANTS RESPOND TO MODERATE UV-B RADIATION?****Gabriella Szalai, Magda Pál, Mihály Dernovics, Kamirán Á. Hamow, Kinga Benczúr, Tibor Janda**

Department of Plant Physiology and Metabolomics, Agricultural Institute, Centre for Agricultural Research, HUN-REN, Martonvásár, Hungary

Presenting author: szalai.gabriella@atk.hun-ren.hu

Rocket, or arugula plant, is widely distributed all over the world. Its leaves contain a lot of secondary metabolites with antioxidant activity [1], and are freshly consumed. Increasing the content of these compounds by different approaches can improve the quality of this salad plant.

The aim of the present study was to investigate how moderate UV-B radiation and salt priming can influence the secondary metabolism in *Eruca* leaves. Seeds were soaked overnight in distilled water (control) or in 0.5 mM NaCl then grown in pots for 60 days under controlled conditions in a plant growth chamber (20/18°C; 8/16 h light/dark period; PPFD: 250 $\mu\text{mol m}^{-2} \text{s}^{-1}$). At the end of this period half of the plants were exposed to moderate UV-B radiation (430 $\mu\text{W cm}^{-2}$). After seven days of UV-B treatment, physiological parameters were measured and samples were collected for biochemical analyses.

For detection of the severity of the stress, physiological parameters were measured. Neither treatment with UV-B nor NaCl priming caused significant effect on the fresh weight of the plants and the chlorophyll *a* fluorescence induction parameters, while a slight, but statistically significant increase was detected in MDA and NO levels in UV-B treated plants. Metabolomic changes were also found after exposure the plants to the UV-B. Targeted and untargeted metabolomics analyses carried out using UPLC-MS/MS and UPLC-IMS/QTOF instruments. The changes in the activity of the main enzymes playing role in secondary metabolite synthesis routes showed that salicylic acid and flavonol biosynthesis of phenylpropanoid pathway were also influenced by the used moderate UV-B treatment.

Keywords: *Eruca*; UV-B stress; metabolomic analysis.

Acknowledgements: This research was funded by project No. TKP2021-NKTA-06 funded by the Ministry of Innovation and Technology from the National Research Development and Innovation Fund, in the frame of the Thematic Excellence Program 2021.

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OP-05

BOOSTING WHEAT TOLERANCE: HOW PRIMING WITH SPERMINE REGULATES THE ANTIOXIDANT SYSTEM DURING BIOTIC STRESS CAUSED BY *FUSARIUM CULMORUM*

Tsvetina Nikolova¹, Elena Shopova¹, Zornitsa Katerova¹, Liliana Brankova¹, Iskren Sergiev¹, Ivo Yanashkov², Dessislava Todorova¹

¹Institute of Plant Physiology and Genetics - Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., Bldg. 21, 1113 Sofia, Bulgaria

²Institute of Soil Science, Agrotechnologies and Plant Protection “Nikola Poushkarov”, Shose Bankya 7, 1331 Sofia, Bulgaria

Presenting author: tnikolova00@bio21.bas.bg

The protective effects of priming of wheat seeds with 5 mM polyamine spermine (Spm) on the plant's transcriptional and biochemical responses during *Fusarium culmorum* infection were examined. Two pathogen-infection approaches were used to investigate the priming's effects in 30-day-old and 8-week-old plants. The pathogen caused severe cellular damage and oxidative stress in mature tissues, through seed inoculation, which were manifested by significant increases in electrolyte leakage, malondialdehyde, and hydrogen peroxide. The infection also caused metabolic alterations, like increased reducing sugars, free proline, and total osmolytes. Simultaneously, the fungus caused significant alterations in important antioxidant enzymes (catalase, peroxidase, superoxide dismutase and glutathione reductase) expressed in biochemical and transcriptional level. However, this pathogen-induced damage was considerably reduced by the 5 mM Spm seed priming. Priming successfully decreased the over-accumulation of reactive oxygen species and most oxidative stress indicators. Additionally, Spm mitigated the pathogen-mediated downregulation of corresponding defence genes at the molecular and enzymatic levels. Overall, our results show that 5 mM spermine priming improves wheat tolerance to *F. culmorum* by enhancing antioxidant efficiency and preserving metabolic equilibrium. Furthermore, spermine's priming activity is plant age-dependent, as older plants show a stronger response to the priming.

Keywords: *Fusarium*; biotic stress; wheat; enzymatic activity; molecular changes.

Acknowledgements: This research is supported by the Bulgarian National Science Fund under contract KP-06-H86/6, dated 06 December, 2024.



OP-06

EFFECTS OF PLANT GROWTH REGULATORS IN TWO WHEAT SPECIES: FROM ABIOTIC STRESS RESPONSES TO FIELD PERFORMANCE

**Vasilissa Manova¹, Yana Koycheva¹, Radoslav Chipilski², Zoia Tsekova¹, Iskren Sergiev¹,
Konstantina Kocheva¹, Elisaveta Kirova¹, Irina Moskova¹**

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Sofia 1113, Bulgaria

²Institute of Plant Genetic Resources, Agricultural Academy, Sadovo 4122, Bulgaria

Presenting author: manovavasilissa@gmail.com

Einkorn (*Triticum monococcum* L.) is an ancient wheat species that has recently attracted the interest of farmers and the food industry because of its rich nutritional profile and inherent tolerance to adverse conditions, which differ from those of bread wheat (*Triticum aestivum* L.). However, the mechanisms underlying the stress response of einkorn remain less understood. Abiotic stress induces the formation of reactive oxygen species (ROS), which at low levels activate cellular defense mechanisms, whereas their excessive accumulation causes oxidative damage to essential cellular components, thus affecting the growth and productivity of crops. Utilization of eco-friendly bioactive compounds that stimulate plant protective mechanisms may help enhance stress adaptation and tolerance in wheat. The aim of the present study was to investigate whether foliar application of the plant growth regulators 4PU-30 (a phenylurea-type cytokinin) and MEIA (β -monomethyl ester of itaconic acid) could improve the response of einkorn and bread wheat to environmental stress factors such as drought, UV-B radiation and salinity. Following spraying with 4PU-30 or MEIA, seedlings were exposed to the respective abiotic stressor, and physiological, biochemical and molecular analyses were performed 72 h later. To assess their response to the applied treatments (regulators, stresses), phenotypic parameters (fresh and dry weight, root and leaf length), stress markers (hydrogen peroxide, malondialdehyde and proline content, electrolyte leakage), pigments (chlorophylls and carotenoids), non-enzymatic antioxidants (total phenols, flavonoids, anthocyanins and free thiol groups), and the activities of ROS-related enzymes such as peroxidase (POX), catalase (CAT), superoxide dismutase (SOD) and glutathione S-transferase (GST) were determined. It was found that the application of 4PU-30 and MEIA positively influenced the response of both wheat species to the tested abiotic factors, mainly associated with a reduction in oxidative stress-induced damage. To further characterize the transcriptional regulation of the antioxidant defense system, the expression levels of genes encoding antioxidant enzymes were also examined. Specific expression patterns were found for some of the studied genes depending on the type of stress factor and on the applied regulator; in general, however, the most pronounced transcriptional changes were observed in genes belonging to the POX and GST gene families. Field experiments further showed that application of 4PU-30 and MEIA had beneficial effects on some morphometric and physiological parameters of plants grown under natural conditions. Additionally, improved germination was observed in seeds harvested from regulator-treated plants. Overall, our results suggest that the applied plant growth regulators may help increase the tolerance of einkorn and bread wheat to oxidative stress, thus providing a basis for their potential application in agricultural practice to protect the studied cereal species from major environmental stressors.

Keywords: Einkorn; bread wheat; oxidative stress; antioxidant defense; plant growth regulators.

Acknowledgements: This research was supported by Bulgarian National Science Fund (BNSF), Grant No KP-06-N56/15.

OP-07**HERACLEUM SOSNOWSKYI CONTROL APPROACHES BASED ON GIBBERELLIN METABOLISM MODULATION**

Tautvydas Žalnierius, Dominykas Laibakojis, Sigita Jurkonienė, Saulė Rapalytė

State Scientific Research Institute Nature Research Centre, Akademijos str. 2, Vilnius, Lithuania

Presenting author: tautvydas.zalnierius@gamtc.lt

The spread of alien species and their destructive impact on native ecosystems is one of the most pressing global issues today. Sosnowsky's hogweed (*Heracleum sosnowskyi* Manden.) is registered in the EU as an invasive species and is known for its environmental and human health hazards. Various measures, including herbicides, are used to control the spread of Sosnowsky's hogweed, but their toxicity and efficiency limit their use. Sosnowsky's hogweed produces seeds once in its lifetime and then dies. Therefore, by preventing seed germination, inducing seedlessness, and embryo abortion through gibberellin metabolism modulation, its spread could be controlled. In this study, we involved the exogenous phytohormones gibberellins and auxins to control seed formation and development in Sosnowsky's hogweed. Results revealed that exogenous GA₃ and other bioactive substances affect fruit development in the central and lateral parts of the terminal and satellite umbels in relation to changes in the hormonal system. The research found that GA₃ can contribute to the development of environmentally friendly measures for controlling the spread of invasive monocarpic hogweed species due to its ability to reduce seed germination, induce seedlessness, embryo abortion, and reduce seed size. These results provided a basis for an environmentally friendly development of RNA-interference technology to silence GA catabolism genes to halt the spread of Sosnowsky's hogweed and mitigate the negative impact of this invasive plant. These results provide a fundamental basis for developing RNA interference-based technology to suppress the spread of Sosnowsky's hogweed in a more environmentally friendly way.

Keywords: Gibberellin; Indole-3-acetic acid; RNA interference; seedlessness, invasive plant.

Acknowledgements: This research was funded by the Research Council of Lithuania (LMTLT) (Project No. S-MIP-25-21).



OP-08

RESTRICTED CAPACITY FOR STATE TRANSITIONS IN THE ANTARCTIC ECOTYPE OF *COLOBANTHUS QUITENSIS* IMPLIES DIFFERENTIAL STRATEGIES TO COLD ACCLIMATION OF ANTARCTIC AND ANDES ECOTYPES

León A. Bravo¹, Beth Szyszka-Mroz², Dimitre A. Ivanov², Alexander G. Ivanov^{2,3}, Norman P.A. Hüner²

¹Instituto de Agroindustria, Departamento de Ciencias Agronómicas y Recursos Naturales, Universidad de La Frontera, Temuco, Chile

²Department of Biology and The Biotron, University of Western Ontario, London, Ontario, N6A 5B7 Canada

³Institute of Biophysics and Biomedical Engineering, Bulgarian Academy of Sciences, Acad. G. Bonchev Str., bl. 21, Sofia 1113, Bulgaria

Presenting author: aivanov@bio21.bas.bg; aivanov@uwo.ca

Excitation energy partitioning measurements indicated that the fraction of absorbed irradiance utilized *via* Photosystem II (PSII) photochemistry (Φ_{PSII}) in cold acclimated (CA) plants of Antarctic ecotype is 1.6-fold higher compared to the Andes ecotype of *Colobanthus quitensis* (Kunth) Bartl. This was accompanied by a higher relative abundance of PSII reaction center protein D1(PsbA). Cold acclimation of both ecotypes resulted in an almost identical increase of Photosystem I (PSI) photochemistry measured via far-red light-induced P700 oxidation ($P700^+$). However, the effective absorption section of PSI in the Antarctic ecotype was 2-fold higher than the Andes ecotype. This corresponds well with the observed higher abundance of Lhca1 in the Antarctic compared to the Andes plants. BN-PAGE profiles of the major chlorophyll-protein complexes also confirmed higher abundance of PSI-related bands in the Antarctic plants. Interestingly, the capacity for State I-State II transitions (Fr) in non-acclimated (NA) and CA Antarctic plants was 55% and 61% lower than the corresponding values in Andes ecotype. In addition, the kinetics of State I-State II transitions were much slower in Antarctic ($t_{1/2} = 12.35 \pm 0.13$ s) compared to Andes ($t_{1/2} = 6.44 \pm 0.02$ s) plants. The restricted capacity for state transitions corresponded with the lower phosphorylation status of Lhcb proteins in cold acclimated Antarctic than Andes plants. The ecotype differences are discussed in relation to the different climatic conditions of the two *Colobanthus* ecotypes.

Keywords: Chlorophyll-protein complexes; cold acclimation; PSII photochemistry; P700; PSI absorption cross section; state transitions.

Acknowledgements: This work was supported by the Natural Sciences and Engineering Research Council of Canada (NPAH) and the National Fund for Scientific and Technological Development of Chile, “Fondecyt 1201824” (LAB).



OP-09

NETWORKING AND OPPORTUNITIES FOR SUSTAINABLE CROP EPITRANSCRIPTOMICS

Bekim Gashi^{1,4}, Valbona Sota^{2,4}, Jesus Pascual Vazquez^{3,4}

¹Laboratory of Plant Physiology, Department of Biology, Faculty of Mathematics and Natural Sciences, University of Prishtina, Eqerem Cabej str, No 51, 10000 Prishtina, Kosovo

²Department of Biotechnology, Faculty of Natural Sciences, University of Tirana, Bulevardi Zogu I, Tiranë 1001, Albania

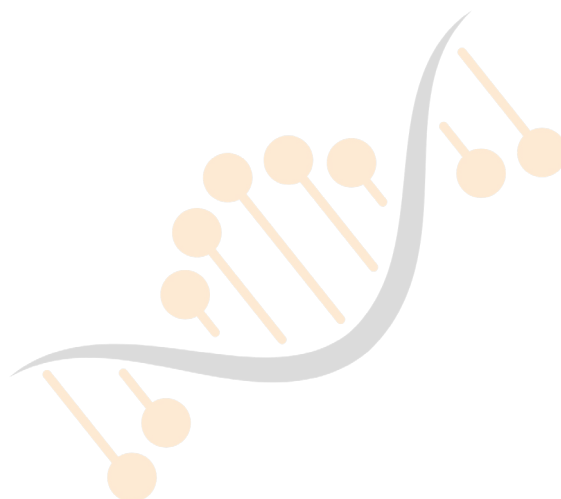
³Department of Organisms and Systems Biology, University of Oviedo, Catedrático Rodrigo Uría s/n, 33071 Oviedo, Asturias, Spain

⁴EPICROPS COST Action CA24152, European Cooperation in Science and Technology (COST), Europe
Presenting author: bekim.gashi@uni-pr.edu

Nowadays, as climate change increasingly affects agricultural systems, international scientific cooperation has become essential for the development of more effective and sustainable solutions. The EPICROPS COST Action (CA24152) addresses this problem by promoting research on epitranscriptomic regulation and non-coding RNAs in crop species, supporting sustainable and climate-resilient agriculture. Participation in EPICROPS gives researchers at all stages of their careers, especially Early-Career Researchers (ECRs), the chance to engage in innovative research, expand their professional networks, and gain hands-on experience in international institutions across participating countries. The Action supports Short-Term Scientific Missions (STSMs), workshops, training schools, and conferences, which promotes knowledge sharing, skill development and teamwork across different fields. ECRs receive focused mentoring, organized networking, and access to advanced experimental and computational methods, which improves their professional growth and helps them connect with the global scientific community. By encouraging interdisciplinary collaboration and outreach activities, EPICROPS builds scientific connections and ensures that all participants can actively work towards the Action's goals. Access to collaborative platforms, open-access data, and dissemination efforts allows researchers, especially ECRs, to increase the scientific and social impact of their work and build expertise in crop science, epitranscriptomics, and sustainable farming practices.

Keywords: EPICROPS; resilience; epitranscriptomics; non-coding RNA; crop resilience; climate change.

Acknowledgements: This publication is based upon work from COST Action: Epitranscriptomics and ncRNAs for climate-change-resilient and sustainable crops, EPICROPS, CA24152, supported by COST (European Cooperation in Science and Technology).



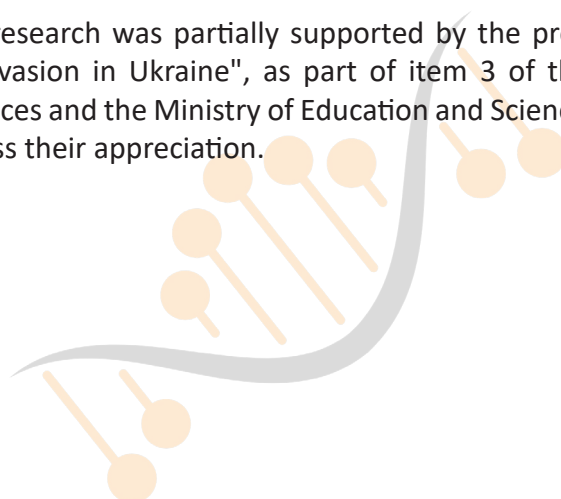
OP-10

THERMAL ADAPTATION AND SEASONAL VARIATION IN ENDOREDUPPLICATION AND POLYTENY IN THE FRUIT FLY *DROSOPHILA MELANOGASTER* (DIPTERA: DROSOPHILIDAE)**Volodymyr Strashnyuk^{1,2}, Yelyzaveta Koptevtsova¹, Evgenij Vakulenko¹**¹Department of Genetics and Cytology, V.N. Karazin Kharkiv National University, Svobody sq., 4, Kharkiv, 61022, Ukraine²Laboratory "Regulation of Gene Expression", Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. G. Bonchev str., B. 21, Sofia, 1113, BulgariaPresenting author: volodymyr.strashnyuk@gmail.com

The fruit fly *Drosophila melanogaster* (Meigen, 1830) in temperate regions is subject to seasonal changes in natural conditions. The limiting factors are environmental temperature and resource availability. Insects demonstrate a wide range of adaptive responses to seasonal changes. In this study, we focused on endoreplication, which plays an important role in fruit fly development. Endoreplication leads to the formation of polytene chromosomes, which are a form of endopolyploidy. This phenomenon underlies postmitotic growth caused by cell expansion. The material for the study was laboratory strains, as well as fruit fly lines obtained from a natural population on the territory of the biological station of the V.N. Karazin Kharkiv National University near the village of Haidary, Kharkiv region in the spring and autumn of 2019 and 2020. Cytomorphometry was used to study polyteny. Laboratory studies have shown a significant increase in polyteny in the salivary glands of fruit fly larvae with decreasing temperature, which is achieved by extending the duration of preimaginal development under such conditions. In the temperature range of 18–28 °C, the difference in ploidy levels was 12.9–39.4%, depending on the genotype. The increase in cell ploidy at low temperatures correlated with an increase in fly size and their stress resistance. Population studies have shown that ploidy patterns in *Drosophila* salivary gland cells undergo seasonal changes. These variations correlate to a certain extent with changes in the direction of natural selection, which undergoes seasonal fluctuations. Endoreplication and polyteny indices reach their highest values after wintering, during which the population collapses and flies undergo selection for stress resistance and viability. During the growing season, the endoreplication index and cell ploidy decrease. We believe that this contributes to population growth by resulting in smaller flies with accelerated development and faster generational change. The seasonal component in the total variation in chromosome polyteny was 21.9%. No sex differences were found for this trait.

Keywords: *Drosophila*; laboratory strains; natural population; developmental polyploidy; endoreplication; fluctuating selection.

