



# 42<sup>ND</sup> ANNUAL MEETING OF THE PLANT PATHOLOGY SOCIETY OF ALBERTA November 4-5, 2021

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## Schedule of Events

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### Thursday, November 4<sup>th</sup> 2021

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9:00 am - 9:20 am Welcome by 2021 PPSA president Dr. Rudolph Fredua-Agyeman and introductions of participants

9:20 am - 9:45 am **Keynote Speaker:**  
Dr. Lone Buchwaldt, