



**SLAMPP**  
NATIONAL SYMPOSIUM  
PLANT HEALTH

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**Proceedings of the  
2<sup>nd</sup> National Symposium of Sri Lanka Association for  
Mycology and Plant Pathology (SLAMPP)**

**Theme: “Towards Sustainable and Eco-Friendly  
Strategies for Plant Disease Management”**



**25<sup>th</sup> June 2022  
PGIS, University of Peradeniya**



**Plant Health 2022**

**"Towards sustainable and eco-friendly strategies of plant disease management"**

**Proceedings of the Second National Symposium of Sri Lanka Association  
for Mycology and Plant Pathology**

**25<sup>th</sup> of June 2022**

**VIRTUAL SYMPOSIUM**

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**Sri Lanka Association for Mycology and Plant Pathology**

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## **Welcome to Plant Health 2022**

*"Towards Sustainable and ecofriendly strategies of plant disease management"*

Plant Health 2022, the Second National Symposium, organized by the Sri Lanka Association for Mycology and Plant pathology (SLAMPP), is held this year as a virtual event, hosted at the Postgraduate Institute of Science (PGIS), University of Peradeniya. This Symposium is one of the key events in the field of Plant Pathology and Mycology, held in Sri Lanka with the participation of professionals in both academia and Research Institutes.

The causative agents of plant diseases, fungi, Oomycota, bacteria, mollicutes, nematodes, Protozoa, viruses, and viroids, are responsible for an estimated yield loss of 10–15% of the world's major crops annually. Among them, approximately 70–80% plant diseases are caused by a diverse array of pathogenic fungi. We have historically experienced that, without proper protection and disease management, the yield loss of crops would range from 50 to 100%, leading to partial or entire destruction of field and plantation crops. Plant diseases can adversely impact on food security, regional and national economic prosperity and the well-being of humans globally and existence of natural environment. Apart from crop losses, the plant diseases are a major threat to ornamental plants, cut flowers, forest trees and global exchange of plant germplasm and a challenge to the conservation of rare species.

Recently, we have witnessed somewhat sudden and forcible introduction of certain eco-friendly strategies for plant disease management in Sri Lanka. Although it is widely known that the commercial agriculture would not flourish and meet the demand of increasing global population without proper strategies of disease management, using chemical fertilizer and synthetic pesticides. Therefore, it is highly recommended to maintain standards guided by agricultural experts, supported by willingness and awareness of farmers for applying those strategies.

The sustainable approaches of disease management include agricultural practices that use bio-based plant nutritional supplements as well as the correct use of chemical methods in disease management with zero or minimum harm to the human and environmental health. Plant microbiomes, the total microbial communities, and their theater of activities have been the basis to develop bio-based strategies of plant health supplements including biofertilizers, bio-control agents, biopesticides, bio-herbicides and plant immunity enhancers. Considering the recent mutual interests in both national and international scientific communities, policy makers, farmers and other interested parties, this symposium was organized under the theme of "Towards sustainable and ecofriendly strategies of plant disease management".

Founded in 2007, the SLAMPP is a professional Association focused on promoting and disseminating knowledge in Mycology and Plant Pathology in the country and acting as the official mouthpiece of the Mycology and Plant Pathology community. SLAMPP is an Associate Member of the International Society of Plant Pathology (ISPP), ISPP council, the Asian Mycological Association (AMA) and the Association of Asian Societies of Plant Pathology (AASPP).

You are invited to join us in our journey of taking Plant Pathology and Mycology forward, and combat the threats of disease and pests that continue to challenge Plant health in our country as well as globally.

Organizing Committee  
Plant Health 2022

25<sup>th</sup> of June 2022



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## **Keynote Speakers**



**Prof. Pratibha Sharma**

ICAR- Emeritus Scientist

Former Principal Scientist, Head and Professor  
Division of Plant Pathology,  
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New Delhi 110012 India

Professor Pratibha Sharma served at the Plant Pathology Division, Indian Agricultural Research Institute (IARI), ICAR, New Delhi for 45 years, after joining as a Scientist in the Agricultural Research Services, UPSC in September 1976. She also completed her PhD as an in-service candidate. Major areas of her research centered on various aspects viz., Variability in Plant Pathogens, Breeding for Plant Disease Resistance, Biological Control, Integrated Pest Management, and Integrated Disease Management of different agricultural cropping systems.

She specifically worked on fungal and bacterial bioagents, developed biologically effective strains, and demonstrated technology in the farmer's field. Developed a patent and, commercialized the technology on *Trichoderma harzianum* and transferred the technology to the State Agriculture department of Rajasthan. As an ICAR- Emeritus Scientist (2017- 2020) in SKNAU Jobner-Jaipur, she worked on the use of biocontrol agents (ecofriendly) in consortium against major pests of Groundnut and Cumin. Presently she works on Agri start-up on Waste Decomposer technology of ICAR-IARI. She has guided more than 30 MSc and PhD students, published >110 research publications in international and national journals, 17 books, 35 chapters in edited books and organized >20 trainings at national and international level. Prof. Sharma is an author of ICAR textbook on "Biological control of plant pathogens and weeds" (2014), Editor of *Trichoderma- Host Pathogen interactions and applications* (2020) by Springer.

Awarded by professional societies, including Lifetime Achievement Award (2022) by S. K. Chaudhary Educational Trust's and Krishi Vigyan Kendra, Madhubani- Bihar, President (2021-22) by Indian Phytopathological Society, New Delhi, Dr. Y. L. Nene outstanding Plant Pathology Teacher award (2019), National Crystal Agri award by Krishi Anusandhan and Kisan Vikas Foundation (2014), New Delhi, Sharda Lele award (2014) by Indian Phytopathological Society and by Indian Society of Mycology and Plant Pathology. Prof. Sharma has been elected as Fellows of many societies including Indian Phytopathological Society (2004), Indian Society of Ornamental Horticulture (2011), Society for Applied Biotechnology (2011), Doctor's Agricultural and Horticultural and Horticultural Development Society (2018), and served as expert member of ICAR, Department of Science and Technology and Department of Biotechnology, Academic council, Research advisory committee.



**Dr. Romina Gazis**

Assistant Professor, Department of Plant Pathology, Director Plant Diagnostic Clinic, Tropical Research and Education Center, University of Florida, FL, USA

Dr. Romina Gazis is an Assistant Professor in the Department of Plant Pathology, University of Florida. During her PhD at the University of Maryland and two postdoctoral fellowships at Clark University and University of Tennessee, her research areas were quite diverse. Her dissertation focused on plant health-promoting fungal endophytes. At Clark University as a postdoc fellow, she conducted research on fungal systematics and evolutionary biology. At the University of Tennessee, she used population genetics and genomic approaches to investigate the evolutionary dynamics and disease ecology of the pathogen/vector system involved in Thousand Cankers Disease of walnut trees.

Her long-term research goal is to understand the biology behind different plant diseases, affecting local industries and natural landscapes and use this knowledge to develop efficient and long-term disease management strategies. At Tropical Research and Education Center, University of Florida (TREC), Dr. Gazis has a 60% extension and 40% research appointment. She is the primary research Plant Pathologist for tropical and subtropical ornamental plants in South Florida and, the Director and Lead Diagnostician of the Plant Diagnostic Clinic at the TREC, Homestead, Florida.

The primary focus of her position has been the research/extension work oriented towards the needs of stakeholders; more specifically, the development of a statewide research and extension program which manages diseases affecting tropical and subtropical ornamental and landscape plants and fruit crops. This program emphasizes education and dissemination of science-based information about ornamental plants and tropical fruit production, disease and pest management, and best practices for pesticide use. Its development and implementation, guided by the needs of farmers, allied industry representatives and county extension agents, occurs in the following main areas: (1) Improving disease management and therefore increasing the sustainability of ornamental plants and tropical fruit production, and (2) Developing a statewide educational program to improve disease management of tropical and subtropical ornamental plants and fruit crops. Her work includes direct research on empirical disease biology topics as well as associated outreach and education programs to enhance awareness about pathogens, emergence and management of diseases, and potential use of locally adapted natural enemies in disease management.

## **Abstracts of the Keynote addresses**

## **Advances in the biological control strategies**

Pratibha Sharma\*

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The evolution of biological control strategies has made the microbial technology as an asset to the organic and Integrated Pest Management systems. The trend towards a biological system management is to develop an integrated relationship between the biological system of the agroecosystem with the crop production system. There had been need based changes in the development of biocontrol strategies. The interrelationships between biological entities – pest, and microbial community with a particular ecosystem play a key role in developing management strategies. There is a change in basic assumptions in the strategies and the knowledge being incredibly old has evolved at various steps. Prioritization of biocontrol research and application is an utmost requirement of all plant protection managers since, biocontrol directly or indirectly has become a vital component. Use of single strain, composite of strains and consortium of different microbial genera makes different formulations. The role of industry needs to be emphasized and should also come out with their viewpoints There is also a need to develop a second generation of BCAs besides the most promising and common tested BCAs. Commercial use, development and application of microbial technologies need to be strengthened under different environmental conditions. The production of commercial formulation of biologically active microorganisms needs to be upgraded keeping in view the cost of developing, testing, registering, and marketing of these products.

## **Tropical plant pathology: challenges and opportunities**

Romina Gazis\*

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Located in the subtropics but with a tropical climate, south Florida receives an average of 13 hours of sunshine per day and 58 inches of annual precipitation. Annual average temperature is 23°C and the annual low temperature typically stays above 18°C. These conditions make south Florida ideal for the development of a large scale and diverse horticulture industry. Second in production of ornamental plants only to California, south Florida leads the country in production of tropical foliage. Moreover, Florida is the only continental U.S. state where a diverse array of tropical fruits can be grown at commercial scale. Only increasing in numbers with each year, nurseries produce tropical foliage, woody ornamentals, tropical fruit trees, and palms. There are more than 1,500 nurseries in Miami-Dade County alone. Landscaping businesses, including pest control companies, are booming due to the importance of aesthetics in the tourism industry and the recognized value of plants to human health. Recent demand for ornamental and specialty crop production suggests significant growth potential for the plant production industry. On the other hand, these prime environmental conditions that allow rapid plant growth year-round are also conducive to fungal establishment, infection, growth, persistence, and spread. Therefore, high disease pressure and new disease introductions, represents constant challenges for local growers. Furthermore, the increasing demand for organically farmed produce, while seldomly available in south Florida, is extremely hard to satisfy under the constant threat imposed by a persistent reservoir of disease-causing agents. This talk covers fungal diseases which affect three categories of staple Floridian crops: tropical fruits, landscape, and ornamentals. Data for disease incidence, host association, and severity was gathered from state-wide submission records to the Plant Diagnostic Clinic located at the Tropical Research and Education Center and from activities conducted through the University of Florida research and extension program. The current and future socio-economic impact of selected pathosystems will be discussed.

# **Abstracts**

## **Technical Session I**

## Evaluating the use of arbuscular mycorrhizae, from *Panicum maximum* grass, as a bio-fertilizer supplement to paddy cultivation in Sri Lanka

P. C. U. Wanninayake<sup>1\*</sup>, P. W. I. M. Chandrasekara<sup>1</sup>, S. Hettiarachchi<sup>1</sup> and M. A. P. W. K. Malaviarachchi<sup>2</sup>

