ICGEB Meeting and Courses 2023

ICGEB WORKSHOP

TRENDS IN MICROBIAL SOLUTIONS FOR SUSTAINABLE AGRICULTURE

13 – 15 September 2023. Belgrade, SERBIA

BOOK OF ABSTRACTS

ICGEB WORKSHOP

Trends in microbial solutions for sustainable agriculture

13 – 15 September 2023. Belgrade, Serbia

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ICGEB Workshop

Trends in microbial solutions for sustainable agriculture

13 – 15 September 2023. Belgrade, SERBIA

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ICGEB MEETINGS & COURSES 2023

WORKSHOP
„Trends in microbial solutions for sustainable agriculture“

BOOK OF ABSTRACTS

September 13 – 15, 2023
Belgrade, Serbia

CONTENTS

Message from the scientific organizers

Dear colleagues and friends,

It is our great pleasure to welcome you to the Workshop "Trends in microbial solutions for sustainable agriculture" to be held in Belgrade, Serbia, September 13-15, 2023. This meeting is organized by the University of Belgrade - Faculty of Biology, Belgrade, Serbia, and co-organized by the International Centre for Genetic Engineering and Biotechnology (ICGEB), Trieste, Italy, and our friends from the Serbian company FERTICO. The Workshop is organized with the support of the Federation of European Microbiological Societies (FEMS), the Ministry of Science, Technological Development and Innovations of the Republic of Serbia and the Serbian Society for Microbiology. We thank the sponsors for their recognition of the importance of the event, their participation and support. We are pleased to inform you that Workshop Belgrade, Serbia - Book of Abstract will appear in the Collection of the Faculty of Biology, published by the University of Belgrade - Faculty of Biology.

This Workshop is dedicated to our colleague and friend Professor Djordje Fira, who was one of the chairs of the Scientific and Organizing Committee. Unfortunately, he is no longer with us. Professor Fira, with his ideas and enthusiasm, had an innovative approach to the use of bacteria in biological pest control and its application in sustainable agriculture. We are sad but joyful because we had the opportunity to live, work and learn from the man who was professor, head of department, colleague and friend. Professor Fira was the embodiment of a good, honest, sincere and careful person. We had the privilege of sharing our lives with professor Fira - your presence we miss, your memories we treasure.

This Workshop, covering all major topics of the use of microbial solutions in sustainable agriculture, is in perfect harmony with the Serbian Smart Specialization Strategy, the European Green Deal and FOOD 2030 research and innovation policy in the field of applied microbiology. Indeed, the environmental and ethical unsustainability of the continued use of chemical pesticides, coupled with the need for yield increases due to population growth and the simultaneous reduction of land under food crops, is leading to a general awareness of the need to drastically reduce the use of chemical pesticides, as well as radical changes in current agricultural practices. Interest in biological control of phytopathogens has particularly increased in the last decade, mainly because of the importance of using environmentally friendly alternatives to the extensive use of chemical pesticides to control pest diseases.

The extensive microbiome research in the field of plant microbiome structure and function, the pivotal role of plant-associated microbes in plant health and productivity, and the new "state-of-the-art" methods available today should expand our knowledge and pave the way from laboratory data to practical applications in sustainable agriculture. Plant-associated microbial communities play a key role in biotic and abiotic stress tolerance as well as nutrient acquisition and carbon and nitrogen cycling. The idea of developing environmentally friendly biofertilizers and other agricultural biotechnologies, along with molecular studies of plant resistance to biotic and abiotic stresses, the study of interkingdom signalling between plants and plant-associated bacteria, with special attention to emerging phytopathogens, will be the main activities and outcomes of the Workshop.

We strongly believe that the Workshop is an excellent place to exchange and combine scientific ideas among experts and participants, with great opportunities to start new international collaborations and joint scientific projects. We have received an overwhelming response to our call, with numerous talented applicants, more than 160 participants from 41 countries (Argentina, Bangladesh, Brazil, British Indian Ocean Territory, Burundi, Cameroon, China, Colombia, Côte d'Ivoire, Croatia, Ecuador, Egypt, Ethiopia, Ghana, Greece, Hungary, India, Iraq, Italy, Kenya, Libya, Malawi, Mexico, Montenegro, Namibia, Nigeria, Pakistan, Papua New Guinea, Peru, Russian Federation, Serbia, Slovenia, Somalia, South Africa, Sri Lanka, Sudan, Tunisia, Turkey, United Republic of Tanzania, Zimbabwe, and the United States) to compete for the limited number of available grant awards. In addition to the invited speakers' presentations, the programme also includes poster presentations by a number of early career scientists and PhD students, many of whom are supported and funded by ICGEB and FEMS grants (we selected more than 30 fellows from 16 countries). We were honoured to welcome 30 speakers from 15 countries (Austria, Belgium, Croatia, Germany, Hungary, India, Italy, Japan, Romania, Saudi Arabia, Serbia, Slovenia, South Africa, Spain and the Netherlands). We thank all participants for their scientific commitment, which will contribute significantly to the success of the Workshop.

We hope you enjoy the Workshop programme and find it stimulating and informative. We also hope that you will enjoy the beauty of Belgrade and the Serbian hospitality. We sincerely wish you health, love and happiness and look forward to the new meetings.

Sincerely,

Ivica Dimkić, PhD
Scientific & Organizing Committee
Chairperson

Vittorio Venturi, PhD
Scientific & Organizing Committee
Co-Chairperson

General information

SYMPOSIUM VENUE

The meeting will be held in the Great Hall of the Municipality of Stari Grad, Makedonska 42, Belgrade, Serbia, and in the Hotel Palace 4*, Topličin Venac 23, Belgrade, Serbia.

REGISTRATION OF PARTICIPANTS

Registration desk will be opened on Wendsday, September 13 from 08:00 to 9:00 in front of the Great Hall of the Municipality of Stari Grad and on Thursday, September 14 from 08:30 to 9:00 in front of the hall „Beogradska Panorama" of the Hotel Palace. Daily updated information about the workshop sessions and social events will be available at the registration desk. All participants and accompanying persons are requested to wear their accreditation badges during the scientific sessions and social events of the workshop.

LANGUAGE

The official language of the workshop is English.

SOCIAL EVENTS

A group photo in front of the Vojvoda Vuk monument in Topličin venac square (17:45), a „Poster Party" (18:00) and a Welcome reception (19:30) will take place in and around the Palace Hotel on Wendsday, September 13.

The Gala Dinner will be held at the Botanical Garden "Jevremovac", Takovska 43, Belgrade, on Thursday, September 14, from 19:00 to 23:00.

For the last day, Friday, September 15, is planned a visit to FERTICO company with lunch break (8:00) and a visit to Belgrade Fortress "Kalemegdan" with professional guide and free evening (17:30). All participants are asked to register at the registration desk for the facultative Danube and Sava cruise, which will be charged extra (19:00).

INFORMATION FOR PRESENTERS

Oral presentations will be held on September 13 in the Great Hall of the Municipality of Stari Grad and on September 14 and 15 in the hall „Beogradska Panorama" of the Palace Hotel. LCD projections will be available during all sessions. Please send your PowerPoint presentation to workshopserbia@gmail.com and katarina.kruscic@bio.bg.ac.rs no later than September 10.

Posters will be displayed during the „Poster Party" on Wednesday, September 13 from 18:00-20:00 in a garden at the Palace Hotel. Poster presenters are asked to be at their posters and available for discussion. They should mounted their posters at 17:30 and to dismount them immediately after the „Poster Party".

Tuesday, September 12, 2023

Arrival of participants and accomodation

Wednesday, September 13, 2023

Venue - Great Hall of the Municipality of Stari Grad, Makedonska 42

08:00-09:00 **Registration of participants (COFFEE & SNACKS)**

09:00-09:40 **WELCOME ADDRESS**

- **Ivica Dimkić, Vittorio Venturi & Slavoljub Vukićević**
- **Jelena Begović**, Serbian Minister of Science, Technological Development and Innovation
- **Lawrence Banks**, Director General of ICGEB, Italy
- **Luciano Catani**, Science Attaché, Embassy of Italy in Belgrade, Serbia
- **Ljubiša Stanisavljević**, Dean of the Faculty of Biology, University of Belgrade, Sebia

09:45-10:15 **Ivica Dimkić (University of Belgrade - Faculty of Biology, Belgrade, Serbia):** Smart biofertilizers as powerful organomineral-microbial soil improvers

Session I: Biopesticides, biofertilizers & biostimulants – biological control approach (Part 1)

Chaired by: **Marc Ongena, Ken-ichi Yoshida**

10:15-10:45 **Ken-ichi Yoshida (FEMS Ambassador, Kobe University, Japan):** Co-inoculation of *Bacillus velezensis* strain S141 and *Bradyrhizobium* strains promotes nodule growth and nitrogen fixation

10:50-11:20 **Marc Ongena (University of Liège, Belgium):** Ecological and biocontrol functions of secondary metabolites produced by *Bacillus velezensis*

11:25-11:55 **Dennis Obonyo Ndolo (ICGEB, Cape Town, South Africa):** Potential of microbial biopesticides in addressing trade challenges

12:00-13:00 **LUNCH BREAK**

Session II Biopesticides, biofertilizers & biostimulants – biological control approach (Part 2)

Chaired by: **Monica Höfte, Jovana Grahovac**

13:00-13:30 **Monica Höfte (Faculty of Bioscience Engineering, Ghent University, Belgium):** Potential of cyclic lipopeptide-producing *Pseudomonas* strains in biocontrol

13:35-14:05 **Jovana Grahovac (University of Novi Sad - Faculty of Technology, Serbia):** Microbial products for agriculture – challenges of technology transfer to commercial application

14:10-14:40 **Marina Soković (Institute for Biological Research "Siniša Stanković" - National Institute of Republic of Serbia, University of Belgrade):** Biosolution against microbes for safety and sustainable agriculture

14:45-15:15 **Antonio Ferrante (University of Milano, Italy):** Application of biostimulants for counteract abiotic stresses

15:20-15:50 **Aleksandra Bešlić & Rebeca Fuzinatto Dall' Agnol (Fertico DOO, Serbia & Simbiotica S.r.l., Italy):** The future prospect of bacterial fertilizers: from lab to market

15:50-16:15 **COFFEE BREAK**

Session III Microbial products for sustainable agriculture (Part 1)

Chaired by: **Ivica Dimkić, Vittorio Venturi**

16:15-16:45 **Vincenzo Michele Sellitto (Biolchim spa, Italy/University of Life Sciences "King Mihai I", Faculty of Agriculture - Timisoara, Romania):** Living soil: a new paradigm for an innovative agriculture. A practical case to phytoparasitic nematodes biocontrol using probiotics and prebiotics approach

16:45-17:15 **Wolfgang Vogt (Agrobiota, Germany):** Diverse functions of microorganisms in agriculture: which function is fulfilled when, or all at once?

Venue - Garden and Hall Club of the Hotel Palace, Topličin venac 23

17:45-17:55 **Group photo in front of the monument to Vojvoda Vuk on Topličin venac square**

18:00-19:30 **POSTER PARTY with Serbian beverages**

19:30-23:00 **Welcome Reception**

Thursday, September 14, 2023

Venue - Hall "Beogradska Panorama" of the Hotel Palace, 6th floor, Topličin venac 23

08:30-09:00 **Registration of participants (COFFEE & SNACKS)**

09:00-09:30 **Vittorio Venturi (ICGEB, Trieste, Italy):** Contact-independent bacterial signalling in the plant beneficial microbiome

Session IV Plant–microbe interactions

Chaired by: **Ines Mandić Mulec, Alessio Mengoni**

09:30-10:00 **Alessio Mengoni (University of Florence, Italy):** When biodiversity preservation meets biotechnology: the challenge of developing synthetic microbiota and the case of symbiotic nitrogen-fixing bacteria

10:05-10:35 **Cayo Ramos (Universidad de Málaga-Spanish Research Council, Málaga, Spain):** The virulence factors orchestra of the bacterial pathogen of woody hosts *Pseudomonas savastanoi*

10:40-11:10 **Ines Mandić Mulec (Biotechnical Faculty, University of Ljubljana, Slovenia):** Unveiling the role of bacterial sociality in designing plant growth promoting inoculants

11:30-12:00 **Stéphane Compant (Austrian Institute of Technology, Tulln, Austria):** Bacterial-fungal interactions and impacts on plant growth and health

12:05-12:35 **Gergely Maróti (Institute of Plant Biology, Hungary/University of Public Service, Faculty of Water Sciences, Hungary/Seqomics Biotechnology Ltd.):** Molecular-level investigation of algal-bacterial interactions and their utilization for plant biostimulation

12:40-13:40 **LUNCH BREAK - Restaurant Klasik of the Hotel Palace**

Session V Plant–microbe interactions and the role of synthetic microbial communities

Chaired by: **Daniele Daffonchio, Aleš Lapanje**

13:40-14:10 **Vittoria Catara (University of Catania, Italy):** Microbiome-assisted selection of endophytic bacteria for the sustainable cultivation of tomatoes in greenhouse

14:15-14:45 **Djordje Bajić (Technical University Delft, The Netherlands):** Using fitness landscapes to engineer microbial communities

14:50-15:20 **Daniele G. Daffonchio (King Abdullah University of Science and Technology, Saudi Arabia):** Bacterial VOCs contrast rice blast infection by inhibiting fungal virulence rather than growth

15:25-15:55 **Ákos T. Kovács (Institute of Biology, Leiden University, The Netherlands):** *Bacilli* in the rhizosphere: interaction and evolution within the biofilm

16:00-16:30 **Aleš Lapanje (Jozef Stefan Institute, Ljubljana, Slovenia):** Engineering microbial communities by the colloid biology approach

16:35-17:05 **COFFEE BREAK**

Session VI Microbial products for sustainable agriculture (Part 2)

Chaired by: **Vittorio Venturi, Ivica Dimkić**

17:05-17:30 **Vincent Lefebvre du Prey (ITAKA S.r.l., Italy):** The place of microorganisms in the transition to new farming systems with reference to research, agronomy and IT, using greenhouse vegetables as example

17:35-18:05 **Vesna Dragičević (Maize research Institute Zemun Polje Belgrade, Serbia):** The effects of bio-fertilizer application on grain yield and quality of maize grown in long-term monoculture

Botanical Garden "Jevremovac", Takovska 43

19:00-23:00	**GALA DINNER**

Friday, September 15, 2023

Studentski trg 3, Belgrade (in front of Faculty of Phylology)

08:00-13:00	**Visit to FERTICO enterprise with lunch break (Indjija, Serbia)**

Venue - Hall "Beogradska Panorama" of the Hotel Palace, 6th floor, Topličin venac 23

Session VII Microbial products for sustainable agriculture (Part 3)

Chaired by: **Vlad Stoian, Marko Vinceković**

13:00-13:30	**Amitava Rakshit (Banaras Hindu University, Varanasi, India):** Seed bio-priming with microbial inoculants: A tailored approach towards improved crop performance, nutritional security, and agricultural sustainability for smallholder farmers
13:35-14:05	**József Fail (Institute of Plant Protection, Hungarian University of Agriculture and Life Sciences, Hungary):** Challenges in the management of some key horticultural pests
14:10-15:40	**Vlad Stoian (Faculty of Agriculture, University of Agricultural Sciences and Veterinary Medicine, Cluj-Napoca, România):** The use of microscopic tools for the increase of resolution in mycorrhizal assessment
15:45-16:30	**Marko Vinceković (Faculty of Agriculture University of Zagreb, Croatia):** New types of biofertilizers: microparticles with biological and chemical active components
16:30-17:00	**COFFEE BREAK**
17:00-17:15	**ORGANIZATIONAL CLOSING REMARKS - Ivica Dimkić & Vittorio Venturi**
17:30	**Sightseeing of Belgrade Fortress "Kalemegdan" with professional guide and free evening**
19:00-20:30	**Optional Danube and Sava Cruise with extra charge - get to know Belgrade from the rivers**

INVITED LECTURES

IL1

Smart biofertilizers as powerful organomineral-microbial soil improvers

Ivica Dimkić[1,3*], **Dejan Lazić**[2,3], **Aleksandar Radulović**[3] **and Gordana Subakov Simić**[1,3]

[1] University of Belgrade, Faculty of Biology, Studentski trg 16, 11158 Belgrade, Serbia
[2] Innovation Center of the Faculty of Biology, Studentski trg 3, 11158 Belgrade, Serbia
[3] BioCombact DOO, Golsvordijeva 32, 11000 Belgrade, Serbia
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Improved management and recycling of plant residues to improve soil health and reduce nutrient (nitrogen, phosphorus, potassium) discharge to the environment is a priority, along with empowered interdisciplinary design processes to create soil improvers that improve the functionality of roots, interacting directly or indirectly with plants, and to enhance plant byproducts and beneficial microbes in sustainable agriculture. The ultimate challenge we have identified is to formulate a range of environmentally friendly and health safe bio-based mixtures (SBF), highly efficient biofertilizers with pesticide character, which are not too technologically complex to produce in scalable quantities and consequently available at acceptable cost. Bacterial isolates from the endosphere of various crops were selected for their plant growth-promoting activity, biocontrol capacities, and environmental compatibility. Differently designed products were used in the treatment of seeds of different plant varieties and crops, with disease severity reduction, crop yield improvement, and community composition evaluated using next-generation sequencing techniques. An untapped opportunity lies in the valorization of new environmentally friendly products containing mineral nutrients, organic matter, and microbes that can contribute to soil fertility and improvement, with emphasis on subsequent reuse and recycling pathways. Advances in synthetic microbiome technology and computational biology indicate that appropriate levels of selected endophytes are maintained in seedlings without affecting the "core" microbiota, and that the severity of plant diseases is reduced. We are interested in further product development, to obtain products with longer shelf life and best sustainability from the point of view of application and efficiency.

Keywords: smart biofertilizers; beneficial bacteria; biocontrol; microbiome; sustainable agriculture**.**

Acknowledgement: This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia; Collaborative Research Programme (CRP) – ICGEB Research Grants Programme; FERTICO D.O.O. - Nitrogenous Fertilizer Manufacturing Enterprise; Innovation Fund of the Republic of Serbia, Science Technology Park Belgrade, and by the Project for the promotion of innovation and technological development in Serbia - StarTech in organization of the National Alliance for Local Economic Development (NALED) funded by Philip Morris Operations A.D. [Contracts Nos.: 451-03-47/2023-01/200178; CRP/SRB19-02; 749/1-23.06.2021; 2522].

IL2

Co-inoculation of *Bacillus velezensis* strain S141 and *Bradyrhizobium* strains promotes nodule growth and nitrogen fixation

Surachat Sibponkrung[1]**, Takahiko Kondo**[2]**, Panlada Tittabutr**[1]**,**
Ken-ichi Yoshida[2*] **and Neung Teaumroong**[1]

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Suranaree University of Technology, Nakhon Ratchasima 30000, Thailand
[2] Graduate School of Science, Technology and Innovation, Kobe University, Kobe 657-8501, Japan
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Co-inoculation of *Bacillus velezensis* S141 with *Bradyrhizobium diazoefficiens* USDA110 into soybean (*Glycine max* (L.) Merr.) resulted in enhanced nodulation and N2-fixing efficiency by producing larger nodules. To understand the role of S141 on soybean and USDA110 symbiosis, putative genes related to IAA biosynthesis were disrupted; revealing that co-inoculation of USDA110 with S141ΔyhcX reduced the number of large nodules. The results suggested that yhcX may play the primary role in IAA biosynthesis in S141 and thus impact soybean growth promotion. On the other hand, disruption of genes related to cytokinin biosynthesis in S141ΔIPI reduced the number of larger nodules, suggesting that IPI to produce cytokinin might also play a role in nodule size. However, not only IAA and cytokinin but also other substances secreted from S141 might act on *Bradyrhizobium* to stimulate nodule formation, resulting in enhanced N2-fixation. Therefore, a better understanding of the PGPR function of S141 would lead to the further development of a better soybean co-inoculum microorganism.

Keywords: PGPR; *Bradyrhizobium*; co-inoculation; soybean; nodulation.

IL3

Ecological and biocontrol functions of secondary metabolites produced by *Bacillus velezensis*

Marc Ongena[1*]

[1] University of Liège, Faculty, TERRA Research center, Gembloux Agro-Bio Tech Faculty, MiPI Lab,
Avenue de la Faculté, 5030 Gembloux, Belgium
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B. velezensis (referred as Bv here below) is a typical soil dwelling plant-associated bacterial species that retains a high potential to protect crops against diseases and therefore represent promising alternatives to chemical pesticides as biocontrol agent. Bv is a strong producer of a range of bioactive secondary metabolites (BSMs) mostly responsible for the two main biocontrol functionalities, which are direct antagonistic activity toward pathogens and the ability to trigger an immune reaction in the host plant leading to a systemically-expressed higher resistance to subsequent infection (induced systemic resistance or ISR). This chemical arsenal includes a range of non-ribosomally synthesized compounds (megaenzymes encoded by giant biosynthetic gene clusters) such as oligopeptides, cyclic lipopeptides and polyketides but also post-translationnaly-modified lanthipeptides and bacteriocins. We will illustrate the chemical diversity of these BSMs and provide an overview of their specific roles in the inhibition of phytopathogens via direct toxicity or indirectly by acting as elicitors of ISR. The impact of multitrophic interactions with the host plant and the auxiliary microflora on the expression of the BSM arsenal in Bv will also be discussed.

Keywords: *Bacillus velezensis*; specialized small molecules; biological control.

IL4

Potential of microbial biopesticides in addressing trade challenges

Dennis Obonyo Ndolo[1*] **and Karen Hope**[1]

[1] Biopesticides Group, International Centre for Genetic Engineering and Biotechnology (ICGEB), Wernher & Beit Bldg (South), UCT Campus, Anzio Road Observatory, 7925, Cape Town, South Africa
* Correspondence: ndolo@icgeb.org

Even though agricultural exports remain a major contributor to the economies of many countries, some countries still experience significant trade challenges due to non-compliance with relevant residue standards (Maximum Residue Limits) in export markets. Such residue violations are common, especially in cases where there is widespread overuse of synthetic chemical pesticides to control late-season pests. This presentation focuses on how the strategic use of microbial biopesticides could help mitigate pesticide residues and ultimately ensure the production of safer and more wholesome food while fostering agricultural trade.

Keywords: microbial biopesticides; maximum residue limits; residue violations; export; trade.

IL5

Potential of cyclic lipopeptides-producing *Pseudomonas* strains in biocontrol

Monica Höfte[1*]

[1] Ghent University, Faculty of Bioscience Engineering, Department of Plants and Crops, Coupure Links 653, B-9000 Gent, Belgium

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Cyclic lipopeptides (CLPs) are metabolites with biosurfactant activity produced and secreted by a range of bacterial genera including *Pseudomonas* and *Bacillus*. CLPs are composed of a hydrophobic fatty acid tail linked to an amphipathic oligopeptide that is partly or completely organized in a cyclic structure. The three main families of *Bacillus* CLPs are surfactins, iturins and fengycins. *Pseudomonas* CLPs are much more diverse and are currently classified in at least 14 different groups based on the length and composition of the oligopeptide and number of amino acids in the cyclic peptide moiety. CLPs are implicated in bacterial motility and attachment to surfaces, including the formation of biofilms and they show antimicrobial activity against a broad range of micro-organisms. Moreover, they play an important role in the interaction of their producers with plants, either as resistance inducers or as phytotoxins contributing to plant pathogenesis. CLPs cause these effects by interacting with the lipids in membranes of microorganisms and plants. While various CLP-producing *Bacillus* strains are commercially available as bioprotectants, CLP-producing *Pseudomonas* strains are less researched and their potential is underutilized. We have been working on CLP-producing *Pseudomonas* strains for many years and have shown that they play an important role in disease suppressive soils, can directly control soilborne pathogens or induce resistance to leaf pathogens. The latest insights in the taxonomy, chemistry, ecology and biocontrol potential of CLP-producing *Pseudomonas* strains will be discussed.

Keywords: *Pseudomonas*; biocontrol; cyclic lipopeptides; disease suppressive soils.

IL6

Microbial products for agriculture – challenges of technology transfer to commercial application

Jovana Grahovac[1*], **Ivana Pajčin**[1], **Vanja Vlajkov**[1], **Marta Loc**[2] **and Mila Grahovac**[2]

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Demand for microbial products in agriculture comes from growing awareness of the public towards health and ecology and also hazards in the unselective use of synthetic agrochemicals. Researchers have academic interests in microbial products as sustainable alternatives for agrochemicals, but they often are not sufficiently motivated to work further on product development, registration and commercialization. There are examples of registered biocontrol agents which are not successfully implemented in the practical application due to lack of field efficacy, as well as examples of highly efficient agents which never reach the market due to issues with the scale-up process. Therefore, one of the key challenges in microbial products technology transfer is outlining a techno-economically feasible production process usable for scale-up at industrial level. In addition to upstream processing, downstream processing as well as the formulation of the final product are important from the aspect of preserving the viability, i.e. the activity of the active components, and the extension of shelf life of microbial products. From the academic point of view, the adoption of regulations on the intellectual property protection and management and the support to scientific institutions in the realization of technology transfer are key steps. For the success of placing microbial products in the market, both multifunctional and highly specific ones, the acceptance of these bioproducts by agricultural producers is a crucial challenge. The experience of our team has shown that involvement of agricultural producers in the product development procedure through citizen science approach can significantly contribute to overcoming this obstacle. Bioprocess development is a complex and time-demanding procedure. Thus it implies the necessity to involve scientists with different backgrounds in a multidisciplinary team and also requires the best possible basis for decision-making at each point of the product development, from the creation of the product idea to the market placement.

Keywords: microbial products; bioprocess development; scale-up; technology transfer.

IL7

Biosolution against microbes for safety and sustainable agriculture

Marina Soković[1*]

[1] Institute for biological research „Siniša Stanković" – National institute of Republic of Serbia, University of Belgrade, Bulevar despota Stefana 142, 11000, Belgrade, Serbia

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As the quest for sustainable agriculture intensifies, microbial solutions are increasingly recognized for their potential in promoting environmentally friendly farming practices. Current trends demonstrate the effective use of microorganisms in enhancing soil fertility, mitigating the negative effects of harmful pests and plant diseases, and bolstering crop yields, all while reducing the dependence on synthetic fertilizers and pesticides. Innovative biotechnological strategies are highlighted for their transformative potential in shifting modern agriculture towards more sustainable and eco-friendly practices. Detailed discussions encompass various types of beneficial microorganisms, such as plant growth-promoting rhizobacteria (PGPR), mycorrhizal fungi, and biopesticides. These are all crucial elements in improving nutrient uptake, stimulating plant growth, and protecting crops from pests and diseases. Further exploration is devoted to the role of microbial inoculants and biostimulants in improving soil structure, water retention, and overall soil health. The significance of microbial diversity within agricultural ecosystems is underscored, illustrating how fostering a balanced microbial community can enhance resilience to environmental stressors and elevate overall crop productivity. The discourse culminates with an examination of cutting-edge research and advanced techniques in microbial-based precision agriculture, such as soil microbiome analysis and the development of targeted microbial formulations designed to meet specific crop needs. In sum, this investigation provides a compelling glimpse into the future of sustainable agriculture, reshaped by the dynamic world of microbial solutions.

Keywords: PGPR; sustainable agriculture; microbial solutions.

IL8

Application of biostimulants for counteract abiotic stresses

Antonio Ferrante[1*]

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Cropping systems are moving towards more environmentally sustainable cultivation strategies. The European Green Deal and the reduction of nutrient losses are targets of modern agriculture. Biostimulants as extrcats form vegetal or raw materials or microrganisms can ensure a reduction in fertilizers application and yield losses.These agronomic tools must be applied to plants with a revised fertilization plan to improve crop sustainability. Biostimulants should avoid yield losses if a reduction in fertiliser is carefully applied. Biostimulants can also be used to counteract abiotic stress. Global climate change has impacted agriculture in several countries. Biostimulants can be used to enhance crop tolerance and avoid significant yield or quality losses. Timing, doses, and distribution modes can affect efficacy. Studies on application timing and abiotic stress occurrence are limited, and integrated approaches should be carried out considering weather forecasting and advanced non-destructive monitoring device utilisation. Biological defense activation should be performed in correspondence with stress incidence. The results demonstrate that biostimulants can play an important role in future agriculture.

Keywords: quality; nutrient losses; stress predictions.

IL9

The future prospect of bacterial fertilizers: from lab to market

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The use of environmental bacteria as bioagents for plant nutrition contributes to increase yield and improve soil health, being a sustainable alternative to reduce the input of chemicals in agriculture. Their ability in producing beneficial molecules and interacting positively with the plant-host helps to maintain a balanced microbial diversity and appropriated physical-chemical characteristics. To assert the potential benefits of a collection of 32 strains, a set of laboratory and field tests were performed from 2019 to 2023, targeting 3 raw crops: wheat, maize and sunflower. Strains were initially tested for their PGPR traits and for their compatibility when in consortium, and the most successful candidates were selected for field trials. Bacterial formulations were applied on seeds before sowing (seed dressing) at a concentration of 10^9 UFC/mL; volumes ranged from 100 to 250 mL/ha, according to the crop. Tests were arranged as follows: two trails on wheat, covering an area of 3,3 ha; seven trails on maize, on 30 ha; and four trails on sunflower, on 14 ha. Results indicated a substantial increase on growth and yield promoted by *Bacillus megaterium* M1.5 in wheat (6%), *B. megaterium* M1.5 + *Glutamicibacter halophytocola* 2a in maize (6 to 16%) and *Herbaspirillim seropediaceae* SIMB8 on sunflower (13% applied alone and 6% with M1.5). *B. megaterium* M1.5 was able to improve yield in all crops tested, and its beneficial activity was observed both alone and in consortium, being a valuable bioagent to be used as biostimulant. Due to their remarkable effect, these bacteria were subsequently formulated and produced on an industrial scale to compose 3 efficient microbial fertilizers branded SPEED4SEED, two of each are already available for commercial use in Serbia and currently undergoing registration in Italy. Soil bacteria can bring huge benefits to agriculture, evidencing the importance of developing and validating promising microbial inoculants.

Keywords: biostimulants; bioagents; microbial inoculants; soil bacteria.

IL10

Living soil: a new paradigm for an innovative agriculture. A practical case to phytoparasitic nematodes biocontrol using probiotics and prebiotics approach

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Over the last decade EU legislation has restricted the usage of chemicals and fumigants applied for the control and management of root-knot nematodes. This trend can be accounted for as an effort to reduce climate warming effects and prevent soil and water contamination. The increase in demand of alternative plant defense strategies followed a rise of studies concerned with the identification of agronomic measures and the application of microbial biocontrol formulations, with the intent of exploiting the community of biological soil antagonists. Several antagonistic microorganisms are suitable for biocontrol, including hyphomycetes like *Pochonia chlamydosporia*. *Pochonia* is a nematophagous fungus egg parasite of root-knot (*Meloidogyne* spp.), cyst nematodes (*Heterodera* spp. and *Globodera* spp.) and false RKN (*Nacobbus* spp.). This species, unlike other nematophagous fungi, is unable to produce any structure to capture nematodes but can infect the eggs and females during the sedentary phase in some species (mature females). The fungus produces resistant structures (chlamydospores) which allows it to survive even in the absence or scarcity of nematodes. *Pochonia chlamydosporia* is distributed worldwide and may also be able to adopt saprotrophic and endophytic lifestyles, colonizing the roots of several plant species inducing a plant defense and resistance mechanisms in fungal, plant, nematode interactions. This multitrophic behavior refers to those microorganisms such as *Pochonia* that do not have an obligatory trophic relationship with the host. It is precisely because of this complex trophic behavior that such fungi have the potential to be effectively employed in biocontrol, bio fertilisation and biostimulation methods. In fact, in a modern context in which soil biodiversity assumes a central role in crop management, knowledge of the intimate relationships between plant, microbiome and soil become of fundamental importance. In this communication we define the concept of living soil, the prebiotic approach and probiotic and finally we propose a practical example of the use of this new technology for the management of phytoparasitic nematodes on a tomato crop.

Keywords: living soil; endophyte; *Pochonia*; soil; prebiotic; probiotics.

IL11

Diverse functions of microorganisms in agriculture: which function is fulfilled when, or all at once?

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A microorganism is a microorganism no matter what you call it; whether you call it a Plant Protection Product (PPP), a biostimulant, or a fertiliser. Each microorganism has a variety of functions or modes of action. Once applied to the plant, it's no longer possible to influence these functions. In vitro, it's possible to influence the result of the microorganism by using different nutrients, different temperature, different culture time, etc. In nature, on the plant (leaf or root) the output can change more than twice within 24 hours. Moreover, other microorganisms already present on the plant have a great influence (in in vitro culture you have only one). With this in mind, it's difficult to describe what type of product is produced. Some bacteria are able to fix N from the air. These are free-living and also endophytic strains. But under what conditions do they fix N and what additional function makes them be called PGPB? It's the whole variety of functions that make them grow plants better. In one product development, a two-year trial on potatoes against *Rhizoctonia* with a *Pseudomonas* strain showed the same reduction in pathogen as the usual fungicide in both years. However, the results showed a high yield increase in one year and not in the other. As a PPP, this strain shows reliable effects, but as a fertiliser it doesn't. Is it a fertiliser or a PPP? What is the main function and what are the side effects? Today, this *Pseudomonas* isolate is approved in Europe and the USA as a PPP against *Rhizoctonia*, with some nice side effects.

Keywords: mode of action; deversity function; plant protection product**.**

IL12

Contact-independent bacterial signalling in the plant beneficial microbiome

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The low cost in next-generation-sequencing and development of computational tools is generating extraordinary insights into the taxonomic and functional composition of microbes associated with plants. Plant associated microbial communities have key roles in biotic and abiotic stress tolerance and nutrient acquisition. Plant-microbe associations occur at the leaf aerial parts in the phyllosphere and at root level in the rhizosphere. The rhizosphere (the nearest soil area to the roots) hosts a rich microbial plant community which provides a series of beneficial outcomes related to plant growth. Plant roots recruit their rhizosphere microbiome from bulk soil and a small number enter colonising the root endosphere and some then move to other plant organs. The phylogenetic conservation of plant microbiomes infers an organized assembly which is directed by mechanisms at large unknown. These involve cell-cell interactions, plant-microbe signalling and root exudate effects. Microbial cell-cell communication regulates metabolic and physiological activities in response to the host, environment and microbial neighbors. Plant microbiomes contain a large number of bacterially produced molecules such as quorum sensing signals, volatiles and secondary metabolites which play cell-cell signaling roles amongst members of the microbiome. Our understanding of the different signal molecules and their roles which are produced in a microbial community, is very much in its infancy. Understanding the chemical languages that shape the plant microbiome will be informative on how these communities contribute to plant health. It will also lead to the development of prebiotic compounds and probiotic competence for a more sustainable agriculture of economically important crops.

Keywords: bacterial signalling; cell-cell interactions; cell-cell communication; microbiome.

IL13

When biodiversity preservation meets biotechnology: the challenge of developing synthetic microbiota and the case of symbiotic nitrogen-fixing bacteria

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Symbiotic nitrogen fixation is one of the main sources of assimilable nitrogen in ecosystems and key for developing low-input agriculture and sustainable food production. However, developing effective bioinoculants from symbiotic nitrogen fixing bacteria (rhizobia) is not trivial, due to the large genomic and phenotypic diversity of rhizobia and their variable effect on the host plant, which limit the possibility of fast identification of elite strains in the collections. In fact, while the intricate core symbiotic machinery required to establish a successful rhizobia-host plant interaction has been mostly elucidated, much remains unknown about the genes required to optimize the interaction and which can determine the symbiotic variability found in nature. We addressed this problem using both in silico and in vitro modelled rhizosphere and nodulation conditions and a variety of natural isolates of the species *Sinorhizobium meliloti* and cultivars of its host plant alfalfa. By performing Genome-Wide Association Studies and comparative transcriptomics, we showed the presence of a large number of genotype-by-genotype (GxG) interactions that can have prominent impacts on symbiotic outcomes. Moreover, GxG interactions with the rhizosphere nonsymbiotic microbiota were identified, providing the basis of knowledge for rational development of synthetic microbiota. These findings may allow to identify genomic markers for selection of elite strains and highlight the importance of investigating natural isolates in the perspective of application as host genotype-specific bioinoculants.

Keywords: nitrogen fixation; symbiosis; rhizobia; legumes.

IL14

The virulence factors orchestra of the bacterial pathogen of woody hosts *Pseudomonas savastanoi*

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Plant pathogens reduce yield and quality of agricultural production and reduce food security, and their control generally involves the use of harmful chemical pesticides. Novel therapeutic approaches to develop alternative treatments include targeting of pathogens' virulence factors, for which design, identifying the molecular key points of pathogenicity is essential. *Pseudomonas savastanoi* strains infecting woody hosts (olive, oleander, ash, broom and dipladenia) are well-established models for the study of the molecular determinants of bacterial phytopathogens. During the past few years, comparative and functional genomics analyses have facilitated our group the identification of pathogenicity determinants that may underlie host range definition and the evolution of virulence in this bacterial group. *P. savastanoi* features exclusively found in the genomes of bacteria isolated from woody hosts include several proteins effectors injected in the plant cytoplasm through a multiprotein "molecular syringe" called type III secretion system, and a genomic island of about 15 Kb involved in the catabolism of phenolic compounds. Other molecular determinants involved in tumor (knot) formation induced by *P. savastanoi* in olive plants include the phytohormones indole-3-acetic acid and cytokinins, the bacterial second messengers cyclic-di-GMP and Ca^{2+}, and quorum sensing molecules allowing communication among *P. savastanoi* cells and other members of the knot microbiome. Global transcriptome analyses of *P. savastanoi* lacking single determinants revealed that a finely tuned coordination between virulence factors choreographs the process of tumor formation. The virulence factors orchestra also counts with the master director GacA, the response regulator of a two-component system controlling the expression of most determinants. Our findings establish the crosstalk existing among the most relevant virulence factors in a group of bacterial pathogens, and reveal key regulators as potential targets for developing novel strategies to control bacterial diseases of woody plants.

