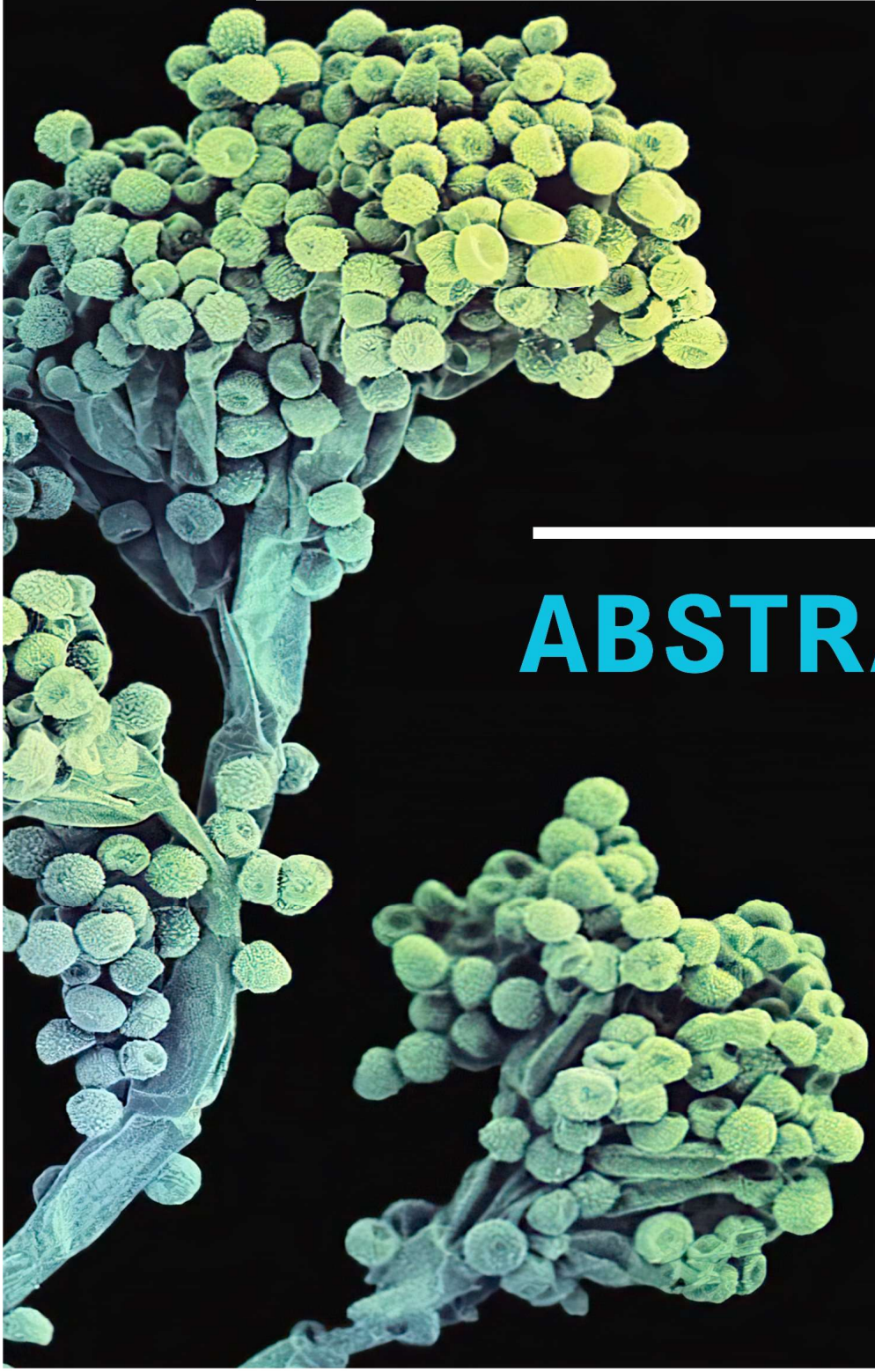
2ND INTERNATIONAL WORKSHOP ON

MYCOREMEDIATION & MYCOTREATMENT



MAY 2025

5TH – 7TH

ABSTRACT BOOK

Université de Sherbrooke
Universidad de Jaén
Universidad Autónoma del Estado de Morelos

WELCOME WORDS

Dear attendees,

We are delighted to welcome you to the **2nd International Workshop on Mycoremediation and Mycotreatment (MYCOREM),** organized by the University of Sherbrooke, the University of Granada, the Autonomous University State of Morelas, SRM Institute of Science and technology and the École Nationale d'Ingénieurs de Sfax. This conference highlights the use and valorization of fungi and their enzymes for bioremediation and other industrial applications.

Our objectives are to foster scientific and technical exchanges around mycoremediation and mycotreatment, promote advancements in these fields, and strengthen collaborations between researchers, industry professionals, and other stakeholders.

We would like to extend our heartfelt thanks to all participants for their presence and commitment. Your participation is essential to the success of this event, and we look forward to discovering your contributions and engaging with you.

We also express our deep gratitude to the University of Sherbrooke and the Groupe de recherche sur l'eau de l'UdeS for their unwavering supports, without which this conference would not have been possible.

We hope this conference will be a source of inspiration and new collaboration opportunities for you. Together, we can advance research and applications of mycoremediation for a more sustainable future.

Welcome to MYCOREM!

Sincerely,

The Organizing Committee

Hubert, Elisabet, Ramon, Vinoth and Tahar





OUR SPONSORS

We extend our sincerest gratitude to our event sponsors for their generous support in making MYCOREM a reality. Your contributions will be invaluable in organizing and executing this event, and we are truly appreciative of your partnership.

To all prospective sponsors, we warmly welcome further contributions and sponsorships to help ensure the success of MYCOREM. For more details on sponsorship opportunities, please refer to the information provided below.

Thank you once again to all our sponsors for your dedication and commitment to advancing the field of mycoremediation.

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PRELIMINARY SCIENTIFIC PROGRAM

May 5th

8:00 - 9:00	**Accreditation**
9:00 - 9:45	*Fungi: morphology, reproduction, nutrition, habitat, and phylogenetic classification* **Elisabet Aranda**, Institute of Water Research, Department of Microbiology, Faculty of Pharmacy, University of Granada, Granada, Spain
9:45 - 10:30	*Polyextremotolerance fungi as unique phenotypes for innovative mycoremediation tools: from molecular to biotech approaches* **Ramon Batista**, Autonomous University State of Morelos, Cuernavaca, Mexico
10:30 - 11:00	**COFFEE BREAK**
11:00 - 11:45	*The enzymatic system in fungi: extracellular, intracellular and advanced oxidation systems* **Elisabet Aranda**, Institute of Water Research, Department of Microbiology, Faculty of Pharmacy, University of Granada, Granada, Spain
11:45 - 12:30	*Fungal enzymes as emerging biocatalysts: A promising platform for the generation of value-added products of industrial significance* **Vinoth Kumar Vaidyanathan**, SRM Institute of Science and Technology, Chennai, India
12:30 - 13:30	**LUNCH BREAK**
13:30 - 14:30	*Ligning modyfying enzymes immobilization and their utilization in continuously operated bioreactors* **Hubert Cabana**, Département de génie civil et de génie du bâtiment, Université de Sherbrooke, Sherbrooke (Qc), Canada
14:30 - 15:30	*Application of fungal enzymes for bioremediation* **Tahar Mechichi**, École Nationale d'ingénieurs de Sfax, Sfax, Tunisia
15:30 - 16:30	The mycelium-based materials – From mushroom to a novel composite **Marc-Antoine Poulin**, Département de génie civil et de génie du bâtiment and Alexis Boisvert, Département de génie mécanique, Université de Sherbrooke, Sherbrooke (Qc), Canada
16:30	**Concluding remarks**

May 6th

OPENING SESSION

8:00 - 9:00 **Accreditation**
Pavillon Marie-Victorin (Bâtiment D7 / D7 Building)

8:45 - 9:00 **Opening Ceremony**
Bienvenue / Welcome to Université de Sherbrooke
Accueil / Opening Remarks

9:00 - 10:00 **CONFÉRENCIÈRE PRINCIPALE / KEYNOTE SPEAKER**
Phenols, pharmaceuticals, plastics and a kingdom supporting us to cope with man-made environmental pollution
Dietmar Schlosser
Helmholtz -Centre for Environmental Research, Leipzig, Germany

10:00 - 10:30 **BREAK & POSTER SESSION**

SESSION 1 - ENVIRONMENTAL STEWARDSHIP USING FUNGI

Session Chair: Hubert Cabana

10:30 - 11:00 *Reducing the Impact of Tire Wear Particles: Fungal Biodegradation, Mycelium Composites, and Toxicological assessments*
Paridhi Singh, Ratul Kumar Das, Satinder Kaur Brar, Raymond Kwong

11:00 - 11:30 *Towards the myco-remediation of halogenated organic compounds: genomic insights of the genus Cladosporium*
Giampiero De Simone, Simona Di Gregorio, David Levin, Laurence Fraissinet-Tachet, Thibault Le Gratiet