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OP-11

INTEGRATION OF ETHYLENE SIGNALING IN DROUGHT STRESS RESPONSES

Irina I. Vaseva¹, Simona Galabova¹, Dominique Van Der Straeten²

¹Institute of Plant Physiology and Genetics-Bulgarian Academy of Sciences, Sofia, Bulgaria

²Laboratory of Functional Plant Biology, Ghent University, Ghent, Belgium

Presenting author: vaseva@bio21.bas.bg

Ethylene is a gaseous plant hormone that controls responses to different environmental and developmental cues. It inhibits vegetative growth through the restriction of cell elongation and promotes fruit ripening and leaf senescence. In *Arabidopsis thaliana*, ethylene is perceived by a family of transmembrane receptors: ETHYLENE RESPONSE 1 and 2 (ETR1, ETR2), ETHYLENE RESPONSE SENSOR 1 and 2 (ERS1, ERS2), and ETHYLENE INSENSITIVE 4 (EIN4), residing in the endoplasmic reticulum. The ethylene signalling cascade is suppressed when the hormone is absent via CONSTITUTIVE TRIPLE RESPONSE 1 (CTR1; a Raf kinase), which inhibits the membrane protein EIN2 by direct phosphorylation. Ethylene binding to the receptors inactivates CTR1, releasing the repression of EIN2. The C-end of EIN2 is cleaved off and moves to the nucleus, where it activates the master ethylene transcription factors EIN3 and EIN3-LIKE 1 (EIL1). EIN3 is constantly degraded in the absence of ethylene, a process that is under the control of two F-box proteins: EIN3 BINDING F-BOX PROTEINS 1 and 2 (EBF1 and EBF2).

Based on this canonical ethylene signalling pathway, we employ a model to study cell type-specific involvement of ethylene in stress responses. It is based on targeted expression of the F-box protein EBF2 that provokes ethylene insensitivity in specific root tissues. *Arabidopsis thaliana* (Col-0) wild type plants were transformed with constructs carrying the coding *EBF2* sequence fused to different cell-type-specific promoters. The success of the targeted EBF2 expression has been confirmed by genetic crosses with the highly sensitive ethylene reporter *pEIN3:EIN3-(3x)GFP (ein3)* and through immunolocalisation of enhanced EBF2 protein levels in particular cell types.

The integration of ethylene signalling in drought protective mechanisms has been evaluated through comparative analyses of the constructed transgenic lines with the ethylene mutants *ein2-1*, *ein3eil1*, *ctr1-1* and the wild type Col-0 plants subjected to water limitation. The experimental data outline the root epidermis and pericycle as central ethylene signalling sites involved in the adaptive response to dehydration. These observations were associated with the accumulation of specific dehydrin types and improved capacity to mitigate oxidative damage.

Keywords: *Arabidopsis thaliana*; cell type-specific hormonal signalling; drought; ethylene; EIN3 BINDING F-BOX PROTEIN 2.

Acknowledgements: This research was funded by the Bulgarian National Science Fund (BNSF); grant number KP-06-N71/12/10 July 2024.



OP-12

ANTICANCER POTENTIAL OF *LEVISTICUM OFFICINALE* IN *IN VITRO* CELL CULTURE MODEL SYSTEMS

Georgi Antov¹, Zlatina Gospodinova¹, Gergana Mihailova¹, Natalia Krasteva², Trayana Kamenska², Svetozar Stoichev², Danail Pavlov³, Antoaneta Georgieva⁴, Stefka Valcheva-Kuzmanova⁴

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Sofia, Bulgaria;

²Institute of Biophysics and Biomedical Engineering, Bulgarian Academy of Sciences, Sofia, Bulgaria;

³Department of Biochemistry, Molecular Medicine and Nutrigenomics, Faculty of Pharmacy, Medical University “Prof. Dr. Paraskev Stoyanov”, Varna, Bulgaria;

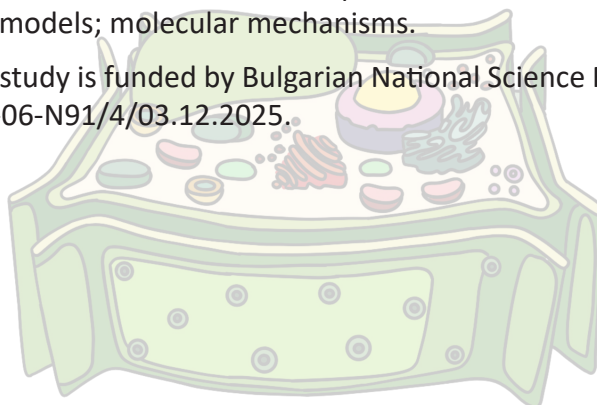
⁴Department of Pharmacology and Clinical Pharmacology and Therapeutics, Faculty of Medicine, Medical University “Prof. Dr. Paraskev Stoyanov”, Varna, Bulgaria.

Presenting author: antov8107@abv.bg

Despite significant advances in cancer diagnosis and treatment, the need for highly selective, low-toxic, and plant-based anticancer agents still remains an urgent priority. *Levisticum officinale* W.D.J.Koch (Apiaceae), a perennial plant with a long ethnopharmacological history, is a rich source of bioactive compounds and secondary metabolites. Although some isolated compounds and extracts show promising properties, to date there is no integrative study that has evaluated the anticancer potential of the total aqueous-ethanolic extract of *L. officinale* across a broad panel of cancer cell lines using both conventional two-dimensional (2D) and physiologically relevant three-dimensional (3D) culture models. For this reason, the present project aims to rigorously characterize the *in vitro* anticancer efficacy and selectivity of the total aqueous-ethanolic extract of *L. officinale* and to elucidate its molecular mechanisms of action in 2D and 3D tumor model systems. The anticancer activity will be investigated against a wide range of human cancer cell lines, with selectivity determined against non-tumorigenic cell lines. Cell viability and proliferation rates, as well as membrane integrity will be assessed via WST-1, Calcein-AM and Trypan blue assays. Morphological changes, induction of apoptosis/autophagy and cell cycle perturbations will be analyzed by phase-contrast and fluorescence microscopy, as well as by flow cytometry. Clonogenic survival and migratory capacity will assess the replicative and metastatic potential. The molecular mechanisms underlying the observed effects will be elucidated by qRT-PCR and protein immunoblotting. All data will be subjected to statistical and bioinformatic analysis. The successful implementation of the project is expected to provide: I) reliable and quantitative data on the anticancer potential and selectivity of the extract against a diverse panel of cancer cell lines, confirmed by a favorable selectivity profile in 2D and supported by efficacy in 3D models; II) elucidation of specific molecular mechanisms focused on the induction of apoptosis combined with suppression of survival and migration signaling. The results of this study will provide a compelling, mechanism-based preclinical rationale for the therapeutic potential of *L. officinale*, providing a valid experimental basis for further *in vivo* evaluations.

Keywords: *Levisticum officinale*; *in vitro* anticancer potential; two-dimensional cell culture; three-dimensional multicellular models; molecular mechanisms.

Acknowledgements: This study is funded by Bulgarian National Science Fund, Ministry of Education and Science, Grand № KP-06-N91/4/03.12.2025.



OP-13

**CELLULAR RESPONSE IN RAPESEED TO EXOGENOUS AMINO ACIDS
UNDER DROUGHT**

**Liudmyla Kozeko^{1,2}, Dmytro Klimchuk¹, Yuri Akimov¹, Yulia Ovcharenko¹, Virgilija Gavelienė²,
Tautvydas Žalnierius², Sigita Jurkonienė²**

¹M.G. Kholodny Institute of Botany, NAS of Ukraine, Tereshchenkivska str. 2, 01601, Kyiv, Ukraine

²Nature Research Centre, Akademijos str. 2, 08412, Vilnius, Lithuania

Presenting author: liudmyla.kozeko@gmail.com

Plant treatment with biostimulants is an environmentally friendly approach to increase stress resistance. Our preliminary studies demonstrated that foliar treatment with amino acids proline and γ -aminobutyric acid (GABA) enhanced tolerance of plants to drought (Jurkonienė et al., 2023; Kozeko et al., 2025). Both amino acids accumulate in plant cells under stressful conditions and play different roles in cellular defence. GABA is a signal molecule in stomata regulation.

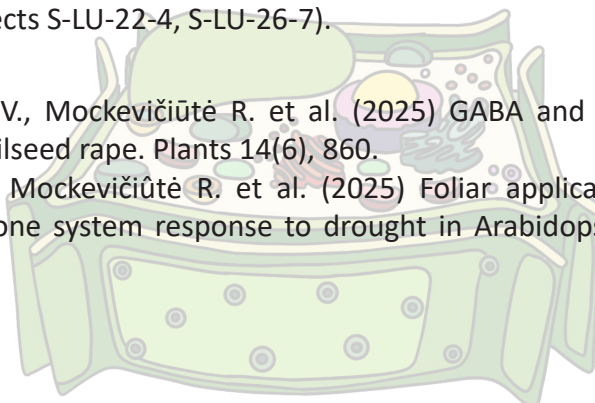
Here, we investigated effects of foliar application of proline and GABA on stomata behaviour and leaf ultrastructure in rapeseed (*Brassica napus* L.) under normal and drought conditions. Two-week-old plants grown under normal conditions were sprayed with 0.1 mM proline, 0.1 mM GABA or their combination, and then half of the plants in each treatment group were subjected to gradual soil drying for 7 days. Control plants were well-watered and sprayed with dH_2O . The upper mature leaves were used for analysis. The micromorphology of the epidermal surface of leaves was studied using scanning electron microscopy (JFC-1100, Jeol, Japan). The epidermal surface of the leaves had cuticular wax, the amount of which increased during drought, especially on the adaxial surface. All variants of amino acid treatments were found to reduce stomatal aperture on both the adaxial and abaxial leaf surfaces under drought, which may reduce stomatal conductance. Remarkably, under normal conditions, all amino acid treatments increased orifice opening on both surfaces, except GABA, which decreased it on the abaxial side. This indicates that regulation of stomata opening by GABA depends on leaf surfaces. Transmission electron microscopy (JSM-1230, Jeol, Japan) revealed changes in the ultrastructure of palisade parenchyma cells under drought after application of amino acids. The most significant changes were induced by GABA treatment under drought – a significant reduction in the size of starch granules, suggesting that exogenous GABA affects starch and sucrose metabolism under water deficit. Thus, the increase in drought tolerance of plants under the influence of these biostimulants may be due to the reduction of stomatal aperture and specific structural changes in leaf cells, mainly in chloroplasts.

Keywords: Proline; γ -aminobutyric acid; *Brassica napus*; drought; stomatal aperture; cellular structure.

Acknowledgements: The research was funded by the Joint Ukrainian-Lithuanian R&D Program of the Ministry of Education and Science of Ukraine (projects M/49-2023, M/85-2026) and the Research Council of Lithuania (projects S-LU-22-4, S-LU-26-7).

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OP-14**NITRIC OXIDE-ASSOCIATED REGULATION OF PLASTIDIAL IRON HOMEOSTASIS****Michel Francis^{1,2}, Maria Gracheva^{2,3}, Helga Zelenyánszki², Katalin Solymosi⁴, Ádám Solti²**¹Doctoral School of Biology, Institute of Biology, ELTE Eötvös Loránd University, Budapest, Hungary²Department of Plant Physiology and Molecular Plant Biology, ELTE Eötvös Loránd University, Budapest, Hungary³Energy Research Center, HUN-REN, Budapest, Hungary⁴Department of Plant Anatomy, ELTE Eötvös Loránd University, Budapest, HungaryPresenting author: michelfrancis97@gmail.com

Nitric oxide (NO) is a gaseous signalling molecule. In plants, NO is involved in the regulation of development processes, senescence, and redox balance, including post-translational protein modifications. Although NO has been proven by multiple studies to affect the Fe homeostasis of plants especially by the regulation of the root Fe uptake processes as an upstream regulator, understanding of how NO-associated processes affect plastidial Fe allocation remains poor. Thus, this research aimed to investigate how the NO signal affects the intracellular, particularly the plastidial Fe allocation. We applied NO metabolism affected *Arabidopsis thaliana* lines. Using high energy (K-edge) micro-X-ray Fluorescence (μ XRF) under cryogenic conditions, we mapped the transition metal distribution in Arabidopsis leaves to discover differences in Fe loci across the applied lines. The results revealed novel insights into the role of NO signals in plastidial Fe accumulation and recycling, providing a basis for future biofortification strategies. To complement these observations at the protein level, we developed a predictive computational pipeline to assess potential nitric oxide-related post-translational modifications, involving cysteine S-nitrosylation and tyrosine nitration, of Ferric Reduction Oxidase 7 (FRO7), which mediates the reduction of ferric chelates to ferrous at the chloroplast inner envelope membrane, thereby facilitating Fe uptake into chloroplasts. As S-nitrosoglutathione (GSNO) functions as the principal cellular NO reservoir and a major physiological regulator of plant iron homeostasis, understanding its specific impact on FRO7 is critical. Using molecular dynamics simulations of a membrane-embedded FRO7 model combined with structure-based scoring, we evaluated residue accessibility, local chemical environment, and reaction geometry. This approach allowed us to identify and prioritize specific cysteine residues with a high likelihood of S-nitrosylation by GSNO and tyrosine residues with a high likelihood of nitration by reactive nitrogen species such as peroxynitrite. The application of NO donor SNAP on isolated chloroplast inner envelope vesicles, containing FRO7 verified the hypothesis that NO signal affects the activity of FRO7. This structural framework clarifies the molecular basis of GSNO-dependent regulation of plastidial iron influx, providing a clear direction for future experimental validation. Together, high-resolution elemental mapping and targeted computational predictions highlight multi-level processes linking signalling, nutrient allocation, and protein regulation.

Keywords: Iron homeostasis; chloroplast; nitric oxide signaling; elemental imaging; protein post-translational modification; structure-based prediction.

Acknowledgements: This work was supported by the grant ADVANCED-25 152773 of NKFIH, Hungary. Á.S. was supported by the János Bolyai Scholarship of the Hungarian Academy of Sciences (BO-00113-23-8). M.F. was supported by the Stipendium Hungaricum Scholarship Programme of Tempus Public Foundation. We acknowledge the European Synchrotron Radiation Facility (ESRF) for provision of synchrotron radiation facilities under proposal number LS-3039.

OP-15

INSIGHTS INTO THE MODE OF ACTION OF NOVEL NAPHTHALIMIDE DERIVATIVES AS SELECTIVE ANTICANCER AGENTS

Zlatina Vlahova¹, Lazar Lazarov¹, Maria Petrova¹, Shazie Yusein-Myashkova¹, Jordana Todorova¹, Maria Schröder¹, Monika Mutovska², Stanimir Stoyanov², Yulian Zagranyski², Iva Ugrinova¹

¹Institute of Molecular Biology “Akad. Roumen Tsanev”, Bulgarian Academy of Sciences, Acad. G. Bonchev Str., bl. 21, 1113 Sofia, Bulgaria

²Faculty of Chemistry and Pharmacy, Sofia University “St. Kliment Ohridski”, 1 J. Bourchier Blvd, 1164 Sofia, Bulgaria

Presenting author: shazi@abv.bg

DNA-targeting small molecules that induce replication stress represent a promising strategy in anticancer drug development. Heterocyclic annulation of 1,8-naphthalimide (NI) offers a rational approach to enhance potency and tumor selectivity. This study reports the synthesis and biological characterization of novel benzofuran-containing and 6-nitro-benzodioxin-containing NI derivatives, focusing on lead compounds 4',6-dinitro-benzofuran-NI (5d) and 6-nitro-benzodioxin-NI (5a).

Both compounds exhibited potent nanomolar cytotoxicity in lung carcinoma models with favorable selectivity over non-malignant MRC-5 cells. Clonogenic assays confirmed that transient exposure led to a sustained, irreversible loss of proliferative capacity. Mechanistically, compounds 5d and 5a caused a marked reduction in EdU (5-ethynyl-2'-deoxyuridine) incorporation during S phase, indicating impaired DNA synthesis and reduced cellular proliferation. They also induced robust γ H2AX (phosphorylated histone H2AX) formation, a sensitive marker of DNA double-strand breaks, confirming the induction of replication stress and DNA damage. Treatment induced cell cycle perturbations (G1 arrest in A549; G2/M accumulation in H1299) and triggered apoptotic pathways. For 5d, p53 nuclear accumulation and autophagy-related stress responses were also confirmed.

Compounds 5d and 5a act as potent inducers of replication stress, triggering DNA damage, checkpoint activation, and apoptosis. These findings establish benzofuran- and benzodioxin-annulated naphthalimides as promising lead structures for the development of anticancer therapies targeting replication vulnerabilities.

Keywords: Naphthalimide; benzofuran; benzodioxin; replication stress; DNA damage; apoptosis.

Acknowledgements: This research was funded by the Bulgarian National Science Fund, grant number KP-06-H61/1.



OP-16**INTEGRATED METABOLOMIC AND DUAL-SEQ ANALYSIS OF NORWAY SPRUCE–
FUNGAL PATHOGEN INTERACTIONS****Amit Roy, Sellamuthu Gothandapani, Amrita Chakraborty**

Faculty of Forestry and Wood Sciences, Czech University of Life Sciences, Prague, Czech Republic.

Presenting author: roy@fld.czu.cz

Forest fungal pathogens are a major threat to tree vitality, contributing to mortality, progressive host decline, and deterioration of wood quality. In coniferous forests, many of these pathogens are dispersed by insect vectors, particularly bark beetles, which can facilitate fungal transmission during colonization of host trees. This study investigated fungal pathogens associated with *Picea abies* and isolated from the gut of the Eurasian spruce bark beetle, *Ips typographus*. The objectives were to characterize these fungi, evaluate their pathogenic potential, and analyze host-pathogen interactions through integrated multi-omics approaches.

Pathogenicity tests conducted on four-year-old *P. abies* seedlings demonstrated pronounced lesion development and vascular discoloration following fungal inoculation. Transmission electron microscopy further confirmed the presence of fungal hyphae within colonized host tissues. Metabolomic analyses showed an elevated accumulation of defense-associated phenolic compounds in infected spruce tissues, indicating activation of host chemical defense responses. Transcriptomic profiling of infected spruce revealed extensive molecular reprogramming, with more than 20,000 differentially expressed mRNAs and over 10,000 differentially expressed long non-coding RNAs. In parallel, RNA sequencing of fungi re-isolated from infection sites identified more than 3,000 differentially expressed genes and over 300 differentially expressed long non-coding RNAs relative to fungi maintained on artificial culture media.

Together, these findings demonstrate that bark beetle-associated fungal pathogens induce substantial anatomical, metabolic, and transcriptional responses in Norway spruce. The study provides new insight into the molecular mechanisms underlying spruce-fungus interactions and establishes a foundation for future functional research on forest host-pathogen dynamics.

Keywords: Tree-pathogen interaction; metabolomics; bark beetles; fungal pathogens; conifers; dual RNA-seq.



OP-17

**REVEALING THE HIDDEN GENETIC DIVERSITY IN NATURAL PLANT
GERMPLASM COLLECTIONS THROUGH DNA BARCODES – CASE STUDIES IN
BREEDING RESEARCH AND CONSERVATION OF ENDEMIC AND
THREATENED SPECIES**

Georgi Bonchev¹, Irina Boycheva¹, Vasilissa Manova¹, Stiliana Simeonova¹, Miroslava Zhiponova², Yordan Georgiev², Iva Doycheva³, Stoyan Stoyanov³, Maria Rosa Marano⁴, Lucila Garcia⁴, Carolina Galli⁵, Miroslav Svec⁶, Michal Hrabovski⁶, Dominika Gahurova⁶

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. Georgi Bonchev str., Bldg. 21, 1113 Sofia, Bulgaria

²Faculty of Biology, Sofia University “St. Kliment Ohridski”, Sofia, Bulgaria

³Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Acad. Georgi Bonchev str., Build. 23, 1113 Sofia, Bulgaria

⁴Instituto de Biología Molecular y Celular de Rosario, Argentina (IBR-CONICET)

⁵Instituto Nacional de Tecnología Agropecuaria, Agencia de Extensión Rural Concarán, San Luis, Argentina.