Keywords: *Pseudomonas savastanoi*; phytopathogen; virulence factor; woody host; olive.

IL15

Unveiling the role of bacterial sociality in designing plant growth promoting inoculants

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Bacteria exhibit a fascinating blend of cooperative and competitive behaviors, making them truly social creatures. Interestingly, they can differentiate between close relatives and less related conspecifics, displaying kin discrimination. This intriguing trait profoundly impacts the fitness and spatial distribution of genotypes in biofilms, as well as their horizontal gene transfer and the stability of cooperation. The rhizosphere microbiome, often referred to as the "second genome" of plants, plays a pivotal role in fostering plant growth, nutrition, and overall health. Despite its importance, we have yet to fully comprehend the social dynamics of the resident rhizosphere microbiota. Moreover, the impact of growth-promoting rhizobacteria (PGPR) on the social behavior of the rhizosphere community remains largely unexplored. To address these questions, we recently conducted a study using the PGP strain *Bacillus subtilis* SQR9 and cucumber plants as our model system. Employing advanced metagenomic approaches and bacterial isolates, we delved into the consequences of SQR9 inoculation on the diversity, connectedness, and social behavior of the rhizosphere bacterial community. Our findings offer valuable insights into the intricate social interactions occurring within the rhizosphere microbial community. Additionally, we shed light on the implications of our research for the development of effective multistrain inoculants, which hold promise in further boosting crop performance while minimizing reliance on chemical interventions in agriculture.

Keywords: *Bacillus subtilis*; bacterial interactions; kin discrimination; swarming; rhizosphere microbiome.

IL16

Bacterial-fungal interactions and impacts on plant growth and health

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Fungi and bacteria cohabit in the plant environment. Separately they have been intensively studied for their impacts on plant growth and health. Except some particular cases, little attention has however been paid to the intricacy of fungi-bacteria interactions. Using different models, we analyzed some associations between fungi and bacteria. The mycorrhiza-like fungus *Serendipita indica* was firstly used. The fungus holds capabilities to enhance plant growth and to confer resistance to different stresses and it is also known to host an endofungal bacterium, *Rhizobium radiobacter*. However, there was still a gap of knowledge on the effect other bacteria can also have on the fungus. We analyzed how co-inoculations of bacteria and *S. indica* influence plant growth and protection against fungal pathogens. Bacteria-fungus associations seem to cooperate in the process of fungal root colonization and establishment, e.g., by increasing fungal sporulation and hyphae expansion. Moreover, some bacteria enter inside the fungus, and synergism between the fungus and some bacteria increases plant protection against fungal pathogens. Another recent discovery was made on a wood fungal pathogen. We determined that a particular fungal species hosts endofungal bacteria. Interestingly, all fungal isolates belonging to this species screened at this moment, contain several taxa of bacteria inside their hyphae, independently from the fungi's isolation source. However, the bacterial endosymbiont's roles are also still unknown. We aim to determine putative persistent hypho-bacteriomes in fungal isolates belonging to the same species across Europe, and from different hosts, and to classify the type of association. We highlight the importance of multipartite microbial interactions between fungi and bacteria and increase our understanding on these associations.

Keywords: plant-microbe-microbe interactions; endofungal bacteria.

IL17

Molecular-level investigation of algal-bacterial interactions and their utilization for plant biostimulation

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Past empirical research has demonstrated that bacterial interaction might enhance algal biomass production and algal biohydrogen evolution. To investigate the mechanisms and microalgal functions activated under bacterial associations different bacterial species were co-cultivated with *Chlamydomonas reinhardtii* cc124 green algae. Bacterial species were isolated from diverse environments including biogas sludge, soil and commercial biostimulant products. Pairwise algal-bacterial combinations were cultivated for five days in synthetic wastewater. We have investigated the effect of bacterial phylogenetic relationship and growth rate on algal growth, biomass yield, nutrient intake, biomolecule and biohydrogen production. Also, we have tested selected stable algal-bacterial combinations in plant biostimulation studies. We have observed, that appropriately selected combinations of green algae and bacteria enhanced tomato growth and stress resistance compared to plants treated solely with algae or bacteria, respectively. Transcriptomic studies on tomato plants revealed differential expression of several genes involved in defense and response to abiotic stresses such as cold and water deprivation relative to the various control treatments. These results show the potential of microalgae-bacteria combinations as improved biostimulants to not only promote growth but also enhance plant's tolerance to stress.

Keywords: tomato; biostimulation; green algae; *Chlorella*; exopolysaccharide.

IL18

Microbiome-assisted selection of endophytic bacteria for the sustainable cultivation of tomatoes in greenhouse

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The rhizosphere is home of several species with beneficial traits influencing plant development and health and counteracting deleterious soil-borne phytopathogens. The analysis of microbial community based on high-throughput amplicon sequencing provided a new window into the study of these plant-microbiota interactions. Due to the potentiality of microorganisms to promote plant growth and biocontrol plant diseases, numerous approaches, including the design of microbial consortia, have been undertaken. We used both the cultivation-dependent approach and amplicon-based metagenomics to study tomato root-associated microbiome in commercial greenhouse conditions. Microbial communities of the seed endosphere, rhizo- and endorhizosphere of tomato seedlings in nursery and of plants grown in greenhouse were analyzed. According to the results, bacterial communities were dominated by the phyla *Proteobacteria*, *Bacteroidota*, *Actinobacteriota* and *Firmicutes* in all microhabitats, although the relative abundance could vary based on the growing conditions and the compartment (rhizosphere, endosphere). The selection of cultivable bacteria enabled us to obtain representative isolates which showed diverse plant growth-promoting features and marked antimicrobial activity *in vitro*. The representative sequences of the OTUs belonging to the rhizosphere and endorhizosphere samples collected in greenhouse were identified as part of the keystone OTUs or core microbiome. Sequences were then aligned to those from the cultivated bacteria. According to the level of similarity (> 97%) resulting from the alignment and the plant growth-promoting phenotypic traits, a set of 10 bacterial endophytes, including the most represented genera, *Bacillus* and *Pseudomonas*, but also less known genera, were selected for further studies. Individual isolates or bacterial consortia were applied as water suspensions by soil drenching immediately after transplanting near the root zone of tomato plants. Most of the bacterial isolates used individually or as bacterial consortia showed a relevant plant growth promotion activity, induction of plant resistance and biocontrol potential against tomato plant pathogens.

Keywords: tomato; endophytic bacteria; microbiome; biocontrol bacteria; bacterial consortia.

IL19

Using fitness landscapes to engineer microbial communities

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Microbial consortia are a promising avenue for the development of sustainable solutions in agriculture and biotechnology. However, rationally engineering microbial ecosystems remains a major challenge. The functions and services that microbial consortia provide depend on an intricate network of interactions between species and their environment, making it extremely difficult to predictively link the composition of a community to its function. Because the number of possible interactions combinatorially explodes with the size of the consortium, the experiments required to characterize these networks become infeasible for even moderately large consortia. Alternative approaches characterize the function of larger communities by coarse-graining their composition to species diversity, thereby losing species-level resolution. To overcome these limitations, we extend the theory of fitness landscapes from quantitative genetics to map the species composition of a community to its function. Using this theory, we quantitatively dissect how species interactions drive the collective function of the community. We then demonstrate that the complex network of interactions underlying community function often distills into simple species-by-ecosystem functional relationships. These are reflected in simple linear trends linking the functional effect of adding a species to the function of the receptor community. These emergent trends can be leveraged to predict the function of arbitrary communities from their species composition with minimal experimental effort. We discuss different approaches to this end, ranging from relatively simple application of linear regressions to more sophisticated machine-learning approaches. Our work illuminates an unexplored path toward the rational engineering of ecological systems, with potential use across agricultural, biotechnological, and environmental applications.

Keywords: microbial consortia; microbial community engineering; fitness landscapes; theory in microbial ecology.

IL20

Bacterial VOCs contrast rice blast infection by inhibiting fungal virulence rather than growth

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Bacterial volatile organic compounds (VOCs) are key mediators of biotic interactions, may act as growth inhibitors, and can interfere with secondary metabolisms. VOCs produced by plant-associated bacteria have been proposed as tools to control fungal pathogens by inhibiting fungal growth. However, growth inhibitors also activate the process of selecting resistant forms that disable the virulence inhibition. We have reverted the growth inhibition paradigm by considering VOCs that may interfere with the virulence process of fungal pathogens rather than killing them. This perspective should transform a harmful pathogen in a neutral saprophyte. We have tested this hypothesis on the melanin-producing fungal phytopathogen *Magnaporthe oryzae*, a worldwide threat for rice. We have selected three bacterial strains from the rhizosphere of rice, whose VOCs did not inhibit *M. oryzae* growth, but could reduce the black pigmentation. Exposure to the VOCs strongly affected *M. oryzae* sporulation and conidia germination, and completely inhibited appressoria formation. By head space solid-phase micro-extraction gas chromatography mass spectrometry, we identified nine volatile molecules that were shared in the VOCs mixtures of the three strains. Among these, 1-butanol-3-methyl reduced of 60% fungal pigmentation and completely inhibit conidia germination *in vitro* when used alone. The inhibition effect was confirmed *in vivo* on rice leaves, with less than 6% lesions incidence in VOCs-exposed leaves, respect to 98% in non-exposed leaves. Optical and scanning electron microscopy of rice leaves inoculated with *M. oryzae* conidia revealed the abolishment of conidia germination and of the tissue invasion by the pathogen. Fungal metatranscriptomic analysis upon exposure to the bacterial VOCs indicated a transcription inhibition of the melanin biosynthesis pathway. VOCs of rice associated-bacteria have the potential of switching off *M. oryzae* virulence, without affecting the disarmed fungal survival and avoiding the selection of resistant forms.

Keywords: VOCs; bacteria; *Magnaporthe oryzae*; rice; virulence inhibition.

IL21

Bacilli in the rhizosphere: interaction and evolution within the biofilm

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To understand how microorganisms, specifically the genus of *Bacilli*, interact with the plant microbiome and how this interaction contributes to plant protection and growth promotion, we apply a series of eco-evolutionary approach to understand the underlying molecular mechanisms of *Bacilli* during interaction with plants. Here, I will present the intricate interaction between *Bacilli* and the *Pseudomonas* genus [1,2] and explain the evolutionary changes observed in *Bacilli* either during interaction with other microorganisms or during plant root colonization [3-5]. Our results pinpoint towards the molecular details of the life of a plant promoting bacterium.

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Keywords: *Bacillus subtilis*; biofilm; plant microbiome; secondary metabolites; evolution.

IL22

Engineering microbial communities by the colloid biology approach

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In nature, microbes form different multicellular structures such as flocs and mats that enable microbes to survive harsh conditions, provide anaerobic niches for the growth of anaerobic bacteria and give possibilities for many interspecies interactions. These structures can be naturally formed when environmental conditions are appropriate. However, it is very difficult to study such structures and even more demanding to construct them in use in different environmental applications. The colloid biology approach relies on the tools that are applying electrostatics on biological objects. Since microbial cells resemble negative surface potential that is attributed to the negative chemical groups present in the membrane and cell wall, the modification of these surfaces enables electrostatic interactions between cells. Therefore, in our laboratories, we were focused on the development of a set of different approaches based on the electrostatic interactions that enable the physical attachment of alive bacterial cells to other cells or on different inanimate surfaces. Using the approach and manipulation of conditions (e.g. ionic strength, flow, the density of cells, etc.) we were able to construct different synthetic polymicrobial structures such as biofilms and aggregates. Using this approach we put cells together and further advanced it by spatially orienting cells by the design. Through studies of the formation and further development of the structures in time, we obtained insights on (i) the self-organisation of structures where strict anaerobes and aerobes became associated, (ii) observing niche separations by the formation of anaerobic and aerobic zones and (iii) rewiring ecological interactions from competition to the cooperation and (iv) developing methods for isolation of spatially distributed consortia for agricultural and remediation purposes.

Keywords: colloid biology; microbial aggregates; spatial distribution of microbes; microbial carriers; encapsulation.

IL23

The place of microorganisms in the transition to new farming systems with reference to research, agronomy and IT, using greenhouse vegetables as example

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The generalised use of chemical products in agriculture led to considerable environmental damages in particular to soils and bio-diversity. Sustainable farming requires to restore in priority soil health whilst ensuring farmer's economical sustainability. The use of Microorganisms is essential to addressing soil fatigue. It is generally accepted that they increase vitality and fertility, prevent pathogen development and overall increase root development. But adding microorganisms to damaged soils may not provide the expected results. The existing microbiome, often unbalanced in favour of pathogens, and key agronomic parameters including carbon level, water management and nutrition deficiency, will condition the soil, thus the plant. This document will look at the understanding of soil conditions and microbiome in a greenhouse environment, the field data that are potentially available to the grower and the methodology he can use to interpret the information, define programmes to act positively on the microbiome, bringing his soils back to production. In most situations, programmes may include specific strains of microorganisms to act on the soil vitality. Regenerative actions have also to be taken over time to address the organic matter level and the presence of pathogens. Questions remain about the impact of such combined programmes on bio-diversity.

Keywords: microorganisms; agronomy; chemical products; environmental damages.

IL24

The effects of bio-fertilizer application on grain yield and quality of maize grown in long-term monoculture

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Monoculture is cropping system with high environmental risk. While maize monoculture is still present in many regions, more sustainable outcomes are required. The experiment with maize monoculture at the Maize Research Institute started in 1972, while period 2013-2019 was comprehended. Crop residues were incorporated on whole area. Experimental factors followed: 1) application of farmyard manure (M) (every 3 years, 60 t ha^{-1}) and without manure (MØ); 2) bio-fertilizer Bioplug (accelerates decomposition of crop residues), applied in amount of 5 l ha^{-1} (BF), 2.5 l ha^{-1} (½BF) and without it (BFØ); 3) mineral fertilizer application: NPK N:P:K= 15:15:15 in autumn (1709 kg ha^{-1}) + Urea in spring (320 kg ha^{-1}) (F1); only Urea in spring (320 kg ha^{-1}) (F2) and without mineral fertilizers (F3). Soil organic matter (SOM), as well as grain yield and concentration of protein, Mg, Ca, Fe and Zn were measured. Both ½BF and particularly BF increased SOM over time, in comparison to BFØ. Greater SOM values were on average present in M (0.7% higher than in MØ), BF (0.5% higher than in BFØ) and urea (0.4% higher than in F3. The average values of grain yield, protein and Zn concentration were 8.7%, 0.4% and 8.7%, respectively greater in S than in SØ, while Mg, Ca, and Fe concentrations were 1.4%, 1.8% and 8.8% lower in S variant. Also, greater average values of grain yield, Mg, Ca, and Zn were achieved in the ½BF (13.1%, 5.5%, 1% and 5.5%, respectively) and in the BF (15.6%, 3.4%, 3.9% and 6.7%, respectively), compared to the BFØ. Results indicate that the bio-fertilizer application in both doses (½BF and BF), particularly in combination with manure, contribute to the SOM increase. Also, BF+F1 could enhance yield potential and concentration of protein, Ca, and Mg, while in BF+F2 absorption of Fe and Zn could be promoted, thus mitigating potentially negative aspects of monoculture system.

Keywords: fertilization; protein concentration; essential elements; soil organic matter; continuous cropping.

IL25

Seed bio-priming with microbial inoculants: A tailored approach towards improved crop performance, nutritional security, and agricultural sustainability for smallholder farmers

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The seed is the most primary requirement of agriculture. Hence, it is crucial to maintain the quality of the seeds through nature-based solutions having multiple benefits. Among the pre-sowing techniques, bio-priming has emerged out to be the most simple, economical, and eco-friendly delivery system of beneficial microorganisms in the agroecosystem. Therefore, comprehension regarding the applicability of bio-priming in restoring soil health, improving plant nutrition, and maintaining crop/seed quality has a major role in achieving the UN-Sustainable Development Goals such as no poverty, zero hunger, good health and well-being, etc. Our efforts to minimize the nonrenewable energy sources and promote the use of microbial resources at the community level need to be re-emphasized. Identification of sustainability indicators like soil enzymes, growth hormones, organic acids, etc. under bio-priming based agroecosystems is imperative for utilizing the potential microbes in the best manner. This work highlights bio-priming as a pragmatic technological option in achieving the tripartite goal of food-nutritional security, environmental stewardship/quality, and agricultural profitability under different agroecosystems.

Keywords: nature-based solutions; crop growth improvement; microbial consortia; soil health; crop quality; sustainability.

IL26

Challenges in the management of some key horticultural pests

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Agricultural products in general and horticultural products in particular face high-quality standards required by retailers and customers: only unblemished fruits, vegetables and ornamental plants are marketable. There is an ever-increasing demand for impeccable products that are also free of pesticide residues. This alone presents great challenge to pest management in horticultural crops. The challenge is magnified by the great diversity of horticultural crops grown, each crop on a relatively small area, leading to a limited interest for developing novel methods of pest control. However, novel IPM strategies of horticultural crops are needed to reduce the reliance on synthetic pesticides. The number of registered plant protection products (PPPs) or available mode of actions in the management of pests is constantly decreasing in the European Union (EU). As the development rate of novel IPM strategies is not keeping up to the phase-out of PPPs, the management of several horticultural pests is increasingly difficult. This challenge is demonstrated by presenting the currently available IPM strategies of some key horticultural arthropod pests. If effective and economically viable IPM strategies are not available, than more and more growers give up crop production. In order to prevent any further reduction in horticultural crop production in the EU, novel IPM strategies must be available on the market.

Keywords: horticultural pests; integrated pest management; plant protection products.

IL27

The use of microscopic tools for the increase of resolution in mycorrhizal assessment

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Arbuscular mycorrhizas are symbionts in the roots of most plants. The hyphal network developed in soil act as an extension of roots. The advantages of symbiosis are represented by an enhanced nutrient absorption, a higher protection to biotic and abiotic stressors and the ability to regulate the nutrient flow at ecosystem level. The arbuscules developed in root cells are specific structures for nutrient exchange between the fungus and the host plants. The main challenge in evaluating the success of symbiosis is the scale and resolution of microscopic assessment. There are several methods which use different scales to highlight the structures and the expansion of mycorrhizas in roots. This must be constantly discussed to find the most suitable method to provide an accurate assessment of the host dependency to symbiosis. The use of mycorrhizal pattern method increase the resolution of microscopic assessment to 1%, which is consistent to a clear image of mycorrhizal development in roots. Each structure – hyphae, arbuscules, vesicles, spores and auxiliary cells are connected to the entry points – and offer a clear overview of the symbiotic mechanism. Based on microscopic images, each structure is coded into a specific color and an entire root segment is converted in virtual map of colonization. All mycorrhizal maps are analyzed through a multi-point analysis to identify the direction of mycorrhizal expansion, the discontinuities in intraradical hyphal networks and the specific structures developed in each point of the root. All data are used to assess the colonization parameters, the strategy of fungi in roots of host plants and the future projection of development. The resolution of microscopic assessment combined with the mycorrhizal maps are an efficient tool to increase the understanding on the symbiotic mechanism.