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Indiscriminate uses of chemical fertilizers in paddy field is acquiring the attention of scientists since it could lead to hazardous health issues and environmental impact directly. Exploration of alternative sources to compensate the chemical fertilizers is necessary to all over the world. Therefore, this study was designed to evaluate the use of root colonized fungi referred to as 'Arbuscular Mycorrhizal Fungi' (AMF), from roots of a weed grass, '*Panicum maximum*' as a bio-fertilizer to cultivation of rice. A field trial was carried out in rice cultivation field at Monaragala, Sri Lanka. Suwadel (*Oriza sativa* L.) was selected as rice variety for the study and a Randomized Complete Block Design was employed with three replicate blocks in 8 different treatment plots per block. The size of a plot was 3 m × 3 m. Different percentages of chemical fertilizer regimes (100%, 50% and 25% fertilizer) and no amendments as controls were applied while AMF inocula were applied with fertilizers as treatments (100% fertilizer +AMF, 50% fertilizer +AMF, 25% fertilizer +AMF, AMF alone). Initial average root colonization of prepared AMF inocula was 75-85%. The harvest of rice and also percentage of AMF colonization in rice roots were quantified. The results revealed that, dry weight of harvest for Suwadel was significantly higher ( $p < 0.05$ ) in treatment of 50% fertilizer with AMF and 100% fertilizer with AMF from all the other treatments. Highest number of seeds was observed from 100% fertilizer but no significant different ( $p > 0.05$ ) from 50% fertilizer with AMF and 100% fertilizer with AMF. Treatments with added AMF inocula alone has shown considerably higher percentages of AMF colonization (average 82% of root colonization) but no significant difference in the harvest observed. Decrease in colonization of AMF was observed with the increase of fertilizer level from 25% to 100%. Increasing synthetic fertilizer may have inhibited AMF colonization hence there may be a negative effect on AMF establishment by fertilizers. According to these results, usage of AMF inocula as a bio-fertilizer combined with 50% of chemical fertilizer is an effective method to improve the sustainable agriculture in Sri Lanka.

**Keywords:** arbuscular mycorrhizal fungi, bio-fertilizers, kuruluthuda, paddy cultivation, suwadel

**Acknowledgment:** Authors are thankful to SRI-PROM project for providing space and funding to research work to improve the sustainable agriculture in Sri Lanka.

## Potential of mycorrhizae from *Panicum maximum* grass as a bio-fertilizer to use for Chili cultivation

P. C. U. Wanninayake<sup>1\*</sup>, P. W. I. M. Chandrasekara<sup>1</sup>, S. Hettiarachchi<sup>1</sup> and M. A. P. W. K. Malaviarachchi<sup>2</sup>

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Excessive usage of chemical fertilizers is a major concern in sustainable agriculture. Therefore, objective of this study is evaluating the use of mycorrhizae as eco-friendly and farmer friendly bio-fertilizer to reduce the indiscriminate usage of chemical fertilizers to agricultural fields. The study was designed to evaluate the use of root colonized fungi called 'Arbuscular Mycorrhizal Fungi' (AMF), obtained from weed grass called '*Panicum maximum*' as bio-fertilizers to cultivation of chili (*Capsicum annum*). Field trial was carried out at field crop research and development institute, Mahailluppallama. Randomized complete block design was employed with three replicate blocks with 6 different treatment of plots per block. 3 m × 3.5 m size plots were used and 35 chili plants were in one plot by providing 60cm × 40cm space requirement. Different percentages of chemical fertilizer regime (100% fertilizer and 50% fertilizer) and no amendments as controls were applied while AMF inocula were applied with fertilizers as treatments (100% fertilizer +AMF, 50% fertilizer +AMF, AMF alone). Initial average root colonization of AMF inocula was 75-80%. Harvest of chili was quantified and percentage of AMF colonization in chili roots was verified. According to results, it was revealed that harvest of the treatment with AMF + 50% fertilizers is significantly higher than ( $p < 0.05$ ) the treatment with added 50% fertilizer only while there is no significant different from 100% fertilizer added treatment. Average root colonization of AMF in 50% fertilizers + AMF treatment was 40% and it may be the reason for this recorded results of harvest. Even though the average AMF colonization level in AMF alone added treatment was 52%, there is no significant difference from others for harvest. Also, significantly higher difference was observed the treatment of AMF + 50% added fertilizers from treatment which only 50% added fertilizers were there. It provide another evidence to reveal positive effect of AMF to increase the harvest of chili when the amount of chemical fertilizers is in low level. Nevertheless, 31% average root colonization was observed in AMF + 100% added fertilizers treatment, there was no significant effect to its harvest. Finally, this study concluded that, AMF with addition of low level of chemical fertilizers are a better substitution for the reduction of indiscriminate uses of chemical fertilizers to chili cultivation. Further investigations are required with farmer's field trials to improve sustainable agricultural system using these AMF bio-fertilizers.

**Keywords:** arbuscular mycorrhizal fungi, bio-fertilizers, chili, *Panicum maximum*

**Acknowledgment:** Authors are thankful to SRI-PROM project for providing space and funding to research work to improve the sustainable agriculture in Sri Lanka.

## **Efficacy of ozone treatment in controlling microbial growth on selected upcountry vegetables**

W. M. S. S. K. Weerasinghe<sup>1\*</sup>, G. D. N. Menike<sup>1</sup>, R. M. R. N. K. Ratnayake<sup>1</sup>, W. M. C. B. Wasala<sup>1</sup>

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Carrot (*Daucus carota* L.), potato (*Solanum tuberosum*) and radish (*Raphanus raphanistrum*) are economically important vegetable crops in Sri Lanka. The major problems associated with commercial cultivation of these crops are excessive postharvest loss during storage and transportation due to mechanical damages and the diseases like bacterial soft rot. Ozone application has been proven a promising method to control diseases thereby reducing the postharvest losses in many vegetables. Ozone is a strong disinfectant used to inactivate microorganisms without leaving any harmful by-products when it is in contact with a product surface. This research aimed to determine the effectiveness of ozone in reducing the pathogenic microbial load of carrot, potato and radish. Two different approaches i.e. aqueous (ozonated water) and gaseous forms of ozone (200mg/hr) were tested against the microbial inactivation on those crops for two different time durations (15 minutes and 30 minutes). Four treatment combinations were evaluated for each vegetable under ambient conditions (30-34 °C, 70-75% RH) and the microbial populations in the samples were examined by the total plate count method using serial dilution technique. As the control treatment crops without ozone treatment was used. All samples were analyzed in triplicate before and after the ozone treatments and results were expressed as colony-forming unit per gram (cfu/g) of the sample. The microbial washing efficiency was calculated by deducting the final microbial load from the initial microbial load and divided by initial microbial load which was expressed as a percentage. The data were subjected to analysis of variance (ANOVA) and means were compared using Duncan multiple range and LSD tests with SPSS statistical software 20.0. For carrot, potato and radish, the lowest mean microbial plate counts after the ozone treatments were  $1.22 \times 10^4 \pm 0.38$ ,  $7.37 \times 10^3 \pm 0.29$  and  $2.062 \times 10^4 \pm 0.55$  cfu/g, respectively and the highest mean washing efficiencies were  $93.46 \pm 0.24\%$ ,  $70.47 \pm 0.32\%$  and  $86.74 \pm 0.52\%$ , respectively after treatment with gaseous ozone for thirty minutes. Thirty minutes after treatment with aqueous ozone, carrot, potato and radish exhibited the lowest mean microbial plate counts of  $5.289 \times 10^4 \pm 0.47$ ,  $1.866 \times 10^4 \pm 0.18$  and  $8.520 \times 10^4 \pm 0.84$  cfu/g respectively and the highest mean washing efficiencies  $71.73 \pm 0.23\%$ ,  $25.27 \pm 0.35\%$  and  $45.22 \pm 0.52\%$ , respectively. Both aqueous and gaseous treatments of ozone were effective in reducing the microbial load on surfaces of all three crops tested where the exposure to gaseous form of ozone for 30 minutes gave better result.

**Keywords:** microbial washing efficiency, ozone treatment

**Acknowledgment:** We would like to show our gratitude to the National Institute of Post-Harvest Management for supporting us during the course of this research and we thank the reviewers for their insights.

## **Environmentally friendly management strategies for cabbage nursery diseases**

C. Ranasinghe<sup>1\*</sup>, M.D.A. Jayasekera<sup>1</sup>, K.P.H. Chathuranga<sup>1</sup> and W.M.A.S. Bandara<sup>1</sup>

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Establishment of a cultivation with good quality planting materials is a basic requirement to obtain a satisfactory yield at the end. Cabbage cultivation is usually started with cabbage seedlings raised in the nursery beds. Diseases caused by soil borne fungal pathogens, damping off and downy mildew, major cabbage seedling diseases in Sri Lanka. Application of fungicides are the most effective way of controlling these diseases. But, under green agriculture concept, it is required to find the alternatives for the fungicides. Therefore, this study was conducted to find the non-chemical cabbage nursery disease management strategies. The experiment was laid out in a Randomized Complete Block Design with three replicates. Four treatments were practiced; soil amended with 0.5 Kg/m<sup>2</sup> biochar, 1 Kg/m<sup>2</sup> biochar, application of recommended fungicides and the application of Bordeaux mixture. Untreated control was maintained for comparison. In biochar applied treatments, biochar was mixed with the soil two weeks before seeding. In fungicide applied treatment Captan was applied one day before seeding and Mancozeb 64% + Metalaxyl 8% was applied at 3,5 and 7 weeks after planting. Bordeaux mixture was also applied at 3, 5 and 7 weeks after planting. Disease incidences of damping off and downy mildew, plant height and number of leaves were recorded before every application. During the experimental period rainfall, relative humidity and environmental temperature was recorded as weather parameters. Finally, fresh and dry weight of the seedlings were recorded. Data was analyzed by ANOVA with SAS software. At 5 weeks after seeding, significantly low disease incidence of damping off was recorded in all treatments than untreated control. At that time significantly low downy mildew disease incidences were recorded in both fungicide and Bordeaux mixture applied treatments than other treatments. But at transplanting there was no significant difference in both disease incidences in all treatments. Highest fresh and dry weights were recorded in 1 Kg/m<sup>2</sup> biochar amended treatment followed by 0.5 Kg/m<sup>2</sup> biochar amended treatment. Further highest plant height was recorded in 1 Kg/m<sup>2</sup> biochar amended treatment, but not significantly difference with other treatments. But there was no significant difference in no of leaves among the all treatments. During the experimental period all the weather parameters were recorded favorable for both diseases. Therefore, soil amended with biochar and application of Bordeaux mixture to the nursery beds is effective in managing the said diseases and help establishing good quality planting materials.

**Keywords:** biochar, bordoux mixture, cabbage seedling, nursery diseases

## ***In-vitro* assessment of Cinnamon bark oil as an effective antifungal agent to control avocado stem end rot caused by *Lasiodiplodia* sp**

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Avocado is one of the most demanding fruits in Sri Lanka. Sellers and consumers are being faced a major issue due to the stem end rot caused by *Lasiodiplodia* sp. Use of chemical fungicides for postharvest treatment is prohibited in Sri Lanka. However, other commonly practicing methods are not effective to complete control of stem end rot. Therefore, this experiment was conducted to evaluate the effectiveness of cinnamon bark oil (50% cinnamaldehyde) as essential oil to control the *Lasiodiplodia* sp. isolated from avocado fruits with typical symptoms. In-vitro trials were conducted as poisoned food bioassay with the concentrations of 0.05, 0.06, 0.07, 0.08, 0.09 and 0.1% with three replicates for each concentration. And the pathogenicity of isolated fungi was confirmed using Koch's postulates. Radial growth of fungal mycelium, growth rate, percentage mycelium inhibition, fungi static and fungicidal effects were tested. Measurements were taken for a period of one week. Control samples achieved the full mycelial growth at second day. Fungal growth was completely inhibited at the concentrations of 0.08, 0.09 and 0.1% throughout the experimental period. Significantly low ( $P < 0.05$ ) radial growth was observed for 0.05, 0.06 and 0.07% compared to control. However, no significant differences ( $P > 0.05$ ) were observed among 0.05, 0.06 and 0.07% treatments. Significantly lower growth rates were observed in above treatments compared to control treatment. Highest growth rate was observed in control treatment as  $40.16 \pm 0.28$  mm/day. Growth rate was below 20 mm/day in other treatments even at end of the experimental period which indicate even at lower concentrations of cinnamon bark oil has growth inhibition effect on *Lasiodiplodia* sp. Percentage mycelium inhibition observed as  $89.06 \pm 3.2$ ,  $95.68 \pm 2.0$  and  $96.86 \pm 5.4$ % for the concentrations of 0.05, 0.06 and 0.07% respectively. However, with the time, inhibition ability was decreased and at 7<sup>th</sup> day it was zero for the concentrations of 0.05 and 0.06%. Concentrations of 0.08, 0.09 and 0.1% showed fungicidal effect on *Lasiodiplodia* sp. under in-vitro conditions where other treatments only retarded the mycelial growth. Therefore, it can be concluded that, cinnamon bark oil could reduce the growth of avocado stem end rot causing *Lasiodiplodia* sp. under *in-vitro* conditions.