⁶Department of Botany, Faculty of Natural Sciences, Comenius University Bratislava, Révová 39, 811 02 Bratislava, Slovakia

Presenting author: georgi.bonchev71@gmail.com

DNA barcoding is an efficient molecular marker tool for taxonomic identification of species, disentangling population diversity and tracking phylogenetic relationships. Classical identification by morphological characters can fail in cases of hidden genetic diversity at within and between-species level. This overlooked diversity is common in cases of cryptic and recently diverged species, ecotype differentiation, within-species cultivars and rare and endemic species.

Here, I will present results on ongoing studies on the applicability of DNA barcoding for:

1. Assessment of cultivar diversity for improved breeding

Better knowledge of pattern of genetic variability of germplasm resources used in agriculture is important to develop genetic passports of valuable genotypes and cultivars. In addition, estimates of genetic distances between germplasm resources are crucial for selecting desirable genotypes for precise breeding strategies. Data on the intraspecific diversity of rosemary (*Salvia rosmarinus*), a species of increasing agricultural importance as a source of plant biostimulants and defense-priming compounds for sustainable crop protection, will be presented with emphasis on correlations with cultivar dependent differential bioactive potential.

2. Exploration of rare, endemic and threatened for Bulgaria species

Rare, endemic and endangered for extinction species often harbor unrecorded, unique genetic lineages and DNA barcoding can help accurately map these lineages, thereby enforcing and improving their conservation status. Data on the genetic diversity of the endemic for Bulgaria and Balkans species *Aethionema rhodopaeum* and *Brassica jordanoffii* (family Brassicaceae) will be presented, together with data on population diversity of orchid species listed as Vulnerable or Endangered in the Bulgarian Red List.

Keywords: Cultivar identification; intraspecific genetic diversity; rosemary, orchids, Brassicaceae.

Acknowledgements: The research is supported by projects N 101086366 CropPrime (HORIZON-MSCA-2021-SE-01) “Stabilizing CROP yield under unfavourable conditions by molecular PRIM(E)ing”; КП-06-H81/10 “Multidisciplinary approaches for the conservation of endangered and endemic species of the Bulgarian flora”, Bulgarian National Science Fund; КП-06-Словакия/4 „DNA-based studies on ecogeographic diversity of endangered orchid species“, Bulgarian National Science Fund.

OP-18

BIOPHYSICAL MARKERS IN LARGE-SCALE ALGAL CULTIVATION

Jana Kvíderová^{1,2}, Jaromír Lukavský¹, Josef Elster^{1,3}

¹Department of Phycology, Institute of Botany CAS, Dukelská 135, 379 82 Třeboň, Czech Republic

²Centre for Biology, Geosciences and the Environment, Faculty of Education, University of West Bohemia, Chodské náměstí 1, 306 14 Plzeň, Czech Republic

³Centre for Polar Ecology, Faculty of Science, University of South Bohemia, 370 05 České Budějovice, Czech Republic

Presenting author: jana.kviderova@objektivem.net

Algal biotechnological applications are crucial for sustainable development and circular economy, since they could be used in various applications, for instance, in waste-water treatment or soil remediation. Moreover, they can produce high-value compounds, e.g., polyunsaturated fatty acids. For large-volume cultivation at semi-industrial and industrial scales, the development of an autonomous regime of cultivation is necessary. Therefore, several biophysical parameters based on non-invasive spectrophotometric and variable chlorophyll fluorescence measurements are proposed to be used to monitor the proposed cultivation.

Spectrophotometric markers are commonly used to monitor algal growth (Andersen 2005):

- A_{750} (A_{720}) – biomass/dry weight content proxy
- A_{680} (A_{682}) – chlorophyll *a* content proxy
- $A_{680}:A_{750}$ ratio – proxy of chlorophyll *a* content per dry weight unit

Variable chlorophyll fluorescence measurements provide information on physiological state and photosynthetic activity, revealing markers that include different parameters based on fluorescence protocol selection (Roháček et al. 2008):

- Maximum quantum yield (F_v/F_m) and effective quantum yield (Φ_{psII}) – basic parameters
- Fast kinetics – OJIP transient – processes within the photosystem II
- Slow kinetics – quenching analysis and rapid light curves – light energy utilization and photoadaptation

The following case studies are presented:

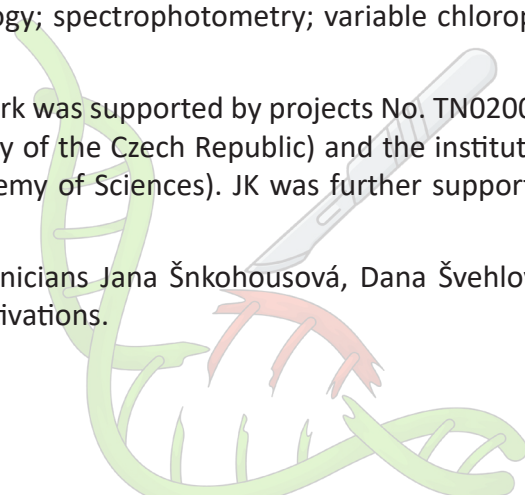
Case study 1: Cultivation of *Chodatodesmus australis* in different light regimes in small-scale photobioreactor (80 mL)

Case study 2: Cultivation of *Koliellopsis inundata* in medium-scale (20 L) and large-scale (250 L) photobioreactors

Keywords: Algal biotechnology; spectrophotometry; variable chlorophyll fluorescence; large-scale cultivations

Acknowledgements: The work was supported by projects No. TN02000044 (JL, JK) and TS02030210 (JE, JK) (Technological Agency of the Czech Republic) and the institutional long-term research plan RVO 67985939 (Czech Academy of Sciences). JK was further supported by the University of West Bohemia.

We are grateful to our technicians Jana Šnkohousová, Dana Švehlová and Veronika Sýkorová for technical support during cultivations.



OP-19

DIFFERENTIAL DROUGHT RESPONSES OF BULGARIAN SWEET, SUPER-SWEET AND FORAGE MAIZE HYBRIDS UNDER FIELD CONDITIONS

Margarita Popova¹, Momchil Paunov¹, Biserka Hinkina², Nataliya Petrovska², Atanas Atanassov³, Miroslava Zhiponova¹

¹Sofia University “St. Kliment Ohridski”, Faculty of Biology, Dragan Tsankov Blvd. 8, Sofia 1164, Bulgaria

²Agricultural Academy, Maize Research Institute Knezha, Knezha 5835, Bulgaria

³Joint Genomic Center, Dragan Tsankov Blvd. 8, Sofia 1164, Bulgaria

Presenting author: margaritnp@uni-sofia.bg

Drought is one of the major constraints on maize productivity and its effects depend strongly on genotype. In the present study, nine Bulgarian maize hybrids belonging to sweet, super-sweet and forage types were evaluated under contrasting field water regimes in order to compare their response to water deficit and identify traits associated with better performance under drought. Seedling emergence, dry biomass, plant height, photosynthetic activity (PI_{total}), and a productivity index were assessed at different developmental stages, and the data were analysed by two-way ANOVA and principal component analysis. Water limitation generally reduced biomass accumulation, plant height, photosynthetic activity and productivity, although the magnitude of these effects varied among hybrids. Forage hybrids maintained comparatively better performance under drought than most sweet and super-sweet hybrids. PCA further separated the experimental variants mainly according to water regime and, secondarily, according to genotype-related differences. Overall, the results demonstrate marked variation among Bulgarian maize hybrids in their response to drought and identify forage hybrids as promising material for further studies on drought adaptation and plant-associated microbiota.

Keywords: Maize hybrids; drought stress; field phenotyping; chlorophyll fluorescence; oxidative stress; productivity index.

Acknowledgements: This work is financially supported by the Scientific Research Fund of Sofia University, project №80-10-96 from 07.04.2026, and by project BG05SFPR001-3.004-0004-C01, Interdisciplinary training of doctoral students to develop knowledge and competencies in various branches of modern research for healthy life and clean technologies (INTERDOC), funded within procedure “Support for development of doctoral projects” of Executive agency “Programme EDUCATION”, co-funded by the European Union.



OP-20

FRIENDS OR FOES? INSECT–MICROBE INTERACTIONS IN FRUIT AGROECOSYSTEMS**Amrita Chakraborty^{1,2}, Paul G. Becher²**¹Faculty of Forestry and Wood Sciences, Czech University of Life Sciences, Prague, Czech Republic²Department of Plant Protection Biology, Swedish University of Agricultural Sciences, Alnarp, SwedenPresenting author: chakraborty@fld.czu.cz

Insect–microbe interactions are fundamental components of agricultural systems, shaping pest behavior, microbial community dynamics, crop protection, and postharvest fruit quality. Understanding these interactions is crucial for revealing how pests colonize crop substrates, how microbes influence insect fitness, and how microbial competitors or pathogens affect food quality. This study investigates the association between the invasive fruit pest *Drosophila suzukii* and the mutualistic yeast *Hanseniaspora uvarum*, focusing on their role in berry colonization and mould suppression. *Drosophila suzukii* develops in ripening fruit and is closely associated with *H. uvarum*, which grows on both ripe and unripe raspberries. The yeast attracts larvae and adult flies, stimulates oviposition, and supports larval development. This yeast–fly association also affects interactions with grey mould (*Botrytis cinerea*), a major postharvest pathogen that affects fruit quality. Although *B. cinerea* reduced larval development and deterred oviposition, its growth was strongly suppressed by the combined activity of *H. uvarum* and feeding fly larvae. These findings demonstrate reciprocal niche construction, in which the fly vectors yeast to fruit, while the yeast improves larval habitat quality and contributes to mould suppression. Overall, this study highlights the importance of insect–microbe interactions as biological drivers of pest success, microbial succession, and fruit quality outcomes. Understanding such interactions may support future strategies that harness naturally occurring antagonistic yeasts to improve fruit shelf life by suppressing mould pathogens, while preventing pest-mediated fruit damage and uncontrolled microbial growth.

Keywords: *Drosophila suzukii*; *Hanseniaspora uvarum*; yeast–fly association; insect–microbe interactions; symbiosis.



POSTER PRESENTATIONS

PP-01

UHPLC-MS/MS METHOD DEVELOPMENT FOR ABSOLUTE QUANTIFICATION OF GIBBERELLINS AND INDOLE-3-ACETIC ACID IN REPRODUCTIVE TISSUES OF *HERACLEUM SOSNOWSKYI*

Dominykas Laibakojis¹, Simonas Balčiūnas², Saulė Rapalytė¹, Jurga Būdienė³, Tautvydas Žalnierius¹, Sigita Jurkonienė¹

¹Laboratory of Plant Physiology, State Scientific Research Institute Nature Research Centre, Akademijos Str. 2, Vilnius, Lithuania

²Department of Analytical and Environmental Chemistry, Vilnius University, Naugarduko Str. 24, Vilnius, Lithuania

³Department of Organic Chemistry, Center for Physical Sciences and Technology, Saulėtekio Ave. 3, Vilnius, Lithuania

Presenting author: dominykas.laibakojis@gamtc.lt

Gibberellins (GAs) are a large group of plant hormones found in trace amounts that regulate plant growth and development. *Sosnowsky's hogweed* (*Heracleum sosnowskyi* Manden.) is enlisted among Invasive Alien Species that are a major concern for the EU. Its ability to destroy native ecosystems and toxicity for human health is worrying. Hogweed spreads only by seeds and dies after reproduction. GAs are crucial for seed development and further spread. GA function and effect also depend heavily on crosstalk with other endogenous phytohormone groups like auxins. Therefore, quantification of endogenous GAs and indole-3-acetic acid (IAA) in *H. sosnowskyi* tissues provides valuable information about the state of GA metabolism in response to various treatments, crosstalk, or changes in environmental conditions. Thus, efficient extraction, purification and determination of minute GA and IAA amounts are essential. This study aims to develop a UHPLC-MS/MS method for quantification of GAs and IAA in *H. sosnowskyi* reproductive tissues, and to optimize an established GA extraction and purification protocol.

In this study, a selective and sensitive UHPLC-MS/MS method for the separation and quantification of 12 different GAs and IAA from 100 mg of *H. sosnowskyi* reproductive tissues was developed. The optimal separation of analytes was achieved on a UHPLC reversed-phase column (ReproSil-Pur Basic-C18, 2.1 mm x 100 mm, 1.9 µm; Dr. Maisch) using a nonlinear gradient of water (A) and acetonitrile (B) mobile phases both containing 0.1 % formic acid at a flow rate of 0.4 mL min⁻¹, from 95:5 A:B (v/v) to 30:70 A:B (v/v) over 13 min with column washing using 0:100 A:B (v/v) until 15 min. Quantification of GAs was performed using a highly selective multiple reaction monitoring (MRM) mode and an isotope dilution method. Furthermore, we modified and optimized an existing GA extraction and purification method, developed by Urbanova et al. (2013) and achieved improved recoveries of GAs.

In conclusion, a robust UHPLC-MS/MS method for quantification of GAs and IAA in *H. sosnowskyi* reproductive tissues and optimized an extraction and purification protocol that allows further investigations into GA-mediated regulation of reproductive development in this species were developed.

Keywords: Gibberellins, indole-3-acetic acid, UHPLC-MS/MS, *Heracleum sosnowskyi*.

Acknowledgements: This research was funded by the Research Council of Lithuania (LMTLT) (Project No. S-MIP-25-21).

PP-02**EFFECT OF UV-C SEED PRIMING ON THE PHENOLIC CONTENT AND ANTIOXIDANT CAPACITY OF DIFFERENT KALE CULTIVARS**

Elena Shopova, Liliana Brankova, Zornitsa Katerova, Lyudmila Dimitrova, Ivo Dimitrov, Dessislava Todorova

Institute of Plant Physiology and Genetics - Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., Bldg. 21, 1113 Sofia, Bulgaria

Presenting author: kostei@abv.bg

This study investigated the effects of UV-C (254 nm) seed priming on the antioxidant capacity and total phenolic content of four kale cultivars (*Brassica oleracea* “Green Curly”, “Red Scarlet”, and “Nero di Toscana”, and *Brassica napus* “Red Russian”). Seeds were exposed to UV-C radiation for varying durations: 0 min (control), 20 min, 40 min, 60 min, 80 min, and 100 min. Antioxidant capacity in kale microgreens was evaluated using Trolox equivalent antioxidant capacity (TEAC) and ferric reducing antioxidant power (FRAP) assays, with both hydrophilic and lipophilic fractions analyzed. The results indicate that water-soluble antioxidants predominate across all cultivars, with a correlation observed between total phenolic content (TPC) and FRAP values. UV-C treatment induces a cultivar-specific antioxidant response, ranging from minimal changes to significant enhancement of antioxidant activity. The most pronounced effect was observed in the Red Scarlet cultivar with high anthocyanin content, in which a substantial increase in both phenolic compounds and antioxidant capacity was recorded. These findings highlight the role of genotype in determining the effectiveness of UV-C in inducing antioxidant metabolites.

Keywords: Seed priming; oxidative stress; antioxidant capacity; kale.

Acknowledgements: This research was supported by the Bulgarian National Science Fund under contract KP-06-N96/5, dated 09 December 2025.



PP–03

ETHYLENE EMISSION IN WINTER WHEAT (*TRITICUM AESTIVUM* L.) UNDER SHORT-TERM COLD STRESS: INFLUENCE OF HERBICIDE AND MICROBIAL BIOSTIMULANT TREATMENTS**Greta Šaltytė¹, Jurga Jankauskienė¹, Rima Mockevičiūtė¹**

¹Laboratory of Plant Physiology, State Scientific Research Institute Nature Research Centre, Akademijos Str. 2, 08412, Vilnius, Lithuania

Presenting author: greta.saltyte@gamtc.lt

The environment stresses can cause up to 50% of crop yield losses. In response to stress, plants activate phytohormone system, among which ethylene is one of the most important regulators of the stress response (Iqbal N., 2017). Ethylene initiates a cascade of adaptive responses in plants under optimal and stressful conditions. Herbicides in the acetolactate synthase (ALS) inhibitor class, such as metsulfuron-methyl and thifensulfuron-methyl, are widely used in cereal crops. Their effectiveness and the physiological response of plants may be influenced by environmental conditions and temperature. Therefore, it is important to consider plant growth conditions when evaluating their effects.

The study was conducted in controlled-environment phytotrons (16 h light / 8 h dark at 23 °C, soil moisture at 80%, and an atmospheric CO₂ concentration of 400 ppm). Extreme environmental conditions were simulated by applying a combination of short-term cold and drought stress. Plants were first exposed to a single cold treatment at –3 °C for 16 h to simulate a late-spring frost, followed by drought stress, which was induced by reducing irrigation until the soil moisture content reached 30% and maintained for 3 days. These conditions enabled a comparison of plant responses under stress following herbicide and microbial biostimulant application.

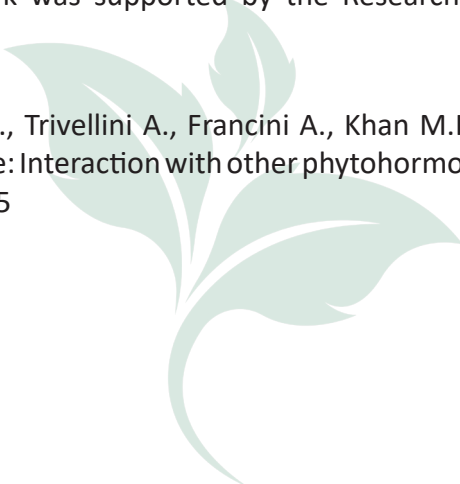
Low temperatures increased ethylene emission in all groups, confirming that cold causes physiological stress in plants. The highest ethylene content was observed in the control plants after short-term cold exposure (53.75 nL g⁻¹ FW h⁻¹). In wheat treated with the microbial biostimulant, ethylene levels were lower than in the control both before and after short-term cold exposure. Following short-term cold exposure, ethylene increased by approximately 50% in the control plants (35.91 to 53.75 nL g⁻¹ FW h⁻¹), whereas the increase was markedly smaller in biostimulant-treated plants (27.29 to 32.63 nL g⁻¹ FW h⁻¹, ~20%), indicating that the microbial biostimulant reduced the stress-induced ethylene response. Therefore, it can be assumed that treatment with microbial biostimulants may modify the plants physiological response to short-term cold stress.

Keywords: Winter wheat (*Triticum aestivum* L.); microbial biostimulant; ethylene.

Acknowledgements: This work was supported by the Research Council of Lithuania PROCLIM project (2025–2028)

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PP-04

UV-C SEED PRIMING AS A TOOL FOR MODULATING THE GLUCOSINOLATE PROFILE OF KALE MICROGREENS

Liliana Brankova¹, Milen Bogdanov², Elena Shopova¹, Savina Stoyanova², Lyudmila Dimitrova¹, Zornitsa Katerova¹, Dessislava Todorova¹, Tsvetina Nikolova¹, Denitsa Petkova²

¹Institute of Plant Physiology and Genetics - Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., Bldg. 21, 1113 Sofia, Bulgaria

²Sofia University "St. Kliment Ohridski", Faculty of Chemistry and Pharmacy, Sofia 1164, Blvd. James Boucher 5

Presenting author: lbrankova@abv.bg

Kale microgreens, owing to their rich nutritional profile and high content of secondary metabolites such as glucosinolates, phenolic compounds, and carotenoids, exhibit considerable potential to enhance the nutritional quality of the human diet. Their incorporation into dietary patterns represents a promising strategy for preventing socially significant chronic diseases. UV-C radiation is a potent elicitor of secondary metabolism, stimulating the synthesis of compounds that perform protective functions in plants and have beneficial effects on human health. In the present study, we investigated the effect of UV-C seed priming at varying durations on the glucosinolate profile of microgreens from four kale cultivars (*Brassica oleracea* "Green Curly", "Red Scarlet", and "Nero di Toscana", and *Brassica napus* "Red Russian"). We observed differences in the baseline glucosinolate profiles among the cultivars, both in composition and in the relative proportions of individual glucosinolates. UV-C radiation was found to induce cultivar-dependent changes in glucosinolate content and may serve as a tool for targeted modulation of kale's phytochemical profile.