Keywords: symbiosis; colonized roots; colonization strategy; multi-point analysis; mycorrhizal maps.

IL28

New types of biofertilizers: microparticles with biological and chemical active components

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One of the global problems and challenges is climate changes which has got a negative impact on nature, environment, human health and life. One of the reasons for climate change is the excessive use of agrochemicals in modern agriculture, which uses 60% of water and 70% of land resources and has a great impact on biodiversity. The goal of this research is to prepare/characterize and apply new biopolymer microparticles filled with a microorganisms and a chemical reagent with a 3in1 effect (nutrition, protection, and time release). Biopolymer microparticles were prepared using the green technology of an optimized encapsulation process and were characterized physiochemically with the help of various spectroscopic and optical methods. New biopolymer microparticles were applied to different plant cultures (lettuce, tomatoes) (open field, hydroponic and foliar). The use of microparticle formulations reduced the use of agrochemicals by 30% and stimulated the biosynthesis of secondary plant metabolites (antioxidants), which have a positive impact on human health and life.

Keywords: biofertilizers; encapsulation; microparticles; nutrition; functional food.

GRANT AWARDED POSTER PRESENTATIONS

GAPP1

Isolation, molecular characterization, and efficacy of native entomopathogenic fungi: *Beauveria bassiana* and *Metarhizium anisopliae* against *Plutella xylostella* (Linnaeus) (Insecta: Lepidoptera: Plutellidae)

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Beauveria and *Metarhizium* are entomopathogenic fungi (EPF), have been previously reported as effective control agent for the *Plutella xylostella*. The pathogenic activity of this fungus against the targeted pest is affected by regional environmental conditions & identifying native EPF species is important strategy for developing potential biopesticides with standard qualities as host specificity, climatic appropriateness, and effective pest population suppression. The aim of the study was to identify *Beauveria* and *Metarhizium* species isolated from soil samples of Western Uttar Pradesh, India. These isolates were identified as *B. bassiana* and *M. anisopliae* after molecular screening using PCR amplification with the ITS region primer. In a contact toxicity experiment, both SVPUAT isolates were tested against 4th instar larvae of *Plutella xylostella* at doses ranging from 1×10^3 to 1×10^8 spores ml^{-1}. The lethal time LT50 and LT90 values of *B. bassiana* with 1 ×108 spores ml^{-1} were 6.61 and 10.95 days, respectively. The lethal time LT50 and LT90 values of *M. anisopliae* with 1×10^8 spores ml^{-1} were 6.47, and 10.91 days, respectively. The present SVPUAT isolates were identified using the ITS-5.8s-rDNA region from GenBank and has showed significant pathogenicity to *Plutella xylostella*. The identification of native EPF species gives an excellent chance to obtain biological control agents with standard properties for developing biopesticides.

Keywords: biological control agent; *Beauveria bassiana*; *Metarhizium anisopliae*; ITS; phylogenetics; *Plutella xylostella*.

GAPP2

Designing synergistic biostimulants formulation containing autochthonous phosphate solubilizing bacteria for sustainable wheat production

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Phosphate solubilizing bacteria (PSB) as bio-fertilizers has enormous potential for sustainable agriculture. Despite this, there is still a lack of information regarding efficient formulation strategies for the development of potential biofertilizers. In this study, we investigated rock phosphate solubilizing *Ochrobactrum sp*. SSR (DSM 109610), *Enterobacter* spp. DSM 109592 and DSM 109593 for an efficient formulation. PSB-inoculated soil supplemented with rock phosphate (RP) showed an increase in plant P (2%), available soil P (4.7%) and alkaline phosphatase (6%) activity. Root architecture improved by PSB inoculation, with higher root length, diameter and volume. *Ochrobactrum* sp. SSR and *Enterobacter* spp. were further used to design bioformulations, using four organic amendments, biochar, compost, filter mud and humic acid. All four carrier materials maintained adequate survival and inoculum shelf life of the bacterium, as indicated by field emission scanning electron microscopy analysis. Filter mud (FM) based bio-formulation was most efficacious and enhanced not only wheat grain yield (4-9%) but also seed P (9%). Moreover, FM based bio-formulation enhanced soil available P (8.5-11%) and phosphatase activity (4-5%). Positive correlations were observed between the PSB solubilization in the presence of different insoluble P sources, soil available P, soil phosphatase activity, seed P content and grain yield of field grown inoculated wheat variety Faisalabad-2008, when di-ammonium phosphate fertilizer application was reduced by 20 %. The present study provides valuable groundwork for designing field scale formulations that can maintain inoculum dynamics and increase shelf life.

Keywords: autochthonous psb; biofertilizers, biostimulants; scanning electron microscopy.

GAPP3

Chickpea seed endophytes: future of sustainable agriculture

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Seed microbiome is likely to have important impacts on plant fitness and productivity but functional potentials of seed microbiome remain poorly understood. It is also suggested that bio-inoculants developed from or compatible with seed microbiome are more likely to produce desired outcomes of sustainable increase in agriculture productivity but few empirical evidences are available. The aim of this study was to identify culturable endophytic microbiome of the chickpea (*Cicer arietinum* L.) seeds (P-362), and their functional attributes. Phylogenetic analysis based on 16S rDNA showed that the dominating seed endophytic bacteria belong to *Bacillus* sp., *Pseudomonas* sp., *Enterobacter* sp., *Staphylococcus* sp. Isolates produced significant amount of Indole-3- acetic acid, ammonia, solubilised different minerals, and also inhibited the growth of chickpea plant pathogen (*Fusarium* sp.) under laboratory conditions. Which directly help the plant growth and development. Several seed endophytes induced significant increase in plant growth and increased tolerance of chickpea plants to the pathogen when tested *in vitro*. Reintroduction of these isolates, resulted in significant increase in plant length, biomass, plant pigments and bio-controlling activity against *Fusarium* sp. These results provide a direct evidence for the presence of beneficial seed microbiome and suggest these isolates could be further developed into potential bioinoculants for improving diseases management and sustainable increase in agriculture productivity.

Keywords: seed microbiome; plant growth promoting endophytic microbes; sustainable agriculture; biocontrol; biofertilizer.

GAPP4

Efficacy of microbial biopesticides on the control of cotton pests in South Africa

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Cotton (*Gossypium hirsutum* L.) is one of the essential fibre crops; however, several factors, including insect pests, affect production. Biopesticides can play a vital role in the integrated programme to address the challenges that limit production and reduce profits for cotton farmers. Two field trials were conducted to evaluate the efficacy of biopesticides compared with insecticides against sucking cotton pests. In the first trial, four biopesticides (Eco-Bb®, Bb endophyte, Bolldex®, Delfin®) were compared with a pyrethroid, Karate® against *Helicoverpa armigera*. The treatments of Karate® and Bolldex® significantly reduced the *H. armigera* population, while the treatment of Eco-Bb® had the lowest number of damaged bolls. The highest average seed cotton yield of 6 400 kg.ha-1 was recorded in the plots treated with Bolldex®. The efficacy of different biopesticides against *H. armigera* varied significantly; however, Karate® and Bolldex® resulted in better control of the pest. In the second trial, a study was conducted to evaluate three biopesticides, Eco-Bb®, Bb endophyte, and Eco-Noc, in comparison with the insecticides Chlorpyrifos® 480 EC, Karate® EC, and Bandit® 350 SC to determine their efficacy against sucking pests, *Jacobiella facialis*, *Aphis gossypii*, *Thrips tabaci*, *Bemisia tabaci*, *Tetranychus urticae* and *Dysdercus* spp. Karate® significantly reduced the *J. facialis* while the biopesticides had some control of the *A. gossypii*. Plots treated with Eco-Bb® and Bandit® 350 SC had the lowest number of *T. tabaci*, and there were no significant differences in the populations of *B. tabaci*. All treatments, except for Bandit®, significantly reduced the number of *T. urticae*. The highest average cottonseed yield of 6 395 kg.ha^{-1} was recorded in plots sprayed with Bandit®. In summary, the efficacy of different microbial biopesticides against cotton pests varied significantly and, in some instances, was on par with the synthetic pesticides. The results suggested that biopesticides may be potentially used in the sustainable control of cotton bollworms as part of integrated pest management programmes. However, further work is required to support this hypothesis.

Keywords: biopesticides; microbial; insecticides; cotton; South Africa.

GAPP5

Differential responses in some quinoa genotypes of consortium of beneficial endophytic bacteria against bacterial leaf spot disease

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Many effective plant-microbe interactions lead to biological changes that can stimulate plant growth and production. This study evaluated the effect of the interaction between quinoa (*Chenopodium quinoa* Willd.) and endophytic bacterial strains on differential responses under biotic stress. Four strains of endophytic bacteria were used to inoculate three quinoa genotypes. Endophytic bacteria were used to evaluate their biocontrol activity against *Pseudomonas syringae* on quinoa plants, which causes leaf spot disease, depending on some different parameters. Quinoa genotype plants were treated with four treatments: pathogenic bacteria only (T1), internal bacteria only (T2), pathogenic bacteria + endogenous bacteria (T3), and untreated as the control (T4). The results indicated that there was a significant difference between chlorophyll content index of infected plants without bioagent (untreated) compared to plants bio-inoculated with endophytic bacteria. The highest mean disease incidence was on the plants without bacterial inoculum (90%, 80%, and 100%) for quinoa genotypes G1, G2, and G3, respectively. The results showed that there were significant differences in the weight of grains / plant, as the value ranged from 8.1 to 13.3 g when treated with pathogens (T1) compared to the treatment with pathogens and endogenous bacteria (T3), which ranged from 11.7 to 18.6 g / plant. Decreases in total aromatic amino acids appeared due to the pathogen infection, by 6.3%, 22.8%, and 24.1% (compared to the control) in G1, G2, and G3, respectively. On the other hand, genotype G3 showed the highest response in the levels of total aromatic and total neutral amino acids. The endophytic strains promoted quinoa seedling growth mainly by improving nutrient efficiency. This improvement could not be explained by their ability to induce the production of amino acids, showing that complex interactions might be associated with enhancement of quinoa seedling performance by endophytic bacteria. The endophytic bacterial strains were able to reduce the severity of bacterial leaf spot disease by 30%, 40%, and 50% in quinoa genotypes G1, G2, and G3, respectively, recording significant differences compared to the negative control. The results indicated that, G1 genotype was superior in different performance indicators (pathogen tolerance index, yield injury %, superiority measure and relative performance) for grain weight /plant under pathogen infection condition when treated with endophyte bacteria. Based on this study, these bacterial strains can be used as a biotechnology tool in quinoa seedling production and biocontrol to diminish the severity of bacterial leaf spot disease.

Keywords: *Chenopodium quinoa*; endogenous bacteria; *Pseudomonas syringa*; amino acids; plant growth-promoting endophytic bacteria (PGPEB).

GAPP6

Co-occurrence networks show rice straw as mulch without microorganisms to recycling agricultural residues decrease network complexity

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Agricultural residues are a great resource of lignocellulosic biomass and microorganisms play a pivotal in their recycle. The benefits of this process include soil fertility, increase in productivity, and soil biodiversity, making microorganisms key players in the agroecosystems. However, knowing how microorganisms relate to each other remains a challenge. Here we investigated the effect of four different rice straw management strategies (1. Rice straw as a mulch; 2. Rice straw as a mulch with the addition of microorganisms' inoculum; 3. Rice straw plus microorganism's inoculum but incorporated into the soil; 4. Burning of rice straw with ashes incorporated into the soil) on soil microbial networks in rice crop at three different sampling times. The productivity, phytosanitary status, physical-chemical soil properties, and bacterial communities were also assessed. The microbial community was identified through 16S rRNA. Co-ocurrence patterns were identified using Sparse InversE Covariance estimation for Ecological Association and Statistical Inference in R®. Correlations with a magnitude > 0.9 or < -0.9 and statistical significance ($p \leq 0.05$) were included in the network analyses. The network topology was identified for each of the treatments, as well as the key taxa and modules. Rice straw as a mulch had the lowest node and edges and positive edges in comparison to the other treatments. The two key taxa identified in this treatment were taxon belonging to *Sphingomonadaceae* and *Sulfuricurvum*. Regarding physico-chemical and productivity parameters rice straw as a mulch had the lowest yield, and pH. These results suggest that rice straw as a mulch is not the best option to recycle rice straw under our study conditions.

Keywords: co-ocurrence networks; soil amendments; rice straw; microbial communities.

GAPP7

Bacterial endophytes contribute to rice seedling establishment under submergence

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

Keywords: coleoptile; endophytes; microbiota; *Oryza sativa*; submergence.

GAPP8

Beneficial effect of plant growth promoting bacteria on maize and their protect potential against phytopathogenic fungi

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Soil microorganisms are essential for soil fertility because they have ability to decompose organic matter, prevent leaching of nutrients and fix nitrogen. Many of them are plant growth promoting microorganisms (PGPM) that can protect plants from pathogens and increase plant growth. The objective of this work was to examine influence of soil isolates on maize growth. Six bacterial isolates were identified and tested (*in vitro*) for plant growth promoting activities (ammonia production, phosphate solubilization, siderophore production, HCN and IAA production), antifungal activity (dual culture method) and their influence on initial growth of maize under greenhouse conditions. After 5 weeks, the following morphological parameters of single-inoculated plants were measured: shoot height (mm), root length (mm), weight of fresh and dry plants (g). Data were analyzed using SPSS software and One-way analysis ANOVA with Tukey´s test was performed. Two isolates were positive on ammonia production, phosphate solubilization and siderophore production. One isolate produced HCN and three were positive for IAA production. The highest percentage inhibition (PGI) were obtained by isolate identified as *Bacillus subtilis* on *Botrytis cinerea* (69.57±4.35%) and *Cercospora* sp. (63.11±1.12%). Five isolates showed a significant difference in shoot height between the treatments and the control (non-inoculated plants). The treatment inoculated with the isolate identified as *Bacillus safensis* showed the best results in shoot height and weight of fresh and dry plants. Four isolates affected root length, with the best result obtained by *Bacillus mojavensis*. This study suggests the possibility of using characterized isolates as bioinoculants to promote growth rate and protect plants from phytopathogenic fungi.

Keywords: plant growth promoting microorganisms; antifungal activity; maize.

GAPP9
Cell-cell signaling in plant associated bacteria

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Many proteobacteria possess LuxR solos which are quorum sensing LuxR-type regulators that are not paired with a cognate LuxI-type synthase. LuxR solos have been implicated in intraspecies, interspecies and interkingdom communication by sensing endogenous and exogenous AHLs as well as non-AHL signals. LuxR solos are likely to play a major role in microbiome formation, shaping and maintenance through many different cell-cell signalling mechanisms. The analysis of 31034 proteobacterial genomes revealed that they are predominant with over 40% of the genomes harboring at least one *luxR* solo gene. Amino-acid sequence homology and mapping of the adjacent genetic loci has allowed the subdivision of the majority of these LuxR solos into 8 sub-groups, suggesting that very likely many more types and sub-groups of LuxR solos exists and are not discovered yet. Bacterial genera such as *Rhizobium*, *Pseudomonas*, *Burkholderia* and *Agrobacterium* carry the highest number of different LuxR solo types. This analysis highlighted that LuxR solos in proteobacteria are exceptionally abundant and many diverse sub-groups exist, having different evolutionary origins, functions, ligand binding properties and ecological context. Some types of solo LuxRs are likely to be specialized in relation to the taxonomy, others to the regulatory mechanisms they might participate in, and other to the specific niche they occupy. Future work needs to validate their role in intraspecies, interspecies and interkingdom signaling in bacterial communities as well as the chemical signals they respond to in order to unravel the function of this major regulatory family in cell-cell interactions of the microbiome establishment and maintenance. Importantly, future works will also focus on the role of a specific sub-group of solo very abundant among plant-associated bacteria and likely to be involved in plant-bacteria interaction. This highlights the importance of these proteins in understanding bacteria sociality and interaction with the host for the design of plant growth promoting inoculants.

Keywords: quorum sensing; LuxR solos; proteobacteria; microbiome; microbe-microbe interaction.

GAPP10

Biofertilization 2.0: Rewriting the rules of plant nutrition with a new approach

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Plant farming face challenges, including soil fertility decline and pest outbreaks, necessitating heavy reliance on synthetic phytopharmaceuticals and fertilizers for efficient production. Biofertilization, using ingenious plant microbiota, shows promise in addressing these issues. However, current strategies have limitations such as dependency on specific microbial strains, inhibition of beneficial microbes by indigenous counterparts, and susceptibility to environmental conditions. Yet, isolating the most effective bacterial strains for plant growth improvement remains a challenging task. To tackle this issue, we propose a new method of isolating and selecting a combination of several dozen of efficient bacterial strains derived from beetroot seeds, roots, or rhizosphere. This approach utilizes random spatial aggregation of bacterial cells caused by electrostatic modification of their surfaces. The aggregation protocols were optimized to obtain diverse sizes of aggregated bacteria that at the end determine complexity of interactions. To assess the efficiency and size distribution of theaggregates, fluorescent microscopy and flow cytometry methods were employed. Obtained aggregates were then encapsulated in alginate beads using microencapsulation technique, and the activity of aggregates incubated in minimal media supplemented with beetroot and soil extracts, separately, was determined spectrophotometrically using resazurin as an indicator. The beads bearing the most active aggregates underwent further analysis of species composition through Denaturing Gradient Gel Electrophoresis (DGGE) and near whole 16S rRNA sequencing. As a result, we achieved aggregation levels >10% with diameters >10 µm. Microscopy confirmed aggregate localization within alginate beads, and distinct spectrophotometric curves were observed for different nutrient sources. DGGE analysis revealed variations in bacterial diversity based on the nutrient source during selection and enrichment procedures. This method effectively enhances isolation of native plant microbiota, enabling rapid and cost-effective selection of the combination of the most effective bacterial strains, that might ultimately promote plants growth.

Keywords: aggregates; bacterial microencapsulation; biofertilization; cell electrostatics; colloid biology.

GAPP11

Novel enrichment strategy for the isolation of beneficial bacterial endophytes from *Oryza sativa* (rice) roots for abiotic stress tolerance

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Rice is a major agricultural and economically valuable crop consumed worldwide. The growth of the world's population and consequent rise in rice consumption require an expansion in this crop's yield. However, agricultural practices are facing challenges due to climate change and there's the need to improve rice resistance towards abiotic stresses like salinity and nitrogen (N) starvation. Microorganisms live in tight association forming ecological interaction webs in nature. Bacterial members of microbiomes undergo direct interactions which influence microbiome composition. Plants intimately associate with a rich and diverse microbiome and plant health heavily depends on it. Plant associated bacteria (PAB) colonize different plant-associated niches and the soil is the major reservoir of microbes showing an enormous microbial diversity. Hence, roots are a hotspot for bacterial colonization and microbial community development. A pattern of separation between microbial communities in root compartments exists and there's a spatial gradient going from the rhizosphere to the internal plant tissues, the endosphere, strongly influenced by plants that actively recruit endophytes, colonizing the endosphere. Bacterial endophytes actively sense the plant environment and adjust their behavior. Endophytes confer fitness advantages to the plant (e.g. N fixation, increasing plant tolerance to environmental stresses and promoting plant growth). Given the recurrent pattern of bacterial recruitment and endosphere colonization under determined environmental and plant growth conditions, we hypothesize that endophytes, selected for their capacity to compete and colonize the endosphere, can confer resistance to plants and promote their growth in non-stressed and in abiotic stress conditions. This study will focus on the identification and characterization of super-endophytic bacteria highly active in colonizing the rice endosphere and focus on their cell-to-cell signaling mechanisms and on the multispecies community they shape. This study will also allow the characterization of endophytic bacteria showing properties conferring resistance to rice plants towards salinity and nitrogen starvation, with possible applications in the industrial/agricultural field as biofertilizers and/or probiotics.

Keywords: super endophytes; plant microbiome; rice; nitrogen starvation; saline soil.

GAPP12

Keystone bacteria in the sorghum rhizosphere microbiome as a strategy to design plant beneficial consortia

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Plants intimately associate with diverse microorganisms, which cooperate and interact together resulting in a stable mixed population called the plant microbiome. Plants recruit specific microorganisms including plant growth-promoting bacteria (PGPB) that live inside and outside the root, performing various beneficial activities for plant growth and health including the biocontrol of potential phytopathogens. The microbial habitat around the root, where soil comprises the highest microbial biodiversity, is referred to as the rhizosphere. The rhizosphere hosts a rich microbial community recruited by the plant via root exudates. Plant roots recruit their microbiome from the soil, but the dynamics of interaction and signaling taking place in the rhizosphere microbiome are still largely unknown. Understanding how bacteria undergo intra- and inter-species signaling in the rhizosphere microbiome will be an important challenge for future studies. In our study, we are interested in Plant growth promoting (PGP) bacteria of the sorghum rhizosphere microbiome. *Sorghum bicolor* is a grass species cultivated as a cereal crop in tropical and subtropical region globally for human consumption, to feed animal and biofuel production. This study aimed to investigate the of keystone/core-microbiome bacterial genera within the sorghum rhizosphere and their networks of interactions, by using 16s rRNA community profiling approach by comparison of the microbial community of different cultivars and geographical locations. The keystone and core rhizosphere bacterial isolates were then isolated and phenotype mapping of specific plantgrowth promoting features were mapped. Synthetic communities of bacterial consortia have been designed that will hopefully promote plants health and growth. Finally, cell-to-cell signaling taking place among the PGP bacteria within the consortia will be studied in order to shed light on the communication mechanisms used by these bacteria in the microbiome context.

Keywords: microbiome; microorganism; *Sorghum bicolor*; rhizosphere; keystone bacteria.

GAPP13

Biological products use in Burundi for sustainable agriculture

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Biological products have been used in agriculture for longtime as inputs to control the plant or soil health. The origin of these products is diverse and their use is decided accordingly. The mots used products are classified in two groups, the first group as biofertilizer and the other group as biopesticides. All the products are from organism like plants, animal and microorganisms like fungi and bacteria. The research on biological product involved in plant growth or protection still not developed. The previous results from research on development of biological products showed that a list of plants that can be used to control some plant diseases and pest. The efficacies of these plants still need to be well studied. The result showed also that bacterial isolates belonging to *Rhizobium* and *Bacillus* genera increased the growth of soy bean after inoculation. The part of plant used are macerated leaves or seeds mixed with water and fermeented for 24 hours. And the application is in the aftenoon when the sun UVs are not so strong. The most plants used are *Azadirachta indica* (leaves), *Melia azedarach* (seeds), the *Tithonia diversiforia* (leaves), *Thevetia neriifolia* (leaves) *Solanum annum* (leaves) and for biofertilizer like *Rhizobium* on Soy bean are under development.

Keywords: biopesticides; biofertilizer; sustainable agriculture.

GAPP14

Characterisation and biocontrol potential of Burundi isolated *Bacillus nakamurai* strain

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Searching for safe and eco-friendly alternatives to chemical pesticides is a general trend worldwide since some decades. Some *Bacillus* strains among other microorganisms already proved their biocontrol potential. Sixteen Bacillus-like bacterial strains were isolated from arable soil samples collected from two different agro-ecological locations in Burundi and were evaluated for their antagonistic activities against six bacterial phytopathogens. One isolate BDI-IS1, however, was active against all the six plant pathogens. Moreover, this isolate BDI-IS1 exhibited an antifungal activity against nine fungal phytopathogens including three isolated from infested leaves samples harvested in Burundi. These antimicrobial activities were comparable to the commercial strain *B. velezensis* QST713. Greenhouse experiments carried with tomato infested by *Alternia solani* revealed that BDI-IS1 reduce the disease incidence by about 40% and better than QST713 (25%). The 16S rRNA sequencing of this isolate showed that it belongs to *Bacillus nakamurai* strains. Culture supernatants of BDI-IS1 exhibited great antibacterial activity, proof that the active compounds are soluble biological secondary metabolites (BSMs). Chemical analyses of culture supernatants by LC/Q-TOF mass spectrometry technique showed that BDI-IS1 produces a vast array of well-known antimicrobial potential bearing BSMs including cyclic lipopeptides (surfactins and iturins A), polyketides (bacillaene), siderophores (bacillibactin), a dipeptide bacilysin and ribosomally synthesized and post-translationally modified peptides-RiPP (plantazolicin). Furthermore, genome mining using web tool Antismash version 6.2 predicted the biosynthesis of amylocyclicin. *In vitro* confrontation tests with metabolite depleted mutants evidenced the role of these BSMs in the mediation of the observed bioactivities. Adaptability of this strain to abiotic stresses including pH and temperature was also shown, proving that it can grow and exert its plant protective property in harsh conditions of pH (acidic) and low temperature (15°C). These findings suggest that this *B. nakamurai* strain isolated from Burundi is worth to be exploited in the biocontrol strategies.

Keywords: Burundi; *Bacillus*; biocontrol potential; plant pathogens.

GAPP15

Role of proteobacterial AHL QS in the plant endosphere

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The plant microbiome represents many diverse microorganisms that interact and colonize different plant-associated niches; one of these compartments is the endosphere, which consists of the inner plant tissues. In this complex multispecies community, many different mechanisms of cell-cell interactions take place among bacteria. Quorum Sensing (QS) is one type of interaction among bacteria, which regulates gene expression in response to cell density via the release and detection of chemical signals, such as the acyl homoserine lactone (AHLs). However, very little information is currently available on the role of QS in the context of complex plant endophytic microbiomes and especially on the role of the AHLs in shaping and establishing this specific community. The endophytic microbiome has a prevalence of Proteobacteria, representing more than 50% of the bacterial community, with gamma-proteobacteria being the most abundant class. We performed a bioinformatics study searching for the presence of complete AHL-QS systems among endophytic bacteria genomes belonging to the protobacteria phylum and publicly available. Among the hits obtained the *Pseudomonas fluorescens* L111 was selected as a model to study the role of AHL QS systems in the rice endosphere, since it harbors two independent AHL-QS systems. *Pseudomonas fluorescens* L111 wild type and the QS systems knock-out mutants have been molecularly and genetically studied to shed light on the organization hierarchy of the two signaling mechanisms and possible target genes. Via *in planta* colonization assays and microbiome 16S rRNA amplicon sequencing analysis, the role of the AHL-QS systems has been unraveled. This study highlights the function of the QS communication systems and AHLs signals in the establishment of a multispecies community in rice plants.

Keywords: plant endosphere microbiome; quorum sensing; acyl-homoserine lactones (AHLs).

GAPP16

Upcycling of lignin from invasive alien plant species (IAPS) using soil bacterial isolates for the production of value-added compounds

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The utilization of black liquor encompasses various practices, including the combustion or gasification of concentrated lignin for electricity generation, as well as the production of chemicals and fuels like methanol, syngas and dimethyl ether. However, limited attention has been given to the production of value-added compounds derived from lignin, particularly through biodegradation or biotransformation processes. Lignin, being the second most abundant biorenewable polymer, contains a significant proportion of aromatic compounds, which can potentially be degraded and transformed into valuable compounds utilizing diverse bacterial isolates found in the environment. In our study, we isolated lignin-degrading bacteria from forest soils and characterized them through 16S rRNA analysis, genome sequencing, and enzyme assays to assess their lignindegrading capabilities. To evaluate lignin degradation, we used three types of substrates pure lignin, neutralized black liquor, and native plant biomass as substrates. Based on the 16S rRNA and genome sequencing of the isolates, we determined that all of them belong to the *Paraburkholderia* genus and the most active strain contains genes that are involved in the lignin degradation pathways. Moreover, by metabolomic approach employing targeted liquid chromatography-mass spectrometry (LC-MS) analysis, we observed the consumption of vanillin and hydroxybenzoic acid, along with the production of hydroxyphenylacetic acid, ferulic acid, 4-hydroxyacetophenone, 4-hydroxycinnamic acid, and acetovanillone. The production of 4-hydroxycinnamic acid was specifically induced when black liquor was utilized as a lignin source for the bacteria. To further elucidate the metabolic pathways relevant to lignin degradation and the production of these compounds, we combined the LC-MS data with genomic analysis of the corresponding enzyme-encoding genes. By integrating genomic and metabolomics analytical methods, we successfully identified the most efficient strain, demonstrating its potential for future application in biotechnological setups.

Keywords: lignin; value-added compounds; degradation; colloid biology; *Paraburkholderia*.

GAPP17

Transcriptional analysis reveals induction of systemic resistance and increased fungal abundance in the rhizosphere of *Chlorella* microalgae-treated tomato

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The application of algae-based products has been reported to promote plant growth and yield of tomato plants, especially by enhancing flowering. However, how microalgae (MA) affect plants at the molecular level remains elusive. The effect of the algal application on the soil microbial community is also unknown. This study aimed to elucidate the effects of live microalgae application on plant photosynthesis and the transcriptome of the unopened flower buds of tomato plants as well as investigate algal effect on soil microbial composition. Microalgae increased leaf temperature differential in tomato leaves but hardly affected photosynthesis. Contrary to our expectations, RNA-seq data revealed remarkable differential expression of several genes participating in responses to abiotic stresses but only a few genes involved in flowering or pollen/ spore development. Late Embryogenesis Abundant (LEA) proteins (mostly dehydrins), oleosins, ethylene, and abscisic-related genes, and Nascent Polypeptide-Associated Complex (NAC)-domain-containing proteins were upregulated. Genes involved in carbohydrate metabolism were also differentially expressed; glycolysis-related genes were upregulated, while those involved in sugar transport were downregulated. Drenching soil with microalgae increased soil microbial diversity with a remarkable enrichment of beneficial fungi and bacteria. This study shows that microalgae treatment primed plants by inducing the expression of genes involved in response to abiotic stress, especially cold and water stress, as well as pathogen attack. It also led to a change in microbial composition with an increase in the abundance of fungi.

Keywords: algae; biostimulant; *Chlorella*; defense; tomato; soil microbial diversity.

GAPP18

Isolation and characterization of *Bacillus* strains from wheat rhizosphere microbiome for their potential plant growth promotion and biocontrol activity against *Fusarium culmorum*

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The rhizosphere microbiome representsthe greatest reservoir of biological diversity known in the world so far. Plants are colonized and live in close association witha large number of microorganisms. Many of them, indicated as PGPR (plant growth-promoting rhizobacteria), play important roles in plant growth and health and resistance to biotic and abiotic stresses. Among the group of PGPR, strains belonging to the *Bacillus* genus have been found in association with a wide range of plant rhizosphere. *Bacillus* species had a high potential, as a promising biocontrol agents and an eco-friendly alternative, to be used against the plant disease, and promote plant growth. In this study, we wereinterested in PGPR of the wheat rhizosphere microbiome. We reportedthe isolation and characterization of 18 strains of *Bacillus*. These strains were screened for plant growth promoting traits and antifungal activity against *Fusarium culmorum*, the causal agent of foot and root rot (FRR) and *Fusarium* head blight (FHB) on wheat. Three of the tested isolated showed the highest antifungal activity against *F. culmorum* and exhibited several PGP traits. Thewheat seed treatment with these strains promoted plant growth (root and shoot weight) in absence as well as in presence of *F.culmorum* inoculation. Furthermore, they improved chlorophyll contents, and phenol peroxidase concentrations, and reduced malondialdehyde and hydrogen peroxide levels in presence offungal inoculation. This study suggested that the selected *Bacillus* strains involved different mechanisms and regulatory functions to enhance plant oxidative systems and regulate mechanisms supporting both growth and fungal disease management. Further studies will explore the dynamics of interaction and signaling taking place in the wheat rhizosphere microbiome with and without *F. culmorum*, which are still largely unknown.

Keywords: *Bacillus;* microbiome; biocontrol; *Fusarium culmorum;* PGP.

GAPP19

Development of low-cost microbial inoculants for sustainable agriculture with increased shelf life

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The progressively increasing demand for organically grown food has resulted in the surge of organic farms. This has led to the augmented use of plant-beneficial microbial inoculants as eco-friendly strategy alternatives to chemical agro-inputs, but the constraint is cost and shelf-life of formulations. Optimization for low-cost formulations (liquid, solid wet, and dry formulations) with increased shelf life of bacteria can be opted as a feasible alternative. Utilization of soil and plant associated microorganisms to develop various inoculants ranging biofertilizers, biostimulants, biofungisides and bioinsecticides has been done. In this regard, Liquid inoculants with several industrial-grade polymeric additives, surfactants, and adjuvants were tested for shelf-life stability. Formulations with 0.1% carboxymethylcellulose, 1% polyvinylpyrrolidone (PVP) and XMPA (surfactant) supported survival of bacteria stored at 30±2°C. Solid wet formulations were tried using two approaches, Alginate beads are excellent to sustain the microbes but were not effective in releasing trapped microbes and this method is relatively costly. On the other hand, the use of agricultural waste for fermentation and formulation is a sustainable and eco-friendly approach. Rice husk and press mud (sugar industry waste) were used for the development of biofilm-based solid wet formulations, but it was challenging for field application. The powder-based dry formulations (wet-able) are excellent to retain microbial count for extended periods and at high temperatures. Spray drying with the addition of whey powder was found effective but it requires specific equipment and is costly, while air drying is a low-cost and effective method. Wet-able powder (WP) air dried biofungicide formulation was tested to preserve bacterial count even after 3 years. Hence WP is most viable formulation due to cost, survival of microbes, and easy field application. Since this method is being successfully used for microbial inoculants (biofertilizer, biofungicide, bioinsecticide and biostimulant) at industrial level.

Keywords: formulation; alginate beads; wet-able powder; sustainable agriculture; adjuvents.

GAPP20

Contribution of local rhizobia to improving soybean (*Glycine max* L.) productivity and restoring soil fertility in Côte d'Ivoire

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Rhizobia are non-pathogenic rhizospheric bacteria that have ability to fix biological nitrogen in symbiosis with legumes. They were used as biofertilizers to improve soil quality and increase agricultural yields. Despite their many agronomic, environmental and economic potentialities, these bacteria are little valued in the cropping systems in Côte d'Ivoire where the available agricultural land is overexploited. This study was undertaken to assess the contribution of local strains of rhizobia on soybean productivity and soil quality on short periods. Rhizobia isolated from soils of Côte d'Ivoire were used to inoculate soybean cv Piramama under field conditions in the Center-West of the country. Nodulation and yield parameters were measured at different phenological stages, and then the impact of bacteria on soil quality was assessed one month after soybean harvest. The field trial showed that the local rhizobia RSC119, RSC309 and RSC508 induced the most nodule and pod formation than the exotic strain *Bradyrhizobim japonicum* IRAT FA3 and the synthetic fertilizer NPK 12 22 22. In addition, these rhizobacteria increased soybean seed yield by 78-250 % compared to unfertilized control. The soil analyzes showed that M.O and N contents were improved by all of bacteria tested. Also, the local bacterium RSC508 was distinguished by its ability to raise the pH of soils one month after soybean harvest, passing from an acidic to neutral pH. Local isolates RSC119, RSC309, RSC504 and RSC508 identified as the most efficient could be recommended in cropping systems involving soybean to restore soil fertility and increase productivity.

Keywords: local rhizobia; inoculum; nitrogen; soybeans; soil fertility.

GAPP21

Preparation and development of carrier-based biofertilizer to improve fenugreek plant growth under salt stress conditions

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Liquid bacterial culture was ideally used for the biofertilizer for plant growth promoter, due some environmental constrains liquid culture was not much effective against stress conditions. Therefore, in the present study, carrier-based biofertilizer are a crucial part of sustainable organic farming in contemporary agriculture practices. In current study the Plant growth promoting bacteria were isolated from root nodules and rhizospheric soil of ground nuts (*Arachis hypaea*). The present deal with the preparation and development biofertilizer using wood ash and cow dung ash as carrier materials. These carrier materials easy to available, eco-friendly and cost-effective compare to other carrier materials. Moreover, this carrier material contains various macro and micronutrient (calcium, sulphate, trace elements, charcoal etc.) and act as bacterial stabilizer. The bacteria such as *Bradyrhizobium* sp., *Rhizobium* sp. *Azotobacter* sp. and *Pseudomonas* sp. were used for the pot study. Results indicated that in the presence of salt stress, the organisms improve the pant growth and plant growth promoting activity. Therefore, the wood ash and cow dung ash carrier material were found to be effective in fenugreek plant as compare to the control and other treatments. The organisms posed various characteristics such siderophore production phosphate, potassium and zinc solubilization potential. In the study gemination rate, survival rate chlorophyll content, root and shoot length was evaluated up to 28th days. The results were statistically significant at ($P < 0.05$). hence based on the results carriers based biofertilizer in more successful fenugreek growth under salt stress compared to the other conventional treatments tested.

Keywords: carrier-based biofertilizer; fenugreek; salt stress.