11:30 - 12:00 *Integrating Phytoremediation and Mycoremediation: The Role of Riparian Forests and Ectomycorrhizal Fungi in Heavy Metal Mitigation in Southern California*
David Banuelas, Natalie Rubio, Jocelyn Sanchez, Daniel Talamantes, Samuel Brown

12:00 - 13:00 **LUNCH BREAK & POSTER SESSION**

SESSION 2 - ENVIRONMENTAL STEWARDSHIP USING FUNGI

Session Chair: Ramon Batista-García

13:00 - 14:00 **CONFÉRENCIÈRE PRINCIPALE / KEYNOTE SPEAKER**
MyCosmos: Enzyme Factory
Satinder K. Brar
York University, Toronto, Canada

14:00 - 14:30 *Wild Metal-Accumulating Mushrooms: A Path to Decontamination?*
Kawina Robichaud

14:30 - 15:00 *Exploring the Impact of Micro-Aeration on the Hydrolytic Stage of Anaerobic Digestion for the Removal of Emerging Contaminants by Native Fungal Communities*
Ángeles Trujillo-Reyes, Antonio Serrano, Cristina Postigo, Elisabet Aranda and **Tatiana Robledo-Mahón**

15:00 - 15:30 **COFFEE BREAK & POSTER SESSION**

15:30 - 16:00 *Innovative Biochar Strategies for Removing Long-Chain Perfluorocarboxylic Acids from Wastewater*
Sepideh Nasrollahpour, Satinder Kaur Brar

16:00 - 16:30 *Bioprospective screening of fungal isolates from polluted environments towards microplastics and related compounds degradation*
Gabriela Angeles de Paz, María del Mar López-Rodríguez, Antonio Blanco, Luna Guirado-Mendoza, Concepción Calvo, Tatiana Robledo-Mahón, Elisabet Aranda

16:30 - 17:00 *Organophosphate flame retardants degradation by White Rot Fungi*
Diana Losantos, Montserrat Sarrà, Gloria Caminal

May 7th

9:00 - 10:00 **CONFÉRENCIÈRE PRINCIPALE / KEYNOTE SPEAKER**
Bioprospecting in Hostile Environments for innovative mycotechnology applications: the St. Lawrence River Case Study
Félix-Antoine B Simard
Biopterre, La Pocatière, Canada

10:00 - 10:30 **BREAK & POSTER SESSION**

SESSION 3 - ENVIRONMENTAL STEWARDSHIP USING ENZYMES

Session Chair: Hubert Cabana

10:30 - 11:00 *Development of a fungal enzyme-based multifunctional biocatalytic system for biodegradation of organic pollutants*
Muhammad Bilal, Grzegorz Boczkaj

11:00 - 11:30 *Comparison of In-Situ and Ex-Situ Laccase Immobilization Techniques in Terms of Sustainability and Durability*
Gulten Yuksek, Hubert Cabana

11:30 - 12:00 *Presence of Organic Contaminants in Hospital Wastewater and Their Continuous Treatment in a Laccase-Based Packed-Bed Reactor*
Komla Alokpa, Sabrina Saibi, Lounès Haroune, Hubert Cabana

12:00 - 12:30 *Utilization of fungal laccase derivated from Trametes hirsuta strain for the remediation of diclofenac in wastewater treatment plant effluent*
Younès el Yagoubi, Pedro Segura, Hubert Cabana

12:30 – 13:30 **DINER / LUNCH BREAK & POSTER SESSION**

SESSION 4 - MYCOTREATMENTS

Session Chair: Lounes Haroune

13:30 - 14:00 *Efficient Gluconic Acid Production via Aspergillus niger*
Vasanth Kumar Vaithyanathan, Devi Sri, Vinoth Kumar Vaidyanathan, Hubert Cabana

14:00 – 14:30 *Therapeutic Potential of V-shaped Lactobacillus planarum and its bioactives against mould pathogens*
Satish Kumar Rajasekharan, Vinoth Kumar V

14:30 – 15:00 **BREAK & POSTER SESSION**

15:00 – 15:30 *Sustainable Production of Chitinase from Crab Shells and Its Application in Phytopathogen Control*
Devi Sri Rajendran, Priyadharshini Bharathi, Vinoth Kumar Vaidyanathan

15:30 – 16:00 *Sustainable Phytase Enzyme Production from Mucor indicus MTCC 6333 Using Agro-Waste for Improved Feed Utilization in Poultry*
Swethaa Venkataraman, Naveen Krishna, Vinoth Kumar Vaidyanathan

CLOSING SESSION

16:30 – 17:00 **Concluding remarks** *(Organizing Committee)*

MAY 5TH, 2025

PRE-WORKSHOP CLASS

Fungi: morphology, reproduction, nutrition, habitat, and phylogenetic classification

Elisabet Aranda*

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Short Description

Fungi are a diverse group of organisms that play crucial roles in ecosystems. They exhibit a wide range of morphologies, from unicellular yeasts to complex multicellular structures like mushrooms. Fungi reproduce through various methods, including spore formation, budding, and fragmentation. They obtain nutrients through absorption, often decomposing organic matter or forming symbiotic relationships with plants. Fungi inhabit diverse environments, from soil and water to extreme conditions. Phylogenetically, fungi are classified into several groups based on genetic and morphological characteristics.

Learning Objectives

Understand Fungal Morphology: Identify and describe the different morphological forms of fungi, including yeasts, molds, and mushrooms.

Reproduction Mechanisms: Explain the various reproductive strategies of fungi, such as spore production, budding, and fragmentation.

Nutritional Modes: Describe how fungi obtain nutrients and their role in decomposition and symbiotic relationships.

Habitat Diversity: Recognize the wide range of habitats where fungi can be found and their adaptability to different environmental conditions.

Phylogenetic Classification: Understand the phylogenetic classification of fungi and the criteria used to classify them into different groups.

PRE-WORKSHOP CLASS

Poliextremotolerance fungi as unique phenotypes for innovative mycoremediation tools: from molecular to biotech approaches

Ramon Batista*

Autonomous University State of Morelos, Cuernavaca, Mexico

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Short Description

This module provides a comprehensive overview of transcriptomics applied to mycoremediation processes under hypersaline conditions, specifically focusing on the degradation of polyaromatic compounds. It covers experimental design, RNA extraction, sequencing technologies, bioinformatics analysis, and interpretation of differential gene expression data to elucidate metabolic pathways involved in PAH degradation by halophilic fungi.

Learning Objectives

Understand the principles and methodologies of transcriptomics in the context of mycoremediation under hypersaline conditions.

Identify key metabolic pathways involved in the degradation of polyaromatic compounds by halophilic fungi.

Design and implement transcriptomic experiments to study gene expression changes during PAH degradation.

Analyze transcriptomic data using bioinformatics tools to interpret differential gene expression.

Apply transcriptomic findings to enhance the efficiency of mycoremediation processes in extreme environments.

PRE-WORKSHOP CLASS

The enzymatic system in fungi: extracellular, intracellular and advanced oxidation systems

Elisabet Aranda*

Institute of Water Research, Department of Microbiology, Faculty of Pharmacy, University of Granada, Granada, Spain

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Short Description

The enzymatic system in fungi is highly versatile and essential for their survival and ecological roles. Fungi produce a variety of enzymes that can be categorized into extracellular, intracellular, and advanced oxidation systems.

Extracellular Enzymes: These enzymes are secreted outside the fungal cells to break down complex organic materials into simpler molecules that can be absorbed. Examples include cellulases, hemicellulases, and ligninases, which degrade plant cell walls.

Intracellular Enzymes: These enzymes function within the fungal cells to facilitate metabolic processes, including glycolysis, the citric acid cycle, and other biosynthetic pathways.

Advanced Oxidation Systems: These systems involve enzymes that generate reactive oxygen species (ROS) to degrade recalcitrant compounds like lignin. Key enzymes include laccases, peroxidases, and lytic polysaccharide monooxygenases (LPMOs).

Learning Objectives

Understand the Role of Extracellular Enzymes: Identify and describe the function of key extracellular enzymes in fungi and their role in nutrient acquisition.

Intracellular Enzymatic Processes: Explain the metabolic pathways facilitated by intracellular enzymes and their importance in fungal physiology.

Advanced Oxidation Mechanisms: Describe the advanced oxidation systems in fungi, including the generation and role of reactive oxygen species in the degradation of complex organic materials.

Enzyme Classification and Function: Classify the different types of fungal enzymes and understand their specific functions and applications in biotechnology and industry.

Ecological and Industrial Relevance: Recognize the ecological significance of fungal enzymatic systems and their applications in industries such as biofuel production, bioremediation, and food processing.

PRE-WORKSHOP CLASS

Fungal enzymes as emerging biocatalysts: A promising platform for the generation of value-added products of industrial significance

Vinoth Kumar Vaidyanathan*

SRM Institute of Science and Technology, Chennai, India

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Short Description

Advancements in pilot-scale production have enabled the translation of lab-scale enzyme processes into scalable bioprocesses, bridging the gap between research and industrial application. Collectively, fungal enzymes due to their catalytic efficiency, specificity, and eco-friendly nature are transforming conventional industrial practices by enabling sustainable, bio-based production pathways aligned with green chemistry principles and the circular bioeconomy. The most prominent fungal enzymes are lignocellulolytic enzymes including cellulases, hemicellulases, laccases, peroxidases, glucose oxidase, naringinase and lipase produced by species such as Aspergillus sp., Trametes sp, Pleurotus sp, and Trichoderma sp. These enzymes are essential for breaking down lignocellulosic biomass into fermentable sugars and other value-added intermediates, enabling their application in nutraceuticals, biorefinery, biofuel, and waste management sectors.

Learning Objectives

Understand the production of industrially significant fungal enzymes: To examine the cultivation of fungi under fermentation using agro-industrial residues, and to understand downstream purification strategies for obtaining high-purity enzyme preparations suitable for diverse industrial uses.

Analyze the role of fungal enzymes in biomass deconstruction and value-addition: To study the catalytic roles of lignocellulolytic enzymes (e.g., cellulases, hemicellulases, laccases, lignin peroxidases, manganese peroxidases) produced by fungi such as Trametes versicolor, Pleurotus dryinus, and Trichoderma viride, and their contribution to efficient hydrolysis of lignocellulosic biomass for applications in biorefineries and agricultural waste valorization.

Explore enzymatic applications for the synthesis of biobased chemicals: To investigate the application of glucose oxidase in gluconic acid production, naringinase and lipase in biosurfactant synthesis.

PRE-WORKSHOP CLASS

Ligning modyfying enzymes immobilization and their utilization in continuously operated bioreactors

Hubert Cabana*

Département de génie civil et de génie du bâtiment, Université de Sherbrooke, Sherbrooke (Qc), Canada

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Short Description

This course explores the principles and benefits of lignin modifying enzymes (LME) immobilization, as well as the techniques for designing bioreactors that utilize immobilized enzymes. Enzyme immobilization involves attaching enzymes to solid supports, allowing for their reuse and enhancing their stability. Bioreactors that use these immobilized enzymes are optimized biological reaction systems for various industrial applications, such as biofuel production, pharmaceutical synthesis, and wastewater treatment.

Learning Objectives

Understand the principles of enzyme immobilization: Students will learn about different methods of enzyme immobilization, such as adsorption, covalent bonding, encapsulation, and cross-linking.

Identify the advantages of enzyme immobilization: Students will discover the benefits of this technique, including enzyme reuse, improved stability, and reduced production costs.

Design bioreactors using immobilized enzymes: Students will be introduced to the basic concepts of bioreactor design, including types of bioreactors (batch, continuous, etc.) and criteria for selecting supports for immobilization.

Apply knowledge to practical cases: Students will analyze real-world examples of bioreactor use in industry, focusing on the challenges and solutions associated with enzyme immobilization.

PRE-WORKSHOP CLASS

Application of fungal enzymes for bioremediation

Tahar Mechichi*

École Nationale d'ingénieurs de Sfax, Sfax, Tunisia

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Short Description

Lignin-degrading enzymes play a key role in bioremediation, particularly in the breakdown of complex pollutants. These enzymes are primarily produced by white-rot fungi and some bacteria, and they are capable of oxidizing lignin — one of the most recalcitrant natural polymers found in plant cell walls. The key Lignin-Degrading Enzymes are Lignin Peroxidase (LiP), Manganese Peroxidase (MnP), Versatile Peroxidase (VP) and Laccases. Due to their nonspecific oxidative nature, these enzymes are powerful tools for degrading environmental pollutants such as Polycyclic Aromatic Hydrocarbons (PAHs), Synthetic dyes, Pesticides & Herbicides. Lignin degrading fungi have a number of applications including Soil remediation, Wastewater treatment, Treatment of industrial effluents, Detoxification of agricultural runoff. To enhance their efficiency and applicability, Genetic engineering of microbes to overexpress these enzymes, Immobilization techniques for enzyme reuse and stability and use of mediators to expand substrate range are involved. In addition to bioremediation lignin degrading enzymes are used in several biotransformation reactions to produce high value-added molecules including: Vanillin, Bio-based polymers, Pharmaceutical precursors and Natural antioxidants.

Learning Objectives

Understanding lignin degrading enzymes classification and mechanisms of activity

Describe the role of lignin degrading enzymes in biological processes involved in bioremediation.

Explain the mechanisms by which enzymes degrade environmental pollutants.

Identify types of environmental contaminants that can be treated using enzyme-based bioremediation.

Analyze case studies where enzymes were successfully applied in cleaning soils, industrial effluents or other hazardous wastes.

Evaluate the limitations and challenges of enzyme stability, availability, and scalability in real-world bioremediation and biotransformation projects.

Describe lignin degrading enzymes use in biotransformation reactions to produce high value-added molecules

PRE-WORKSHOP CLASS

The mycelium-based materials – From mushroom to a novel composite

Marc-Antoine Poulin*

Département de génie civil et de génie du bâtiment and Alexis Boisvert, Département de génie mécanique, Université de Sherbrooke, Sherbrooke (Qc), Canada

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Short Description

Nowadays, the increasing concern for the environmental impact of plastic pollution has led to a growing interest in developing alternatives to petroleum-based materials such as mycelium-based composites. This latter group are self-grown materials, based on agricultural residue fibers that are inoculated with fungi mycelium. During the process, the mycelium forms an interwoven 3-dimensional filamentous network where all fiber particles are bounded together to create a rigid, lightweight and biodegradable composite material. Mycelium-based composites can be specifically produced to replace current products in the industries of packaging or construction.

Learning Objectives

Description of a classical and a laminated fabrication process of mycelium-based materials

Market interest and application sectors of mycelium-based materials

Characterisation of mycelium-based materials from different residual biomass and comparison with commonly used plastics

Characterisation of failure behavior of laminated mycelium-based assemblies

Hands-on experience of fabricated objects using classical and laminated process.

MAY 6TH, 2025

KEYNOTE PRESENTATION

Phenols, pharmaceuticals, plastics and a kingdom supporting us to cope with man-made environmental pollution

Dietmar Schlosser*

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The kingdom Fungi represents one of the most diverse and ancient branches of the tree of life (Li et al. 2021) and harbors about 150,000 known species, with an estimate of up to 13.2 million species based on high-throughput sequencing (Hyde 2022). Fungi are abundant and everywhere, and have successfully conquered a multitude of marine, freshwater and terrestrial habitats where they dwell as saprotrophs, mutualists or parasites. Terrestrial as well as aquatic habitats of fungi are heavily exposed to a plethora of man-made polluting chemicals, including synthetic polymers (plastics) and their additives. Some of these compounds can serve as substrates for fungal growth, whereas many others undergo cometabolism in presence of suitable growth substrates providing carbon and energy. Initial pollutant attack is often initiated by unspecific enzymes originally "intended" to act on naturally occurring substrates, and may occur intra- (often by cytochrome P450 monooxygenases) or extracellularly (via diverse oxidoreductases and hydrolases). Especially extracellular radical-generating oxidoreductases like lignin-modifying peroxidases and laccases, and reactive oxygen species generated in extracellular Fenton-type reactions enable many fungi to attack even particularly recalcitrant pollutant structures very efficiently. The frequent independence of fungi from using pollutants as a growth substrate seems beneficial for bioremediation in these cases where pollutants are only poor growth substrates since they contain only little energy, or because of their limited availability to microbial degraders (Harms et al. 2011). Situations named last e.g. arise when environmental pollutant concentrations are extremely low as for so-called micropollutants (e.g. many pharmaceuticals, and others), or pollutants are only poorly water-soluble and strongly sorb to environmental matrices (like, e.g. certain phthalate esters used as plasticizers). Another characteristic potentially making many fungi well suited for bioremediation relates to their ability to form extended mycelial networks, hereby allowing resource allocation thus withstanding adverse conditions in certain areas or spots of their habitats (Harms et al. 2011). These biochemical and ecological capacities of fungi provide numerous options to potentially apply them successfully in mycoremediation approaches and also in waste recycling, as exemplified for selected fungi, pollutants and applications.

Harms et al. (2011), Nat. Rev. Microbiol. 9(3):177-92, https://doi.org/10.1038/nrmicro2519

Hyde, K.D. (2022), Fung. Divers. 114:1, https://doi.org/10.1007/s13225-022-00507-y

Li, Y. et al. (2021), Curr. Biol. 31(8):1653-1665.e5, https://doi.org/10.1016/j.cub.2021.01.074

Acknowledgement

This work was supported by the Helmholtz-Association of German Research Centres in the frame of the Integration Platform "Tapping nature's potential for sustainable production and a healthy environment" at the UFZ. Funding within the projects FINEST and PUreValue of the Investment and Networking Fund of the Helmholtz Association under grant agreement numbers KA2-HSC-10 and KA-HSC-13, respectively, is gratefully acknowledged.

KEYNOTE PRESENTATION

MyCosmos: Enzyme Factory

Ratul Kumar Das, **Satinder Kaur Brar***

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Nature and nurture are the two ways of applying microbes for bioproduction and remediation respectively. Inherently, microbes are the cell factory of specific intracellular/extracellular enzymes, and their adaptability make them ideal candidate for diverse applications. In the last two decades of research, our lab has exclusively designed and developed microbial-mediated bioproducts and formulations for bioremediations. Value addition approach has been successfully implemented by bioconversion of agro-industrial wastes into platform chemicals (citric acid, fumaric acid etc.); biofuels (bioethanol, biobutanol and biohydrogen), biosurfactants, antibiotic alternatives and bioplasticizers. Industrially important enzymes (laccase, peroxidase, lipases, alkane hydroxylases, cellulases and proteases) have been produced and formulated for real-filed application. In the bioremediation approach, emerging contaminants (microplastics, PAH, pharmaceuticals, endocrine disruptors etc.) present in wastewater, soil and sewage sludge matrices have been studied for their identification, fate, transport, transformation and removal technologies were developed accordingly. Newer concepts such as enzyme-coated biochar, biochar-enzyme-based membrane modules, cold-active enzyme booster technology, jellyfish-inspired technology have been developed and tested for the field trials of bioremediation at different contaminated sites across Canada.

ORAL PRESENTATION

Wild Metal-Accumulating Mushrooms: A Path to Decontamination?

Kawina Robichaud*

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Soil pollution by trace metal elements (TMEs) is a major environmental issue that requires effective and sustainable remediation solutions. Mycoremediation—using the metabolic abilities of fungi—offers a promising approach for cleaning up contaminated sites. This project focuses on identifying and characterizing wild fungal species from Quebec that can hyperaccumulate TMEs, with the ultimate goal of developing in situ remediation protocols.

We selected contaminated sites in the Quebec and Mauricie regions, where we sampled mushroom fruiting bodies and surrounding soils. The samples were analyzed for TME content, and initial results show that some species can accumulate over 50 times more of certain metals in their tissues compared to surrounding soil concentrations. The next step will involve isolating and cloning fungal species with hyperaccumulation potential for lab-based cultivation tests. A pilot mycoremediation experiment will then be conducted, using environmental modifications to optimize metal uptake by the fungi.

Expected outcomes include the creation of a database of TME-accumulating fungal species, the cultivation and preservation of fungal strains with potential for bioremediation, and the development of remediation techniques tailored to local conditions. This project aims to demonstrate the potential of mycoremediation as an ecological and economically viable solution for managing contaminated soils.

ORAL PRESENTATION

Exploring the Impact of Micro-Aeration on the Hydrolytic Stage of Anaerobic Digestion for the Removal of Emerging Contaminants by Native Fungal Communities

Tatiana Robledo-Mahón*, Ángeles Trujillo-Reyes, Antonio Serrano, Cristina Postigo, Elisabet Aranda and Tatiana Robledo-Mahón

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Anaerobic digestion (AD) is a well-established technology for stabilizing waste materials, including sewage sludge generated during wastewater treatment. However, to meet new requirements regarding the removal of emerging pollutants, AD processes need to be reevaluated in light of current water policy restrictions (Directive 2008/98/EC). Among these emerging pollutants, pharmaceutical active compounds (PhACs) are of particular concern due to their transfer through ecosystems when sewage sludge is used as a soil amendment, aligning with circular economy policies. Unfortunately, AD-treated sewage sludge does not adequately remove some commonly consumed PhACs, such as diclofenac, carbamazepine, and ketoprofen. These compounds are part of daily anti-inflammatory and anxiolytic treatments for a global population. In recent years, micro-aeration has been proposed as a pre-treatment strategy to enhance the two-stage AD process. Implemented in the first stage, controlled micro-aeration (maintaining ~ 0.005-5 L O2/L reactor·day) would promote the proliferation of fungal communities. These fungi produce hydrolytic and oxidative exoenzymes, improving hydrolysis capacity compared to strict anaerobic conditions. Additionally, these fungal enzymes may play a role in organic matter decomposition and PhACs degradation, as demonstrated in aerobic bioremediation systems [1]. Despite this, there are no previous studies on the effects of micro-aeration on PhACs degradation or its impact on fungal communities during hydrolysis. The MICROFUNGI project aims to achieve successful PhACs removal by enhancing hydrolysis in a two-stage AD system through micro-aeration [2]. To achieve this objective, we will apply different oxygen flows: 0, 2.5, 5 and 7.55 0.005-5 L O2/L reactor·day. The analysis of microbial communities will be conducted using Illumina MiSeq sequencing, while PhACs will be quantified using UHPLC-TQ-MS/MS. The changes in the fungal community will be determined by the ergosterol profile. The results obtained will provide key operational parameters and identify relevant microbial communities involved in PhACs removal during anaerobic digestion (AD) with micro-aeration. In conclusion, this research will establish a knowledge framework covering the field of AD with mycoremediation, ensuring the safe use of sewage sludge in soils.

Acknowledgments: This project has been funded by the University of Granada PPJIA2023.033.

[1] Conejo-Saucedo, U. et al. 2021. https://doi.org/10.3390/toxics9060115

[2] Huiliñir, C. et al. 2023. https://doi.org/10.1016/j.biortech.2023.129249.

ORAL PRESENTATION

Organophosphate flame retardants degradation by White Rot Fungi

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Organophosphate flame retardants (OPFRs) are widely used alkyl esters of phosphoric acid commonly added to various commercial products, for fire resistance. As they are not chemically bonded, they can easily diffuse into wastewater streams. Conventional treatments are ineffective at removing these contaminants (Reemtsma et al., 2008), so alternative methods should be investigated. Ligninolytic white rot fungi (WRF) could degrade OPFRs in wastewater, as they can constitutively and non-specifically break down a range of contaminants using extracellular ligninolytic enzymes and a versatile intracellular system.

This work aimed to identify fungal candidates capable of removing water-soluble OPFRs and to understand the mechanism and toxicity associated with this degradation. An initial screening of various WRF revealed that Ganoderma lucidum and Trametes versicolor removed over 90% of tributyl phosphate (TBP) and tributoxyethyl phosphate (TBEP) within 4 days, with Pycnoporus sanguineus also displaying effective removal. Chlorinated tris(2-chloroethyl) phosphate (TCEP) removal was only partially removed (47%) by G. lucidum. A subsequent screening with tris(2-chloropropyl) phosphate (TCPP) showed TCPP's greater susceptibility to degradation compared to TCEP, with T. versicolor exhibiting the highest removal efficiency (77%). This, plus the poor degradation of triethyl phosphate by all fungal candidates suggests an inverse correlation between OPFR's polarity and its susceptibility to fungal degradation.

Biomass sorption studies confirmed WRF's ability to predominantly degrade OPFRs. Enzymatic system tests identified the CYP450 intracellular system as responsible for OPFRs degradation. Further tests on each effectively degraded OPFR allowed to identify transformation products (TPs) and potential degradation pathways, involving typical mechanisms of the CYP450 system, like hydroxylation, dealkylation and oxidative dechlorination, as well as rare mechanisms such as dehydrogenation and reductive dechlorination. Despite theoretical predictions of reduced toxicity for the individual TPs, toxicity of the individual solutions increased throughout the degradation process, according to the Microtox assay, suggesting potential synergic effects. Overall, this study provides valuable insights into OPFRs degradation by WRF, with implications for future WW treatment using mixed consortia, emphasizing the importance of reducing generated toxicity.

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ORAL PRESENTATION

Bioprospective screening of fungal isolates from polluted environments towards microplastics and related compounds degradation

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Predictions about plastic waste production suggest that by 2050, plastic waste in ecosystems will exceed 25 billion tons. The degradability of these polymers vary according to their physical and chemical properties including crystallinity, shape, size, chloride atoms, benzene rings and molecular weight. One solution to this problem is the bioprospection of microorganisms with the ability to degrade these compounds, exploring polluted environments containing plastics as a source of potential degrader strains. Fungi are promising candidates for plastic degradation due to their hyphal growth and extracellular oxidation mechanisms, facilitated by the secretion of extracellular and intracellular enzymes. In this study, we aimed to identify and characterize fungal strains capable of degrading either plastics or plasticizers. To achieve this, we evaluated 27 fungal isolates obtained from plastic-rich sources including sewage sludge composting piles, digested sewage sludge and plastic residues. The screening consist in a) solid cultivation in Petri plates using different common plastics as the only carbon source and b) by evaluating the effect of the microplastics over the growth rates in microplate culture over 5 days. For all the experiments, we used Bisphenol A (BPA), Di(2-ethylhexyl) phthalate (DEHP) and Polyethylene (PE) to be tested. The evaluation included optical density, weighed measurement and Atomic Force Microscopy (AFM). Additionally, the strains were tested for their acute toxicity on Daphnia magna and Lepidium sativum. As a result, among the 27 strains, Aspergillus terreus, Bjerkandera sp., Trichosporon faecalis, Circinella sp, Trichoderma asperellum and Fusarium oxysporum exhibited significant differences in the evaluated parameters positively related with the effective degradation of such plastics and plasticizers as well as their low toxic effect for further biotechnological application. However, further studies are currently addressing to enable the application of these strains in real-world polluted scenarios.

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ORAL PRESENTATION

Innovative Biochar Strategies for Removing Long-Chain Perfluorocarboxylic Acids from Wastewater

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Research Background / Problem Statement: Perfluoroalkyl and Polyfluoroalkyl Substances (PFAS) are persistent "forever chemicals" with significant environmental and health risks, particularly in wastewater. Long-chain Perfluorocarboxylic Acids (PFCAs), like PFDA and PFNA, are especially difficult to remove using conventional adsorbents such as activated carbon. This study investigates biochar, a sustainable and cost-effective adsorbent, for PFCA removal, leveraging Deep Neural Networks (DNNs) to optimize the adsorption process.

Objectives: The research evaluates the adsorption efficiency of wood- and compost-derived biochars for PFDA and PFNA removal, using DNN-based modeling to determine optimal conditions for maximum efficiency.

Methodology: Biochars provided by CHAR Technologies Inc. were characterized using Fourier Transform Infrared Spectroscopy (FTIR). Adsorption experiments simulated wastewater conditions, and PFCA concentrations were measured using LC-MS/MS. Compost-derived biochar, showing promising results, was further analyzed using DNNs to optimize the adsorption process.

Results: FTIR analysis identified critical functional groups in the compost-derived biochar, which significantly outperformed wood-derived biochar in PFCA removal. The DNN model identified optimal conditions for maximum adsorption efficiency.

Discussion: The study highlights the superior performance of compost-derived biochar due to its complex structure and abundant active sites. The integration of DNN modeling with experimental techniques demonstrates the potential for improved PFAS remediation strategies.

Conclusions: Compost-derived biochar is an effective and sustainable solution for PFCA removal from wastewater. DNN-based optimization enhances biochar performance, positioning it as a viable alternative to conventional adsorbents.

Recommendations: Further research should explore biochar regeneration, performance in real wastewater, and scalability for integration into existing treatment systems.

ORAL PRESENTATION

Integrating Phytoremediation and Mycoremediation: The Role of Riparian Forests and Ectomycorrhizal Fungi in Heavy Metal Mitigation in Southern California

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San Jose Creek, a major tributary of the San Gabriel River in Southern California, has a long history of industrial pollution, with concerning levels of lead (Pb), cadmium (Cd), and arsenic (As) detected due to stormwater and wastewater discharge. As contamination persists, there is an urgent need for nature-based solutions to mitigate exposure, particularly through the combination of phyto and mycoremeidation. Riparian forests, including willow (Salix lasiolepis) and Peruvian pepper (Schinus molle) trees, are known to uptake heavy metals, yet their effectiveness in this region remains understudied. These trees host ectomycorrhizal fungi, such as Geopora spp., which play a critical role in metal uptake and stabilization, but their specific contributions to San Jose Creek's remediation potential are unknown. This study will quantify heavy metal concentrations in water, soil, and plant tissues at the Confluence of San Jose Creek and the San Gabriel River, assess the extent to which willows and pepper trees accumulate and sequester heavy metals, and identify the fungal communities associated with these trees using metagenomic sequencing. Understanding the role of fungal symbionts like Geopora in mycoremediation will provide critical insights into their potential to enhance phytoremediation, guiding targeted restoration strategies. By integrating these findings into a community-driven approach, this research aims to deploy riparian forests as an effective remediation tool in historically polluted watersheds, offering a scalable solution for reducing heavy metal exposure in impacted communities.

https://dornsife.usc.edu/eri/eri-community-engaged-research-grantees/

ORAL PRESENTATION

Towards the myco-remediation of halogenated organic compounds: genomic insights of the genus Cladosporium

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Halogenated Organic Compounds (HOCs) are man-made pollutants used for decades in different industries. These compounds are persistent in the environment and pose potential risks of chronic ecotoxicity. Polychlorobiphenyls (PCBs) and Hexachlorocyclohexane (HCH) are among the most concerning HOCs, as they contaminate environmental matrices worldwide, with a particular relapse on soils. Non-white rot fungi can easily adapt to polluted soils and rely on a diverse metabolic machinery; thus, their application for real scale bioremediation approaches is of extreme interest. Despite this, little is known about HOCs degradation by non-white rot fungi. The aim of this study was to evaluate the capability of the non-white rot fungal genus Cladosporium in degrading both PCBs and HCH, with emphasis on the genes involved. To do so, a total of 6 Cladosporium strains were isolated from two polluted sites: one with PCBs and the other with HCH. Fungal degradation of the two pollutants was tested separately for each strain, maintaining the same cultural conditions. The growth medium contained 5 g/L of glucose and Aroclor 1254 (a mixture of PCBs) or tHCH (α, β, γ, δ-HCH 1:1:1:1) at the final concentration of 25 mg/L. Furthermore, the genome of each strain was sequenced via PacBio HiFi circular consensus sequencing and assembled using the dedicated tool HiFiasm. Predicted genes were obtained using the Braker pipeline and subjected to a functional annotation against the FungiDB database. Remarkably, PCBs degradation reached peaks of 75% after 5 days. HCH degradation capacity turned to be less efficient, thus, additional tests have been performed to improve and compare it in different growth conditions. Functional annotation identified on average 12,000 genes per strain. Pentachlorophenol monooxygenase and other genes related to PCBs degradation were detected, alongside different genes encoding for non-specific oxidizing enzymes such as laccases and dioxygenases, all potentially involved in HOCs degradation. Multiple dehalogenases were also annotated. The obtained results suggest numerous fungal degradation mechanisms and are pivotal for the development of myco-remediation approaches for the decontamination of HOC-polluted soils.

ORAL PRESENTATION

Reducing the Impact of Tire Wear Particles: Fungal Biodegradation, Mycelium Composites, and Toxicological assessments

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Tire wear particles (TWPs) are a growing environmental concern as microplastic and heavy metal pollutants, particularly zinc, which leaches into aquatic ecosystems during tire degradation. Zinc-laden leachates disrupt food webs and accumulate in organisms, posing ecological risks. While remediation strategies exist, scalable, sustainable solutions remain limited. Fungi show potential for metal bioremediation via biosorption, bioaccumulation, and enzymatic transformation, yet key knowledge gaps remain. These include interspecies fungal interactions in co-culture, their impact on zinc sequestration, and the role of mycelium-based composites (MBCs) in long-term zinc immobilization. Furthermore, the toxicological effects of TWP leachates on zebrafish (Danio rerio) embryos, including gene expression and oxidative stress, are poorly understood.

This study aims to (i) isolate and characterize fungi with zinc adsorption and enzymatic transformation capabilities, (ii) investigate co-culture dynamics to optimize zinc removal and MBC properties, and (iii) assess the toxicity of untreated and fungal-treated TWP leachates in zebrafish.

Twenty-four fungal isolates from a decade-old tire disposal site will be screened using zinc-spiked agar to identify high-tolerance species, followed by liquid culture trials with synthetic TWP leachates to quantify biosorption via ICP-AES. Promising strains will be tested in co-culture for synergistic zinc removal and incorporated into MBCs using lignocellulosic substrates. Zebrafish embryo assays will evaluate the ecotoxicological impact of untreated versus fungal-treated leachates.

This study aims to identify fungal strains with strong zinc bioremediation potential, develop MBCs for zinc sequestration, and assess whether fungal treatment reduces TWP leachate toxicity. By optimizing fungal consortia and composite formulations, we seek to improve zinc sequestration and mitigate ecotoxicological effects. Findings will contribute to nature-based remediation strategies for stormwater management and sustainable materials. Future research should focus on scaling co-culture fermentation, field-testing MBC stability, and integrating fungal-based solutions into waste management policies.

Keywords: Tire wear particles, fungal bioremediation, enzymatic zinc transformation, mycelium-based composites, co-culture, ecotoxicology

MAY 7TH, 2025

KEYNOTE PRESENTATION

Bioprospecting in Hostile Environments for innovative mycotechnology applications: the St. Lawrence River Case Study

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The fungal kingdom is vast and contains many different species with a wide variety of useful properties, supported by enzymes and their associated metabolites (primary and secondary). These properties are, among others, inherent to their genetic plasticity and ability to rapidly adapt to new hazardous and difficult to colonize environments. Therefore, fungal bioprospection in hostile environment have tremendous potential for solving urgent global issues (environmental sustainability, medicine, food security, etc.).

To offer innovative solutions to the companies that approach us to develop new eco-responsible biotechnological processes, we have undertaken bioprospecting campaigns in hostile environments, particularly the St. Lawrence estuary. Indeed, its physicochemical parameters (variation in osmotic pressure and depth, low temperature, strong current, pH, etc.) suggested the possibility of discovering new fungi (psychro-, halo-, osmo- and/or contaminant-tolerant).

Sampling campaigns on this emblematic Quebec River have enabled us to acquire several fungal tools that are very useful for 1- biomonitoring the St. Lawrence estuary against various stresses (global warming, environmental contaminants, exotic invasive species, etc.) and 2- isolating fungal species with great biotechnological potential. Among these, it is possible to highlight specimens which are particularly effective for producing various biomolecules of interest, such as biocides (Epicoccum nigrum), melanin (Cadophora sp.), mycoprotein (Paradendriphiella salina), mycomaterials (Isaria farinosa), degradation of contaminants (Lulwoana sp., Pestalotiopsis linearis), and many others. Thus, the fungal biodiversity of the St. Lawrence River is a good example that supports the idea that hostile environments are real gold mines for the development of innovative bioprocess.

ORAL PRESENTATION

Development of a fungal enzyme-based multifunctional biocatalytic system for biodegradation of organic pollutants

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Industrial and human activities frequently produce and release a wide variety of environmental contaminants that negatively affect water quality and pose a major threat to aquatic life and humans. To maintain environmental sustainability, it is crucial to eliminate such contaminants. Enzyme-based biocatalytic processes offer advantages of low energy input, non-toxicity, mild operating conditions, minimal sludge generation, and applicability over a wide range of pollutants. However, free enzymes suffer due to their low stability and reusability, which limit their applicability. In this study, the main research objectives are to design immobilized laccase-based robust bio-catalytic systems, and their exploitation to degrade a diverse set of emerging pollutants. A novel biopolymeric hybrid material was synthesized by combining attapulgite and sodium alginate (ATP/Alg). Laccase from Trametes versicolor was covalently cross-linked and immobilized on the surface of ATP/Alg. The immobilization efficiency and laccase loading achieved with ATP/Alg at a 2:1 ratio was 91.4% and 104 mg/g, respectively. By employing SEM-EDX, XRD, FT-IR, TGA/DTG, and zeta potential, the ATP/Alg hydrogel composite was characterized both with and without immobilized Lac, confirming successful fabrication coupled with effective Lac attachment. In contrast to the aqueous form, ATP/Alg/Lac exhibited superior tolerance over a wide window of pH and temperatures. The degradation capacity of 2,4-dichlorophenol was assessed using the ATP/Alg/Lac biocomposite. It is noteworthy that ATP/Alg/Lac effectively eliminated 2,4-DCP, allowing its complete decomposition in the aqueous solution within a 30-minute timeframe. Furthermore, it exhibited encouraging reusability characteristics, retaining 96.18% and 70%, respectively, of its initial biodegradation capacity after three and six reuse cycles, respectively. Lac-2,4-DCP complex revealed a docking score of -4.418 kcal/mol, and an MMGBSA score of -32.31 kcal/mol with making H-Bond type interaction among ALA-103, SER-225 residues. The immobilized laccase derivative also exhibited a promising efficacy in catalyzing the degradation of a variety of other emerging pollutants. Altogether, the developed system holds noteworthy potential in catalyzing similar pollutants given easy separation, green bio-fabrication, and little energy consumption.

Keywords: Environmental biotechnology; Biocatalysis; Emerging pollutants; Enzyme immobilization

ORAL PRESENTATION

Presence of Organic Contaminants in Hospital Wastewater and Their Continuous Treatment in a Laccase-Based Packed-Bed Reactor

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Hospital wastewater can contribute significantly to the contamination of municipal wastewater. Due to inadequacy of conventional wastewater treatment facilities to effectively remove trace organic contaminants (TrOCs), these compounds can eventually reach environmental water bodies. Fungal laccases (EC 1.10.3.2) have shown promising potential to remove TrOCs from aqueous media. However, the potential use of free laccase in wastewater treatment is hampered by a few drawbacks such as enzyme washout, lack of stability, or the need of large amounts of enzyme.

In this study, we first investigate the presence of TrOCs in an untreated HWW from the CIUSSS de l'Estrie – CHUS (Hôpital Fleurimont), Sherbrooke (Qc, Canada). Second, laccase from Trametes hirsuta was covalently immobilized on an amino-functionalized silica microspheres attached to polyethylene packings (PE-AFHMS-GLA-Lac) which were used as bed material in a plug-flow type packed-bed reactor (PBR) for continuous removal of TrOCs from the HWW. The PBR was fed with previously filtered HWW on a 0.45µm PTFE filter, in unspiked form and spiked form at 1µg L–1 with acetaminophen (ACT), ibuprofen (IBP), naproxen (NPX), ketoprofen (KTP), mefenamic acid (MFA), indomethacin (IDM) and carbamazepine (CBZ). The occurrence of the TrOCs in the HWW was investigated using a non-targeted UPLC-MS, whereas a targeted UPLC-MS/MS was used to assess their removal.

After 6h of the PBR operation in a batch mode with spiked HWW at a 324-min hydraulic retention time (HRT) using 1215 U of PE-AFHMS-GLA-Lac at pH 6.8±0.5 and 22±1°C, more than 94 and 92% removals were achieved for ACT and MFA, respectively whereas KTP and trimethoprim (TPM) were removed at 35 and 20%, respectively. Using the PBR in a continuous mode at a 180-min HRT with unspiked HWW, more than 98% of ACT was removed. NPX, CBZ, IBP, and ofloxacin were removed at 71, 69, 65, 59, and 52%, respectively but carbendazim, TPM, atenolol, and caffein were the least removed molecules at 27, 34, 39, and 42%, respectively. The different removal rates observed suggest the contribution of various mechanisms such as biocatalysis, adsorption, and oxidative radical reactions. The oxidative efficiency of this laccase biocatalyst makes it suitable for use in the wastewater treatment industry. In addition, the treatment reduced the toxicity of the treated HWW, as the latter did not significantly affect the mobility of Daphnia magna, unlike untreated HWW.

ORAL PRESENTATION

Comparison of In-Situ and Ex-Situ Laccase Immobilization Techniques in Terms of Sustainability and Durability

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Laccase, a biocatalyst predominantly derived from white-rot fungi, has significant potential for environmentally sustainable treatment. However, its large-scale application is hindered by instability, high production costs, and sensitivity to environmental fluctuations. Immobilizing laccase onto solid matrices has been studied to enhance its stability, reusability, and resistance to operational stressors. In-situ immobilization integrates enzyme production and binding into a single step, whereas ex-situ immobilization involves attaching commercial, purified, or crude pre-produced enzymes onto functionalized supports. This study aims to identify a more sustainable and scalable methodology by evaluating catalytic efficiency, operational durability, and economic feasibility. In the ex-situ method, three different enzyme sources were used: commercial laccase from Trametes versicolor, partially purified laccase from Pleurotus dryinus, and crude fermentation broth. After activating silica-coated supports with glutaraldehyde at various pH levels, immobilization was carried out via covalent bonding. The process lasted 48 hours at 20°C, continuously shaking in dark conditions. For the in-situ method, P. dryinus was cultivated in liquid media supplemented with laccase inducers, including copper, ethanol, methanol, hexane, and industrial hexane waste. Mesoporous silica-coated plastic packings were directly introduced into the fermentation flasks, enabling real-time enzyme binding. The cultivation was conducted under both sterilized and unsterilized conditions at 26°C, 150 rpm for 22 days. Enzymes immobilized through the ex-situ technique demonstrated excellent stability across a broad range of pH and temperature conditions, retaining more than 50% of their activity after five months at 4°C. In contrast, free enzymes degraded rapidly under similar storage conditions. The in-situ method reached a maximum activity of 1.25 U/mg packing under sterilized conditions, with industrial waste hexane proving an effective inducer. Meanwhile, using high-activity crude broth (~15,000 U), the ex-situ method achieved up to 346 U/g packing with a 60% immobilization yield. Even at low enzyme loading (15 U), 1.8 U/g activity was obtained with 74% efficiency. According to the experimental results, in-situ immobilization enables operational simplicity with lower enzyme activity. It reduces chemical usage through integrated enzyme production, whereas ex-situ approaches provide enhanced catalytic efficiency, greater stability, and effective use of crude enzyme sources. Both strategies align with green chemistry principles and offer scalable, practical pathways for advancing environmental biocatalysis, though they require further studies for optimization.

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ORAL PRESENTATION

Utilization of fungal laccase derivated from Trametes hirsuta strain for the remediation of diclofenac in wastewater treatment plant effluent.

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The release of trace organic contaminants (TrOCs) into the environment from WWTPs is a major environmental problem. Remediation of TrOCs by traditional oxidation processes such as ozonation, photolysis or chlorination leads to the formation of numerous known transformation products which, in turn, increase the toxicity of the solution, with a low reduction of the organic total carbon (TOC). However, as well as being a 'friendly' and 'forward-looking' process, the use of fungal laccases would probably reduce it, with promising TOC reduction, thanks to the formation of as yet unidentified high-molecular-weight transformation products. The Trametes hirsuta strain seems promising for bioremediation of TrOCs in complex matrices. However, multiple genes responsible for its secretion are present, even within a single organism. These different laccases possess small physico-chemical differences and could potentially modify characteristics such as their stability against various denaturing agents or their effectiveness in eliminating particular contaminants.

The aim of this presentation is to improve the understanding of the impact of laccase on TrOCs by characterizing the particles formed and identifying the transformation products resulting from the reaction between diclofenac and fungal laccase.