**Keywords:** anti-fungal properties, essential oils, plant extracts, postharvest diseases

## Characterization of powdery mildew pathogens of pumpkins and screening for resistant varieties

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Powdery mildew infection is frequently reported wherever pumpkins are grown in Sri Lanka. Two powdery mildew pathogen species, *Podosphaera xanthii* and *Golovinomyces cichoracearum* have been reported to cause the disease on pumpkins around the world. The identification of the exact pathogen species causing the disease is very important for developing resistant varieties. Objectives of the present study was to determine the exact pathogen species causing powdery mildew in Sri Lanka and to screen popular and commercial pumpkin varieties for the resistance to identify the resistant varieties. Powdery mildew infected pumpkin samples were collected from Makandura Agriculture department green house and Sabaragamuwa University greenhouse areas. Conidia of the pathogen were collected and inoculated to seven weeks old leaves of 10 varieties of pumpkin plants grown in the greenhouse of the Sandalanka Central collage, Giriulla. Koch's postulates were completed confirming that the pathogen is indeed causing the disease. Adaxial leaf surfaces were initially had small white color mycelia and later the entire leaf was covered with white powdery mycelia and conidia making white to brown and irregular colonies. Pathogen identification was conducted based on morphological characterization. The conidial length was in the range of 27 – 36  $\mu\text{m}$  (average = 32.23  $\mu\text{m}$ ) and the width was in the range of 12.88 - 18.93  $\mu\text{m}$  (average =16.0  $\mu\text{m}$ ). The length: width ratio was 1.72 - 2.35 (average =2.02). The foot cell dimensions length was in the range of 34-48  $\mu\text{m}$  and the width was in the range of 7.5-12  $\mu\text{m}$ . Clear fibrosin bodies were observed in conidia. Based on these characteristics especially the presence of fibrosin bodies in conidia, the powdery mildew pathogen was identified as *P. xanthii*. Seeds of popular commercial pumpkin varieties (n = add number of varieties) were planted in the greenhouse and in the field with three replicates. After 5days from the inoculation, adaxial surface of fully expanded first leaf was inoculated with *P. xanthii* conidia using a paint brush. Non inoculated controls were kept aside. The disease progress was monitored and percent leaf area infected and disease severity was estimated using the Townsend–Heuberger formula. Level of powdery mildew resistance was rated based on its Percent Disease Severity Index (DSI). Tukey's test was performed to compare the differences among treatment means. In the present experiment it was found that pumpkin variety "Mora" variety showed zero disease severity index indicating that the variety "Mora" was highly resistant to the pathogen. Arjuna and Krishna varieties were highly susceptible and all the other varieties belonged to the susceptible category. It can be concluded that the pathogen causing powdery mildew on pumpkin in the Giriulla area is *P. xanthii* and the variety "Mora" can be used in future breeding programs.

**Keywords:** *Podosphaera xanthii*, Powdery mildew, resistant varieties

## Identification of the fungal pathogen causing leaf blight of *Cinnamomum zeylanicum* Blume and *in vitro* screening of Tebuconazole sensitivity

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Leaf Blight Disease (LBD) is a major nursery disease of cinnamon (*Cinnamomum zeylanicum* Blume) and the disease causes serious foliar damage in seedlings, especially under high humidity and shade conditions. It also causes considerable damage to mature cinnamon bushes through leaf necrosis, defoliation and dieback of newly emerging flush. However, not much research has been conducted on LBD in Sri Lanka despite cinnamon being one of the key export crops. Therefore, the main objectives of the present study were to identify the causative agent of the LBD in Sri Lanka and to determine the minimum inhibitory concentration of Tebuconazole, one of the commonly used fungicides, to mitigate the pathogen growth *in-vitro*. LBD symptomatic leaves were collected from cinnamon fields in the Matara district representing 4 categories; tender leaves with initial LBD spots, tender leaves with regular lesions, mature leaves with regular lesions, and mature leaves with irregular lesions. Infected tissues from each category were cultured on PDA and percentages of emerging fungal colonies were determined. Pure cultures of all the fungal isolates were tentatively identified using morphological traits up to the genus level. From the cultures of tender leaves, *Rhizoctonia* and *Colletotrichum*-like isolates frequently appeared in 60% and 33.3% respectively. However, in the cultures of mature leaves, *Pestalotiopsis*-like isolates were detected more frequently (22 – 35%). To determine the exact causal agent, Koch's postulates were performed using representative isolates from each genus on detached tender cinnamon leaves. Actively growing mycelial plugs were inoculated on one side of the damaged leaf and the other side was inoculated with plain PDA plugs. Leaves were incubated in a moist chamber for 03 days and symptom development was observed. Typical LBD symptoms were developed when mycelial plugs of *Colletotrichum*-like species were introduced. *Rhizoctonia* and *Pestalotiopsis*-like species produced brown, circular or irregular lesions while *Colletotrichum*-like species showed a unique concentric ring pattern which is similar to the original symptoms of LBD. The potential pathogen species causing LBD was confirmed to be a *Colletotrichum*-like species. For species identification DNA of two *Colletotrichum*-like fungal isolates was extracted and ITS gene sequencing confirmed that the two isolates were *C. gloeosporioides* and *C. horii* with 100% similarity to the vouchered accessions of the GenBank. *In vitro* fungicide, sensitivity assay was conducted using a concentration gradient of the fungicide Tebuconazole. It was found that Tebuconazole could mitigate 100% growth of both *Colletotrichum* spp. at 5 ppm concentration *in-vitro*.