GAPP22

The impact of different plant nutrition systems on yield and yield components of sweet maize

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An optimal nutrient supply is necessary to achieve high and stable yields. The plant uses large amounts of nutrients from the soil by biomass and yield. Because of that, people should compensate for those losses. This study aims to compare the impact of different plant nutrition systems on the yield and yield components of sweet maize. The experiment was performed in Maize Research Institute „Zemun Polje", in the growing season 2021. We analyzed the influence of three fertilization levels on grain yield, ear length, the diameter of ear and shelling percentage of ZP355su and ZP418su sweet maize genotypes. Three fertilization levels include F1 – mineral fertilization; F2 – mineral fertilization + microbiological seed treatment; F3 – mineral fertilization + microbiological seed treatment + foliar fertilization. The experiment was conducted using irrigation and the plots were maintaining by hoeing. Under F3 treatment, the ZP355su hybrid gave a yield of 10.02 t ha-1 and the ZP418su hybrid yielded 8.28 t ha-1. Compared to the F1 treatment, yield increase for ZP 355su and 5% for ZP481su was 7%. The F2 and F3 treatments expressed an equally positive effect on the ear length, the diameter of the ear and shelling percentage of the ZP355su hybrid, while in the ZP418su these traits reached the highest values under the F3 treatment. Compared to the F1, under the F3 treatment, the shelling percentage, the ear length and the ear diameter, of the ZP418su, increased by 8%, 5,3% and 8,7%, respectively. Hybrid ZP355su had 4% greater shelling percentage, 5,4% longer ear length and 8,3% bigger ear diameter in F3 when compared to F1 treatment. By applying only mineral fertilization, all tested parameters, in both hybrids, had the lowest values. The combined application of mineral, microbiological and foliar fertilization had the best influence on the yield of both genotypes.

Keywords: sweet maize hybrids; yield; mineral fertilization; microbiological seed treatment; foliar fertilization.

GAPP23

Solving the *Pseudomonas fuscovaginae* quorum sensing puzzle

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Pseudomonas fuscovaginae (*Pfv*) is one of the main phytopathogens associated with brown sheath rot, a complex disease that causes severe rice yield loses worldwide. *Pfv* virulence is dependent on quorum sensing (QS), a form of cell-cell communication that enables bacteria to coordinate population behavior in response to cell density. Similarly to other QS systems commonly found among Proteobacteria, the two QS systems of *Pfv,* designated as PfsI/R and PfvI/R, rely on signaling molecules - acyl homoserine lactones (AHLs), which are produced by the two LuxI family synthases (PfsI/PfvI) and bind to the cognate LuxR family transcriptional regulators (PfsR/PfvR). However, both QS systems have so far shown to be active exclusively *in planta*, otherwise being under stringent negative control of a regulator designated as RsaM, regardless of the population cell density. RsaM possesses a characteristic domain not found in other protein families and is not predicted to be able to bind DNA, RNA or AHLs. This study aims to elucidate the mechanism/s underlying the repressive effect of RsaM on the *Pfv* QS systems, starting with the identification of putative interacting partners of RsaM. Understanding how this novel QS regulator exerts its function, and especially, under which conditions the *Pfv* QS becomes switched on would be helpful for completely unveiling its pathogenicity mechanism and the ecological purpose *in natura* of the above QS-driven regulation. Future results would be of considerable importance both for the fields of QS regulation and plant-microbe interactions.

Keywords: *Pseudomonas fuscovaginae*; quorum sensing; RsaM.

GAPP24

The presence of ice nucleation active *Pseudomonas syringae* in the Danube River basin

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Pseudomonas syringae (*Psy*) is a widespread complex of plant pathogenic bacteria that can provide nutrients by causing frost damage to plant tissue. They synthesize the ice nucleation protein InaZ, which is anchored to the outer membrane of the bacterial cell. It enables ice formation by bringing water molecules into an "ice-like" arrangement at temperatures as high as -2°. The aim of our study was to investigate the ability of *Psy* isolates to nucleate ice. The ice nucleation activity (INA) test was performed on 51 *Psy* representatives from the Danube River Basin in Serbia, which is an important irrigation source. The isolates were exposed to decreasing temperatures from -2° to -10° in a decline of 1°. The INA tests revealed 39 active strains (76.5% of the collection). Depending on the ice-forming temperature, three different INA types were observed: warmer than -4° (type I), -4° to -7° (type II), and colder than -7° (type III). Based on the average freezing temperatures, seven isolates belonged to type I, 14 to type II and 18 to type III. The highest temperature at which ice formed was -3°C. Our study showed the presence of INA bacteria in the irrigation water source, which may increase the risk of frost damage to plants. Frost damage causes significant losses to frost-sensitive plants, so its prevention is essential for crop health.

Keywords: *Pseudomonas syringae*; plant pathogen; ice nucleation activity; frost damage.

GAPP25

Screening of AHL lactonase activity in *Bacillus* spp. strains isolated from different natural samples

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Through evolution, patogenic bacteria developed mechanism to increase their virulence to hosts by production of *quorum sensing* (QS) signaling molecules, primarily acyl homoserine-lactones (AHLs). Since the discovery, QS phenomenon has been promising target in biocontrol strategies involving *quorum quenching* (QQ) mechanisms. The great potential in this area lies in detection, characterization and possible apllication of AHL-lactonases from Bacillus spp. The purpose of this study was to test presence and activity of AHL-lactonases in natural *Bacillus* spp. isolates in order to find prospective strains which could be possibly used in QQ-mediated control of AHLproducing phytopathogens. The collection of 149 Bacillus spp. isolates, orginated from soil, manure and straw, was screened for the presence and activity of AHL-lactonases using well-diffusion method. Violacein production by biosensor strain *Chromobacterium violaceum* CV026 was indicator for AHL-presence and degradation. PCR method was performed for the detection of *aiiA* gene in selected isolates, with primers aiiA1-F and aiiA2-R. Degradation of AHLs in medium was observed by the loss of violacein purple colour around wells. Total of 52 strains showed AHLlactonase activity, which was confirmed by detection of a single 793 bp long band of amplified *aiiA* gene. Furthermore, difference in AHL-lactonase production was percieved in *Bacillus* spp. isolates from different natural samples. The highest number of AHL-lactonase producing Bacillus spp. were found in soil samples (65 %), followed by manure samples (33 %), while only one strain from straw demonstrated AHL-lactonase activity. The results suggest that AHL-lactonase is common in Bacillus spp. exeptionally in soil isolates.

Keywords: *Bacillus* spp.; biocontrol; *quorum quenching*; AHL-lactonase activity.

GAPP26

Herbicide-resilient bacterial consortia in plant growth promotion

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Modern agriculture relies heavily on agrochemicals that can adversely affect non-target organisms, including microorganisms. In terms of agricultural sustainability, herbicideresilient plant growth-promoting (PGP) bacteria that can improve crop yield are critical. Previously, we isolated 122 high herbicide concentration tolerant (HHCT) bacteria from soils contaminated with S-metolachlor, a widely used herbicide. Initially, *in vitro* experiments were conducted to screen potential candidates for further evaluation. Our goal was to identify HHCT bacteria with high PGP potential, and the ability to improve soil quality and biocontrol activity. Subsequently, the most promising candidates were grouped into three consortia and evaluated *in pot* for their effects on corn seed germination, early-stage plant development, and root colonization. Experiments were conducted with seeds of commercial maize hybrids under controlled conditions, with and without the presence of the herbicide S-metolachlor. The results showed that the consortia positively affected seed germination and promoted higher germination rates. The bacterial isolates showed the ability to colonize the root system of maize, with significant effects of the consortia on shoot and root biomass and shoot and root length. However, it was observed in some consortia that the presence of herbicides resulted in plant damage. Markedly, the presence of the herbicide S-metolachlor reduced or reversed the beneficial effects of bacterial consortia. This could be due to incomplete biotransformation of the herbicide leading to the formation of toxic intermediates. The selection of bacterial consortia that not only promote plant growth but also have the ability to fully biotransform herbicides is critical. In conclusion, the isolated HHCT bacteria showed promising PGP potential and formed an effective consortium to promote maize seed germination and early-stage development. However, careful consideration of the selection of bacterial consortia is required to ensure their compatibility with herbicides and to minimize negative impacts on plant health and environmental sustainability.

Keywords: PGP; soil; maize; herbicides; agriculture.

GAPP27

Suppressive soils as reservoir of bacteria with biocontrol properties

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Crops are exposed to a wide range of soil-borne phytopathogens, particularly oomycetes and fungi, which are difficult to control. However, certain microorganisms present in soil can compete with these pathogens and provide general (microbialcommunity-based) disease suppressiveness or specific disease suppressiveness (microbial-population-based) by efficiently controlling one or a few phytopathogens, which corresponds to soil suppressiveness *sensu stricto*. These biocontrol properties are important when considering phytopathogens such as *Fusarium graminearum*, which is increasingly damaging crops in the context of an on-going climate change. Both types of suppressiveness are likely to involve many different microbial populations with plantbeneficial traits. Therefore, the objective of our study was to isolate and characterize potential biocontrol agents from rhizospheres of wheat (*Triticum aestivum*) grown in soils suppressive to *F. graminearum* disease, previously identified in Serbia. Bacteria were isolated using standard plating techniques, subsequently subjected to a dual-culture assay with *F. graminearum* to assess their efficacity. The genomes of the most promising isolates were sequenced and screened for known biocontrol and plant-growth promotion genes. *In silico* analysis was complemented by functional *in vitro* characterization. Genome analyses confirmed the presence of genes encoding for biocontrol metabolites, such as phenazine, 2,4-diacetylphloroglucinol, pyrrolnitrin, hydrogen cyanide and 2- hexyl-5-propyl-alkylresorcinol. In addition, the bacterial genomes contained genes associated with plant-growth promotion, some of which were confirmed by functional characterization. Overall, our findings show that suppressive soils in Serbia can be a source of biocontrol bacteria with modes of action widely evidenced in different types of suppressive soils.

Keywords: suppressive soils; *Fusarium graminearum*; biocontrol agent.

GAPP28

Plant growth promoting *Halomonas* from roots of halophytes as a strategy to improve crop resistance to soil salinity

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Increased soil salinity has numerous negative effects on plant growth and poses a significant threat to agricultural productivity worldwide. The diversity of endophytic bacterial communities found in various indigenous halophytes such as *Salicornia europaea, Suaeda maritima*, and *Camphorosma annua*, may reveal beneficial bacteria with various plant growthpromoting (PGP) properties. Members of *Halomonas*, the largest genus of the family Halomonadaceae, are rod-shaped, gram-negative, moderately halophilic bacteria that have been increasingly isolated from various saline environments, including halophyte tissues. As part of the halophytic microbiota, halotolerant plant growth-promoting endophytic bacteria can contribute to plant host growth, productivity, and fitness under abiotic and biotic stresses. The use of halotolerant PGP bacteria represents a good alternative strategy to mitigate the effects of salt stress in crops and to recover saline soils. The aim of this study was to isolate halotolerant bacteria from halophyte roots, and evaluate their PGP ability and their effect on germination parameters of selected crops. The endophytic bacterial communities in the roots of halophytes from natural saline soil in Slano Kopovo (Serbia) were analyzed using culture-dependent techniques. The genus *Halomonas* was the predominant among the halotolerant root-associated bacteria of analyzed halophytes. We have isolated seven *Halomonas* sp. strains, six of which were closely related (>99,86% similarity) to *Halomonas songnenensis*, a moderately halophilic bacterium from saline and alkaline soils. Obtained isolates were tested at different sodium chloride concentrations for a range of PGP traits at different concentrations of sodium chloride. The selected strains were used to inoculate seeds iand monitor of the germination process of barley, sunflower, and wheat under conditions of elevated salinity. Most of the isolates tolerate the NaCl concentration up to 18% and showed multiple PGP features with some of them retained even in the presence of additional salt concentrations. The germination test results showed that *Halomonas* seed inoculation positively affected barley germination parameters in conditions with and without added salt. The observed traits can be further exploited to improve plant growth and augment the salt tolerance of agricultural production on saline soils.

Keywords: halophytes; *Halomonas*; endophytes.

GAPP29

Characterization of outer membrane vesicles of plant growth promoting bacteria *Paraburkholderia phytofirmans* PsJN

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Plant growth promoting bacteria (PGPB) are microorganisms present in plant rhizosphere that stimulate plant growth, as well as prime plants against abiotic and biotic stress. PGPB are also located in plant intercellular space and can directly interfere with plant pathogens. Many different mechanisms of interaction between plants and PGPB has been shown, but the interaction through outer membrane vesicles (OMVs) has only been proposed. OMVs are membrane-enclosed nanostructures produced by Gram-negative bacteria. Size of OMVs can range from 20 to 300 nm, depending on the strain. In this research, we determined optimal method for isolation of OMVs produced by *Paraburkholderia phytofirmans* PsJN, characterized the size range of OMVs, their concentration and origin. OMVs from PGPB PsJN have been isolated us\ing two methodological approaches: 1) differential centrifugation, ultrafiltration, and purification using Optiprep density gradient, and 2) OMV isolation by the commercial ExoBacteria OMV Isolation Kit that uses an affinity-based column system. Size and concentration of isolated OMVs were analyzed using Nanoparticle Tracking Analysis (NTA). As the OMVs are derived from outer membranes of bacteria, their origin has been proven using an essay for detection of lipopolysaccharides. In addition, protein profiles of OMV from PsJN were visualized by SDS-PAGE. Comparing different methods for isolation and purification of vesicles from PsJN we concluded that for our PGPB strain the commercial ExoBacteria OMV Isolation Kit provides the highest yield. Quantification of OMV using NTA of OMVs produced by *P. phytofirmans* PsJN, isolated with the commercial kit, showed a mean particle numeration of 4.05*109 particles/ml. Size of OMVs isolated from PsJN ranged from 50 to 200 nm. Detection of lipopolysaccharides confirmed that isolated vesicles originated from outer membranes of PsJN. These findings are the important first step in our research of the role of OMVs in PGPB interaction with plants.

Keywords: PGPB; extracellular vesicles; outer membrane vesicles; OMV.

GAPP30

Development of a PCR-based method in the detection and identification of Tobacco Mosaic Virus (TMV) and Potato Virus Y (PVY) in tobacco

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Morphological techniques are relied upon heavily at Tobacco Research Board in Zimbabwe for the detection of Potato Virus Y (PVY) and Tobacco Mosaic Virus (TMV). However, these methods have pitfalls in their detection. The surge in the use of PCR-based markers has provided accurate genotypic information, giving a precision lacking with phenotypic identification which is influenced by other factors such as abiotic stress and phytotoxicity. Reverse transcriptase- Polymerase Chain Reaction (RT-PCR) protocols were developed for the identification of PVY and TMV. Internal specific primer pairs used detected DNA fragments of the virus' genomes; four hundred and eighty bp of the coat protein for PVY, four hundred and twenty two bp for the protein movement gene and four hundred and ninety six bp for the virus genome for TMV. Effectiveness and reliability of the protocols was analysed by sensitivity comparisons of double stranded RNA extraction method (dsRNA), Double Antibody Sandwich-Enzyme Linked Immunosorbent Assay (DAS-ELISA) and RTPCR at weekly intervals after viral infection of tobacco samples. Least effective was dsRNA as it detected PVY after three weeks of infection. DAS-ELISA was more sensitive than dsRNA as it detected viruses after one week of infection up to six weeks for PVY and three weeks for TMV. However, RT-PCR could detect viral infection throughout the duration of the experiment. Cost-benefit analysis of dsRNA, ELISA and RT-PCR was done. RT-PCR was seen to be slightly expensive than ELISA and more to dsRNA. The use of RT-PCR is recommended for application since it has improved sensitivity and not much significance was observed in comparison with dsRNA and ELISA as far as cost-benefit analysis is concerned.

Keywords: Potato Virus Y (PVY); Tobacco Mosaic Virus (TMV); RT-PCR; DAS-ELISA; dsRNA.

GAPP31

Bioprospecting of plant growth promoting rhizobacteria from medicinal plants of garhwal region

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The rhizomicrobiome is important for plant health and can influence the structure of the microbial community. Plants depend upon beneficial interactions between roots and microbes for nutrient availability, growth promotion, and disease suppression. Plant roots gets associated with microbial communities both inside the roots and in the rhizosphere, and these root-associated microbiomes play pivotal roles in plant nutrition and productivity by influencing their physiology and development. Medicinal plants provide an enormous bioresource potential use in modern medicine and agriculture, yet their microbiome is largely unknown. Although the soil microbiota is known to play a key role in the resulting plant-soil feedbacks, the proximal mechanisms underlying this phenomenon remain unknown. Plant microbiome studies with medicinal herbs may unravel a distinct spectrum of microbes that can tolerate, or even enhance, the production of novel classes of compounds and their variable applications. We have isolated different bacterial isolates from rhizospheric soil of an important medicinal plant *Ajuga parviflora* and *Bergenia ciliata* displaying growth promoting attributes like production of siderophores, indole acetic acid, phosphate solubilization, calcite solubilization, nitrogen fixation etc. and supplementation of these bioinoculants to plants such as *Cicer arietinum* and medicinal plants *Andrographis paniculata* and *Vinca rosea* was investigated. Bioinoculation led to increase in root length, shoot length, fresh weight, dry weight, relative water content and total chlorophyll content. Development of shoot and root system was also prominent due to multiple branching in shoots and roots and appearance of numerous root hairs was also observed. This may lead to uncovering a distinct set of microbes that can open new avenues for promoting plant growth, biocontrol activities, and stress management for the development of eco-friendly sustainable agriculture.

Keywords: microbiome; medicinal plants; plant-microbe interactions; sustainable agriculture.

GAPP32

Comparison of bacterial communities in soil samples with and without tomato bacterial wilt caused by *Ralstonia solanacearum* species complex

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Ralstonia solanacearum is one of the most notorious soil-borne phytopathogens. It causes a severe wilt disease with deadly effects on many economically important crops. The microbita of disease-suppressive soils are thought that they can contribute to the disease resistance of crop plants, thus, evaluation of the microbial community and their interaction characteristics between suppressive soil (SS) and conducive soil (CS) will help to understand resistance mechanism. To do this, the bacterial community structure, correlation analysis with soil chemical properties, interaction network of SS (nearly no disease in three years), and CS (suffered heavy bacterial wilt disease) were analyzed. A higher bacterial community diversity index was found in **SS**, the relative abundance of *Nocardioides, Gaiella* and *norank_f_Anaerolineaceae* were significantly more than that of the CS. Moreover, the relative abundance of main genera *Bacillus, norank_o_Gaiellales, Roseiflexus,* and *norank_o_Gemmatimonadaceae* were significantly more than that of the CS. Redundancy analysis at the genus level indicated that the available phosphate played a key role in the bacterial community distribution, and its role was negatively correlated with soil pH, organic matter content, alkali-hydrolyzable nitrogen, and available potassium contents. Interaction network analysis further demonstrated that greater diversity at the genus level existed in the SS network and formed a stable network. Additionally, the species of *Mycobacterium, Cyanobacteria,* and *Rhodobiaceae* are the key components that sustain the network stability. Seven clusters of orthologous groups exhibited significant differences between SS and CS. Moreover, 55 bacterial strains with distinct antagonistic activities to R. solancearum were isolated and identified from the healthy tomato plant rhizosphere soil of the CS. Our findings indicate that the bacterial diversity and interaction network differed between the CS and SS samples, providing a good foundation in the study of bacterial wilt.