Keywords: Functional foods; glucosinolates; phytochemicals; seed priming; kale; microgreens.

Acknowledgements: This research was supported by the Bulgarian National Science Fund under contract KP-06-N96/5, dated 09 December, 2025.



PP-05

COMPREHENSIVE STUDY OF THREE WHEAT CULTIVARS SUBJECTED TO DROUGHT

Dilyana Doneva¹, Violeta Peeva¹, Svetlana Misheva¹, Katelina Prokopova¹, Gabriella Szalai², Magda Pal², Alexander Angelov¹, Vladimir Aleksandrov¹

¹Institute of Plant Physiology and Genetics, BAS, “G. Bonchev” str., bl.21, Sofia, Bulgaria

²HUN-REN ATK, Brunszvik u. 2, H-2462 Martonvásár, Hungary

Presenting author: donevadiliana@gmail.com

Wheat (*Triticum aestivum* L.) is a staple crop for much of the population. It is the world's leading cereal crop, recognized for its nutritional value, extensive cultivation and global consumption. According to FAO data, in the 2025/26 marketing year, global wheat production is forecast to reach nearly 810 million tons. On the other hand climate change, through rising temperatures and altered precipitation patterns, poses a major threat to global agriculture and food security. The availability of water is a key factor determining the structure of ecosystems, the functioning of economies and the well-being of human populations. Thus drought stands out as a critical abiotic stress factor, threatening not only agricultural yields but also the sustainability of food systems and humanity's nutrition globally. It significantly influences both morphological and phenological traits of the plants. In this study morphological features and functional efficiency of the photosynthetic apparatus were compared in spring and winter wheat varieties by measuring leaf area, plant biomass, total chlorophyll content, chlorophyll a fluorescence, thermoluminescence and metabolomics. In our experiments spring cultivar Virradat demonstrated the highest drought tolerance. The other spring cultivar (Pirkadat) reacted to the water limitations with decreased photochemical efficiency but succeeded to preserve its growth and developmental processes. The winter wheat genotype Enola was affected by the drought but recovered well from the stress. We concluded that all three cultivars used different strategies to cope with the drought stress.

Keywords: Wheat; drought; photosynthetic performance; metabolic profile.

Acknowledgements: The authors thank the BULGARIAN NATIONAL SCIENCE FUND, grant number KP-06-H81/1, for financing this study.



PP–06

CHARACEAN *NITELLOPSIS OBTUSA* CELL RESPONSES TO YTTRIUM AND SELECTED LANTHANIDES

Brigita Gylytė¹, Rolandas Karitonas¹, Sigita Jurkonienė¹, Vaidevutis Šveikauskas¹, Vira Khoma², Jūratė Vaičiūnienė³, Vitalija Mickevičiūtė⁴, Oksana Stoliar², Levonas Manusadžianas¹

¹SSRI Nature Research Centre, Vilnius, Lithuania

²Ternopil Volodymyr Hnatiuk National Pedagogical University, Ternopil, Ukraine

³Centre for Physical Sciences and Technology, Institute of Chemistry, Vilnius, Lithuania

⁴Vilnius University, Life Sciences Center, Vilnius, Lithuania

Presenting author: brigita.gylyte@gamtc.lt

Rare earth elements (REEs), including the lanthanides and yttrium (Y), are increasingly used in modern technologies and are considered emerging aquatic contaminants. However, their behaviour in exposure media and cellular-level effects in aquatic plants remain insufficiently understood. This study investigated the responses of internodal cells of the characean alga *Nitellopsis obtusa* to praseodymium (Pr), neodymium (Nd), dysprosium (Dy), and Y. We assessed REE accumulation in cells and depletion from the medium, distribution between the cell wall and the cell interior, bioelectrical responses, and inhibition of K⁺, Mg²⁺-specific H⁺-ATPase activity.

Accumulation and cell fractionation. During 24 h exposure to 0.5 mM of the respective element, REE accumulation in *N. obtusa* cells and the decrease in dissolved REE concentrations in the medium followed a two-phase pattern: rapid uptake within approximately 9 h, followed by a relatively steady state between 12 and 24 h. Cell-accumulated REE concentrations ranged from 24 to 33 mg g⁻¹ DW and did not differ significantly among elements. However, the 12 h bioconcentration factors were 256, 1037, 334, and 376 L kg⁻¹ for Y, Pr, Nd, and Dy, respectively, indicating 3–4-fold higher Pr accumulation intensity. Most of the measured REE content in whole cells was associated with the cell wall. The cell interior, consisting of vacuolar sap, cytoplasm, and organelles, accounted for up to 20% of the total accumulated content. The highest internal concentrations were found for Y and Dy, reaching 2.1 ± 1.0 and 2.2 ± 0.1 mg L⁻¹, respectively, followed by Nd at 1.3 ± 1.2 mg L⁻¹. Pr in the cell interior was below the LOD of 0.005 mg L⁻¹.

Bioelectrical response. Nd induced the strongest 90-min resting potential (RP) depolarisation, 53 ± 13%, and the highest residual depolarisation after rewash (RD), 35 ± 16%, indicating the weakest recovery from REE-induced disturbance. These values differed significantly ($p < 0.05$, ANOVA) from the corresponding values for Y (RP) and for Y, Pr, and Dy (RD).

K⁺, Mg²⁺-specific H⁺-ATPase. Enzyme activity was measured in the microsomal fraction of the cell. The IC₅₀ values for Y, Nd, Dy, and Pr were 0.2, 0.4, 0.6, and 2.3 mM, respectively. The IC₅₀ for Pr, the least inhibitory among the four REEs, differed significantly from those of the other REEs.

Overall, REE interaction with *N. obtusa* cells was dominated by rapid association with the cell wall, whereas membrane-level responses revealed element-specific functional toxicity. The mismatch between high Pr bioconcentration and weak ATPase inhibition suggests that REE accumulation and physiological effects are governed not only by total accumulated mass but also by element-specific interactions with cellular compartments and membrane-associated targets in this macrophytic alga.

Keywords: Rare earth elements; *Nitellopsis obtusa*; bioaccumulation; cell wall; cell interior.

Acknowledgements: This work was funded by the Joint Lithuanian-Ukrainian R&D programme (project No. S-LU-24-13) supported by the Research Council of Lithuania and the Ukrainian-Lithuanian R&D programme (Project No. M48/2024) supported by the Ministry of Education and Science of Ukraine.

PP-07**EVALUATION OF THE EFFECT OF TWO PLANT GROWTH REGULATORS – 4PU-30 AND MEIA, ON EINKORN (*TRITICUM MONOCOCCUM* L.) AND WINTER WHEAT (*TRITICUM AESTIVUM* L.) IN FIELD CONDITIONS, USING DIFFERENT APPROACHES****Hristo Nikolov¹, Radoslav Chipilski², Zoia Tsekova³, Irina Moskova³**¹Bulgarian Academy of Sciences, Space Research and Technology Institute, 1113, Sofia, Bulgaria²Agricultural Academy, Institute of Plant Genetic Resources “Konstantin Malkov”, 4122, Sadovo, Bulgaria³Bulgarian Academy of Sciences, Institute of Plant Physiology and Genetics, 1113, Sofia, BulgariaPresenting author: hristo@stil.bas.bg

The aim of the study was to evaluate the effects of two plant growth regulators - 4PU-30 (a phenylurea-type cytokinin) and MEIA (β -monomethyl ester of itaconic acid) - on einkorn (*Triticum monococcum* L.) and winter wheat, cv. Enola (*Triticum aestivum* L.), under field conditions. Evaluation was performed using a combination of conventional biochemical and biometric analyses, as well as physical methods – more specifically spectrometric measurements in optical and NIR ranges of the spectrum.

The crops were cultivated in accordance with standard agronomic practices under typical seasonal environmental conditions at the experimental field of the Institute of Plant Genetic Resources (IPGR) in Sadovo during the 2024–2025 growing season.

The experimental scheme consisted of the following variants, applied to both crops – control plants (not treated); plants treated with 4PU-30; plants treated with MEIA.

In early April, the plants were leaf sprayed with water solutions of the investigated growth regulators, 4PU-30 and MEIA. This treatment was repeated following the onset and completion of the heading stage. At the end of the grain-filling stage, morphophysiological measurements were performed on the flag leaves of the plants, and samples were collected for biochemical analysis (determination of pigment content) In parallel, during the mentioned plants growth stages large number, narrowband (10 [nm]/sample) spectrometric measurements were performed in situ.

A positive effect of 4-PU30 and MEIA was established on einkorn and wheat plants under standard field conditions. The growth regulator-treated variants exhibited increased chlorophyll content compared to the control. A beneficial effect of the growth regulators was also observed regarding relative water content and plant phenotype (leaf width and leaf area)

An additional focus of this study was to leverage narrow-waveband spectrometric measurements to obtain spectral reflectance data for monitoring wheat and einkorn plants. To achieve this, previously validated in prior experiments, a remote sensing (RS) approach was applied.

The high-confidence results from the spectrometric measurements strongly align with the obtained biochemical and biometric data, as reflected in the values recorded in both the green visible spectrum and the near-infrared region. The two applied approaches demonstrate a positive correlation, validating spectrometric methods as a reliable criterion for assessing the impact of various treatments on plants.

Keywords: Einkorn; winter wheat; MEIA; 4PU-30.

Acknowledgements: This research was supported by Bulgarian National Science Fund (BNSF), Grant No KP-06-N56/15.

PP-08

RESPONSE OF THE CROP PLANT *SOLANUM LYCOPERSICUM* AND THE RESURRECTION PLANT *HABERLEA RHODOPENSIS* TO INFECTION CAUSED BY PHYTOPATHOGENIC FUNGI

Katya Georgieva, Zornitsa Stoyanova, Eva Popova, Vasilissa Manova, Rossitza Rodeva, Bistra Mihailova, Gergana Mihailova

Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. G. Bonchev Str., Bl. 21, 1113 Sofia, Bulgaria

Presenting author: katya@bio21.bas.bg

Plant pathogenic fungi cause substantial yield losses in many economically important crops and negatively affect biodiversity. While considerable attention has been directed toward diseases in agricultural species, the role of wild and native plants in pathogen spread remains insufficiently explored. Resurrection plants, with their unique ability to survive desiccation to an air-dry state, represent valuable models for studying mechanisms of desiccation tolerance to improve the drought resistance of crop species. Although their tolerance to dehydration is relatively well characterized, little is known about whether this exceptional tolerance to abiotic stress is accompanied by increased resistance to biotic stress, particularly fungal pathogens. In this study, the response of *Solanum lycopersicum* and *Haberlea rhodopensis* to inoculation with *Botrytis cinerea*, *Alternaria solani*, and *Fusarium oxysporum* was assessed based on visible symptoms and chlorophyll fluorescence measurements. Detached leaves of *H. rhodopensis* and two tomato cultivars (Rozova magia and San Marzano) were inoculated by 4-mm mycelial discs of 7-days actively growing fungal colonies. Both plant species reacted with the most pronounced symptoms to the *Botrytis* infection. Tomato leaves were completely damaged and died in four days, whereas in *H. rhodopensis*, the infection remained limited to necrotic spots. Measurements above these spots showed that the quantum efficiency of photosystem II, Y(II), declined by 30% on day 6 and by 55% on day 12. The reduced photochemical activity of PSII was accompanied by increased dissipation of excess excitation energy, protecting the PSII reaction center from overexcitation. Notably, the quantum yield of non photochemical quenching, Y(NPQ), increased more strongly than the fraction of energy that is passively dissipated as heat and fluorescence, Y(NO). In tomato, the infection with *A. solani* resulted in a reduction of approximately 60% in Y(II) and a five fold increase in excitation pressure. Although *F. oxysporum* caused milder symptoms than *A. solani*, tomato leaves died seven days after inoculation with both pathogens. *F. oxysporum* did not affect *H. rhodopensis*. The inoculation with *A. solani* led to a 15% decline in PSII photochemical activity and a 2.5 fold increase in non photochemical quenching. Both tomato cultivars showed a similar response to all pathogens and cv. Rozova magia was selected for further investigations. Overall, the results clearly demonstrate a higher resistance of *H. rhodopensis* to the tested fungal isolates compared to *S. lycopersicum*.

Keywords: Resurrection plant; tomato, fungal pathogens; artificial inoculation; photosynthesis.

Acknowledgements: This research was funded by the Bulgarian National Science Fund, grant number KP-06-H96/3.

PP–09

COMBINED EFFECT OF TEMPERATURE AND LIGHT ON THE ANTIOXIDANT ACTIVITY OF THE NEWLY ISOLATED STRAIN *SCENEDESMUS SP. JV-25***Nadya Agayn-Stoyanova, Juliana Ivanova, Veronika Deleva**

Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Experimental Algology Department, Acad. G. Bonchev Str. 21, 1113, Sofia, BULGARIA

Presenting author: nadia199000@abv.bg

Microalgae are considered a promising source of natural antioxidants, whose biosynthesis is strongly influenced by environmental factors. This study evaluates the combined effect of temperature and light intensity on the antioxidant capacity of the newly isolated strain *Scenedesmus sp. JV-25*.

Cultivation was performed in a controlled laboratory system at four temperatures (18°C, 24°C, 28°C, and 31°C) under two light regimes (5000 and 2×5000 lx). Samples were pre-frozen prior to analysis. Additionally, fresh samples (without pre-freezing) were analyzed after cultivation at 24°C and 5000 lx. Polyphenols and flavonoids were determined using standard spectrophotometric methods, while total antioxidant capacity was evaluated using FRAP (Ferric Reducing Power assay) and ABTS (radical scavenging activity).

The results revealed a statistically significant interaction between temperature and light intensity affecting antioxidant potential. Fresh biomass exhibited the highest antioxidant activity, reaching 82.64 $\mu\text{mol TE g}^{-1} \text{ DW}$ (FRAP) and 50% inhibition (ABTS). In frozen samples, the maximum flavonoid content (17.88 mg QE $\text{g}^{-1} \text{ DW}$) and FRAP activity (47.99 $\mu\text{mol TE g}^{-1} \text{ DW}$) were observed at 28°C and 5000 lx. Increasing light intensity at the same temperature decreased FRAP activity but enhanced ABTS radical scavenging capacity (up to 34.86% inhibition), suggesting differential regulation of antioxidant mechanisms. The obtained values indicate a comparatively high antioxidant potential relative to green microalgae.

In conclusion, *Scenedesmus sp. JV-25* demonstrates high metabolic plasticity and significant potential as a source of natural antioxidants. Optimization of cultivation conditions and sample preparation is crucial for enhancing the biosynthesis of these bioactive compounds.

Keywords: Microalgae; *Scenedesmus sp. JV-25*; antioxidant activity; temperature; light intensity.



PP-10

TOXICITY OF BINARY MIXTURES OF SULFAMETHOXAZOLE AND PRASEODYMIUM TO *LEMNA MINOR*. PHYSIOLOGICAL AND OXIDATIVE STRESS MARKERS AND PEROXIDASE INDUCTIONS**Rolandas Karitonas¹, Eimantas Antanas Lapukas²**¹State Scientific Research Institute Nature Research Centre, Vilnius, Lithuania²Vilniaus University, Vilnius, LithuaniaPresenting author: rolandas.karitonas@gamtc.lt

The increasing release of pharmaceuticals and rare earth elements into freshwater raises concerns about combined effects on aquatic plants. We tested seven mixture proportions of sulfamethoxazole (SMX) and praseodymium (Pr) (SMX:Pr, in percents): 0:0; 6.25:100; 12.5:50; 25:25; 50:12.5; 100:6.25; 100 (1.5 mg L⁻¹) : 100 (74.2 mg L⁻¹) on *Lemna minor*, measuring relative frond and root growth rates (RFGR, RRGR), chlorophyll a+b, carotenoids, H₂O₂, MDA, guaiacol peroxidase (GPOD), and pyrogallol peroxidase (PPOD) after 7 days of exposure. The control values were 0.283 ± 0.007 day⁻¹ (RFGR), 0.649 ± 0.020 day⁻¹ (RRGR), 8.86 × 10⁻⁵ ± 5.88 × 10⁻⁶ mg g⁻¹ FW (chlorophyll a+b) and 2.19 × 10⁻⁵ ± 5.19 × 10⁻⁷ mg g⁻¹ FW (carotenoids), 437 ± 23 mM g⁻¹ FW H₂O₂, 3.76 ± 0.12 mM g⁻¹ FW (MDA), 0.0896 ± 0.0027 nkat mg⁻¹ protein (GPOD) and 5.76 ± 0.446 nkat mg⁻¹ protein (PPOD). The 100:100 mixture caused severe inhibition of RRGR by 95% to 0.035 day⁻¹; RFGR by 54% to 0.131 day⁻¹, maximal lipid peroxidation (MDA = 9.51 ± 0.23 mM g⁻¹ FW), and enzyme activations (by 3.7-fold for GPOD = 0.334 ± 0.016 nkat mg⁻¹ protein; by 1.5-fold for PPOD = 8.38 ± 0.53 nkat mg⁻¹ protein). An intermediate mixture (SMX:Pr, 12.5:50) produced an H₂O₂ peak of 538 ± 21 mM g⁻¹ FW, indicating an early ROS burst. Correlations identified MDA as the principal damage marker: strong negative links with root (r = -0.94), frond growth (r = -0.78) chlorophyll a+b (r = -0.71), and carotenoids (r = -0.68) as well as positive links with GPOD (r = 0.65) and PPOD (r = 0.58), suggesting damage-driven enzyme induction. The weak H₂O₂ - MDA correlation (r = -0.42) reflects temporal decoupling of H₂O₂ generation and accumulating lipid damage. Roots' sensitivity to MDA suggests direct uptake and meristem vulnerability. Indeed, *L. minor* roots is the primary route of entry for SMX and Pr, therefore, lipid peroxidation likely begins there first and subsequently affects root growth. Since harmful lipid peroxidation is associated with excessive reactive oxygen species and H₂O₂, the weak correlation of MDA and H₂O₂ at the end of the experiment may indicate that a substantial portion of hydrogen peroxide was consumed during lipid peroxidation and for the activation of guaiacol and pyrogallol peroxidases. GPOD showed greater stress-induced induction than PPOD, respectively, 3.7- and 1.5-fold increase, confirming guaiacol peroxidase as the more sensitive early oxidative stress biomarker. Mixture component analysis revealed that higher proportion of SMX in the mixture (100:6.25), causes moderate growth inhibition and pigment loss, while higher proportion of Pr in the mixture (100:0.625) produces severe root growth suppression, marked frond growth, pigment reduction and MDA increase. High SMX+Pr (100:100) produced synergistic toxicity effects. Thus, mixture proportions determine ecological impact, with combined high concentrations of toxicants eliciting oxidative stress-mediated impairment in *L. minor* and identifying root growth as the most sensitive endpoint and MDA as the most reliable cumulative damage marker.

Keywords: *Lemna minor*; sulfamethoxazole; praseodymium; oxidative stress; root growth sensitivity; peroxidase activity.