**Keywords:** cinnamon, Koch's postulates, *Colletotrichum*, *Rhizoctonia*, *Pestalotiopsis*

# **Abstracts**

## **Technical Session II**

## First report of *Curvularia dactyloctenicola* causing leaf spots of *Zea mays* and *Sorghum* sp. in Sri Lanka

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The dematiaceous hyphomycetous genus *Curvularia*, belongs to the family Pleosporaceae (Pleosporales, Dothideomycetes) comprised of endophytes, saprophytes and pathogens on both poaceous and non-poaceous hosts. Graminicolous *Curvularia* species were known to cause severe diseases on economically important crops worldwide. *Curvularia dactyloctenicola* was first reported on *Dactyloctenium aegyptium* in Thailand causing oval or fusiform, brown to brown-red leaf spots often surrounded by yellow halos. Although twelve (12) *Curvularia* species have been identified so far from Sri Lanka, *C. dactyloctenicola* has never been reported from the country. During the current study, diseased leaf samples of *Zea mays* and *Sorghum* sp. (traditional indigenous cereal landrace in Sri Lanka- "Swayanjatha") were collected from Galle, Hambanthota and Matale districts. Fungi were isolated using the single spore isolation technique. Colony characters were observed on three different culture media: Potato Dextrose Agar (PDA), Corn Meal Agar (CMA) and Malt Extract Agar (MEA). Fungal isolates were characterized based on micro-morphological and molecular data. The nuclear ribosomal internal transcribed spacers 1 and 2 with 5.8S region (ITS), glyceraldehyde-3-phosphate dehydrogenase (GAPDH) and translation elongation factor 1- $\alpha$  (TEF1) loci were sequenced from the fungal isolates and used in the multi-locus phylogeny. Phylogenetic analyses were performed based on maximum parsimony and maximum likelihood criteria. Three isolates from *Z. mays* and a single isolate from *Sorghum* sp. were identified as *C. dactyloctenicola* based on the morphological characters and resulted phylogram. Pathogenicity of fungal isolates was confirmed following the Koch's postulates. Characteristic leaf spots were developed in five days of inoculation on test plants and non-inoculated control plants were asymptomatic. The same fungus was recovered from inoculated plants confirming Koch's postulates. *Curvularia dactyloctenicola* has been reported as a pathogen of *Saccharum officinarum* in China following the first report from Thailand. To our knowledge, this is the first report of *C. dactyloctenicola* on *Z. mays* and *Sorghum* sp. (traditional – "Swayanjatha"). Thus, the current study updates the host-fungal association records of the species.

**Keywords:** dematiaceous hyphomycetes, pathogen, phylogeny, Poaceae

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## A case report on the blast disease outbreak in 'Maha' season 2021 in rice

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*Pyricularia oryzae*, the causative organism of the blast disease, is one of the most destructive pathogens in rice. Rice blast causes 30% annual yield loss all over the world. A severe blast disease outbreak was reported from 6 districts in Sri Lanka during the 'Maha' season 2021. Rice fields, located in the affected districts, were visited and field observations were recorded. Disease samples were collected for further analysis. Standard protocols were used in assessment of disease severity and, isolation and identification of the pathogen. From seventeen farmers interviewed, only four had previous experience in severe blast disease incidence, despite most of them had years of experience in rice farming. This indicates infrequent incidence of blast disease outbreaks in the past. All farmers reported abrupt changes of weather conditions during the panicle emergence stage of the cultivars, whereby gloomy weather resulting low temperature (17-28 °C) and high humidity (60-85 %) that coincided the outbreak. Affected cultivars were BG 359 (Kandy), BG 300, "Attakkari" (Kilinochchi, Anuradhapura), BG 1/94 (Moneragala, Ampara, Kandy), BW 367 (Gampaha) and BG 357 (Ampara). The initial symptom was panicles with white unfilled grains ("Sudu Karal") that appeared randomly in the fields. The symptoms spread and fields were completely affected within 48 h after onset of symptoms, resulting in 100% yield loss. Black-grayish to brown, dry and 2-5 mm long lesions were observed at the base of the infected panicles. Low to moderate level of blast symptoms on leaf and sheath were observed in BW 367 and BG 359 however, 'neck' was the most susceptible to the blast pathogen. Eight of the farmers out of the 17 interviewed applied systemic fungicides at the initial symptomatic stage but none of them were able to recover the crop indicating that curative application of fungicides was not effective in disease controlling. Fourteen pathogen isolates were obtained from leaf, leaf collar and panicle showing typical blast symptoms and the isolates were morphologically characterized. All cultures had white to gray, fluffy, raised colonies with a significant variation in pigment production on oatmeal agar. Upon florescent light treatment, blast isolates sporulated with hyaline, bottle shape, 3-celled, conidia. These strains will be further characterized using molecular markers. Since neck blast causes high yield losses, and all cultivars studied were highly susceptible, there is an urgent need for screening protocols and development of resistant germplasm for neck blast.

