CANADIAN PHYTOPATHOLOGICAL SOCIETY

ATLANTIC REGION MEETING 2024

KENTVILLE RESEARCH AND DEVELOPMENT CENTRE,
AGRICULTURE AND AGRI-FOOD CANADA,
32 MAIN STREET, KENTVILLE, NS

Atlantic Region Meeting
November 21, 2024
Kentville Research and Development Centre, Kentville, NS

INTRODUCTION

Welcome to Kentville and to the 2024 Canadian Phytopathological Society (CPS) Atlantic Region Meeting. It is our pleasure to host this year's CPS regional meeting, and we are looking forward to a stimulating afternoon of scientific discussion and fellowship.

There are nine oral presentations and two poster presentations on various topics in phytopathology scheduled for the afternoon. A dinner will take place at Old Orchard Inn, 153 Greenwich Rd South, Exit 11, Highway 101, Wolfville NS following the meeting.

We are pleased to welcome **Dr. Shuanglong Huang,** Department of Agriculture, Province of Prince Edward Island as our keynote speaker. Dr. Shuanglong Huang, is a new scientist at PEI Department of Agriculture and his presentation is entitled: From west to east: advancing crop health through collaborative research.

Many thanks to all who are attending the meeting. We trust we will be able to get together on a regular basis in future, and on that note, plans are developing for Linda Jewell and Dawn Bignell to host the 2025 meetings in Newfoundland and Labrador.

This booklet contains abstracts of oral and poster presentations in the order that they will be presented. All abstracts will subsequently be published in an upcoming edition of the Canadian Journal of Plant Pathology. The research work represented by these papers forms an important part of the on-going development of phytopathological research in Atlantic Canada.

Rick Peters, Atlantic CPS Rep
Shawkat Ali, Chair, Local Arrangements Committee

The Canadian Phytopathological Society Atlantic Region Meeting 2024

Thursday, 21 November 2024
Apple Orchard Room, KRDC, Agriculture and Agri-Food Canada, Kentville, NS

13:00-13:10 **Introduction/Welcome:**
Dr. Rick D. Peters, CPS representative – Atlantic region
Geoff Mercer, Director, Research, Development and Technology, Kentville Research and Development Centre, Kentville, NS

Session A: Host-Microbe Interactions
Moderator: Shawkat Ali, AAFC-KRDC

13:10-13:40 **Keynote Speaker: Dr. Shuanglong Huang. From west to east: advancing crop health through collaborative research.** *Department of Agriculture, Province of Prince Edward Island.*

13:40-14:00 **Microbial community composition in the stems and rhizosphere of potato plants displaying early dying syndrome. Tudor Borza,** *Department of Plant, Food, and Environmental Sciences, Dalhousie University, Truro, NS.*

14:00-14:20 **Exploring the Bacterial Communities of 'Honeycrisp' Apples in Atlantic Canada. Shayne McLaughlin**, *Kentville Research and Development Centre, Kentville, NS.*

14:20-14:40 **Deep sequencing and transcriptomics analysis of potato wart. Xian (Sean) Li,** *Canadian Food Inspection Agency, Charlottetown Laboratory, PE.*

14:40-15:00 **Apple (*Malus domestica*) rootstock selection of microorganisms induces differential microbiome architecture resulting in a disease suppressive state. Xavier Godin**, *Kentville Research and Development Centre, Kentville, NS.*

15:00-15:30 Nutrition Break/Poster Presentations

Posters: **Effects of fumigation and crop rotation on soil microbial diversity and *Verticillium* spp. abundance in fields with intensive potato cultivation. Tudor Borza,** *Department of Plant, Food, and Environmental Sciences, Dalhousie University, Truro, NS.*

Assessing the viability of potato wart (*Synchytrium endobioticum*) sporangia using Fluorescent In-Situ Hybridization (FISH) techniques. Qifan (Evelyn) Yang, *Canadian Food Inspection Agency, Charlottetown Laboratory, PE.*

Session B: **Pathogen Detection and Alternative Disease Management**
Moderator: Rhea Amor Lumactud, Dalhousie University

15:30-15:50 **Screening for resistance to powdery mildew in *Cannabis sativa*. David Joly,** *Université de Moncton, Moncton, NB.*