PP–11

BIOCHEMICAL RESPONSES OF WINTER WHEAT TO APPLICATION OF MICROBIAL BIOSTIMULANT AND SELECTIVE HERBICIDE

Dessislava Todorova¹, Liliana Brankova¹, Elena Shopova¹, Zornitsa Katerova¹, Tsvetina Nikolova¹, Lyudmila Dimitrova¹, Sigita Jurkonienė², Jurga Jankauskienė², Iskren Sergiev¹

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., Bldg. 21, 1113 Sofia, Bulgaria

²State Research Institute, Laboratory of Plant Physiology, Nature Research Centre, Akademijos str. 2, 08412 Vilnius, Lithuania

Presenting author: dessita@bio21.bas.bg

To manage weeds that hinder crops productivity, a common routine in the current agricultural practices is the application of selective herbicides. Although this technique eliminates weeds, crop plants also might undergo different physiological and metabolite alterations caused by the used herbicides. To boost crop metabolism that will provide better response to the herbicide usage, various methods are being explored, including preliminary application of stimulants of organic origin. This study investigates the efficacy of leaf priming with microbial biostimulant ProbioHumus on wheat plants (*Triticum aestivum* L., cv. Sadovo 1) to modulate physiological responses against following treatment with selective herbicide Ergon. Investigation of non-enzymatic and enzymatic antioxidants was performed 14 days after herbicide spraying. We established that the herbicide application caused an increase in the activity of catalase and peroxidase enzymes along with a substantial reduction in the superoxide dismutase activity. No alterations were detected in the glutathione reductase activity, but glutathione S-transferase activity raised significantly after herbicide treatment. Although non-enzymatic antioxidants were decreased by Ergon application, noticeable increase was detected in antioxidant capacity of treated plants both in FRAP and TEAC lipo- and water-soluble fractions. Probiotic applied alone did not provoke considerable alterations in parameters studied but adjusted the wheat plant's defense system to mitigate herbicide undesirable effect on antioxidant protection. Based on these results, it could be assumed that the leaf priming of wheat with microbial biostimulant ProbioHumus may be a promising strategy for boosting plants antioxidant defense under treatment with selective herbicide.

Keywords: *Triticum aestivum* L.; microbial biostimulant; herbicide; antioxidant capacity.

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PP-12

**MODULATION OF WHEAT PHYSIOLOGICAL RESPONSES TO HERBICIDE
BY APPLICATION OF MICROBIAL BIOSTIMULANT**

**Iskren Sergiev¹, Elena Shopova¹, Liliana Brankova¹, Zornitsa Katerova¹, Tsvetina Nikolova¹,
Lyudmila Dimitrova¹, Jurga Jankauskiene², Sigita Jurkoniene², Dessislava Todorova¹**

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., Bldg. 21, 1113 Sofia, Bulgaria

²State Research Institute, Laboratory of Plant Physiology, Nature Research Centre, Akademijos str. 2, 08412 Vilnius, Lithuania

Presenting author: iskren@bio21.bas.bg

Herbicides are the primary method used to manage weeds in modern agriculture. Despite advances in agricultural practices, the usage of selective herbicides in some cases may cause the production of reactive oxygen species (ROS) and oxidative stress in crops. Application of different bioactive organic products, which can enhance plant growth and mitigate herbicide-induced stress in target crops, is of special interest. Wheat seedlings (*Triticum aestivum* L., cv. Sadovo 1) grown under controlled conditions as a soil culture, were sprayed with microbial biostimulant ProbioHumus, and 24 hours later the plants were treated with selective herbicide Ergon, according to the manufacturer's instructions. Growth and biochemical traits were examined in the above-ground part of plants 14 days after the herbicide application. Wheat growth and content of leaf pigments were not considerably influenced by the herbicide. However, Ergon increased the lipid peroxidation and leakage of electrolytes, while reduced H₂O₂ content. The herbicide augmented total osmolytes and reducing sugars but decreased free proline content. Compared to the control, the probiotic application by itself did not alter the studied parameters but substantially adjusted the herbicide-altered traits. It could be concluded that the preliminary application of the microbial biostimulant ProbioHumus might be used as a promising strategy for counteracting the herbicide-induced stress in common wheat.

Keywords: Winter wheat; herbicides; biostimulants; stress markers.

Acknowledgements: This study was supported by Bulgaria-Lithuania joint research program of BAS (2025-2027), Project ID: IC-LT/02/2025-2027.



PP-13**TISSUE-SPECIFIC MANIPULATION OF ETHYLENE SIGNALING IN *ARABIDOPSIS THALIANA* AFFECTS THE TRANSCRIPTIONAL DYNAMICS OF STRESS-INDUCIBLE GENES UNDER DROUGHT AND RECOVERY****Simona Galabova¹, Irina I. Vaseva¹, Dominique Van Der Straeten²**¹Institute of Plant Physiology and Genetics at the Bulgarian Academy of Sciences, Sofia, Bulgaria²Laboratory of Functional Plant Biology, Ghent University, Ghent, BelgiumPresenting author: sgalabova@bio21.bas.bg

Efficient recovery from water deficit is as critical for plant survival as drought tolerance itself, yet the spatial regulation of these processes remains poorly understood. This study investigates how tissue-specific ethylene signaling coordinates the molecular mechanisms driving both moderate drought responses and subsequent recovery in *Arabidopsis thaliana*. Using RT-qPCR, we profiled the transcriptional dynamics of key antioxidant and osmoprotectant genes across a panel of ethylene signaling mutants (*ein2-1*, *ein3eil1*, *ctr1-1*) and transgenic lines featuring ethylene desensitization in distinct root cell types: epidermis and lateral root cap (*pA14:EBF2*, *pLRC1:EBF2*), the quiescent center (*pQ6:EBF2*), endodermis (*pE30:EBF2*), pericycle (*pS1:EBF2*), cortex (*pCOR:EBF2*), and vasculature (*pS2:EBF2*) [1].

Our findings reveal that downregulation and activation of stress-responsive genes is highly tissue-specific. Under dehydration, *pLRC1:EBF2* and *pS1:EBF2* exhibited the most dramatic responses, showing up to 67-fold and 189-fold increase of the drought-specific *P5CS1* splice form and *CAT2* transcripts, respectively, while *SOD1* was uniformly suppressed. The post-drought recovery phase unveiled a distinct trend of genotype-specific transcriptional shifts. The lines with ethylene-desensitized quiescent center and epidermis displayed major spikes in *PER57*, *PER44*, and *P5CS1* expression during recovery, a trend sharply contrasting with their downregulation in *pS1:EBF2* and *pCOR:EBF2* transgenic lines.

These results support the concept of drought resilience as a tightly orchestrated, tissue-specific process where ethylene signaling acts as a spatial modulator. This work provides new insights into the complex regulatory networks governing plant stress responses, demonstrating how localized hormonal signaling shapes the broader transcriptional landscape during environmental fluctuations.

Keywords: *A. thaliana*; drought stress; recovery; antioxidant enzymes.**Acknowledgements:** This research was funded by the Bulgarian National Science Fund; grant number KP-06-N71/12/10 July 2024.**References:**

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PP–14

EFFECTS OF CHEMICALLY DIVERSE ELICITORS ON OXIDATIVE STATUS
AND SPECIALIZED METABOLISM IN MEDICINAL PLANTS

Maria Stasińska-Jakubas, Barbara Hawrylak-Nowak

University of Life Sciences in Lublin, Department of Botany and Plant Physiology, Akademicka 15,
20-950 Lublin, Poland

Presenting author: maria.jakubas@up.edu.pl

Medicinal plants are an important source of specialized metabolites with diverse biological activities and considerable industrial value. The use of exogenous metabolic modulators, including chemical elicitors, represents a promising and sustainable strategy for activating plant defense responses and enhancing the biosynthesis of desirable bioactive compounds. This study aimed to evaluate the effects of chitosan lactate (150 mg/L), sodium selenite (10 mg/L), and salicylic acid (100 mg/L) on the accumulation of specialized metabolites, physiological responses, and selected indicators of oxidative stress in controlled pot cultivation of three medicinal plant species (*Melissa officinalis*, *Hypericum perforatum*, *Chelidonium majus*) [1-3].

The results demonstrated species- and elicitor-dependent modulation of plant metabolism, resulting in significant changes in bioactive compound accumulation that depended on the metabolite class. Elicitation effectively increased the levels of polyphenolic compounds (phenolic acids, flavonoids) [1,2] and selected isoquinoline alkaloids [3] while also enhancing the antioxidant activity of plant extracts [1-3]. In two of the analyzed species, *M. officinalis* and *H. perforatum*, these metabolic alterations were associated with changes in redox status, including increased H_2O_2 and $O_2^{\cdot-}$ accumulation, enhanced membrane lipid peroxidation, elevated free proline levels, and increased antioxidant enzyme activities [1,2]. Although reductions in selected photosynthesis-related parameters, including chlorophyll *a* fluorescence and photosynthetic pigment content, were observed in some cases, the application of elicitors generally did not adversely affect plant biomass or induce visible phytotoxic symptoms [1-3].

These findings provide new insights into the impact of chemically diverse elicitors on the accumulation of specialized metabolites and the physiological condition of medicinal plants. Furthermore, they support the use of elicitation as an effective strategy for the production of high-quality medicinal plant raw materials under controlled pot cultivation conditions.

Keywords: Specialized metabolites; medicinal plants; chitosan; selenium; salicylic acid; oxidative stress.

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PP–15

**HERACLEUM SOSNOWSKYI FRUIT DEVELOPMENT DISRUPTION BY
SPRAY-INDUCED GENE SILENCING OF GIBBERELLIN 2-OXIDASE****Saulė Rapalytė, Dominykas Laibakojis, Tautvydas Žalnierius, Sigita Jurkonienė**

State Scientific Research Institute Nature Research Centre, Akademijos str. 2, Vilnius, Lithuania

Presenting author: saule.rapalyte@gamtc.lt

The invasive species *Heracleum sosnowskyi* causes great threat to biodiversity, ecosystems and human health. While *H. sosnowskyi* maintains high seed productivity, the most common method to combat its spread remains the application of chemical herbicides. Since EU aims to halve pesticide use by 2030 under the Farm-to-Fork strategy, there is a need to look for more eco-friendly alternatives for fighting the spread of such species. A promising tool for weed management is spray-induced gene silencing (SIGS) based on RNA interference (RNAi) – a natural eucaryotic cellular defense mechanism against viruses. This process is built on post-transcriptional gene silencing upon exogenous double stranded RNA (dsRNA) entrance to the cell. Such approach could be utilized to silence *H. sosnowskyi*'s genes crucial for fruit development. Plant hormones gibberellins (GAs) play a key role in fruit development by stimulating cell elongation and division, regulating seed germination, inducing flowering. Gibberellin 2-oxidases (GA2ox) regulate GA catabolism by converting biologically active GAs to inactive products. While specific *H. sosnowskyi* GA2ox (HsGA2ox) role in fruit development remains unclear, findings from model plant species suggest these genes as potential silencing targets to obtain malformed and seedless *H. sosnowskyi* fruits. Thus, we aim to apply SIGS to silence the most abundant HsGA2ox to induce seedlessness.

Initially, *H. sosnowskyi*'s terminal umbel samples from 3 different plants were collected at -1, 0, 2, 3, 4, 5, and 8 days after anthesis (DAA). 100 mg of frozen plant material was used for total RNA extraction, which was further digested with DNase and used for cDNA synthesis to evaluate HsGA2ox gene expression levels. Primers for 13 different HsGA2ox genes were designed and applied in real-time quantitative PCR (RT-qPCR). After discovering the most expressed HsGA2ox during analyzed time period, we performed analysis to identify the most effective gene regions for RNAi. Such targets were amplified from cDNA with specific primers and cloned into the L4440 vector containing two T7 promoters. dsRNA was synthesized *in vivo* using *E. coli* HT115 strain and extracted with ethanol precipitation method. To encapsulate dsRNA and protect it from degradation for application in the field we synthesized layered double hydroxide (LDH) nanoparticles.

HsGA2ox gene expression analysis revealed that HsGA2ox1 was the most expressed at 2 and 3 DAA while HsGA2ox7 expression peaked at 5 DAA, making them the most expressed genes. After analyzing which regions of these genes could be the most effective for SIGS, we generated specific primers and after successfully cloning them into the vector we were able to synthesize specific ~300 bp dsRNAs. We determined dsRNA:LDH ratio needed for RNA encapsulation to be roughly 1:70 (w/w). The plants will be treated with LDH encapsulated HsGA2ox1 and HsGA2ox7 dsRNAs separately and combining them together. This research will investigate whether such HsGA2ox dsRNA constructs could successfully induce seedlessness in invasive *H. sosnowskyi* and provide a promising environmentally friendly biopesticide.

Keywords: *Heracleum sosnowskyi*; gibberellins; spray-induced gene silencing; RNA interference.**Acknowledgements:** This research was funded by the Research Council of Lithuania (LMTLT) (Project No. S-MIP-25-21).

PP–16

**INSIGHT INTO PHOTOCHEMICAL AND PHYSIOLOGICAL TRAITS
OF TWO WHEAT VARIETIES UNDER WATER DEFICIT**

**Violeta Peeva, Dilyana Doneva, Svetlana Misheva, Katelina Prokopova, Alexander Angelov,
Vladimir Aleksandrov**

Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. G. Bonchev Str., Bl. 21, 1113 Sofia, Bulgaria

Presenting author: vnp@abv.bg

Wheat productivity is highly sensitive to climatic and environmental variations. Thus, increasing incidences of drought associated with global warming are posing serious threats to food security. Drought stress during early growth stages is extremely unfavorable and severely limits the yields. Understanding varietal physiological responses to drought stress is critical for breeding climate-resilient genotypes with improved drought tolerance. For the aim of our study young seedlings from two winter wheat (*Triticum aestivum* L.) varieties—Katya and Zora, were subjected to drought for seven days, followed by rehydration. The experiments were conducted in pots in climatic chamber with controlled conditions. The photosystem II (PSII) function was assessed by chlorophyll a fluorescence (OJIP transients), thermoluminescence emissions and pigment content analysis. Under drought, Katya maintained functional PSII integrity with stable quantum efficiency and increased chlorophyll content, while Zora exhibited chlorophyll degradation. Fresh and dry weight declined significantly in Zora; recovery occurred after rehydration. Chlorophyll fluorescence revealed that varietal divergence was localized to the O–J phase of PSII photochemistry, indicating differences in reaction-center behavior confirmed by thermoluminescence emission. Katya demonstrated preserved PSII reaction-center density, balanced energy partitioning, homogeneous PSII populations and superior recovery capacity. Conversely, Zora showed reaction-center depletion, elevated energy dissipation, impaired electron transport beyond QA, and persistent PSII heterogeneity even after rehydration. Drought tolerance in the studied genotypes was associated with the maintenance of PSII structural integrity, efficient photochemical function and rapid recovery mechanisms. Our study demonstrated that the early PSII photochemistry kinetics and reaction-center stability provide valuable selection criteria for breeding programs, targeting drought resilience under changing climate conditions.

Keywords: Wheat; drought; photosynthesis; rehydration.

Acknowledgements: This research was funded by Bulgarian National Science Fund; grant number KP-06-H81/1.



PP–17

**ALTERATIONS IN THE CONTENT OF PHENOLIC COMPOUNDS AND FLAVONOIDS
IN WINTER WHEAT AFTER PHYTOPATHOGENIC FUNGUS INFECTION IS
MODULATED BY SPERMINE SEED PRIMING**

Zornitsa Katerova¹, Tsvetina Nikolova¹, Liliana Brankova¹, Elena Shopova¹, Iskren Sergiev¹, Ivo Yanashkov², Dessislava Todorova¹

¹Institute of Plant Physiology and Genetics - Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., Bldg. 21, 1113 Sofia, Bulgaria

²Institute of Soil Science, Agrotechnologies and Plant Protection “Nikola Poushkarov”, Agricultural Academy, Shose Bankya 7, 1331 Sofia, Bulgaria

Presenting author: zornitsa@bio21.bas.bg

The infection with soil-borne pathogen *Fusarium culmorum* causes significant losses worldwide of the productivity of wheat (*Triticum aestivum* L.). At the cellular level, the contagion with phytopathogen is accompanied by the occurrence of adverse oxidative events in the plant organisms, in which reactive oxygen species (ROS) are formed. The ROS over-accumulation disrupts key physiological processes and negatively affects the vital status of the plants. To counteract the stress outcomes infected plants trigger their endogenous defense system, which involves enzymatic and non-enzymatic antioxidants, including the production of secondary metabolites with antimicrobial activity. Seed priming with environmentally friendly compounds is gaining attention for its ability to enhance plant resilience under biotic stress. Our study investigated the possibility of seed priming with the polyamine spermine, a naturally occurring plant growth regulator, to modulate the content of phenolic compounds and flavonoids in the Bulgarian wheat cultivar Sadovo-1 under a direct seed surface infection or soil contamination with *Fusarium culmorum*. Our results showed that the phytopathogen treatment significantly decreased the content of secondary metabolites with antioxidant activity at early stage of infection. Later, when the disease symptoms developed, phenolics and flavonoids tended to increase with a considerable rise in seed-infected plants. Spermine seed priming facilitates a more robust defense response against *F. culmorum* evidenced by adjusted content of phenolic compounds and flavonoids in injured plants of both disease stages.

Keywords: *Fusarium culmorum*; secondary metabolites; seed pre-treatment; spermine.

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PP–18

PATHOGENIC POTENTIAL OF *FUSARIUM OXYSPORUM* ISOLATES FROM TOMATO AND WHEAT ROOTS, ASSESSED ON DETACHED TOMATO FRUITS**Zornitsa Stoyanova¹, Agnieszka Jamiołkowska², Weronika Kurska², Ewa Zalewska³**¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., Bldg. 21, 1113 Sofia, Bulgaria²Department of Plant Protection, University of Life Sciences in Lublin, 7, Akademicka Str., 20-069 Lublin, Poland³Department of Vegetable and Herbs, University of Life Sciences in Lublin, 50A, Doświadczalna Str., 20-280 Lublin, PolandPresenting author: agnieszka.jamiolkowska@up.edu.pl

Fungal diseases caused by soil- and seed-borne pathogens are one of the most common problems in tomato cultivation. *Fusarium oxysporum* Schltdl. is a root-infecting fungus but can also cause tomato fruit rot especially after harvest.

The aim of this study was to prove virulence of selected *Fusarium oxysporum* isolates from roots of tomato and wheat on detached tomato fruits.

Material and methods: Eight *F. oxysporum* isolates (Fox) were involved in the study from roots of tomato (PL7, P6, PK8, PK9, PK12) and winter wheat (PO22a, PO34, PO50), deposited in the microbial collection of the Department of Plant Protection, University of Life Sciences in Lublin (Poland). The pathogenicity test was performed on detached fruits of plum shaped tomato (*Solanum lycopersicum* L.), cv. Slivka red. The isolates were cultivated in liquid Synthetic Nutrient Deficient Agar (SNA) for 13 days. Six tomato fruits for each *F. oxysporum* isolate were cleaned, disinfected and pricked, and then each wound was inoculated with 20 µl of fungal suspension. The fruits serving as a control were wounded and dripped with 20 µl of sterile distilled water. After treatment, fruits were kept in closed plastic containers placed in a humidity chamber in the light at room temperature (20-22°C). Daily observations were performed on symptom development for 10 days when the size of lesions was measured. Macroscopic observation on the type of mycelial growth was made 21 days after inoculation.

Results: Two days after inoculation, white mycelium developed on the surface of the tomato fruits on and around the inoculation hole. On the 10th day, most of the infected fruits showed symptoms of rot. The spots were round to oval, concave, with white mycelium on the surface of the spot and cracking of the epidermis depending on disease severity. Lesion measurements showed that the isolates caused infection on the fruit with varying severity. The spots caused by the PK6 isolate had the largest average size of 35.5 x 33.2 mm, while those caused by the PO22a isolate had the smallest size of 4.0 x 3.7 mm. Generally, the spots provoked by isolates obtained from wheat roots were smaller and limited by a dark necrotic ring. There was no disease development on the control fruits. Re-isolations from diseased fruits and taken from them seeds showed development of fungal mycelium typical for each isolate, confirming the purity and identity of the fungal cultures being studied. No fungal growth was observed from the control fruits and their seeds.