**Keywords:** cultivars, neck blast, *Pyricularia oryzae*, rice

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## Characterization of freshwater lignicolous ascomycetes collected from Kelani River

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Lignicolous freshwater fungi are important in biological diversity and functioning of freshwater ecosystems. Among them, freshwater ascomycetes occurring on decaying wood and other herbaceous materials collected from aquatic ecosystems are well known for their hyper diversity and potential applications. The discovery of fungal species or strains with higher level of lignocellulolytic enzymes are important in industries including animal feed, pulp, paper, textile, detergent production, biofuels and other sectors. The main objective of this study was to identify fungi and evaluate their lignocellulolytic activities from selected freshwater ecosystems of Kelani River, Sri Lanka. Soft decorticated submerged wood samples were randomly collected from Kelani River. Single spore isolation was carried out to obtain pure fungal cultures. Qualitative analysis of enzyme activities was carried out using Carboxymethyl cellulose agar (CMC), Tannic acid agar (TAA) and 2, 2-azino-bis 3-ethylbenzthiazoline-6-sulphonic acid (ABTS) agar plate assays. The standard fungal barcode, nuclear ribosomal internal transcribed spacers 1 and 2 including the 5.8S region (ITS) was sequenced for nine (09) lignocellulolytic fungal isolates. Phylogenetic analyses were performed to confirm the phylogenetic placement of the species and six (06) fungi were identified up to species level. Results confirmed that, out of the fifteen (15) isolates; eleven (11), five (5) and fourteen (14) isolates were positive for cellulases, overall polyphenol-oxidases and laccases activities respectively. This study reveals the high biodiversity and bio-prospective potential of lignicolous fungi associated with freshwater ecosystems in Sri Lanka which require extended studies in future.

**Keywords:** cellulases, laccases, lignocellulolytic, polyphenol-oxidases, submerged wood,

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## **Cultural, micro-morphological and pathogenic variability among *Curvularia* isolates from *Zinnia elegans***

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Spot disease caused by *Curvularia* sp. makes considerable damage to leaves and flowers of zinnias (*Zinnia elegans*, Family Asteraceae) affecting the aesthetic value of the plant. Diseased samples were collected from nursery-grown zinnia plants from three randomly selected localities in the country. Four *Curvularia* sp. isolates designated as CAp (locality: Anuradhapura), CGd, CWj (from two sites in Peradeniya) and CMw (from Mawanella) were obtained from diseased zinnia leaves/ray florets. These isolates were characterized for cultural, micro-morphological and pathogenic variations. The fungal colonies on PDA were powdery to velvety with light brown to dark brown pigmentation and formed concentric zonation with time. By 6 days of incubation at 30 °C, the colony diameter of isolates varied from 7.8 cm (CAp) to 8.7 cm (CWj). Mycelia were branched, septate hyaline to dark brown. Conidiophores arising singly or in groups were septate, branched, pale brown to dark brown and length up to 250 µm. Conidia which arose singly or as clusters on apex of the conidiophores were 2-4 septate with 3-5 cells, straight to curved, mostly ellipsoidal, sometimes clavate, length 15 µm to 28 µm, pale brown to dark brown, with flat and dark hila and all isolates showed both monopolar and bipolar germination. Degree of pathogenicity among isolates gave inconsistent results on the host plant. When spot-inoculated on lower surface of unwounded zinnia leaf explants, the lesion diameters exhibited by isolates CMw, CGd, CAp and CWj were 1.2 cm, 1.1 cm, 0.8 cm and 0.4 cm, respectively, at 6 days after inoculation whereas smaller lesion diameters were shown by all four isolates when inoculated on the leaf upper surface. On potted plants spray-inoculated with conidial suspensions, the diseased areas shown by isolates after 6 days were 16.6% (CMw), 36.6% (CGd), 11.0% (CAp) and 25.0% (CWj). Based on the molecular characterization, isolate CWj was identified as *Curvularia senegalensis*. The molecular characterization of the other three isolates will lead to a better understanding about the species of *Curvularia* damaging zinnias in Sri Lanka.

**Keywords:** leaf spot, morphology, pathogenicity

## **Phylogenetic relationships of phytoplasma strains associated with major phytoplasma diseases in Sri Lanka with their close relatives based on 16S rRNA sequences**

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Sugarcane White Leaf Disease (SCWL) and Weligama Coconut Leaf Wilt Disease (WCLWD) are two devastating phytoplasma diseases prevailing in Sri Lanka. Though host plants have belonged to different families, respective phytoplasmas species have been taxonomically classified under the 16SrXI phytoplasma group. Many phytoplasma diseases infecting sugarcane and coconut that show similar symptoms have been reported in Asian and African countries. Therefore, the present study was done to find out the identity, subgroups, and phylogenetic relationship of the 16S rRNA gene of SCWL and WCLWC phytoplasmas and their close relative phytoplasmas diseases reported around the world. Fourteen phytoplasma isolates 16S rRNA gene sequences ranging from 857bp to 1554bp including eight SCWL and sugarcane grassy shoot (SCGS) sequences, one WCLWD sequence, three different coconut phytoplasma sequences, and two different grass phytoplasma sequences reported in Sri Lanka were retrieved from NCBI-GenBank database. The sequence identity was estimated by the Clustal Omega program to determine the similarities. The phylogenetic tree was constructed with 1000 bootstrap replicates by using the Neighbor-Joining method of MEGA application. The WCLWD phytoplasma shared 97.1% sequence identity with SCWL all Sri Lankan, China, and Thailand isolates which belong to the 16SrXI-B subgroup. It was the highest percentage identity among WCLWD phytoplasma and SCWL phytoplasmas that suggested two phytoplasma species should belong to different phytoplasma subgroups as per the IRCPM classification guideline. The WCLWD phytoplasma shared 96.99%, 91.84%, and 91.72% respective sequence identities with coconut root wilt, St.Paul wilt, and Awka wilt phytoplasma isolates belonged to 16SrXIV, 16SrXXII-B and 16SrXXII-A group and subgroups. Results confirmed that WCLWD phytoplasma is distantly related to other coconut phytoplasma diseases reported in the world. Further, WCLWD shared 95.67% identity with two grass phytoplasma isolates in Sri Lanka that belong to the 16SrXIV-A subgroup. All SCWL and SCGS isolates were clustered with WCLWD phytoplasma and coconut root wilt phytoplasma in the same clade of the phylogenetic tree which implied a high level of evolutionary relationship among those phytoplasmas. All the other coconuts and grass phytoplasma isolates were clustered in two separate clades in the phylogenetic tree. The present study confirmed, that two phytoplasma diseases prevailing in Sri Lanka have a close identity and phylogenetic relationship based on the 16S rRNA gene. Further studies based on the sequence identity of conserved loci of the phytoplasma genome are needed to determine how both phytoplasma and their relatives divergent from each other.