Two main laccases were observed and purified called Yn and Yg. Peptide fingerprinting analysis suggested that Yn and Yg was constituted mainly of laccase Q02497 and laccase A0A6M5CX58, respectively. Robustness tests, based on tolerance and stability, showed that both laccases were affected in a relatively similar way. Determination of kinetic constants (KM), catalytic constant (kcat) and kinetic efficiency (K=kcat/KM) for the transformation of diclofenac indicates a lower KM and kcat for laccase Yn but relative similar K constant compared to Yg. Numerous diclofenac dimers and trimers have been identified, differ from one another mainly in the number of hydroxylations, decarboxylations and unsaturations. The high hydrophobicity and low solubility of these oligomers cause them to precipitate, followed by agglomeration, through hydrophobic interactions, to unsoluble particles. The formation of these insoluble particles reduces bioavailability for biota, thus explaining the reduced toxicity observed in the literature, even at low initial diclofenac concentrations and in a complex matrix such as effluent of WWTPs.

ORAL PRESENTATION

Efficient Gluconic Acid Production via Aspergillus niger

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Gluconic acid (GLA) is a mild acid with many industrial applications in chemical, pharmaceutical, textile, agriculture and food industries. The production of gluconic acid at industrial scale has been carried out using Aspergillus niger based fermentation process. The fungal based fermentative GLA production processes have the following drawbacks (1) complex production media which are costly and give impurities, (2) fermentation broth contains many byproducts formed such as colored substances and other organic acids, (3) overall production time usually requires several days, (4) yield of gluconic acid is lower and (5) extensive purification steps are needed. Considering the above mentioned problems of the fermentative GLA manufacture process, many efforts have been through to progress an enzymatic method by the glucose oxidase (GOD) and catalase. Still, no account of economically viable and stable enzymatic route for the production of GLA is available. In A. niger, glucose is transformed into gluconic acid by an enzymatic complex consisting of GOD and catalase. In our previous work, we utilized low-cost resource like municipal biosolids for the production of GOD up to 6600 U/L (Kumar et al., 2020). In the present study, we utilized the partially purified Aspergillus niger GOD enzyme to optimize the production of gluconic acid from glucose. The novelty of the present study was employing a three-phase partitioning (TPP) technique for the purification of glucose oxidase from Aspergillus niger based submerged fermentation process. At optimum conditions of 28°C and 80% (w/v) ammonium sulphate along with a 1:1 ratio of t-butanol to crude extract, the fold purification of GOD is enhanced by 11.8-fold with an activity recovery of 82 %. The purified GOD along with commercial catalase used for the conversion of glucose into GLA. The 1:1 ratio of liquid enzyme preparations were mixed with 0.5 M glucose solution at pH 6.0 and temperature of 28°C. After 3 h of incubation time, the obtained mixture contains GLA, GLA was quantified using high performance liquid chromatography. These results can be inevitable to improvise the biocatalytic method to be an inexpensive, economical and attractive technology for better conversion and find application in industrial sector.

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ORAL PRESENTATION

Therapeutic Potential of V-shaped Lactobacillus planarum and its bioactives against mould pathogens

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Probiotic Lactobacilli may produce bioactive metabolites that represent promising alternatives to antibiotics for combating food-borne and drug-resistant pathogens. The current study characterizes antagonistic capacity of V-shaped Lactobacillus planatrum cells generated in response to acidic pH stress. We investigate the inhibitory effect of cell-free filtrates from the V-shaped structured L. planatrum cells against hyphae formed by postharvest mold pathogens. We identify several new antibiofilm metabolites by liquid chromatography–mass spectrometry (LCMS) profiling that is secreted during V-shaped cell formation that function as adenylate cyclase inhibitor. We suggest that functionalized probiotic cells and postbiotics produced by them may effectively mitigate pathogenic microorganisms that provide a staple basis for developing functional and medicinal foods.

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ORAL PRESENTATION

Sustainable Production of Chitinase from Crab Shells and Its Application in Phytopathogen Control

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In this study, crab shells have been utilized as an abundant waste resource, for sustainable chitinase production using Trichoderma viride, a well-known chitinase-producing fungus. This eco-friendly approach offers a cost-effective alternative for enzyme production while promoting sustainable practices in industrial biotechnology. The enzyme activity of optimized media was found to be 29.66 U/ml containing 10% glucose syrup, 10% crab shell, 0.5% casein, 1% yeast extract, with 5% of inoculum at the optimum time of 48 h, 30 °C, and pH 6.0. Analysis of the fermented broth revealed the successful synthesis of chitinase, confirmed by Fourier transform infrared spectroscopy. Furthermore, the chitinase enzyme was purified using chromatographic techniques to obtain a highly active and stable enzyme preparation. The kinetic parameters such as Vmax, km, Kcat, and Kcat/ km values of chitinase are observed as 13.85 ±1.09 µM, 10.221± 0.7 µM, 542.4± 2.36 µ M/sec, and 53.06±0.2 sec-1 respectively. An improved zone of inhibition of 10 mm, 8 mm, and 12 mm against Phoma medicagnis, Fusarium oxysporum, and Rhizoctonia Solani, respectively was observed. The obtained chitinase can find applications in various industries, including agriculture, food processing, and waste management. The integration of waste utilization and enzyme production underscores the importance of a circular bio-economy for a more sustainable future.

ORAL PRESENTATION

Sustainable Phytase Enzyme Production from Mucor indicus MTCC 6333 Using Agro-Waste for Improved Feed Utilization in Poultry

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Dietary fibers play a vital role in animal feed by promoting gut health, increasing fecal bulk, and maintaining salinity. However, the bioavailability of essential minerals and nutrients is significantly impeded by phytate, a major phosphorus storage compound that binds with minerals, leading to poor digestion, malnutrition, and environmental pollution. Dephytinization is therefore necessary to improve nutrient absorption. This study explored various agro-wastes, including wheat bran, rice bran, chickpea husk, and black gram husk, for their potential in phytase enzyme production. A thermostable phytase enzyme was produced from Mucor indicus by optimizing media components and immobilization parameters. Refining culture conditions—specifically temperature, pH, inoculum age, inoculum size, carbon source, and nitrogen source—enhanced enzyme activity from 16.78 U/ml to 177.78 U/ml. The optimal conditions for enzyme production were identified as 50°C and a pH of 5.5. Kinetic analysis revealed a Vmax of 17.85 µmol/min. Dephytinization of broiler and layer feed using the immobilized enzyme liberated 35.45 mg/g and 58.46 mg/g of phosphorus after 24 hours of incubation. These findings highlight the effectiveness of utilizing agro-waste substrates and optimizing culture conditions for the efficient production of phytase, contributing to improved mineral bioavailability and enhanced nutritional quality of animal feed.

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POSTERS

POSTER PRESENTATION

Unraveling microplastic biodeterioration mechanisms: A comparative analysis of whole-cell fungi cultures and enzymatic approaches

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Microbial attachment, as well as extracellular and intracellular enzymatic action on its surface, are the primary mechanisms of microplastic biodegradation. This study aims to investigate at these underlying processes and compare the efficacy of whole-cell fungal systems of white rot fungi, Trametes hirsuta, against crude enzyme extract in achieving efficient microplastic transformation. Microscopic analysis indicated considerable cellular proliferation around microplastics, implying the successful microbial attachment on microplastic surface. The contribution of extracellular enzymes was examined through the activity of hydrolases and oxidases over a period of 2.5 weeks. Laccase, manganese peroxidase, esterase, xylanase, carboxymethyl cellulase and glucosidase exhibited increased activity in the presence of microplastics. Additionally, further enhancement was observed while using weathered microplastics and in presence of biosurfactant, rhamnolipids. This suggests that surface modifications and reduced hydrophobicity could improve the availability of enzymes. Further, there was an increase of about 4.9% in weight average molecular weight (Mw) value of microplastics after 90 days of incubation with whole-cell culture. However, the inhibition of intracellular enzyme CYP450, also achieved similar Mw change, 5.3% implying that they did not contribute to primary biodeterioration of microplastics. In contrast to whole-cell culture, crude enzyme extract showed no Mw changes. This highlights the importance of synergistic action of microbial attachment and enzymatic activity. Overall, this study is a firsthand move towards developing feasible fungal treatment strategies to tackle microplastic pollution.

POSTER PRESENTATION

Comparative Secretome Exploration of Two Cuban Native Strains of Ganoderma Genus: Applicable for Biodegradation of Environmental Pollutants

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Environmental pollution with xenobiotics is a major ecological concern on the planet. Nowadays, research on pollutants, their origin and the development of innovative treatment technologies are priorities of science. Fungal bioremediation by means of white rot fungi is a promising technology exploiting their capacities of degrading pollutants. The ligninolytic machinery and biotechnological potential of well-adapted autochthonous strains belonging to genus Ganoderma remains underexplored. In our previous study, strains Ganoderma weberianum B-18 and Ganoderma sp. UH-M were the most promising strains for the degradation of dyes and Polycyclic Aromatic Hydrocarbons. Moreover, the diversity of genes encoding ligninolytic enzymes was determined, it was also demonstrated that laccase isozymes produced by these strains played a key role in the degradation of xenobiotics (Torres-Farradá, et al, 2017; 2018). Up to now, secretome analysis of WRF in presence of pollutants is limited, especially in Ganoderma species. Exploring the secretome of these strains could reveal novel hydrolyzing enzymes valuable for the degradation of pollutants. The objective of this study is: To analyze the laccase gene expression and the secretome of strains Ganoderma weberianum B-18 and Ganoderma sp. UH-M grown on SB-U medium and during the biological treatment of dye Remazol Brilliant Blue (RBBR) and naphthalene by laccase transcripts analysis and comparative secretome approach. The laccase transcripts were amplified with specific primers and the secretome analysis was performed by SDS-PAGE and LC-MS/MS. The combination of these methodologies allowed us to detect that laccase genes are differentially regulated. The genes lac 2, lac 4 and lac 6 from strain B-18 and genes lac II and lac VII from strain UH-M were constitutively expressed; however, the presence of pollutants induced the synthesis of the laccase isoforms encoded by genes lac 1, lac 3 from B-18 and lac V from UH-M. The comparative secretome analysis performed in the presence of RBBR and naphthalene, allowed the detection of new enzymes, confirming that these xenobiotics switched on the expression of enzymes in order to degrade these recalcitrant pollutants. The characterization of the secretome of these Ganoderma sp. strains can bring new insights into the mechanisms of the complex machinery of the WRF. The set of proteins detected are suitable for the degradation of a wide range of lignocellulosic-related compounds.

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2. Torres-Farradá, G, Manzano León AM, Rineau F, et al. Biodegradation of Polycyclic Aromatic Hydrocarbons by native Ganoderma sp. strains: identification of metabolites and proposed degradation pathways. Appl Microbiol Biotechnol. 2019 Sep; 103 (17): 7203-7215.

POSTER PRESENTATION

Hyphal interactions in soil remediation with Tricholoma vaccinum and Schizophyllum commune

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Mycelial networks form enormous surface areas providing three-dimensional interaction structures with soil particles, soil pollutants, microorganisms and plant roots, and play crucial roles in mycoremediation. To develop strategies for soil remediation we work with soil from our testfields on a former uranium mining site near Ronneburg, Thuringia, Germany. Frequently such radionuclide/heavy metal contamination plumes are co-contaminated with polycyclic aromatic hydrocarbons (PAH). Working with two basidiomycete species, we applied a series of treatments, combining different media with soil, metals and PAHs (phenanthrene, pyrene and fluoranthene), glass beads and saw dust. On these substrates, we inoculated five different genotypes of our white rot model organism Schizophyllum commune, including a laccase overexpressing and a strontium adapted version, or four variants of the ectomycorrhizal basidiomycete Tricholoma vaccinum. in addition, co-inoculations with ectomycorrhizosphere bacteria and the host tree, Picea abies, were performed.

Using qPCR and RNA-sequencing, this set-up allows to study the role of hydrophobins that are known to cover aerial hyphal surfaces and aldehyde dehydrogenases involved in fungal phytohormone biosynthesis in soil interactions, detoxification, metal tolerance and mycorrhization. Evaluation of growth rates were combined with fluorescence microscopy of Fura-stained metals and autofluorescent PAH molecules, showing uptake and transport of metals and adsorption and transport of PAH molecules in vacuoles of T. vaccinum and S. commune. Hyperaccumulation, increased septation, hyphal death and release of vesicles led to the hypothesis that PAH tolerance mechanisms of S. commune include the hyperaccumulation into certain hyphae, which are compartmentalized by septa and subsequently degenerated to release the toxins. On PAH-contaminated soil, the laccase gene lcc2 was up-regulated in the laccase overexpressing strain, and the transporter gene mfs4 enciding a member of the major facilitator superfamily (MFS) proteins was down-regulated in the Sr-adapted strain. These results indicated active PAH tolerance mechanisms for S. commune and provide a basis for further investigations into possible degradation capacities. The mycoremediation potential of white rot fungi like S. commune combined with ectomycorrhizal fungi such as T. vaccinum thus can be applied to remediate co-contaminated soils.

POSTER PRESENTATION

Innovative Biochar Strategies for Removing Long-Chain Perfluorocarboxylic Acids from Wastewater

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Problem Statement: Perfluoroalkyl and Polyfluoroalkyl Substances (PFAS) are persistent "forever chemicals" with significant environmental and health risks, particularly in wastewater. Long-chain Perfluorocarboxylic Acids (PFCAs), like PFDA and PFNA, are especially difficult to remove using conventional adsorbents such as activated carbon. This study investigates biochar, a sustainable and cost-effective adsorbent, for PFCA removal, leveraging Deep Neural Networks (DNNs) to optimize the adsorption process.

Objectives: The research evaluates the adsorption efficiency of wood- and compost-derived biochars for PFDA and PFNA removal, using DNN-based modeling to determine optimal conditions for maximum efficiency.

Methodology: Biochars provided by CHAR Technologies Inc. were characterized using Fourier Transform Infrared Spectroscopy (FTIR). Adsorption experiments simulated wastewater conditions, and PFCA concentrations were measured using LC-MS/MS. Compost-derived biochar, showing promising results, was further analyzed using DNNs to optimize the adsorption process.

Results: FTIR analysis identified critical functional groups in the compost-derived biochar, which significantly outperformed wood-derived biochar in PFCA removal. The DNN model identified optimal conditions for maximum adsorption efficiency.

Discussion: The study highlights the superior performance of compost-derived biochar due to its complex structure and abundant active sites. The integration of DNN modeling with experimental techniques demonstrates the potential for improved PFAS remediation strategies.

Conclusions: Compost-derived biochar is an effective and sustainable solution for PFCA removal from wastewater. DNN-based optimization enhances biochar performance, positioning it as a viable alternative to conventional adsorbents.

Recommendations: Further research should explore biochar regeneration, performance in real wastewater, and scalability for integration into existing treatment systems.al application.

POSTER PRESENTATION

Valorisation of olive mill solid waste by Anthracophyllum discolor and Stereum hirsutum for phenols and volatile fatty acids recovery

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The management of the olive mill solid waste (OMSW), a by-product generated during the extraction of olive oil in two phase processes, is one of the main challenges due to the enormous volume generated, i.e., 4 tons of OMSW per ton of olive oil. However, implementing a proper valorisation strategy would make possible to change OMSW from being treated as a waste to a potential source of benefits. For example, the OMSW is a rich source of valuable phenolic compounds, such as tyrosol or hydroxytyrosol, or volatile fatty acids (VFA), which can be used as building-block molecules for diverse bioprocess. Phenolic compounds are of significant industrial interest due to their antioxidant and/or antimicrobial properties. However, their presence in untreated OMSW is problematic due to their association with phytotoxicity. Thus, the recovery of these compounds would mean a double benefit since, in addition to obtaining economically interesting compounds, their presence in the OMSW would be reduced, thereby reducing their phytotoxicity.

This study aimed to assess the efficacy of two fungi, Anthracophyllum discolor and Stereum hirsutum, to recover valuable compounds from OMSW, such as enzymatic extracts, VFA, and/or phenolic compounds, under the stimulation of extracellular enzymes production in solid-state fermentation conditions for 30 days and simultaneously determine its phytotoxic effects on tomato plants. The laccase activity of A. discolor (15-day) was improved by 88.7%, and S. hirsutum (20-day) was improved by 32.3% with Cu-Mn addition. The Cu-Mn addition also resulted in an increase of 97.2% in MnP activity by S. hirsutum. Regarding the phenol release from OMSW into the liquid fraction, S. hirsutum exhibited a higher release capacity than A. discolor, achieving a concentration up to 6001 ± 236 mg gallic acid eq L-1. Also, S. hirsutum generated the highest concentration of VFA (1627±325 mg L-1), mainly isobutyric acid and acetic acid. Using treated OMSW, the solid fraction showed a slightly lower phytotoxicity than the raw OMSW for both strains. However, it was insufficient to avoid severe phytotoxicity effects in the germination of Solanum lycopersicum at OMSW doses higher than 10% (w/w).

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POSTER PRESENTATION

Laminated mycelium composites – A novel design approach

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In recent times, the increasing concern about the environmental impact of plastic pollution has sparked a growing interest in developing alternatives to petroleum-based materials, such as mycelium-based composites. In these composites, the filamentous mycelium network of fungi acts as a binding matrix, gradually consolidating residual agricultural fibers together as it grows. As such, a rigid, lightweight, and biodegradable composite material is thus created with minimal energy input or waste production. Mycelium-based composites can be specifically produced to replace current products in industries such as packaging, interior design accessories, and even construction, including materials for thermal and acoustic insulation. Moreover, an innovative approach of manufacturing these composites was developed using a lamination process. Specifically, these laminated boards gradually merge over time through their respective mycelial networks. To enhance material strength and expand design options, various potent growth stimulators were applied at the laminated interfaces to foster a dense mycelial concentration. Results were obtained using 3-point flexion bending tests, which were used to assess the internal energy per unit volume stored by the resulting mycelial growth at the laminated interface.