15:50-16:10 **Potential application of chemical and rRNA probes for assessing the viability of potato wart (*Synchytrium endobioticum*) sporangia**. **Jiacheng (Eric) Chuan,** *Canadian Food Inspection Agency, Charlottetown Laboratory, PE.*

16:10 -16:30 **Development and validation of approaches for the detection, propagation, and synthesis of Hop Latent Viroid: toward the development of a biotechnological tool for functional genomics in cannabis. Y. Moutahir**, *Université de Moncton, Moncton, NB.*

16:30 -16:50 **Uncovering the diversity of *Fusarium oxysporum* formae speciales affecting *Cannabis sativa*. L. Roy,** *Université de Moncton, Moncton, NB.*

16:50-17:00 **Closing remarks:** Rick Peters

17:30-20:00 **Dinner:** Old Orchard Inn, 153 Greenwich Rd South, Exit 11, Highway 101

Note to presenters: please ensure that your presentation is given to the audio/visual coordinator prior to the start of the meeting.

From west to east: advancing crop health through collaborative research. S. HUANG, W. G. D. FERNANDO, G. PENG, AND S. LIU. *Department of Agriculture, Province of Prince Edward Island, 5th Floor, Jones Building, 11 Kent Street, P.O. Box 2000, Charlottetown, PE C1A 7N8 Canada; (W.G.D.F.) Department of Plant Science, University of Manitoba, Winnipeg, MB R3T 2N2, Canada; and (G.P.) Agriculture and Agri-Food Canada, Saskatoon Research and Development Centre, Saskatoon, SK S7N 0X2, Canada*

Global climate change is increasingly reshaping agricultural ecosystems, leading to more unpredictable environmental conditions and heightened risks from crop diseases and pests, thereof imposing profound implications for crop health. The effort to protect crop health becomes more challenging especially for a country like Canada that is tremendously diverse in abiotic and biotic factors from coast to coast, and in some cases, this may require region-specific solutions. This diversity, however, also offers us valuable opportunities to share some unique stories with different audiences that might serve as a source of inspiration or as a piece of information transferable to new scenarios in one region or another. Towards this end, a few case studies with collaborative efforts focusing on major crops canola, soybean, corn and wheat diseases and abiotic stresses conducted in western Canada will be deliberated. Moving to the east specifically in Prince Edward Island (PEI), two prominent case studies with collaborative efforts accentuating the development of a pest prediction model and an integrated pest management (IPM) system tailored to the region's needs will also be discussed. Premised on collaborative efforts, a potential future research framework centering on economically significant crop diseases in PEI and the broader Atlantic region will be highlighted, aiming to enhance crop health and resilience in the face of climate change.

Microbial community composition in the stems and rhizosphere of potato plants displaying early dying syndrome. T. BORZA, T. AL-MUGHRABI, S.Y. SHIM, R. LACMATUD, K.I. AL-MUGHRABI AND B. PRITHIVIRAJ. *Department of Plant, Food and Environmental Sciences, Faculty of Agriculture, Dalhousie University, 50 Pictou Road, Cox Institute, Truro, NS B2N 5E3, Canada; and (S.Y.S, K.I.M) Department of Agriculture, Aquaculture and Fisheries, 39 Barker Lane, Wicklow, NB E7L 3S4, Canada*
Potato plants exhibiting Potato Early Dying disease (PED) undergo premature senescence, usually followed by a rapid demise. The main pathogens responsible for PED are *Verticillium dahliae, V. nonalfalfae* and *V. albo-atrum*. The more severe symptoms observed in PED, compared to the potato wilt caused by *Verticillium* species, are due to cumulative effects caused by the presence of other potato pathogens such as root-lesion nematodes (*Pratylenchus*), fungi like *Colletotrichum* and *Fusarium*, and soft-rot bacteria. To better understand the complex microbiome responsible for PED, soil found in the proximity of the root system of healthy plants and of diseased plants as well as stems from healthy and diseased plants were analyzed by quantitative PCR and by amplicon-targeted next generation sequencing (Illumina and PacBio). Samples were collected from four locations in New Brunswick, from fields that in the previous year had rotating crops, either barley or oat. Comparative analysis of the bacterial, fungal and eukaryotic diversity and abundance in soil samples from healthy and diseased plants revealed minimal differences; only bacterial alpha diversity was found to be influenced by the plant health status. *V. dahliae* was found to be abundant in the soil found in the proximity of both healthy plants and diseased plants. Though all stems from diseased and healthy plants were found to be infected by *V. dahliae*, the abundance of *V.dahliae* was found to be significantly higher in infected plants. Additionally, several other fungal species contributing to PED, including *Alternaria alternata*, *Colletotrichum coccodes* and *Plectosphaerella cucumerina*, were identified in stems.

Exploring the bacterial communities of 'Honeycrisp' apples in Atlantic Canada.

M.S. MCLAUGHLIN, S.N. YURGEL, P.A. ABBASI AND SHAWKAT ALI. *Department of Plant, Food, and Environmental Sciences, Faculty of Agriculture, Dalhousie University, Truro, NS, Canada; (P.A.A., S.A) Kentville Research and Development Centre, Agriculture and Agri-Food Canada, Kentville, NS, Canada; and (S.N.Y.) United States Department of Agriculture (USDA), Agricultural Research Service, Grain Legume Genetics and Physiology Research Unit, Prosser, WA, United States of America*

The maintenance and manipulation of the beneficial plant microbiome is seen as a new frontier in eco-friendly disease management. However, the fruit microbiome is highly variable and can be influenced by both biotic and abiotic factors. Thus, understanding how these factors influence microbial communities will be necessary in order to unlock the microbiome for sustainable disease management. We demonstrate that changes in external environmental conditions between growing seasons as well as growing sites significantly influence the structure and composition of the bacterial communities of 'Honeycrisp' apples from seven different orchards in the Atlantic Maritime Ecozone, and demonstrate significant changes in microbial communities due to organic and conventional management practices and compare the relative impact of these three factors on the communities of both core and peel tissues. While the bacterial communities of peel tissues were highly variable between growing seasons, core bacterial communities were relatively stable, underscoring the need to include multiple tissue-types in future fruit microbiome research. Nineteen bacterial genera were identified as CORE bacterial genera of the ‘Honeycrisp’ apple microbiome, appearing in over 75% of all samples. Finally, we describe the core microbiome of the describe microbial intrakingdom co-cooperation networks of apple core and peel tissues, respectively, and discuss the key microbial taxa influencing these networks.

Deep sequencing and transcriptomics analysis of potato wart. X. (SEAN) LI, J. (ERIC) CHUAN, Q. (EVELYN) YANG, D.L. HAMMILL AND P. ROSS. *Canadian Food Inspection Agency, Charlottetown Laboratory, PE C1A 5T1, Canada*

Potato wart (*Synchytrium endobioticum*) is the most important quarantine pathogen of potato with sporadic occurrence in Latin America, Europe, North America, Asia, Africa and Oceania, facing worldwide regulatory actions after discovery. Its obligate biotrophic nature and other technical difficulties limit the progress on whole genome sequencing (WGS) of *S. endobioticum.* To date, ASM653595v1 is the best genome assembly uploaded to GenBank/NCBI which still contains as many as 786 scaffolds. The other two genome assemblies also contain fragmented and incomplete sequences (contigs). A recent study (van de Vossenberg et al., 2023) compared the WGS datasets for pathotype 18 originating from the Netherlands and pathotype 6 from PEI, Canada indicated that the comparative genomics study is still challenging due to the lack of sufficient sequence data and sequencing depth for *S. endobioticum*. Here we present some preliminary analysis of the datasets on deep and transcriptomic sequencing of potato wart. DNA and RNA were extracted from four warty tubers (ct Russet Burbank) infected by *S. endobioticum*, and one healthy RB tuber. Both DNA and RNA were sequenced using the Illumina NovaSeq Platform (Genome Quebec). An average of 1.38 (DNA) and 1.18 (RNA) billion raw sequences were obtained from sequencing libraries. Atria (v4.1.0) was used to trim the adapters sequences and remove sequencing duplications, resulting in an average of 363 (DNA) and 791 (RNA) million unduplicated clean sequence reads for further analysis. Using Salmon (v1.4.0) to quantify the clean data against the *S. endobioticum* transcriptome, we have identified 7219 core genes that expressed in all potato wart positive RNA samples, with further assembly and annotation are in progress.

Apple (*Malus domestica*) rootstock selection of microorganisms induces differential microbiome architecture resulting in a disease suppressive state. X. GODIN, S. YURGEL, V. LEVESQUE, K. FULLER, T.A. FORGE, R. LUMACTUD AND S. ALI. *Department of Plant, Food, and Environmental Sciences, Faculty of Agriculture, Dalhousie University, Truro, NS, Canada; (V.L.,K.F.,S.A.) Agriculture and Agri-Food Canada, Kentville Research and Development Centre, Kentville, NS, Canada; (S.Y.) United States Department of Agriculture (USDA), Agricultural Research Service, Grain Legume Genetics and Physiology Research Unit, Prosser, WA, United States of America; and (T.A.F.) Agriculture and Agri-Food Canada, Summerland Research and Development Centre, Summerland, BC, Canada*

In Nova Scotia, apple trees are considered a highly profitable crop. To meet consumer choice and to replace old apple trees, farmers have frequently had to update their existing orchards. During this renovation process, producers typically uproot the old trees and replant the new tree in the same tree row at the same sites as the previous orchards. This results in the young trees having to deal with the buildup of numerous pathogenic organisms remaining from the prior generation. This causes stunted growth in the early years of establishment and delays fruit bearings. There is increasing evidence that rootstocks from the Geneva series compared to the Malling series have a higher tolerance to the Apple Replant Disease complex (ARD). We hypothesised that this resistance is in part the result of a specific microbiome architecture induced by different recruitment strategies between rootstocks. To elucidate this we conducted an experiment in which rootstocks from the Geneva and Malling series were planted in old orchard soils with known ARD issue. Before and after the growing period, several plant growth parameters were measured. For microbiome analysis DNA was isolated from washed root samples and subjected to amplicon-targeted sequencing for bacteria and fungi. This allowed us to evaluate the microbial population and architecture in relation to plant performance. The Geneva series was found to perform better than the Malling series in ARD soil. This could in part be explained by the differential recruitment strategies that were observed between genotypes.

Screening for resistance to powdery mildew in *Cannabis sativa*. Y. MOUTAHIR, A. LANDRY, K. KJOLBRO AND D.L. JOLY. *Université de Moncton, 18 avenue Antonine-Maillet, Moncton, NB E1A 3E9, Canada*

Cannabis powdery mildew, primarily caused by *Golovinomyces ambrosiae*, poses a significant threat to *Cannabis sativa*, impacting yield, quality, and profitability. This study aimed to identify sources of resistance to powdery mildew by screening a diverse collection of over 50 cannabis cultivars under controlled inoculation conditions. Resistance-associated phenotypes were evaluated across cultivars, and highly resistant individuals were selected for molecular analysis, focusing on known susceptibility genes such as MLO1, to differentiate between resistance and loss-of-susceptibility mechanisms. Preliminary results identified several promising cultivars, including a subset for which resistance correlated with MLO1 gene inactivation. However, MLO1 inactivation did not consistently indicate resistance, highlighting variability in resistance/susceptibility mechanisms across genetic backgrounds. These cultivars provide valuable candidates for breeding programs aimed at enhancing powdery mildew resistance in cannabis. This research will contribute to sustainable cannabis cultivation by facilitating the selection of disease-resistant lines, reducing reliance on chemical controls, and supporting resilient crop production.

Potential application of chemical and rRNA probes for assessing the viability of potato wart (*Synchytrium endobioticum*) sporangia. J. CHUAN, Q. YANG, M. ANNETT, N. MATHESON, A. PROUD, M. ANTOUN AND X. LI. *Canadian Food Inspection Agency, Charlottetown Laboratory, PE C1A 5T1, Canada*