Conclusions: Experiments showed that all isolates from tomato roots were pathogenic for detached tomato fruits. The most virulent were the isolates from tomato roots PK6, PK12 and PK8. Isolates from wheat roots were able to cause infection, but with limited development. In the presence of Fox infection in the soil in monocultural tomato cultivation, there is a real threat of developing the disease rot on the fruits before and/or after harvesting the produce, while after a wheat predecessor this risk is significantly lower.

Key words: *Fusarium oxysporum* Schltdl.; *Solanum lycopersicum*; tomato fruit rot; fungal pathogen; virulence.

Acknowledgements: This research was supported by IAEA fellowship event EVT2505575 under project BUL5020.

PP–19

**SCREENING OF BIOSTIMULANTS TO INCREASE CELLULOSE CONTENT
IN FLAX FIBERS**

**Sigita Jurkonienė¹, Virgilija Gavelienė¹, Rima Mockevičiūtė¹, Elžbieta Jankovska-Bortkevič¹,
Tautvydas Žalnierius¹, Vaidevutis Šveikauskas¹, Liudmyla Kozeko²**

¹Laboratory of Plant Physiology, State Scientific Research Institute Nature Research Centre, Akademijos str. 2, 08412 Vilnius, Lithuania

²Laboratory Department of Cell Biology and Anatomy, M.G. Kholodny Institute of Botany of the National Academy of Sciences of Ukraine, Tereshchenkivska Str. 2, 01601 Kyiv, Ukraine

Presenting author: sigita.jurkoniene@gamtc.lt

Common flax (*Linum usitatissimum* L.) is an important source of high-quality natural fiber, whose main structural component is cellulose. Increasing the cellulose content of flax fibers is of particular interest for improving fiber quality and expanding its applications in the textile and other fiber-based industries. The aim of this study was to identify a biostimulant capable of enhancing cellulose accumulation in flax fibers and to investigate the physiological and biochemical mechanisms underlying this effect. Based on literature data and our previous research, several biologically active compounds, including proline, γ -aminobutyric acid (GABA), polyamines, and phytohormones, were selected for screening. Twelve-day-old flax seedlings were treated exogenously with the selected compounds. Plant growth and development were evaluated at seven-day intervals following treatment using morphometric, microscopic, biochemical, and molecular analyses. Changes in total monosaccharide and cellulose contents were also determined during the 56-day growth period. Initial morphometric analyses demonstrated that GABA applied at a concentration of 10 mM was the most effective treatment for promoting the accumulation of both fresh and dry biomass. Subsequent analyses showed that GABA (10 mM) significantly enhanced total monosaccharide accumulation and increased cellulose content in flax during the 56-day growth period. These findings suggest that GABA-induced changes in carbohydrate metabolism may contribute to enhanced cellulose biosynthesis and accumulation in developing flax fibers. Further studies are being conducted to elucidate the molecular and physiological mechanisms underlying the observed response.

The results identify GABA as a promising biostimulant for improving cellulose accumulation in flax and provide a basis for further development of biostimulant-based cultivation strategies aimed at obtaining high-quality flax fiber.

Keywords: *Linum usitatissimum*; biostimulants; GABA; total monosaccharide content; cellulose.

Acknowledgements: This project has received funding from the Research Council of Lithuania (LMTLT), Lithuania – Ukraine program, agreement No S-LU-26-7 and has been performed in a cooperation with M.G. Kholodny Institute of Botany of the National Academy of Sciences of Ukraine.

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PP-20

EVALUATION OF LOCAL MELON GENOTYPES FOR RESISTANCE TO APHID (*APHIS GOSSYPPII* GLOVER) AND ZUCCHINI YELLOW MOSAIC VIRUS (ZYMV) AND DEVELOPMENT OF A RESISTANT LINE

Yıldız Dogan¹, Abdullah Unlu¹, Musa Kirisik¹, Kubra Yildiz¹, Mine Unlu¹, Levent Keskin¹, Muhammad Azhar Nadeem², Faheem Shehzad Baloch²

¹Batı Akdeniz Tarımsal Araştırma Enstitüsü Müdürlüğü, Antalya, Türkiye

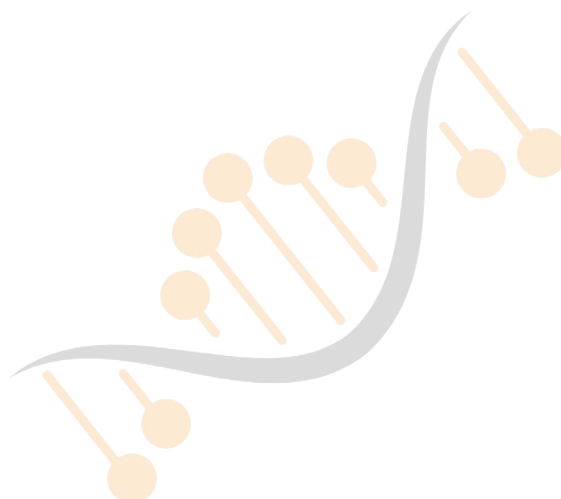
²Department of Biotechnology, Faculty of Science, Mersin University, Mersin, Türkiye

Presenting author: balochfaheem13@gmail.com

There are many pest species and diseases that limit melon production. Aphid (*Aphis gossypii*), a polyphagous pest, and Pumpkin yellow mosaic virus (ZYMV) are among the most economically damaging species. The most effective way to combat aphids and ZYMV is to use of resistant varieties. However, limited progress has been made in breeding studies so far. In this regard, breeding studies aimed at providing plants with resistance to this pest and disease are very important. Within the scope of this project, it is aimed to select resistant plants and obtain resistant lines by mechanical inoculation and molecular analysis against this pest and disease in local melon genotypes. In the study, resistance will be transferred by backcross breeding method and can also be tested molecularly using one ZYMV primers and one aphid primer. In addition, POGP and SRAP marker techniques were used to reveal the genetic relationships among 184 local melon genotypes in the BATEM melon gene pool. Of the 22 SRAP primer pairs, 14 were found to be polymorphic, and 12 of the 22 POX primer pairs were polymorphic and were used in the analysis of 184 local melon genotypes. Population structure analysis revealed the genetic relationship among the evaluated genotypes.

Keywords: Melon;, *Aphis gossypii*; ZYMV; resistance breeding; molecular markers; Vat.

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PP–21

INTEGRATIVE miRNA–mRNA REGULATORY SIGNATURES IN BRAF-STRATIFIED METASTATIC MELANOMA

Iliyan Pochileev, Ivanka Dimova

Department of Medical Genetics, Medical University Sofia, Bulgaria

Presenting author: iliyan.pochileev@gmail.com

Introduction: Metastatic melanoma is characterized by pronounced molecular heterogeneity driven by the accumulation of genetic and epigenetic alterations. A key determinant of this heterogeneity is the mutational status of gene BRAF, which defines dependency on specific signaling pathways and influences tumor progression as well as therapeutic response. In addition to classical oncogenic drivers, microRNAs (miRNAs) represent an essential regulatory layer that modulates transcriptomic programs involved in tumor–microenvironment interactions, angiogenesis, and cellular plasticity. However, their integrated role in BRAF-stratified metastatic melanoma remains insufficiently characterized.

Materials and Methods: Formalin-fixed, paraffin-embedded (FFPE) biopsy material was analyzed under inclusion criteria requiring >30% tumor cellularity. A total of 8 samples were included: 3 healthy skin controls and 5 metastatic melanoma samples, all *BRAF*-mutant. Differential expression analysis between tumor and control groups was performed using qRT-PCR in the context of BRAF status. Expression profiling of 827 miRNAs and 769 mRNAs was conducted using the NanoString platform, followed by normalization and fold-change analysis. In parallel, multiplex omics-based profiling of selected miRNA and mRNA transcripts was performed. Statistical significance was set at $p=0.01$ – 0.05 , with fold-change thresholds applied to identify biologically relevant alterations. Integrative bioinformatic analysis was used to assess miRNA–mRNA regulatory interactions.

Results: Metastatic melanoma samples demonstrated significant overexpression of hsa-miR-296-5p, hsa-miR-146a-5p, hsa-miR-30b-5p, and hsa-miR-29b-3p ($FC>2$), accompanied by marked downregulation of *FGFR1* ($FC=0.47$), *NOTCH1* ($FC=0.40$), *CXCL12* ($FC=0.28$), *PDGFRB* ($FC=0.41$), and *ANGPTL4* ($FC=0.38$) ($FC<0.5$; $p=0.01$ – 0.05). The observed inverse expression pattern was preserved within BRAF-mutant samples, suggesting the presence of a coordinated post-transcriptional regulatory mechanism, likely mediated by miRNAs. Correlation analysis confirmed the existence of synchronized miRNA-mediated repression of genes functionally involved in angiogenesis, stromal signaling, and regulation of cellular plasticity.

Discussion: The obtained data demonstrate a coherent miRNA regulatory module contributing to transcriptomic remodeling in metastatic melanoma. The upregulated miRNAs appear to target functionally related genes involved in receptor tyrosine kinase signaling pathways, chemokine cascades, and tumor–stroma communication. Importantly, the preservation of this pattern across BRAF-mutant samples suggests that miRNA-mediated regulation operates independently of classical oncogenic drivers, representing a parallel layer of control in tumor adaptation. The reduced expression of *FGFR1* and *PDGFRB* indicates suppression of growth factor signaling, while decreased *NOTCH1* and *CXCL12* suggest alterations in differentiation and cellular migration processes. Downregulation of *ANGPTL4* further supports changes in vascular and metabolic adaptation within the tumor microenvironment. These findings highlight the potential of miRNA networks as a key regulatory layer and underscore the need for further functional validation studies to elucidate their mechanistic role in melanoma progression.

Keywords: miRNA; metastatic melanoma; BRAF; microRNA biomarkers; nanostring technology.

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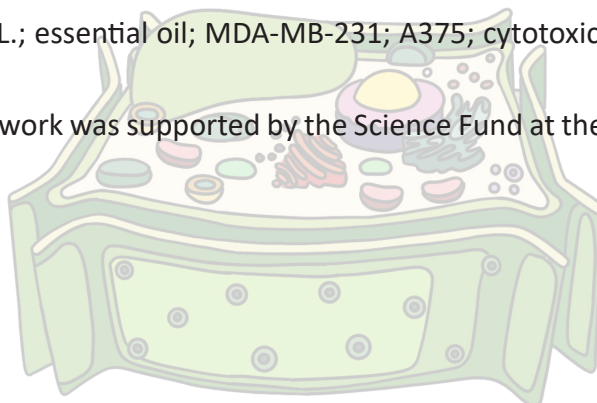
PP-22

EFFECTS OF *LAURUS NOBILIS* L. ESSENTIAL OIL ON CELL VIABILITY AND PROLIFERATION OF BREAST AND SKIN CANCER CELL LINES**Zlatina Gospodinova¹, Georgi Antov¹, Silvia Stamova², Emilia Georgieva³**¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, "Acad. Georgi Bonchev" Str., Bl. 21, Sofia 1113, Bulgaria²Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Medical University of Varna, 55 Marin Drinov str., 9002 Varna, Bulgaria³Training sector "Medical Laboratory Assistant", University of Varna, 55 Marin Drinov str., 9002 Varna, BulgariaPresenting author: zlatina.go@abv.bg

Essential oils (EOs) are aromatic secondary metabolites with a major role in plant protection against microorganisms and insects, communication between plants, and signaling in response to environmental stimuli. These substances with complex composition exhibit various pharmacological properties, including antioxidant, antibacterial, fungicidal, virucidal, anti-diabetic, anti-inflammatory, and others. Essential oils have attracted growing interest as potential promising candidates in anticancer research due to their ability to affect multiple molecular pathways involved in tumor development. Aromatic medicinal plant *Laurus nobilis* L. (Lauraceae), is an evergreen shrub or tree native to the Mediterranean but widely cultivated in many regions worldwide. The plant possesses a broad spectrum of biologic activities, and is commonly used in cooking, cosmetics, traditional medicine and pharmacy thanks to the volatile oils and polyphenols it contains. The objective of the present study was to assess cytotoxic and cytostatic potential of essential oil from *Laurus nobilis* on *in vitro* model system of breast cancer cell line, MDA-MB-231, and malignant melanoma cell line, A375. Non-cancerous skin cell line BJ was used in the analysis to evaluate the selectivity in the action of the *L. nobilis* EO. Effects of the plant substance on cell viability and growth were examined by MTT assay performed in parallel with cell morphology observation under light microscope, after 24 and 72 h treatment period in a wide range of concentrations varying from 0.0625 to 2%. Anticlonogenic capacity of the herb EO was investigated through colony-forming assay after 7 days treatment duration. The obtained results detected strong and selective cytotoxic activity and significantly higher antiproliferative potential of the essential oil towards both cancer cell lines, more pronounced against MDA-MB-231 cells. A slighter effect on cell survival and proliferation rates was found in the normal BJ cell line in comparison to the cancer cells. Light microscopy observations indicated considerable morphological alterations and significantly reduced density of cancer cells monolayer even in the lowest applied concentration. The clonogenic ability of breast and melanoma cancer cells was also strongly inhibited after treatment with *L. nobilis* EO, highlighting the strength of the substance's antiproliferative potential. Future studies will be focused on more detailed investigations of mechanisms of anticancer action of the plant EO in the studied model system.

Keywords: *Laurus nobilis* L.; essential oil; MDA-MB-231; A375; cytotoxic potential; antiproliferative effect.

Acknowledgements: This work was supported by the Science Fund at the Medical University - Varna under project No 24027.



PP-23

DIFFERENTIAL EXPRESSION OF MICRO RNAS IN SYMPTOMATIC AND ASYMPTOMATIC *PETUNIA HYBRIDA*

Stoyanka Jurak, Veselin Baev, Elena Apostolova-Kuzova, Rumyana Valkova, Galina Yahubyan, Mariyana Gozmanova

University of Plovdiv, 24, Tzar Assen Str., 4000 Plovdiv, Bulgaria

Presenting author: tanyajurak@uni-plovdiv.bg

MicroRNAs (miRNA) are key post-transcriptional regulators of gene expression and play important roles in plant growth, development, and responses to biotic stress. To investigate miRNA expression changes associated with virus related symptoms in *Petunia hybrida*, small RNA libraries from symptomatic and asymptomatic samples were analyzed through high-throughput sequencing. MicroRNA identification in *P. hybrida* was performed based on sequence homology with known microRNAs from representatives of the family *Solanaceae* deposited in miRBase.

The results revealed that symptomatic *P. hybrida* samples exhibited altered miRNA expression profiles compared with asymptomatic samples. Among the identified microRNAs, miR399a showed the strongest upregulation, whereas miR319a, miR482a, miR396a, and miR162 were downregulated.

To validate the sequencing results, RT-qPCR analysis was performed for selected miRNA, including miR399a, miR396a, miR398a-3p, and miR166a. Relative expression levels were calculated using the $2^{-\Delta\Delta Ct}$ method, with U6 used as an endogenous control. The RT-qPCR analysis confirmed the small RNA sequencing results.

The specific changes observed in the miRNA expression profile of symptomatic *P. hybrida* samples compared with asymptomatic samples suggest the involvement of these regulatory molecules in the host plant response to viral infection. Future analyses will focus on the *in silico* identification of target genes of the differentially expressed miRNA and the evaluation of their expression.

Keywords: *Petunia hybrida*; miRNA; high-throughput sequencing.

Acknowledgements: The research was supported by National Science Fund under project KP-06-H76/10.



PP–24**NATURAL ANTI-BIOFILM AGENTS FROM *ACHILLEA* SPP.: EFFECTS ON BIOFILM FORMATION, MOTILITY, AND BACTERIAL VIABILITY**

Tsvetelina Paunova-Krasteva¹, Petya D. Dimitrova¹, Viktoria Ivanova², Antoaneta Trendafilova²

¹The Stephan Angeloff Institute of Microbiology, Bulgarian Academy of Sciences

²Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences

Presenting author: pauny@abv.bg

Biofilms represent microbial communities which, due to their structural organization, are inherently protected against external stress factors and exhibit high resistance to antimicrobial agents. Their resistance to antibiotics is achieved through a combination of a physical barrier, metabolic heterogeneity, and the activation of specific resistance-related genes. Biofilm inhibition is considered a key strategy in combating chronic infections, as disruption of the polymeric matrix and suppression of intercellular communication (quorum sensing) can enhance susceptibility to therapeutic agents. The present study focuses on extracts derived from representatives of the genus *Achillea*. Plant extracts were prepared via sequential extraction using solvents such as chloroform and methanol at room temperature. The experimental design included the application of these extracts to inhibit biofilm formation in five Gram-negative and Gram-positive bacterial strains. The crystal violet assay demonstrated dose-dependent inhibition in several of the tested strains. To further assess virulence-associated traits in Gram-negative bacteria, a bacterial motility inhibition assay was performed. Compared to untreated controls, a measurable inhibitory effect on motility was observed in some of the strains. In addition, a Live/Dead assay was conducted to evaluate bacterial viability. The obtained results demonstrate the effectiveness of the applied extracts, highlighting their potential as a promising source of natural antibiofilm agents with therapeutic applications in the control of biofilm-associated infections.

Keywords: Biofilms; plant extracts; bacterial viability; bacterial motility.

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PP–25

MICROALGAL CARBONIC ANHYDRASE FOR BIOSEQUESTRATION OF EXCESS ATMOSPHERIC CARBON DIOXIDE

Ivan Iliev, Liliana Gigova, Tanya Toshkova-Yotova

Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Department of Experimental and Applied Algology, Acad. G. Bonchev Str., bldg. 21, 1113, Sofia, Bulgaria

Presenting author: ivaniliev@bio21.bas.bg

Development of efficient technologies for capturing and utilizing excess carbon dioxide (CO₂) in the atmosphere is essential for mitigating global warming. Carbonic anhydrase (CA), an enzyme capable to significantly accelerate the reversible hydration of CO₂, has emerged as a promising catalyst for biotechnological carbon sequestration.

Photosynthetic microalgae are among the most efficient biological systems for atmospheric CO₂ fixation and sustainable CA producers. Elevated CO₂ levels in microalgal cultivation lead to improved photosynthetic performance and biomass productivity, while suppressing CA expression and enzymatic activity. This limitation can be addressed through a two-stage cultivation strategy, that optimizes biomass accumulation during the first stage (CO₂-enriched conditions) and promotes biosynthesis/activity of the target enzyme in the second stage (low CO₂).

In this study, the green microalgae *Chlorella kessleri* and *Coelastrella* sp. BGV were cultivated under intensive laboratory conditions using air supplemented with 2% CO₂ until the mid-exponential growth phase was reached. Then, the addition of CO₂ was discontinued, and the cultures were grown under ambient air. Carbonic anhydrase activity was determined at the end of the first cultivation stage and after 6, 24, and 48 h of cultivation in the second stage. The highest CA activity was recorded for *Chlorella kessleri* grown for 24 h under ambient air conditions, reaching 7.6 ± 0.4 WAU mg⁻¹ protein, which corresponds to a 2.2-fold increase compared to the culture supplied with 2% CO₂.

These demonstrate that *Chlorella kessleri* represents a reliable source of carbonic anhydrase, the enzyme with considerable potential for future biotechnological applications, particularly as an efficient biocatalyst for capturing the increasing amounts of CO₂ in the atmosphere, thus participating in unwanted climate change mitigation. Furthermore, the successful implementation of the proposed two-stage cultivation strategy provides an effective approach to enhance the production/activity of microalgal carbonic anhydrase and is a valuable contribution to the development of sustainable, environmentally friendly carbon capture and utilization biotechnologies.

Keywords: Green microalgae; carbonic anhydrase (CA); biosequestration.



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HIGH-PRESSURE DISINTEGRATION OF *CHLORELLA VULGARIS* AND *PORPHYRIDIVM AERUGINEUM* FOR THE EXTRACTION OF BIOACTIVE COMPOUNDS

Veronika Deleva^{1,2}, Juliana Ivanova¹, Nadya Agayn-Stoyanova¹, Gloria Georgieva²

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Sofia, Bulgaria

²FSB Products Ltd., Sofia, Bulgaria

Presenting author: delevaveronika93@gmail.com

Microalgae are valuable sources of proteins, carbohydrates, lipids, pigments, antioxidants, and plant growth-regulating compounds. However, efficient extraction of intracellular components requires disruption of cellular structures. This study evaluates high-pressure homogenization as a disintegration method for two taxonomically and morphologically distinct microalgae: the green microalga *Chlorella vulgaris* RJ1 and the red microalga *Porphyridium aerugineum* HINDAK 1983/2.

Microalgal cultures are treated at pressures of 300, 500, and 700 bar for 5 or 10 min, using biomass concentrations ranging from 0.5 to 4.0 g/L dry matter. Cell disruption efficiency strongly depends on pressure and biomass concentration, whereas treatment duration has a less pronounced effect. Disintegration efficiency reaches 97.08% for *C. vulgaris* and 90.04% for *P. aerugineum* at 700 bar, 10 min, and 4.0 g/L biomass.

A treatment regime of 700 bar for 5 min is selected for the preparation of disintegrates for subsequent biological assays. This regime provides high disruption efficiency while limiting the temperature increase to approximately 37°C, thereby reducing the risk of degradation of temperature-sensitive bioactive compounds. The results demonstrate that high-pressure homogenization is an effective method for releasing intracellular metabolites from microalgal biomass.

Keywords: Microalgae; cell disintegration; high-pressure homogenization; *Chlorella vulgaris*; *Porphyridium aerugineum*; intracellular metabolites; biostimulants.



PP–27

MUTUAL EFFECTS OF MULCHING ON THE RHIZOSPHERE AND CROP YIELD OF POTATO AND BUSH BEAN

Dimitar Danchev¹, Slavil Peykov¹, Momchil Paunov¹, Ana-Maria Nedelcheva¹, Desislava Mantovska¹, Grigor Zehirov², Krasimir Rusanov³, Mila Rusanova³, Aneliya Raycheva¹, Miroslava Zhiponova¹

¹Faculty of Biology, Sofia University “St. Kliment Ohridski”, Sofia, Bulgaria

²Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Sofia, Bulgaria

³Agrobiointitute, Agricultural Academy, Sofia, Bulgaria

Presenting author: zhiponova@biofac.uni-sofia.bg

Utilizing natural resources and regenerative practices is a key element in the sustainable agricultural production of food. In our work, we estimated the impact of straw cover of the soil (mulch) on the rhizosphere and the yield of two crop species – potato (*Solanum tuberosum*) and bush bean (*Phaseolus vulgaris*). The physicochemical parameters and microbiome of the crops' rhizosphere zone in mulched and non-mulched variants were investigated and compared to a control soil without plants. The respective effect of mulching on the tuber and bean yield quantity and quality was explored. The overall results highlighted a positive correlation between mulching and the soil redox potential (ORP), and bacterial and fungal diversity. Upon mulching, the potato tuber yield and starch content were significantly increased, while in beans, the yield was not changed; however, the amino acid level was enhanced. In both species, mulching upregulated the soluble sugars. Curiously, mulching led to a drop of total phenolics in potatoes and increased the germination potential of beans. Our data revealed both common and differential effects of crop type and mulching on the plant rhizosphere and yield parameters.

Keywords: Regenerative agriculture; soil redox potential; soil metagenomics; *Solanum tuberosum* tubers; *Phaseolus vulgaris* beans.

Acknowledgements: This research was funded by the Bulgarian National Science Fund, Ministry of Education and Science, Grant No. KP-06-N86/4/06.12.2024.



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CHEMICAL PROFILE AND ANTIBACTERIAL ACTIVITY OF *CORIANDRUM SATIVUM* L. ESSENTIAL OIL AGAINST CLINICALLY RELEVANT RESPIRATORY BACTERIAL PATHOGENS

Polina Petrova¹, Sylvia Stamova², Nadezhda Ivanova³, Neli Ermenlieva⁴, Emilia Georgieva⁵

¹Faculty of Pharmacy, Medical University of Varna, 55 Marin Drinov str., 9002 Varna, Bulgaria

²Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Medical University of Varna, 55 Marin Drinov str., 9002 Varna, Bulgaria

³Department of Pharmaceutical Technologies, Faculty of Pharmacy, Medical University of Varna, 55 Marin Drinov str., 9002 Varna, Bulgaria

⁴Department of Microbiology and Virusology, Medicine Faculty, Medical University of Varna, 55 Marin Drinov str., 9002 Varna, Bulgaria

⁵Training sector "Medical Laboratory Assistant", University of Varna, 55 Marin Drinov str., 9002 Varna, Bulgaria

Presenting author: sylvia²stamova@yahoo.com

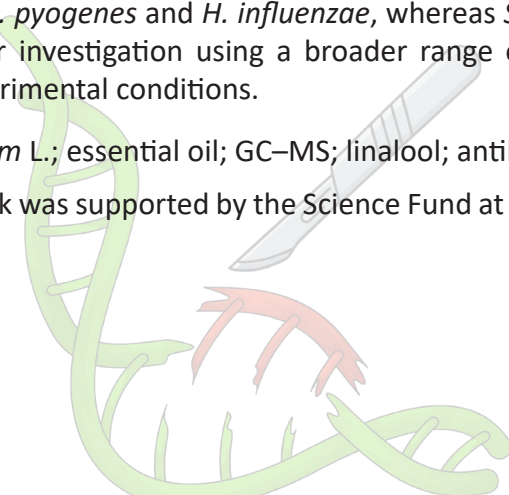
Antibiotic resistance remains a major challenge in modern medicine and has intensified research into natural products with potential antimicrobial activity. *Coriandrum sativum* L. essential oil is of particular interest because of its rich content of bioactive volatile compounds. The aim of the present study was to characterize the chemical composition of coriander essential oil by gas chromatography–mass spectrometry (GC–MS) and to evaluate its antibacterial activity against selected respiratory bacterial pathogens.

GC–MS analysis identified 14 volatile compounds, accounting for 99.79% of the total chemical composition of the oil. Linalool was the predominant constituent (79.50%), followed by α -pinene (8.48%), geranyl acetate (2.60%), γ -terpinene (2.50%), camphor (2.34%), limonene (1.41%), and p-cymene (1.07%). The resulting chemical profile was characteristic of linalool-rich essential oil obtained from the fruits of *Coriandrum sativum* L. Antibacterial activity was assessed by broth microdilution using twofold serial dilutions. The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) were determined against the reference strains *Streptococcus pyogenes*, *Haemophilus influenzae*, and *Staphylococcus aureus*. The highest susceptibility was observed for *S. pyogenes* and *H. influenzae*, with MIC and MBC values of 0.6% for both species. For *S. aureus*, the MIC and MBC were 2.5% and 5%, respectively, indicating lower susceptibility to the tested essential oil.

The findings demonstrate species-dependent differences in susceptibility to *Coriandrum sativum* L. essential oil. Under the conditions of the present study, the oil exhibited more pronounced antibacterial activity against *S. pyogenes* and *H. influenzae*, whereas *S. aureus* was less susceptible. These results support further investigation using a broader range of bacterial strains and more extensively standardized experimental conditions.

Keywords: *Coriandrum sativum* L.; essential oil; GC–MS; linalool; antibacterial activity; MIC; MBC.

Acknowledgements: This work was supported by the Science Fund at the Medical University - Varna under project No 24027.



PP–29

SHOOT MULTIPLICATION AND ANTIOXIDANT ENZYME ACTIVITY OF *ARNICA MONTANA* L. CULTURED IN A TEMPORARY IMMERSION SYSTEM UNDER ELICITATION WITH SALICYLIC ACID

Mariana Sichanova, Maria Petrova, Maria Geneva, Kamelia Miladinova-Georgieva, Margarita Dimitrova, Lyudmila Dimitrova

Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Sofia, Bulgaria

Presenting author: m.sichanova@abv.bg

Background: Establishing *in vitro* cultures of medicinal plants is crucial, as it facilitates rapid, controlled multiplication of valuable species, preserves their genetic material, and generates beneficial bioactive compounds regardless of climatic conditions or seasons. *Arnica montana* L. (Asteraceae) is a medicinal plant species with increasing market demand requiring the development of an effective method for its mass propagation. TIS RITA® is a system that combines the advantages of both solid and liquid media and provides maximum gas exchange and increased nutrient uptake.

Objectives: The study aimed to investigate the influence of salicylic acid addition to liquid MS medium on physiological and biochemical parameters in *in vitro* propagated *Arnica montana* using TIS RITA®. It is assumed that the rate of shoot propagation will be significantly increased and the system will ensure the production of healthy plants of high quality.

Methods: Control shoots were grown in liquid MS nutrient medium supplemented with 0.5 mg/L BAP and 3% (w/v) sucrose without elicitor. The stock solution of SA was filter-sterilized through a 0.22 µm syringe Millipore filter (Minisart® Sartorius, Goettingen, Germany) and then added to the autoclaved liquid MS medium containing 0.5 mg/L BAP aseptically at the concentrations of 50, 100, or 200 µM. In order to measure enzymatic antioxidant activity in *A. montana* plantlets, a number of spectrophotometric methods were used.

Results: The results of the study show that the addition of SA has a beneficial effect on the morphological parameters of *A. montana* grown in a Temporary Immersion System, enhancing its antioxidant capacity. The developed method for mass multiplication of *A. montana* significantly enhances the *in vitro* process and could be utilized to produce planting material with antioxidant properties, thereby contributing to the conservation of the species.

Keywords: Micropropagation; temporary immersion system; salicylic acid; antioxidant enzymes.



PP–30

APPLICATION OF SiO₂-NANOPARTICLES ON SHOOT PROLIFERATION AND ANTIOXIDANT METABOLITES OF *ARNICA MONTANA* L.

Mirena Chakarova, Maria Geneva, Kamelia Miladinova-Georgieva, Mariana Sichanova, Lyudmila Dimitrova, Margarita Dimitrova, Maria Petrova

Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. G. Bonchev Street, Bldg. 21, 1113 Sofia, Bulgaria

Presenting author: dmirena@abv.bg

Arnica montana L. is a perennial herbaceous plant typical of high mountain areas of Europe. The plant is protected in Belgium, France, Germany, Italy, Poland, and some regions of Switzerland. *A. montana* has been known in European folk medicine for centuries, and the bioactive compounds identified to date are sesquiterpene lactones of the pseudoguaiane type, phenols, glucuronides, flavonoids, essential oils, fatty acids, etc. The specificity of the chemical composition of the species defines it as a medicinal plant, used traditionally to treat various ailments due to its antifungal, antibacterial, antiseptic, anti-inflammatory, and antioxidant properties. Silica is the common form of Si in soil. Nanoparticles are relatively more soluble and reactive due to their small size (1-100 nm) and large surface area. SiO₂NPs reduce the uptake and accumulation of heavy metals, increase the levels of macro- and microelements, reduce the accumulation of free radicals, stabilize the photosynthetic apparatus, mitigate oxidative damage to the plasma membrane, and improve the antioxidant defense system. The present study aims to assess the effect of SiO₂ nanoparticles on the *in vitro* propagation of *A. montana* and synthesized antioxidant metabolites in the obtained shoots. For shoot induction and proliferation, nodal explants were used. Stem explants (1 cm) were excised from three-month *in vitro* shoots and placed onto MS medium supplemented with various concentrations (50, 100, and 200 mg/L SiO₂NPs).

The SiO₂NP included in the nutrient medium positively affected the morphogenetic potential of *A. montana*. An increase in the number of shoots per explant was noted at all tested SiO₂NP concentrations compared to the control. The best results were obtained using MS medium supplemented with 50 mg/L SiO₂NP. The antioxidant activity measured by ferric reducing antioxidant power (FRAP), as well as the content of metabolites with antioxidant power in the shoots of *A. montana*, increased with SiO₂NP treatment.

Keywords: Multiplication; nanoparticles; total phenolics; total flavonoids; water-soluble antioxidant metabolites; lipid-soluble antioxidant metabolites; FRAP.

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**EFFECTS OF PLANT EXTRACTS ON THE DIVERSITY OF *FUSARIUM* SPECIES –
FIELD EXPERIMENT****Weronika Kursa¹, Agnieszka Jamiołkowska¹, Aleksandra Nucia², Sylwia Okoń²**¹Department of Plant Protection, University of Life Sciences in Lublin, Lublin, Poland²Institute of Plant Genetics, Breeding and Biotechnology, University of Life Sciences in Lublin, Lublin, PolandPresenting author: weronika.kursa@up.edu.pl

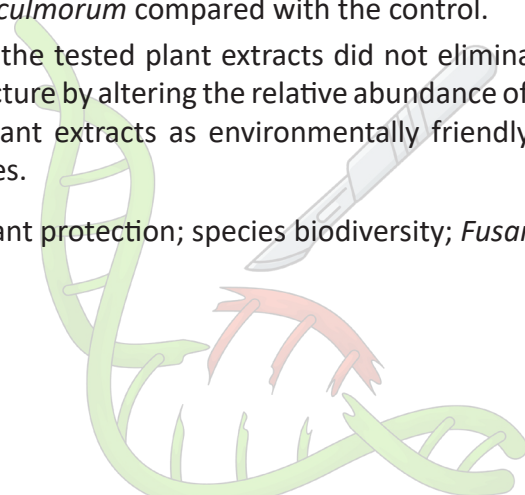
The increasing importance of biological plant protection strategies has stimulated the search for environmentally friendly solutions to suppress plant pathogenic fungi while maintaining the microbial balance of agroecosystems. The aim of this study was to evaluate the effect of three foliar applications of hemp inflorescence extract (H) and a mixture of extracts obtained from hemp inflorescences, sage leaves, and tansy leaves (M) on the species composition of *Fusarium* communities colonizing the stem base of winter wheat (*Triticum aestivum* L.) cv. 'Venecja'. Water-treated plants served as the absolute control (C), whereas tebuconazole (250 g L⁻¹, a triazole fungicide) was used as the relative control (F). Field experiments were conducted during the 2022/23 and 2023/24 growing seasons in Skrzynice Kolonia (Lublin Voivodeship, Poland).

Fungal isolates were initially identified based on their morphological characteristics using classical mycological analysis. Mycological analysis resulted in the recovery of fungi colonizing the stem base of winter wheat, of which 53.87% belonged to the genus *Fusarium*, represented by 26 isolates. A total of 26 representative isolates were subsequently subjected to molecular identification through sequencing of the ITS rDNA and translation elongation factor 1-alpha (TEF1) regions, followed by phylogenetic analysis using the maximum likelihood method and comparison with reference *Fusarium* sequences deposited in GenBank. Among the 26 *Fusarium* isolates, the following species were identified: *F. avenaceum*, *F. culmorum*, *F. oxysporum*, *F. poae*, *F. sporotrichioides* and *F. tricinctum*.

Fusarium avenaceum remained the dominant species irrespective of the protection strategy applied in both growing seasons. *F. poae* was the co-dominant species in the 2022/23 season, whereas *F. oxysporum* became the second most abundant species in the 2023/24 season across all experimental treatments. The most pronounced changes in *Fusarium* community composition were observed in the 2022/23 season, when *F. culmorum* and *F. sporotrichioides* were not detected in the hemp extract (H) and mixed-extract (M) treatments. Moreover, the mixed extract (M) reduced the abundance of *F. oxysporum*. These changes were accompanied by an increase in the relative abundance of *F. avenaceum* compared with the control treatment. In the 2023/24 season, no complete elimination of any *Fusarium* species was observed. However, both plant extract treatments markedly reduced the relative abundance of *F. culmorum* compared with the control.

These findings indicate that the tested plant extracts did not eliminate the *Fusarium* complex but modified its community structure by altering the relative abundance of individual species. The results highlight the potential of plant extracts as environmentally friendly alternatives to conventional chemical protection strategies.

Keywords: Plant extracts; plant protection; species biodiversity; *Fusarium*; *Triticum aestivum*.



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INTEGRATIVE MULTI-OMICS REVEALS MOLECULAR MECHANISMS UNDERLYING ROSEMARY EXTRACT-INDUCED DEFENSE PRIMING

Lucila Garcia¹, Catalina Feuli¹, Dimitre Ivanov², Carolina Galli³, Alan Roy Blanc¹, Villalobos-Piña Liara¹, Georgi Bonchev⁴, Veselin Petrov², Kiril Mishev⁴, Pavel Kerchev⁵, Tsanko Gechev², María Rosa Marano¹

¹Instituto de Biología Molecular y Celular de Rosario, Rosario, Argentina (IBR-CONICET)

²Center of Plant Systems Biology and Biotechnology, Plovdiv, Bulgaria (CPSBB)

³Instituto Nacional de Tecnología Agropecuaria, Agencia de Extensión Rural Concarán, Argentina

⁴Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Sofia, Bulgaria

⁵Estación Experimental del Zaidin, CSIC, Granada, España

Presenting author: alanrblanc@gmail.com

Plant-derived biostimulants and bioprotectants are sustainable alternatives for integrated disease management, yet the molecular basis of *Salvia rosmarinus* (rosemary) aqueous extracts (ARE) against phytopathogens remains largely uncharacterized[1,2]. In this work, we implemented an integrative multi-omics pipeline—combining DNA barcoding, untargeted LC-MS/MS metabolomics, high-throughput transcriptomics, and shotgun proteomics—to systematically decode the chemical diversity and biological mechanism driving ARE bioactivity.

DNA barcoding successfully authenticated the analyzed rosemary accessions. To pinpoint the specific metabolites associated with disease protection, we performed comparative untargeted metabolomic profiling between a broad-spectrum protective reference extract and cultivars with differential protective capacities. Comparative multivariate statistical analyses identified 27 distinct metabolic features strictly correlated with the bioprotective phenotypes. These differential markers spanned a wide chemical diversity, from highly polar low-molecular-weight compounds to medium-polarity polyphenols and hydrophobic terpenoids, all of which were significantly downregulated or absent in non-protective cultivars.

Principal Component Analysis (PCA) of global transcriptomes revealed that under oxidative stress, ARE-treated plants clustered closely with untreated controls, indicating a powerful buffering effect not achieved by purified rosmarinic acid (RA) alone. While both treatments activated basal defenses, ARE triggered a much broader molecular reprogramming; in the presence of oxidative stress, ARE stimulated growth-related pathways via auxin signaling, while simultaneously repressing senescence and preventing ROS-induced programmed cell death. Proteomic analyses supported these findings, showing distinct protein accumulation patterns that aligned with the transcriptomic defense and growth signatures. Overall, our results indicate that ARE bioactivity relies on the coordinated action of multiple bioactive metabolites rather than RA alone, providing new insights into rosemary-induced defense priming and laying the foundation for the rational, matrix-based design of standardized botanical formulations for sustainable agriculture.

Keywords: *Salvia rosmarinus*; pPriming; biostimulant; untargeted metabolomic; multi-omic integration.