Keywords: *Ralstonia solanacearum;* tomato bacterial wilt; soil samples**.**

GAPP33

Metagenomics analysis of artisanal cheeses: importance for sustainable development and food security in the face of climate change

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Artisanal cheeses play a crucial role in both culinary traditions and local economies around the world. However, the sustainability and future of these unique cheese varieties are under significant threat due to the challenges posed by climate change. As global temperatures rise, extreme weather events become more frequent, and traditional agricultural practices face disruption, it becomes imperative to employ advanced scientific techniques such as metagenomics analysis to understand and mitigate these risks. Metagenomics, a culture-independent approach, allows for the comprehensive study of microbial communities present in artisanal cheeses, providing valuable insights into their biodiversity, composition, and functional potential. By characterizing the microbial diversity and metabolic activities, metagenomics aids in identifying key microorganisms involved in cheese fermentation and ripening processes. Furthermore, it offers a deeper understanding of the impact of environmental factors on the cheese microbiome and its response to changing climatic conditions. The application of metagenomics analysis in artisanal cheese production contributes significantly to sustainable development and food security. Firstly, it helps preserve and promote traditional cheese-making practices by identifying microbial strains unique to specific geographical regions. This knowledge allows for the conservation of indigenous microbial communities, safeguarding the distinct flavors and qualities of artisanal cheeses. Secondly, metagenomics enables the development of innovative strategies to enhance the resilience and adaptability of cheese production systems in the face of climate change. By identifying stress-tolerant microorganisms and their functional traits, it becomes possible to select or engineer microbial consortia that can withstand changing environmental conditions. This resilience is crucial for maintaining cheese quality and ensuring a stable supply of artisanal cheeses even during challenging climatic periods. Overall, metagenomics analysis of artisanal cheeses plays a vital role in ensuring the long-term sustainability, diversity, and availability of these cherished culinary products. By leveraging this powerful scientific tool, we can tackle the challenges posed by climate change, foster local food security, and support the livelihoods of cheese producers, all while preserving the unique cultural and gastronomic heritage embedded within artisanal cheese-making traditions.

Keywords: metagenomics; artisanal cheeses**;** food security.

GAPP34

Effects of lactic acid bacteria on physicochemical properties and residual nitrite of the fermented meat product

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The accumulation of nitrite, one of the main causes of a lot of human diseases, and meat-processing industry and research institutions are testing various strategies to reduce elevated salt concentrations in products while maintaining quality. A decrease in sodium chloride could have an impact on several qualities, either directly or indirectly. Nonetheless, the majority of the current work focuses on the technological issues that salt reduction entails. Lactic acid bacteria (LAB) strains were evaluated in traditional fermented products to determine their nitrite-reducing capability in diverse production techniques in order to generate high-quality fermented meat products. LAB was extracted and identified using 16S rDNA sequencing from traditional meat fermented products. The effects of the isolated bacteria, physicochemical properties, and nitrite-decreasing capacity during meat fermentation were investigated by comparing non-activated and activated treatments at 12°C in the fermentation chamber. The two screened LAB strains were discovered as *Pediococcus pentosaceus* and *Staphylococcus carnosus* which adapted well. The novel fermented product habitat had a high acidifying capacity, with the original activated treatment > modified activated treatment > non-activated treatment, respectively. Additionally, the fermented products' moisture content, essential amino acid content, non-essential amino acid content, lightness, springiness, and water-holding capacity were all increased. However, the product's protein, fat, ash, pH, redness, hardness, gumminess, and chewiness reduced. When it came to sensory evaluation, the original *Staphylococcus carnosus* product outperformed the other experimental groups and the control group in terms of flavor, scent, and acceptability. All of these findings suggest that incorporating starting culture into fermented products is a feasible and effective method of obtaining nutritional and healthy meat products, and improving nitrite residue control.

Keywords: starter culture; lactic acid bacteria; meat fermentation; nitrite residue; meat product.

GAPP35

Identification of rhizosphere bacteria associated with dolichos bean (*Lablab purpureus*) in Namibia

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Dolichos lablab (*Lablab purpureus*) is a multipurpose drought tolerant legume crop which is grown globally. It is known to have a significantly high protein structure when compared to other legumes and its uses are various ranging from human consumption as a vegetable, improving soil fertility, as forage, and for pulse. It is of great benefit to subsistence farmers especially those in marginal areas. In Namibia, there is a knowledge gap when it comes to information on the rhizosphere bacteria found in *Dolichos lablab*. Beneficial microbes, especially rhizobium, offer a sustainable solution for improving nutrient uptake in legume crops and the research community understands a substantial amount about the processes involved in nitrogen fixation in legumes. The present study had the aim to identify the native bacteria found in the rhizosphere of *Dolichos lablab* accessions from 2 regions in Namibia. From Mashare in the Kavango East region and from Windhoek in the Khomas region of Namibia. Isolation involved the use of general media (Luria Bertani agar and/or tryptic soy agar) and selective media such as *Rhizobium* and Yeast Extract mannitol (YEM) Congo red until pure colonies were obtained. The Isolated 85 strains of bacteria were identified by 16S rRNA gene sequencing analysis. Among the identified genus were *Bacillus*, *Acetobacter*, *Streptomyces*, *Exiguobacterium*, *Stutzerimonas*, *Rhizobium*, *Burkholderia* and *Pseudomonas*. The study indicates the potential of these plant growth promoting rhizobacteria for inoculums production or biofertilizers for enhancing growth and nutrient content of beans and other crops under field conditions. The study is the first report of *Dolichos lablab* rhizosphere associated bacterial in Namibia.

Keywords: rhizosphere bacteria; *Lablab purpureus*; dolichos; 16S rRNA; Namibia.

GAPP36

Influence of physical parameters on antinutritional factors of *L. mutabilis* sweet flour subjected to solid fermentation with *A. niger* and *T. viride*

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Agroforestry residues can be a very usable feed source for monogastric cattle, however, the same components of these residues and the presence of certain metabolites (flavonoids and polyphenols) can become anti-nutritional factors (ANF) that prevent the correct assimilation of nutrients by cattle. To find the best parameters to obtain the greatest reduction of these ANF in *Lupinus mutabilis* Sweet foliage meal by solid-state fermentation with *Aspergillus niger* J1 and *Trichoderma viride* M5-2 strains, two laboratory-scale experiments were conducted. Two completely randomized designs were used, with 22 factorial arrangements and three replicates. The factors were pH and humidity for the first experiment and temperature and particle diameter for the second experiment. As a result, changes were obtained in the physicochemical properties of the flour studied, with a notable decrease in fiber, flavonoids and polyphenols, being better those obtained with *A. niger* J1, decreasing polyphenols up to 87% and flavonoids by 63%.

Keywords: temperature; pH; *T. viride*; *A. niger*.

GAPP37

Bacteria from saline soil as promising biocontrol agents against *Fusarium oxysporum*

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The use of microbial inoculants as an alternative to chemical pesticides holds significant value in modern agriculture, addressing the growing demand for sustainable and environmentally friendly pest management strategies. *Fusarium oxysporum* is a plant pathogenic fungus that poses substantial challenges to global agriculture, mostly due to its broad host range and persistence in soil. Therefore, the aim of this research was to find potent bacterial isolate(s) for managing infection caused by this plant pathogen. Twelve bacterial isolates (coded as PAM1.1 – PAM1.12), obtained from saline soil were used in this research. Their ability to produce protease and cellulase, as well as their antifungal potential against *F. oxysporum* were tested *in vitro*. DNA of the most effective isolate was extracted using the CTAB extraction protocol and amplified with primer pair P0/P6 targeting bacterial 16S rRNA gene. For identification, the obtained sequences were compared to those available in the National Center for Biotechnology Information (NCBI) database. Out of twelve tested isolates, PAM1.3 showed the highest proteolytic activity, while the production of cellulase was not detected. This isolate was concurrently the most efficient in suppressing *F. oxysporum*, with an inhibition percentage of 51.7%. Based on the NCBI BLASTn analysis of the 16S rRNA gene sequence, isolate PAM1.3 was identified as *Stenotrophomonas lactitubi,* showing 99.29% identity with *S. lactitubi* strain NR_179509.1. The results of this research indicate that saline soils could be a valuable reservoir of beneficial bacteria with biocontrol properties.

Keywords: *Fusarium oxysporum*; biocontrol; *Stenotrophomonas lactitubi*; saline soil.

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ICGEB MEETINGS & COURSES 2023

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PPP1

Biochemical and molecular approaches in agriculture after heavy metals (copper & cadmium) exposure

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Soil contamination with heavy metals has become a world-wide problem, leading to the loss in agricultural productivity. Plants have a remarkable ability to take up and accumulate heavy metals from their external environment and it is well known that high levels of heavy metals affect different physiological and metabolic processes. *Solanum melongena* were chosen for studying the toxic effect for two elements (cadmium & copper) on some biochemical & molecular characteristic, so cadmium concentration (0,5, 10, 15) ppm were determined from Cd (NO3)2 in addition to control (D.W.) as triplicate for each concentration and Solanum buds irrigate with age (30) day and after implantation in fertilized soil and the irrigation continue for 60 days. On the other hand, Copper concentration was (0,10, 15, 25) ppm which previously prepared from (Cu(NO3)2) in addition to control group represented by D.W. Regarding soil, pH, E.C., nitrate, soil texture values were determined to detect the soil characteristics before and after agriculture. The results have confirmed at the end of the experiment and from plant species analysis that copper concentration and cadmium concentration in irrigation water were lead to change in ROS, SOD, CAT, MDA, GSH-px, total protein, total sugar, proline, chlorophyll & moisture content in *Solanum*. Molecular results showed these concentrations, which have no effect on DNA status i.e. that no damage to DNA in plant with concentration increasing and this compatible with no significance effect on alsgene amplification and more amplified was in 15 mg/l for Cu & 10mg/l for Cd. Heavy metals don't reveal any considerable effect on plant species according to their concentrations. Non response in some biochemical parameters returns to ccumulation of heavy metal and chelating mechanism by plant species. Molecular markers confirmed that these concentrations of heavy metals don't have considerable effect.

Keywords: agriculture; biochemical markers; molecular markers; heavy metals; DNA.

PPP2

Novel research project – BioPhysFun for advancement of characterization of *Trichoderma* as biological control agent

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The use of chemical fungicides is detrimental for soil and aquatic ecosystems. Although highly effective, they raise concerns about impact on the environment. Therefore, the need to reduce the use of chemical fungicides and protect the soil and water, pose the challenge to find novel, eco-friendly, but also more efficient, means of crop protection from phytopathogenic fungi. We are aiming to utilize the native *Trichoderma* species, as biological control agent, for controlling or eliminating the phytopathogenic fungi. Our research will be based on cell nano-surgery of fungal cell wall using ultrashort laser pulses and subsequent patch clamping on the released protoplast membrane. This will enable the studies of activity and potency of specific class of antimicrobial short peptides, peptaibols, released by *Trichoderma* species, which act by forming ion channels in pathogen membrane as one of the main mechanisms of their fungicide action. The result of this project will be the developed instrumentation and unique method based on nano-surgery and patch clamp for studies of ionic channels in native fungal plasma membrane. Testing a range of autochthonous *Trichoderma* sp. isolates will enable formulation of their combinations that work best. The selected Trichoderma strains with the highest standardized peptaibol activity in specific combination of conditions, can be used as a starting point for development of new, more potent biocontrol agents.

Keywords: *Trichoderma*; biological control agents; peptaibols; cell nano-surgery; patch clamp.

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PPP3

Circular economy principles integration in production of *Bacillus*-based microbial products for sustainable agriculture

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The necessity to adopt sustainable practices is present not only in agriculture as the basis of the food production system, but as well in other food chain elements, with a special emphasis on food industry and consumers as the largest generators of food-based waste with high organic burden posing a threat to environmental safety. Since food industry waste has been generated at high rates and it could not be safely disposed without prior treatment, the scientific community has engaged in finding possible solutions for its valorization. Rich nutritional composition of food industry waste corresponds with microbial nutritional profile and requirements, offering a palette of possible bioconversion technologies. From the aspect of sustainable agricultural practice, the most perspective one relates to production of microbial products – biopesticides, biostimulants and soil amendments as value-added products by using food industry waste as substrate in the proposed circular economy approach. The aim of this study was to investigate the possibility to apply different food industry effluents as medium basis for production of *Bacillus*-based microbial products for agricultural applications, including distillery, winery, sugar industry, meat industry, dairy industry and fruits and vegetables processing effluents. The antimicrobial activity of the produced biocontrol agents was investigated against *Xanthomonas euvesicatoria*, pepper bacterial spot causer, and *Aspergillus flavus*, aflatoxigenic corn pathogen, while the seed germination activity was investigated in pepper and corn. The results have shown a great potential of the investigated food industry effluents in this type of circular economy approach, considering that application of winery, sugar industry and dairy industry effluents resulted in higher biocontrol and seed germination activity compared to nutrient broth as the commercial synthetic medium. Further research will include optimization of cultivation conditions to maximize plant-beneficial activities and to offer scale-up ready bioprocess solution for potential industrial adopters, simultaneously with development of microbial product formulation.

Keywords: food industry waste; biocontrol; seed germination; dairy industry effluents; winery effluents; sugar industry effluents.

PPP4

New biofertilization approach based on the microbial multispecies biocatalytic aggregates

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Soil quality is decreasing worldwide, due to erosion and intensive use of farmland and as crop species are not adapted to the reducing nutrient content, we need to develop novel soil fertilization approaches, which will deliver the appropriate nutrients into the root system. Using microbes in the form of multi-species bio-catalytic aggregates, which simulate naturally occurring microbial "hotspots" in the rhizosphere, we are planning to induce a high metabolic activity to support nutrient leaching and other beneficial traits which support plant growth. Using classical and advanced bacterial isolation techniques we have obtained and are still collecting the optimal combinations of plant growth stimulating bacteria (mineral weathering and phosphate solubilizing) which can be combined with nitrogen fixing, biocontrol and polysaccharide producing strains to prepare complex beneficial communities. Real-time quantitative PCR, genome sequencing and RNAseq will be used to characterize the community potential and activity, fluorescence time-lapse microscopy to describe their structure and chemical analyses to demonstrate the effect the microbes have in the substrate.We have obtained numerous strains that have the potential to promote plant growth and are functionally characterizing their genomes. We have developed several approaches to bind the cells together into artificial structures like 2D biofilms or 3D aggregates and pack them into different types of organic or inorganic delivery systems, i.e. cell carriers, that help initiate the formation of bacterial hotspots in the substrate close to the root surface.We are developing an artificial consortium of beneficial bacterial strains that will collectively promote the growth of plants through different mechanisms, nutrient leaching, N2 fixation, water retention and biocontrol and are examining the most optimal ways of delivering them to the vicinity of the root surface. Our main focus is to study the robustness, persistence and functional evolution of the artificially constructed bacterial systems in real agricultural systems.

Keywords: bacterial consortia; cell aggregation and immobilization; nutrient leaching; N2 fixation; biocontrol.

PPP5

Trichoderma species: biofertilizers and biocontrol agents for agricultural sustainability

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Modern agricultural practice is burdened with increasing caution related to the use of chemical inputs in the form of fertilizers and pesticides. These concerns have been followed by a growing interest in more ecological approaches to nutrient supplementation and disease control management. As an option, biofertilizers and biocontrol agents gather the scientific community, aiming to deeply explore and emphasize “the solutions from nature”. In that context, the objective of our research was to isolate *Trichoderma* spp. from different environments (agricultural, forest soil, and pine bark) and test their biocontrol and plant growth promoting potential. A total of 13 *Trichoderma* spp. were isolated, and confrontation tests were established with *Botrytis cinerea*, *Fusarium graminearum*, and *Fusarium oxisporum*. Biochemical characterization involved the assessment of several plant growth promoting characteristics (production of indoles, siderophores and enzymes). The confrontation test revealed different mechanisms employed in newly formed interactions. The mycelia of six *Trichoderma* isolates completely overgrew the tested pathogens, expressing very high antagonistic activity (≥75%), and emphasizing the competitive character of the isolates. The presence of an inhibition zone, a sign of antibiosis as a probable mode of action, was recorded within *F. oxisporum* interactions with several *Trichoderma* isolates. Biochemical characterization confirmed the production of cell-wall degrading enzymes (cellulase, β-glucosidase and N-acetyl-β-glucosaminidase) which are correlated to mycoparasitism. Biochemical assessments confirmed the plant growth promoting nature of *Trichoderma* isolates, which were capable of producing indoles, siderophores and enzymes involved in P cycle (phosphatase and naphthol-AS-BI- phosphohydrolase). The obtained results suggests several newly isolated *Trichoderma* spp. as effective plant growth promotors capable of expressing direct and indirect mechanisms of growth stimulation and acting as promising biocontrol agents against significant crop pathogens. Further analyses demand assessment of field conditions performance and deeper insight into the mechanisms involved in mutualistic interactions with plants and antagonistic interactions with phytopathogens.

Keywords: *Trichoderma* spp.; biocontrol agents; *Botrytis cinerea*; *Fusarium* spp.; plant growth promotion.

PPP6

Comprehensive treatment and analysis of fishpond sediments as a source of organic fertilizers

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Agricultural fertilizers are essential to enhance proper growth and crop yield. Chemical fertilzers endanger ecosystems, soil, plants, and animal and human lives. This has increased interest in biofertilizers which are products that contain living microorganisms or natural compounds derived from organisms such as bacteria, fungi, and algae that improve soil chemical and biological properties, stimulate plant growth and restore soil fertility. Fishpond wastewater and sediment were collected, treated with a natural coagulant, and dried. Greenhouse evaluation of the dried sediment was carried out using maize seeds on soil primed with the sediment. Growth parameters such as plant height, leaf length, root length, plant weight, and percentage yield were determined. The nitrate, total nitrogen, phosphate and total phosphorus content of the biofertilizer were also determined. Plant height in maize showed a 35.8 - 44.4% increase in fertilized maize and a 20.3 - 39.3 % increase in the leaf length. There was a 38.8 - 66 % increase in root length and a 23.9 – 36.5 % increase in plant weight in fertilized maize. The nutritive component was 59.80 mg/Kg, 655.56 mg/Kg, 103.87 mg/Kg and 426.60 mg/Kg, respectively. For a period of 21 days of active growth, the growth rate of maize primed with biofertilizer was 0.93 cm/day with a total yield of 100 % while the control was 0.68 cm/day with a total yield of 50 %. A dried biofertilizer that can improve plant growth, weight, and percentage yield without having to deal with smelly manure looks promising and seems to be a better alternative to explore in agriculture.

Keywords: biofertilizer; fishpond; wastewater; maize.