**Keywords:** phylogenetic relationships, phytoplasma, sequence identity, Sugarcane white leaf disease, Weligama coconut leaf wilt disease, 16S rRNA gene

## Isolation of a potential rock phosphate solubilizing *Aspergillus* sp. towards the development of biofertilizer amendment

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Phosphorus (P) is one of the key bio elements that limits agricultural production. Although Sri Lanka is endowed with rock phosphate deposits, practical means to utilize this resource are limited. Phosphate-solubilizing fungi (PSF) play an essential role in increasing the bioavailability of phosphorus in soil for plants. In this study microorganisms which were isolated from eight soil samples in Kaikawala area, Matale District, Central province, Sri Lanka by spread plate method were further analyzed for its phosphate solubilizing capability. One fungal isolate was identified as phosphate solubilizing fungus based on the extent of halo zones present on Pikovskaya's agar (PVK) plates containing 0.5% tricalcium phosphate as an insoluble phosphorus source. This isolate was further purified and transferred into three media combinations with uniform conditions except for, PVK medium with 0.5% apatite from the Eppawala Rock Phosphate (ERP) as the phosphate source, PVK agar with 0.5% tricalcium phosphate as the phosphate source of the positive control, and PVK agar without any P source as the negative control. DNA sequences of the internal transcribed spacers with 5.8S region of ribosomal DNA (ITS1-5.8S-ITS2) was analyzed with universal primer pair ITS4-F/ITS5-R to determine the identity of the species. Fungal isolate PSF01 showed a phosphate solubilizing activity on the PVK medium with ERP having the solubilization index (SI) of  $1.07 \pm 0.03$  cm whereas that of on PVK medium with tricalcium phosphate was  $1.07 \pm 0.01$  cm thus not showing a significant difference between the two ( $p > 0.05$ ). However, the SI in the negative control without the phosphorous source was  $1.00 \pm 0.00$  cm and it did not form a halo zone compared to the media containing phosphorus ( $p < 0.05$ ). Fungal strain is fast growing with initially white but quickly changing to black colonies after producing conidial spores on potato dextrose agar with distinctive conidial heads and pale-yellow lower surface characterized as *Aspergillus*. Sanger DNA sequencing identified the fungus as *Aspergillus niger* (99%). Consequently, our preliminary study indicates the importance of screening more soil samples to identify PSF for sustainable agricultural applications in the future.

**Keywords:** *Aspergillus niger*, biofertilizer, phosphorous solubilization

## **Biocontrol potential of two Bacilli isolates against sheath blight of rice caused by *Rhizoctonia solani***

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Rice (*Oryza sativa* L.) is an essential food crop grown worldwide that feeds more than half of the world population. Rice sheath blight (Causal organism: *Rhizoctonia solani* Kühn) is one of the three major diseases in rice, which accounts for considerable yield losses throughout the rice growing areas of the world. No sheath blight resistant commercial varieties of rice have been reported so far. Agrochemicals are mainly used to control rice sheath blight. Apart from the usage of agrochemicals, alternative approaches for disease control such as bio control agents is being practiced by some countries around the world. *R. solani* Kühn was isolated from sheath blight infected Bg 379-2 variety and a pure culture was obtained. Eleven different bacterial endophytes were isolated from a rice plant of variety Bg 409 and cultured on Nutrient Agar (NA) medium. Bacterial isolates were tested *in-vitro* to determine their antagonistic activity against *R. solani* using dual culture plate technique. From these two endophytic bacteria were selected based on antagonistic ability. Those two bacterial isolates were named as Bg B01 and Bg B02, and the former was found to be the most effective antagonistic bacterial isolate against *R. solani* Kühn. The *in-vitro* results showed, they were bacilli according to microscopic observations. Bg B01 isolate was a catalase positive, gram negative bacterium. The percentage growth inhibition of *R. solani* Kühn was found to be 30.28 % by the second day of dual culturing and observed inhibition until 8<sup>th</sup> day (28.89%). The pathogen produced a light pink color pigment with Bg B01 isolate whereas Bg B02 isolate did not produce such a pigment. Bg B02 isolate was found to be a catalase negative, gram positive bacterium and percentage growth inhibition of *R. solani* Kühn was 24.81 % on the second day of culturing. However, further field experiments are needed.

**Keywords:** antagonism, biocontrol, endophytic bacteria, Rice sheath blight

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## Isolation, identification and development of *Trichoderma* as a biocontrol agent against tea root pathogens

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Biopesticides are attracting global attention as a safer strategy to manage diseases because of its advantages associated with the environmental safety, target-specificity, efficacy, biodegradability and suitability in the integrated disease management (IDM) programs. This study was aimed to isolate, identify, characterize, and evaluate effective *Trichoderma* spp. against primary root pathogens of tea *Poria hypolateritia* (red root rot), *Rosellinia arcuata* (black root rot), and *Phellinus noxius* (brown root rot). Soil was collected from 10-20 cm depth in a tea field from 10 different locations covering different tea growing regions and 10 g of soil from each location was plated on Dicloro Rose Bengal Chloramphenicol medium (DRBC). A total of 149 *Trichoderma* isolates were obtained. Among them 72 isolates showed antagonistic activity against all the three root pathogens in dual cultures. Based on higher Percentage Inhibition of Radial Growth PIRG% (>75%) and spore count (>107/mL water), three isolates were selected for further evaluations. The growth of the selected isolates was evaluated at different temperature and pH ranges. Higher growth of the isolates was observed at temperature 25 °C to 28 °C and pH at 4.5 to 5.0. Compatibility of the isolates to common agrochemicals used in tea, Dolomite, CuO, Hexaconazole (EC) and Glyphosate, was tested in vitro. Dolomite and Glyphosate did not significantly affect growth of *Trichoderma* while CuO and Hexaconazole (EC) suppressed the growth of *Trichoderma* isolates. The selected isolates are potential biocontrol agents that can be combined into IDM approaches of root pathogens of tea.

**Keywords:** Biopesticides, Black root rot, Brown root rot, IDM, Red root rot

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