Acknowledgements: The research is supported by projects N 101086366 CropPrime (HORIZON-MSCA-2021-SE-01) “Stabilizing CROP yield under unfavourable conditions by molecular PRIM(E)ing”

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PP–33

THE EFFECT OF ENVIRONMENTAL CHANGE ON THE SUBALPINE BIRCH FOREST OVER TIME IN KAZBEGI MUNICIPALITY, CENTRAL GREATER CAUCASUS**Natalia Togonidze^{1,2}, Nani Goginashvili¹, Margalita Bachilava¹, Nika Gigauri¹, Jana Ekhvaia^{2,3}**¹Scientific-Research Centre of Agriculture, Tbilisi, Georgia²Institute of Botany, Ilia State University, Tbilisi, Georgia³Institute of Ecology, Ilia State University, Tbilisi, GeorgiaPresenting author: natalia.togonidze.1@iliauni.edu.ge, +995 599726771

Changes in climatic conditions, land use and social transformations significantly affects the natural vegetative cover in Georgia. Land degradation, deforestation, soil erosion and loss of biodiversity closely linked to ongoing social changes have been constantly observed in high-mountain regions. The northern slopes of the Central Caucasus Mountains in Kazbegi Municipality were formerly covered by birch forest (*Betula litwinowii* Doluch.), typical of cold climatic conditions characteristic of subalpine zones. Kazbegi, among Georgia's high-mountain regions, is particularly interesting due to its long history of anthropogenic impact, which caused degradation of extensive forest stands. However, in the recent decades, birch forests have naturally reforested, largely attributed to reduced uncontrolled sheep grazing in the municipality, along with its close relationship to global climate change. This study aims to evaluate temporal changes in the condition of subalpine birch forests in the Central Greater Caucasus and identify the main factors driving these changes. The condition of subalpine birch forest was evaluated for a 15-year period. Initial research was conducted in 2010 and repeat assessment was conducted in 2025. The obtained results indicated that significant changes have occurred in the Kazbegi birch forests over the last decade. Key tree characteristics, such as height, diameter at breast (DBH), age and diversity of plant species were changed. For instance, birch seedlings that were only a few centimeters in size in 2010 reached approximately 5-6 meters in height and about 7-8 cm in diameter after 15 years period. During successional stages of reforestation, some plant species appear, while other ones disappear. In addition, distribution of the forest was assessed. The treeline in the Central Greater Caucasus exhibited upward vertical migration shift over time. The upper limit of birch range was recorded at 2560 m. in 2002, 2685 m. in 2015 and 2698 m. in 2025. These findings highlight the importance of continuous monitoring of the treeline ecotone in relation to global climate change. Such monitoring will provide valuable insights into the condition of vegetation cover in this region.

Keywords: Subalpine birch forest; Global climate change; *Betula litwinowii* Doluch.; treeline ecotone.**Acknowledgements:** This work was implemented with the financial support of Shota Rustaveli National Science Foundation of Georgia (Grant # YS-24-2579).

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PRELIMINARY INSIGHTS INTO cpDNA VARIATION AND PHYLOGEOGRAPHY OF *FAGUS* TAXA IN THE CAUCASUS AND ADJACENT REGIONS

Jana Ekhvaia^{1,2}, Marco Cosimo Simeone³, Tamar Jolokhava¹, Arsen Bakhia¹, Tornike Pankvelashvili¹, Mariam Osepashvili¹, Mariam Kikvidze¹, Khabagin Aliev⁴, Oleksiy Yanchukov^{1,6}, Natalia Togonidze^{2,5}, Aleksandre Gavashelishvili¹

¹Institute of Ecology, Ilia State University, 45 Ilia Chavchavadze Avenue, Tbilisi, Georgia

²Niko Ketskhoveri Institute of Botany, Ilia State University, 1 Botanikuri Street, Tbilisi, Georgia

³University of Tuscia, 4 Via Santa Maria in Gradi, Viterbo, Italy

⁴Mountain Botanical Garden of Federal Dagestan Research Center of the RAN, 45 Gadzhieva Street, Makhachkala, Russian Federation

⁵Scientific Research Center of Agriculture, 6 Marshal Gelovani Avenue, Tbilisi, Georgia

⁶Zonguldak Bülent Ecevit University, İncivez, Üniversite Cd., Zonguldak, Türkiye

Presenting author: zhana.ekhvaia@iliauni.edu.ge

Chloroplast DNA variation was analyzed to assess genetic diversity, population structure, and demographic history of *Fagus* taxa in the Caucasus and adjacent regions. A total of 440 individuals from the Black Sea and Caspian Sea basins were examined using two concatenated cpDNA intergenic spacers (*trnK-trnQ* and *petA-psbJ*, 2,352 bp). Overall, 18 polymorphic sites defined 14 haplotypes, revealing high haplotype diversity ($Hd = 0.804$) and moderate nucleotide diversity ($\pi = 0.0017$). Genetic diversity differed between basins: the Black Sea basin showed higher variability ($Hd = 0.752$; $\pi = 0.0019$), whereas the Caspian basin exhibited reduced diversity ($Hd = 0.585$; $\pi = 0.0009$) and an excess of singleton mutations. Demographic analyses indicated contrasting histories. The Caspian basin displayed a unimodal mismatch distribution and significantly negative Fu and L_i 's statistics, consistent with recent expansion. In contrast, the Black Sea basin showed a multimodal distribution and positive neutrality tests, suggesting long-term stability or population subdivision. In conclusion, these results support distinct demographic patterns for Black and Caspian Sea beech populations, likely reflecting multiple glacial refugia and basin-specific postglacial dispersal pathways.

Keywords: *Fagus* taxa; Caucasus; Black and Caspian Sea populations; contrasting biogeographical history; postglacial dispersal pathways.

Acknowledgments: This work was implemented with the financial support of the Shota Rustaveli National Science Foundation of Georgia (Grant # FR- 22- 889).



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ASSESSMENT OF GENETIC VARIABILITY IN THE ENDEMIC SPECIES *BRASSICA JORDANOFFII* AND *AETHIONEMA RHODOPAEUM* USING DNA BARCODING MARKERS

Irina Boycheva¹, Georgi Bonchev¹, Stoyan Stoyanov², Iva Doycheva²

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. Georgi Bonchev str., Build. 21, 1113 Sofia, Bulgaria

²Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Acad. Georgi Bonchev str., Build. 23, 1113 Sofia, Bulgaria

Presenting author: i.i.boycheva@gmail.com

Natural and herbarium collections are subject to rigorous taxonomic revision and curation by experts in botany, ecology, and taxonomy. However, the need for higher resolution in distinguishing closely related taxa, as well as the possibility of detecting hidden genetic variability within species, necessitates the application of molecular DNA methods. These approaches to taxonomic identification of organisms have significant potential for improving identification, monitoring, systematics, and the management of plant biodiversity in Bulgaria.

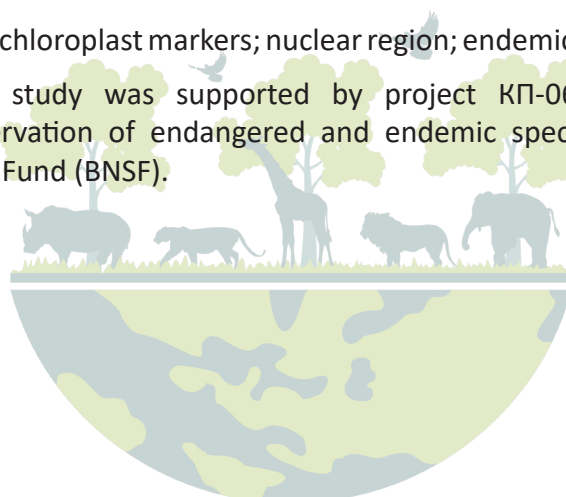
The present study aims to assess patterns of inter- and intraspecific genetic divergence between the Bulgarian endemics *Brassica jordanoffii* and *Aethionema rhodopaeum* using DNA barcoding method. Three populations of *Aethionema rhodopaeum* from Western Rhodopes and three populations of *Brassica jordanoffii* from the Pirin Mountain were analyzed based on nuclear (*ITS* region) and chloroplast (*trnH-psbA* and *rbcl*) barcode markers.

All three markers were able to differentiate specimens at species level, but the resolution power at within-species level varied for different markers. *trnH-psbA* provided the highest resolution and clear separation of populations. *ITS* showed moderate to high informativeness, while *rbcl* exhibited limited discriminatory capacity. The combination of all three markers was shown to increase the taxonomic resolution power. BLAST analysis against NCBI assigned *Aethionema rhodopaeum* to *Aethionema thomasianum* (99,6%, *ITS*), and *Brassica jordanoffii* to *Brassica oleracea* (95%, *trnH-psbA*) or *Brassica elongata* (97,5%, *ITS*). The lack of full sequence identity to available database records supports the genetic distinction of the analyzed accessions as separate species.

The results highlight the potential of DNA barcoding in facilitating classical approaches for studying diversity of analyzed taxa. The absence of DNA barcode records of these species in genebank repositories implies the importance of actions towards the development and enrichment of genetic reference libraries for the Bulgarian local and often unique biodiversity with emphasis on rare and endemic species.

Keywords: DNA barcoding; chloroplast markers; nuclear region; endemic species; genetic divergence.

Acknowledgements: The study was supported by project КП-06-H81/10 “Multidisciplinary approaches for the conservation of endangered and endemic species of the Bulgarian flora”, Bulgarian National Science Fund (BNSF).



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PHYSIOLOGICAL AND BIOCHEMICAL DETERMINANTS OF DROUGHT AND SALINITY TOLERANCE ACROSS THREE GRAIN LEGUME SPECIES

Angeliki Petraki¹, D. Vlatsos¹, K. Soi¹, A. Motsenigou¹, N. Vidali¹, D. Sifneos¹, E. Barilli², V. Vassileva³, A. Balestrazzi⁴, D. Chachalis¹

¹Benaki Phytopathological Institute, Laboratory of Weed Science, 8 St. Delta Street, 14561, Kifissia, Attica, Greece

²Agencia Estatal Consejo Superior de Investigaciones Científicas, Institute for Sustainable Agriculture, Avenida Menéndez Pidal, 14004, Córdoba, Spain

³Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Acad. G. Bonchev Str., Bldg. 21, 1113, Sofia, Bulgaria

⁴University of Pavia, Department of Biology and Biotechnology 'L. Spallanzani', via Ferrata 9, 27100 Pavia, Italy

Presenting author: ag.petraki@bpi.gr

Salinity and drought are among the most critical abiotic constraints limiting grain legume productivity in Mediterranean agroecosystems. Increasing climatic pressures and rising food demand (feeding 9 billion people) are challenging crop establishment and yield stability. At the same time, programs such as the European Green Deal require different and alternative production systems, and thus also crops. In some areas this means that alternative farming practices will have to be developed and implemented. Collectively, these factors present both challenges and opportunities, as they have the potential to enhance the speed, efficiency, and efficacy with which drought- and salinity-tolerant varieties can be developed. Morphological and physiological traits, biomass, root architecture, and the protective enzyme system were measured in Spanish and Greek accessions of *Lathyrus* species (*L. sativus*, *L. ochrus*, *L. clymenum*, and *L. tingitanus*) under growth chamber and greenhouse conditions. Bulgarian accessions of faba bean (*Vicia faba*) and chickpea (*Cicer arietinum*) were evaluated under greenhouse conditions. All species were screened under both drought and salinity stress. The genotype Luna (*L. sativus*, Spain) combined low lipid peroxidation (MDA: 0.063 ± 0.007 mmol g⁻¹ FW) with high canopy vigour (NDVI: 0.47 ± 0.02) and moderate H₂O₂ (1.07 ± 0.40 μmol g⁻¹ FW), while breeding line L316 (*L. ochrus*) showed the lowest MDA (0.026 ± 0.007 mmol g⁻¹ FW), indicating effective membrane protection. The response of the protective enzyme system and potential cell membrane damage under salinity differed markedly between varieties. In chickpea, flower number was unaffected by irrigation treatments in both genotypes tested, indicating maintained reproductive development under water deficit. Salt stress significantly altered root system architecture ($p \leq 0.05$) in faba bean. These findings demonstrate that drought and salinity tolerance in grain legumes varies by genotype and by trait across grass pea, chickpea, and faba bean, and delineate candidate accessions combining drought and salinity tolerance, providing a foundation for breeding programs targeting stress-resilient cultivars in Mediterranean cropping systems.

Keywords: Salinity; drought; legumes.

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ISOLATION AND GENETIC IDENTIFICATION OF BACTERIAL AND FUNGAL STRAINS FROM BURNT MEADOWS AND PASTURES IN BULGARIA

Mariana Radkova, Aneta Lyubenova, Katerina Stefanova, Miglena Revalska, Anelia Iantcheva

Agrobiointitute, Agricultural Academy, Blvd. “Dragan Tzankov” 8, Sofia 1164, Bulgaria

Presenting author: marianaradkova@yahoo.com

In recent years, as a result of global warming, the number of fires has increased dramatically both globally and in Bulgaria. According to the European Forest Fire Information System (EFFIS), in the summer of 2024 Bulgaria was ranked second after Cyprus in terms of the number of fire occurrence. Fires cause direct damage to plant and soil resources, in both forest and adjacent grassland ecosystems. Meadows and pastures are a major source of fodder for livestock, help preserve plant diversity, and maintain soil water balance. These ecosystems, like forests, can store significant amounts of carbon. The purpose of our study is the isolation and identification of bacterial and fungal strains from burnt meadows and pastures in Bulgaria. Our final goal is to select the highly active strains with application for recovery of burnt/degraded grassland areas. Genetic identification by 16S rRNA (V1-V9 region) and ITS (ITS 1/ ITS 4) was performed.

The bacterial strains isolated from the burnt locations Lovech and Suhindol (Central North Bulgaria) demonstrated high homology with NCBI annotated strains from the genera *Bacillus*, *Priestia* and *Pseudomonas*. The isolated fungal strains include *Mortierella* spp., *Trichoderma* spp., *Aspergillus* spp., *Penicillium* spp., *Fusarium* spp. and representatives of *Mucorales*. The potential of the new isolates to impact plant development and soil recovery will be evaluated in the future.

Keywords: Fire; pastures and meadows; 16S rRNA; ITS.

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IDENTIFICATION AND CHARACTERIZATION OF FUNGAL SPECIES TO ASSESS THE EFFECT OF MULCHING IN POTATO AND BEAN CULTIVATION

Zornitsa Stoyanova¹, Ivelina Nikolova¹, Dimitar Danchev², Miroslava Zhiponova², Georgi Bonchev¹, Vasilissa Manova¹

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Sofia, Bulgaria

²Faculty of Biology, Sofia University “St. Kliment Ohridski”, Sofia, Bulgaria

Presenting author: z.b.stoyanova@abv.bg

The application of regenerative agricultural practices, such as mulching, helps improving soil fertility and limiting weed development. Organic mulches have proven benefits for soil health and for reducing water loss. At the same time, they provide a suitable substrate for the development of micromycetes, thus creating a specific microenvironment around plants depending on the associated microorganisms. Microscopic fungi represent one of the most diverse and ecologically significant groups of organisms on Earth. They are heterotrophic eukaryotes and constitute a natural part of the plant microbiota, contributing to normal plant development, although some of them may cause diseases in agricultural crops. The aim of the present study was to identify and characterize the culturable micromycetes obtained from potato and bean plants grown with two types of organic mulch, either straw or grass, in field trials at two different locations: a mountain region near Momchilovtsi, Smolyan, and an urban location in Dragalevtsi, Sofia, and to compare them with fungi isolated from control plants grown without mulch. The material used in this study, consisting of soil and whole plants, was collected between June and October 2025. Fungi were characterized using classical mycological and modern molecular methods. The fungal isolates were cultivated in Petri dishes on three culture media: Bengal Red Agar, Sabouraud 4% Glucose Agar, and Potato Dextrose Agar, at a temperature of 24 °C. For DNA barcoding analyses, DNA was isolated from fungal mycelium collected from the surface of well-developed colonies. More than 300 fungal isolates were obtained, of which 115 pure cultures were selected on the basis of their morphological and cultural characteristics for subsequent molecular identification. DNA barcoding analysis was performed by amplification and sequencing of the ITS region using the ITS1–ITS4 primer pair. The largest number of micromycete isolates was obtained from the soil and underground parts of the experimental plants. In potato, a multitude of morphologically distinct isolates was observed in the material from Momchilovtsi, whereas in bean, greater morphological diversity was detected in the samples from Dragalevtsi. The fungal isolates obtained from the aboveground plant parts from both fields were also morphologically heterogeneous. The data indicate differences in the amount and morphological diversity of culturable fungal isolates depending on the host plant and geographical location. Their analysis will allow assessment of the influence of mulch on potato and bean cultivation through the identification of micromycetes, including phytopathogenic fungi, associated with these plants. This knowledge could expand our ecological and taxonomic understanding of culturable fungi from soil and plant substrates and contribute to the more informed and targeted use of regenerative practices in agriculture, leading to more effective and sustainable plant protection management.

Keywords: Regenerative agriculture; micromycetes; cultivable fungi; mulching; DNA barcoding.

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IMPACT OF METAL NANOPARTICLES ON TOMATO PHYSIOLOGY, ANTIOXIDANT RESPONSES AND FOOD CHAIN CONTAMINATION

Mirsade Osmani¹, Marilena Carbone²

¹Faculty of Food Technology, University Isa Boletini in Mitrovica, Ukshin Kovačica str, 28000 Mitrovica, Kosovo

²Department of Chemical Science and Technologies, University of Rome “Tor Vergata”, Via della Ricerca Scientifica 1, 00133 Rome, Italy

Presenting author: mirsade.osmani@umib.net

The increasing accumulation of metal nanoparticles (MNPs) in agricultural soils, driven primarily by industrial pollution and anthropogenic activities, is a serious concern because of their effects on plant physiological functions and the potential risks to public health. This study investigates how MNPs in contaminated soils affected the photosynthetic performance and antioxidant defense of tomato (*Solanum lycopersicum* L.) grown in the northern region of Kosovo. Soil and plant samples were analyzed to determine and characterize MNPs using advanced analytical techniques, including scanning electron microscopy (SEM) coupled with energy-dispersive X-ray spectroscopy (EDX), X-ray diffraction (XRD), and X-ray fluorescence (XRF). The results revealed relatively high concentrations of MNPs, including Cd, Cu, Pb, Ag, Ti, and Zn, in all analyzed samples, particularly in locations closest to the Trepça industrial complex. The findings also indicate differential responses to MNP accumulation across plant tissues. In highly contaminated areas, an increase in intercellular CO₂ concentration (*C_i*) was observed, accompanied by decreases in stomatal conductance (*g_s*), photosynthetic rate (*A*), and transpiration (*E*), indicating limitations in photosynthesis and inefficient CO₂ utilization. Furthermore, reductions in Φ PSII and *F_v'*/*F_m'* suggest damage to photosystem II. In contrast, plants from moderately polluted areas exhibited partial recovery of these parameters, suggesting reduced stress or adaptive responses. In parallel, increased flavonoid levels and enhanced antioxidant activity (FRAP, DPPH, and ABTS assays) were observed in polluted areas, indicating activation of protective mechanisms against oxidative stress induced by nanoparticle exposure. Overall, these findings highlight the risks posed by MNPs in soils to plant physiological health and productivity, emphasizing the need for monitoring and mitigation strategies to ensure sustainable crop production.

Keywords: Metal nanoparticles; tomato; food safety; photosynthetic performance; antioxidant activity.

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AFTERWORD

Closing Remarks

The **Second International Conference on Functional Biology** continues the dialogue initiated with the first FunBio meeting, bringing together researchers, students and professionals from different fields to exchange knowledge, discuss new ideas and strengthen scientific connections.

The contributions collected in this Book of Abstracts highlight the diversity and interdisciplinary character of functional biology. They also demonstrate how advances across different areas of biological research can contribute to addressing major challenges related to environmental change, sustainable agriculture, biodiversity, biotechnology and food security.

We warmly thank all participants, invited speakers, partners and colleagues whose enthusiasm and involvement have made FunBio2 possible. We hope that the discussions and connections established during these days in Varna will develop into new collaborations, joint projects and lasting scientific partnerships.

We look forward to continuing the FunBio tradition and welcoming you again at the next edition of the **International Conference on Functional Biology**.

Until we meet again at FunBio3!



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