PPP7

Outer membrane vesicles of plant beneficial bacterial strain *Paraburkholderia phytofirmans* PsJN make a contact with *Arabidopsis thaliana* roots

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Extracellular vesicles (EVs) are recognized as important mediators of intercellular communication in both eukaryotes and prokaryotes. These lipid membrane – coated spherical nanoparticles carry proteins, nucleic acids and other cellular products, and facilitate exchange of these biomolecules among cells within an organism, but also between cells of different organisms, belonging to different species and even kingdoms. Outer membrane vesicles (OMVs), EVs produced by Gram-negative bacteria, are a significant mediator of microbial communication, involved in biofilm formation, virulence, and modulation of host immunity. OMVs of both pathogenic and plant beneficial bacteria have been shown to elicit plant immune responses. Investigations on the modes of OMV-plant cells interactions are still in their infancy, but gain rising attention. Aiming to monitor the interaction between OMVs of *Paraburkholderia phytofirmans* PsJN, a plant growth promoting bacteria, and *Arabidopsis thaliana* roots, we isolated OMVs from bacterial culture in mineral medium, using an ion-exchange chromatography system. Isolated OMVs were labeled with lipid binding fluorescent dye Vybrant™ DiD and unbound dye was removed by washing vesicles on ultrafiltration columns. The same dye concentration in phosphate buffer saline, equivalently washed, was used as a control. *A. thaliana* roots, grown on Murashige and Skoog medium, were incubated with DiD-OMVs or control dye/buffer mixture, washed and observed under confocal laser scanning microscope. Red signals were observed in root hairs and epidermis in DiD-OMV treated plants, while in control-treated roots the same signals were missing. The results indicate direct contact of bacterial vesicles with epidermis and root hairs, which are indispensable for nutrient acquisition and plant-microbe interactions in rhizosphere. Further investigation will address the questions of the nature of OMV-plant cell interaction, including potential delivery of OMVs cargo into host plant cells. Considering that OMVs are increasingly recognized as promising tools in biomedicine, exploring their potential for agronomical applications would be highly appreciated.

Keywords: outer membrane vesicles (omvs); *Paraburkholderia phytofirmans* PsJN; intercellular communication.

PPP8

Investigating the secondary metabolites associated with biocontrol activity of *Bacillus nakamurai* BDI-IS1 against northern corn leaf blight

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The biocontrol potential of *Bacillus nakamurai* strain BDI-IS1 (BDI-IS1) agent against northern maize leaf blight (NLB) was investigated by in vitro and in vivo bioassays and experiments under field conditions in Burundi. The genome of BDI-IS1 was sequenced and its metabolomic profile was used to generate knock-out mutants. The aim of this study is to evaluate the antagonistic activities of mutants of BDI-IS1 against NLB in order to identify the role of secondary metabolites produced by this bacterium. Two independent bioassays were performed in vitro to compare the effects of seven mutants including iturin, surfactin, bacillibactin, bacillaene, bacilysin, plantazolicin and non-ribosomally produced peptides (sfp) and the wild type against the mycelial growth of *Exserohilum turcicum*. In addition, the mutants iturin, surfactin and bacillibactin were also applied on roots and leaves of maize plants under greenhouse conditions to assess their biocontrol potential against NLB. Our results show that BDI-IS1 mutants, which were impaired in iturin and sfp production, significantly lost their efficacy to inhibit mycelial growth of *E. turcicum* (by >85%). In planta, the protection indexes (PI) of root treatments with the three mutants were almost twice that of leaf treatments. Furthermore, the PI of plants treated in roots or leaves with BDI-IS1 mutants defective in surfactin or bacillibactin were significantly lower than that of the wild type. These results suggest a possible role of iturin in direct antagonism of NLB, whereas surfactin and bacillibactin would be involved in indirect antagonism of BDI-IS1 by inducing plant defence responses. Further experiments are under progress to evaluate the differential expression of maize defence genes against NLB in response to BDI-IS1 treatments. These further knowledge on the mode of action of BDI-IS1 will help in developing sustainable crop protection strategies against a major disease in Burundi.

Keywords: biocontrol; metabolites; *Bacillus nakamurai* BDI-IS1; northern leaf blight; induction plant defence responses; protection index.

PPP9

Screening of the effect of synthetic and complex cultivation media on *Bacillus* spp. volatilome profile

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Beneficial bacteria of the genus *Bacillus* are well known as biocontrol and plant growth promoting (PGP) agents, being represented both in scientific research and commercial agricultural applications. Among different mechanisms of action, including competition for nutrients and space and production of wide spectra of diffusible extracellular metabolites, *Bacillus* strains also show capability to produce volatile organic compounds (VOCs), which express plant-beneficial activities even without direct plant-microbe interaction. Considering that this type of biological plant treatment could be of interest especially in close production systems (i.e. greenhouses), its potential should be further explored, mostly in terms of bioactive compounds identification in the mixture of volatiles produced by different *Bacillus* strains that are responsible for a specific plant-beneficial action. Furthermore, bacterial volatilome is highly dependent on the cultivation conditions, as well as on the interactions with plants and other (micro)organisms present in the same ecological niche. The aim of this study was to perform volatilome screening of two *Bacillus* strains with previously proven biocontrol and PGP properties grown on different cultivation media by using headspace solid-phase micro-extraction followed by gas chromatography coupled with mass spectrometry (GC-MS). The results have shown a vast of VOCs synthesized by *Bacillus* spp. in the categories of hydrocarbons, alcohols, ketones, organic acids, esters etc. Furthermore, significant differences were observed in volatilome profile of *Bacillus* spp. grown on different media, including nutrient broth as the commercial synthetic medium in comparison to complex media based on industrial effluents, such as whey and raw glycerol from biodiesel production. As confirmed in this study, *Bacillus* spp. volatilome profile could be manipulated by adjusting the strain cultivation conditions to maximize plant-beneficial action. However, further research is required to better understand signal-based, genetic and metabolic background of VOCs synthesis in *Bacillus* spp. to make a step closer towards their application in sustainable agriculture.

Keywords: GC-MS; volatile organic compounds; biocontrol; plant growth promotion; industry effluents.

PPP10

Audible sound impact on soil microbes

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Modern civilization is producing alarming levels of noise which often has an opportunity to influence soil microorganisms. Nevertheless, its effects on soil microbial communities are unexplored. To investigate the effects of noise of different sound types we tested classical music, construction and road works noise and regular and irregular rhythm played on drums at the environmentally relevant sound levels and time intervals. For that purpose, three different sound amplifiers with separate sound sources (laptop or cell phone) were connected to the speaker driver. To decrease sound leak drivers were placed in a box within a box with thick walls of expanded polystyrene. The distance between the speaker driver and the soil sample was 20 cm and the sound pressure level was 90 dB. All tested sounds impacted soil biofilm forming ability but differently between soil (automorphic vs hydromorphic soil types) and land use types. Agricultural fields were less impacted by noise compared to forest soils. Interestingly only regular rhythm stimulated all tested samples. Such results are the first evidence that noise can change the functional properties of soil microbial communities and that its effects are dependent on the type of sound and rhythm.

Keywords: soil; biofilm; sound; noise; rhythm.

PPP11

A model for predicting the optimal consortium in sustainable agriculture

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Microbial consortia are essential for nutrient mobilization, disease control, plant defense, stress tolerance, postharvest fruit management, and overall ecosystem management. Learning how to predictably assemble microbial consortia that optimize a desired function has the potential to develop sustainable alternatives to chemical pesticides and fertilizers. Community-function landscape models are a novel and promising computational method for predicting which consortium composition will optimize a given function from a large subset with limited experimental effort. In this study, 19 sugar beet bacterial isolates were used to form 171 pairwise combinations tested in triplicate for phosphate solubilization, and indole-3-acetate, siderophore, lipase, and pectinase production. These results were used to parameterize a community-function landscape model and to quantitatively predict the five tested functions for all 3-, 4-, and 5-member consortia that can be formed from our 19 isolates (4,845 consortia in total). Based on these predictions, we selected the six best performing consortia for field testing. We first selected three consortia based on the total sum of the five functions. These included cocultures of *Bacillus subtilis* KO3-18 and *Mixta theicola* KO3-44, *Bacillus halotolerans* C3-16/2.1 and *M. theicola* KO3-44, and *B. subtillis* KO3-18 and *Pseudomonas oryzihabitans* KO3-19. We selected three additional consortia that maximized the minimum value of all 5 functions, indicating their functional versatility. These consortia included *B. subtilis* KO3-18, *M. theicola* KO3-44, and *Curtobacterium pusillum* ED2-6, and the consortium of *B. subtilis* KO3-18, *P. oryzihabitans* KO3-19, and *M. theicola* KO3-44 with or without *C. pusillum* ED2-6. The selected consortia were applied to sugar beet seed, and then planted in experimental fields to evaluate their effectiveness in promoting plant growth and development. Ongoing work includes monitoring and evaluating the performance of these consortia. By combining *in vitro* experiments, community-function landscape models and field trials, our work is blazing a new trail in finding sustainable solutions for agriculture.

Keywords: sugar beet; microbial consortia; community-function landscape models.

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PPP12

Plant extracts and essential oils as potential alternative biocontrol products

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Contamination of plants and seeds with microorganisms and insects is one of the main problems in the production and distribution of various agricultural products, as well as raw herbal material for the preparation of herbal remedies. Biological control of plant pathogens is a potential alternative to the use of chemical pesticides, which have already been proved to be harmful to the environment. Plant extracts and essential oils (EOs) of aromatic and medicinal plants, as well as certain bacteria, showed a significant capacity for biocontrol. To find an effective biological control agent, we investigated the possibility of applying the essential oils and plant extracts in the control of selected fungi isolated from medicinal plants and their seeds. *In vitro* tests were performed in order to select the most active EOs and extracts in relation to the most common phytopathogenic fungi. Essential oils and extracts with the highest antifungal potential were included in further, in vivo, tests as potential fungicides through various treatments: evaporation in special chambers, foliar spray in the field, soil treatment. EO of origano, savory, cinnamon, clove and rosemary were the most effective as well as extracts of horseradish root, kale leaf and nettle leaf. The best effect was shown by combinations of individual oils and extracts through synergistic action. It was observed that oil vapours in combination with plant extracts reduced the level of fungal infection, delayed conidial germination and germ-tube elongation of tested phytopathogenic fungi. Similar results were obtained by treating the seeds of selected plants with a combination of EOs and extracts without affecting the germination of the seeds. The investigation of their insecticidal activity (fumigant, repellent and contact) is ongoing. These researches support the new tendencies of replacing chemical pesticides with natural products.

Keywords: medicinal plants; plant pathogens; biocontrol; essential oils; plant extracts.

PPP13

Characterization of the rhizobiome' bacteriobiota of different *Artemisia* species and analysis of the biocontrol potential of autochthonous endophytes and plant extracts

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This study focused on characterizing the diversity of endophytic and rhizospheric bacteria and their potential for plant growth promotion, biocontrol, and bioremediation obtained from seven native *Artemisia* plants (*A. alba* - AL, *A. annua* - AA, *A. austriaca* - AM, *A. caerulescens* - ACE, *A. campestris* - ACM, *A. santonicum* - AS, and *A. vulgaris* - AV). Alpha diversity analysis revealed a significant difference between the rhizosphere (R) and roots (K), with higher diversity observed in the rhizosphere. Especially, the samples of AA showed high Shannon indices, while the lowest alpha diversity was observed in ACM. Beta diversity analysis at genus level of roots revealed three distinct groups: 1. ACMK and AVK; 2. ACEK and ALK; 3. AAK, AMK and ASK. The roots were particularly represented by taxa such as the *Rhizobium* group (ACMK), *Clostridium* (ACEK and ALK), *Streptomyces* (ALK), *Paenibacillus* (AVK), *Sphingomonas* (AAK, ACEK and AMK) and *Flavobacterium* (ASK and AVK). Interestingly, the highest abundance of *Pseudonocardia*, *Mycobacterium*, *Pedobacter*, *Devosia* and *Chryseobacterium* was characteristic of AMK, while *Micromonosporaceae*, *Actinoplanes*, *Cellulomonas*, *Chitinophaga* and *Cutibacterium* dominated in ASK. Of the total 212 isolates obtained by culturing on different media, 61 isolates were selected for further study. In plant growth promotion tests, *Rossellomorea arthrocnemi* (ACMK) showed the best tolerance to salt stress, *Rouxiella aceris* (ASK) was identified as the most potent producer of IAA and phosphate solubilizer, and *Pseudomonas helmanticensis* (AAK) stood out as the best producer of siderophore. Ten isolates from the genera *Olivibacter*, *Raoultella*, *Paenibacillus*, *Rhizobium*, *Pseudomonas*, and *Priestia* proved to be the most efficient nitrogen-fixing bacteria. In the enzymatic activity tests, the isolates *Bacillus mojavensis* (AMR) and *Paenibacillus xylanilyticus* (AMK) showed the best production of proteases, xylanases, mannanases, pectinases and glucanases, simultaneously. Tests on the bioremediation potential of toxic metals such as cobalt, lead, cadmium, and mercury identified 16 isolates with remarkable capabilities. Finally, the isolates *B. mojavensis* (AMR) and (ACER), as well as extract from AA, showed the most promising antifungal results. Synergistic effects were observed with *Pseudomonas capeferrum* (ACMR) in combination with extracts of AA and AM, and with *Pseudomonas bijieensis* (AAK) in combination with extracts of AA, AM and AL.

Keywords: *Artemisia*; bacteriobiota; endophytes; PGP; biological control.

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PPP14

Maize bacteriobiota and the use of bacterial formulations and smart compost systems in sustainable agriculture

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The synergy of beneficial microorganisms and organic fertilizers emerges as a potent approach for sustainable agricultural management. Our study focused on optimizing plant performance and boosting crop yields through either coating seeds with bacterial formulations or co-sowing with composting material during planting. We conducted a comprehensive analysis of the bacteriobiota, encompassing high-throughput amplicon and whole-genome sequencing, together with computational biology. Our research spanned three types of compost, untreated and treated seeds with *Glutamicibacter* and *Bacillus* species, and the roots of the plants. Beta diversity analysis reveals substantial differences in taxa presence between treated and non-treated seeds. We observed that pure compost (CT1) and compost with post-composting biochar addition (CT2) exhibited substantial similarities in relative abundances and alpha diversity, marked by high Shannon indices. Conversely, compost with biochar integrated during composting (CT3) demonstrated diminished alpha diversity. Beta diversity analysis also differentiated CT3 from the grouped CT1 and CT2. Notably, *Microtrichaceae* exhibited a statistically significant differential abundance favoring CT3 over CT1 and CT2, accompanied by a slight increase in the relative abundance of *Bacillus* and *Geobacillus* genera. In relation to roots, treatment led to an elevated relative abundance of the *Serratia* genus, particularly prominent in sample T6, where its relative abundance exceeded 75%. Although T6 showcased the lowest alpha diversity, these plants displayed a significant increase in seed weight, surpassing untreated samples (T2 and T3), treated-but-not-fertilized samples (T8), biochar-grown samples (T7), and untreated, unfertilized samples (T9). This gain in seed weight translated into enhanced yield per plot, kilograms per plot, and kilograms per hectare metrics, with T6 securing the second position after T8. Notably, T8 demonstrated a statistically significant increase in the relative abundances of *Bradyrhizobium* and *Mesorhizobium* genera, alongside a decrease in *Serratia* abundance. T8 also exhibited an elevated Shannon index compared to T6 samples. In summary, our study achieved a statistically significant increase in maize yield compared to the negative control (6724 kg/ha) through the application of a bacterial formulation and in a combination with CT3 compost (average values 7828 – 16.4% and 7636 kg/ha – 13.6%, respectively).

Keywords: maize bacteriobiota; smartbiofertilizers; bacterial formulations; sustainable agriculture.

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PPP15

Mycorrhizal symbiosis and diversity in fruit crops in Himalayas

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Mycorrhizal associationscan benefit fruit crops which preserve plant variety and ecological function. Soil fertility has decreased as a result of a number of factors, including climate change, overuse of chemicals and tillage. Mycorrhizal fungi are a complex group of organisms that interact with plants in a variety of symbiotic relationships in a wide range of terrestrial habitats and are primarily responsible for supplying soil nutrients and dissolved minerals to plants. Arbuscular mycorrhizal fungi (AMF) are important for improving soil fertility. The study addressed the variety of AMF in the Himalayas and how they affect the soil's physical, chemical, and biological properties. An overview includes information on the diversity of AMF including temporal variation (hyphae, vesicules and or arbuscules), seasonality of P and Zn nutrition and its interaction with AMF colonization and how they contribute to improving soil fertilityin the mycorhizosphere of apple, litchi, citrus, mango and apricot. Notably, *Glomus, Gigaspora, Acaulospora, Sclerocystis* and *Scutellospora* were the predominant genera besides *Endogone* species. The elevated prevalence of *Glomus* species, which showed a more marked frequency of occurrence in the Himalayan regions across diverse fruit crops, was particularly remarkable. We highlighted various beneficial effects of AMF, including carbon sequestration, growth performance, microbial activities, and improved bio-physiolocal characteristics. The synthesis of organic acids and glomalin, which provide defence against soil erosion, bind heavy metals, improve carbon sequestration, and strengthen soil macro-aggregation, demonstrates the beneficial effects of AMF on soil. AMF interact with bacteria that produce alkaline phosphatase, an enzyme in the soil linked to the availability of organic phosphorus. Together, these various AMF activities play a critical part in improving the soil fertility condition of fruit crops. This comprehensive learn of how AMF works not only deepens our understanding but also inspires more research efforts, ultimately assisting in the maintenance and preservation of soil health and productivity.

Keywords: mycorrhizosphere; root colonization; soil enzymes; spore population.

PPP16

Development of Woolee products as an environmentally friendly brand of organic wool fertilizer

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The Lika region is the territorially largest region in Croatia and is known mainly for its protected national and natural parks - the Plitvice Lakes are the most famous. It is ideally located between the coastal areas and the inland and is connected by a main road. An additional advantage is its proximity to the wider Southeast European market. However, Lika has a problem with environmental pollution due to improper disposal and storage of wool in the wild. Sheep breeders do not have a wool market - more than 200 tons per year only in the Lika region and about (more than 1300 tons in Croatia) are illegally disposed of in nature or landfills. There is no processing of raw wool in Croatia. The wool in the karst area has not been processed and is not profitable for the existing textile markets. In addition, the rural karst areas are affected by extreme depopulation. Lika is one of the five most sparsely populated areas in the EU and is socio-economically stagnant, although tourism is the main source of income for the region. The development of the Woolee of organic wool-based fertilizer eco-friendly brand was a solution to this problem. Wool is 100% recyclable and can bring profit as well as benefit the environment. The future of clean technology and green biotechnology is well underway as the cost of producing expensive substances is offset by the benefits of low-cost, efficient and environmentally sustainable recycling processes. Wool has a variety of uses and applications, such as: a) Wool pellets and other products: Agriculture, industry and construction; b) Lanoline and keratine: medical and pharmaceutical industries, cosmetics, personal care and baby care with a triple bottom line. Before Woolee, Croatia and the surrounding countries did not take advantage of the opportunities of wool, which can create new green jobs, sustainable agriculture and a cleaner environment. Accordingly, the potential of Woolee products shows the sustainable perspectives of future developments.

Keywords: wool pellets; sheep breeders; organic wool based fertilizers.

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