Potato wart is caused by *Synchytrium endobioticum*, an obligate biotrophic and soil-borne fungus that does not produce hyphae but instead forms winter sporangia, each containing 200-300 motile zoospores. Under suitable conditions in the spring, these overwintering resting sporangia germinate, releasing uninucleate zoospores that infect potato meristems, leading to altered metabolic processes. *S. endobioticum* can survive in soil for over 40 years in the form of resting sporangia, even without plant hosts, making it a significant quarantine pathogen for potatoes. This pathogen has sporadic occurrences in Latin America, Europe, North America, Asia, Africa, and Oceania, prompting worldwide regulatory actions upon discovery. The thick-walled winter sporangia are the dormant structures that facilitate the fungus's dispersal and establishment of new infections. These sporangia are generally spherical to ovoid, with thick walls adorned with irregularly shaped wing-like protrusions. The sporangium wall contains fatty acids and wax esters, which protect the viability of *S. endobioticum* in the soil. Determining the viability of potato wart sporangia after prolonged dormancy under environmental conditions is crucial, particularly for those sporangia that retain the ability to invade and replicate in susceptible potato varieties. This research proposes to identify effective solutions for testing the viability of resting sporangia using chemical and biological staining methods, including fluorescein diacetate (FDA), Propidium Iodide (PI), Trypan blue, rRNA fluorescent probe hybridization, and so on. Currently, we successfully observed *S. endobioticum*-specific rRNA probes binding only to viable sporangia under a fluorescent microscope. Those staining methods show a potential to determine sporangia viability effectively.

Development and validation of approaches for the detection, propagation, and synthesis of Hop Latent Viroid: toward the development of a biotechnological tool for functional genomics in cannabis. Y. MOUTAHIR AND D.L. JOLY. *Université de Moncton, 18 avenue Antonine-Maillet, Moncton, NB E1A 3E9, Canada*

Many viruses have emerged as valuable tools in plant functional genomics, providing a unique means to study gene function through virus-induced gene silencing. In Cannabis sativa, no efficient tool has been developed to the point of being adopted by the scientific community. This study focused on validating methodologies for the reliable detection, propagation, and synthesis of HLVd (Hop Latent Viroid), laying the groundwork for its use in cannabis biotechnology. We first compared existing detection protocols based on RT-qPCR, enabling the accurate identification of HLVd in diverse tissue types under varying conditions. Subsequently, we tested various inoculation methods to propagate, maintain, and amplify the viroid in Cannabis sativa, trying to ensure consistent infection rates. Finally, synthetic biology approaches were employed to produce in vitro HLVd RNA, allowing precise control over viroid concentration and which will facilitate future gene-silencing experiments. Validation of these methods demonstrated reproducible viroid detection and infection in cannabis plants, including using in vitro synthesized HLVd. This could provide a novel tool to explore gene function in Cannabis sativa, with potential applications in metabolic engineering, stress tolerance, and pathogen resistance.

Uncovering the diversity of *Fusarium oxysporum* formae speciales affecting *Cannabis sativa*. L. ROY AND D.L. JOLY. *Université de Moncton, 18 avenue Antonine-Maillet, Moncton, NB E1A 3E9, Canada*

The soil-borne fungus *Fusarium oxysporum* poses a serious threat to *Cannabis sativa*, leading to wilt, root rot, and stem necrosis in affected plants. However, despite its agricultural impact, little is known about the specific *F. oxysporum* formae speciales (ff. spp.)—groups of isolates that specialized to infect particular host plants—responsible for disease in cannabis. This study seeks to clarify the identity of various *F. oxysporum* strains isolated from cannabis by combining classical isolation methods with molecular and phylogenetic analysis. Using targeted sequencing of regions including the translation elongation factor 1-alpha (TEF1-α) and known SIX (Secreted in Xylem) effectors, we analyzed *F. oxysporum* isolates from cannabis, together with reference strains from other formae speciales. Phylogenetic reconstruction allowed us to group isolates into clusters and assess genetic relatedness to other formae speciales. Preliminary results revealed a polyphyletic assemblage of distinct lineages with pathogenicity toward cannabis, and two distinct effector profiles, suggesting multiple potential formae speciales. In the near future, pathogenicity tests will be conducted on a range of *C. sativa* cultivars and other species to evaluate host specificity. These findings will contribute to the accurate identification of *F. oxysporum* in cannabis, enabling more targeted management practices.

POSTER PRESENTATIONS

Effects of fumigation and crop rotation on soil microbial diversity and *Verticillium* spp. abundance in fields with intensive potato cultivation. T. BORZA, P.S. SHUKLA, T. AL-MUGHRABI, S.Y. SHIM, R. LACMATUD, K.I. AL-MUGHRABI AND B. PRITHIVIRAJ. *Department of Plant, Food and Environmental Sciences, Faculty of Agriculture, Dalhousie University, 50 Pictou Road, Cox Institute, Truro, NS B2N 5E3, Canada; and (S.Y.S, K.I.M) Department of Agriculture, Aquaculture and Fisheries, 39 Barker Lane, Wicklow, NB E7L 3S4, Canada*

Potato Early Dying disease (PED) significantly impacts potato yields worldwide, causing substantial economic losses. The primary pathogens responsible for PED are *Verticillium* species with *V. dahliae* being the most prevalent. Identifying effective soil management methods to control Verticillium is crucial for mitigating these losses. Soil samples from eight fields, previously rotated with barley and oat, half of which were fumigated, were analyzed using quantitative PCR and amplicon-targeted next generation sequencing (Illumina and PacBio NGS) across five time points over two growing seasons. Data indicated that fumigation had minimal impact on *V. dahliae* incidence and abundance early in the first growing season, with no significant differences observed later in the study. These findings suggest crop rotation and fumigation, either alone or in combination, are not very effective in controlling *V. dahliae.* NGS revealed that fumigation initially reduced bacterial alpha diversity, but the effect gradually diminished by the end of the growing season. Similar trends were observed for the alpha diversity of fungal and eukaryotic communities. The incidence of *V. dahliae, V. tricorpus* and *V. klebahnii* increased from May to September, especially in fumigated fields, where their initial incidence was lower. Notably, *V. albo-atrum* was detected only in the last time point of the first growing season. In addition to *Verticillium* taxa, other fungal species such as *Colletotrichum coccodes*, *Fusarium solani*, *F. graminearum* and *F. oxysporum*, *Botrytis* sp., *Alternaria alternata*, *Plectosphaerella cucumerina*, and *Gibellulopsis piscis* were found to be ubiquitous in soil samples, irrespective of the sampling time, crop rotation and fumigation.

Assessing the viability of potato wart (*Synchytrium endobioticum*) sporangia using Fluorescent In-Situ Hybridization (FISH) techniques. Q. (EVELYN) YANG, J. (ERIC) CHUAN, M. ANNETT, N. MATHESON, A. PROUD, M. ANTOUN AND X. (SEAN) LI.
Canadian Food Inspection Agency, Charlottetown Laboratory, PE C1A 5T1, Canada
Potato wart (*Synchytrium endobioticum*) reduces crop yield and poses an economic impact, hence listed as a federal quarantine pest in Canada. It is also listed as a selected agent in the US, and A1 quarantine pest in EPPO countries. *S. endobioticum* is a soil-borne fungus that causes host cells to proliferate and form cauliflower-like warty galls on potato tubers. Under stress conditions, *S. endobioticum* produce resting sporangia. Upon warm and moist conditions, each sporangium can release 200-300 overwintering zoospores into the soil. The zoospores infect host cells and produce summer sporangia. These summer sporangia then release more zoospores to infect neighboring cells. When the potato tuber decays, it releases resting spores into the surrounding soil. In adverse conditions, these resting spores can remain dormant but viable for decades, and will germinate when unknown causes stimulate them. Currently, a reverse transcriptase qPCR assay was developed for testing the viability of the resting spores (van de Vossenberg et al., 2023). Unfortunately, these methods have some challenges in regard to repeatability and reproducibility in different labs. It has been important to determine whether a potato wart spore is dead, alive, or alive but dormant. Fluorescence in-situ hybridization (FISH) is a molecular technique which has been used to determine the viability of bacterial and fungal pathogens. The objective of this study is to explore the feasibility to develop FISH techniques to assess the viability of PW resting sporangia. To date, preliminary experiment indicated that chitinase and esterase could effectively break open the thick, triple-layered sporangium cell wall. A fluorescently labeled oligonucleotide probe could enter the sporangium and bind to the target rRNA of *S. endobioticum*. Under a fluorescence microscope, the viable winter sporangia cracked open by the chitinase and esterase enzymes were visualized by binding rRNA with designed fluorescent-labelled probe.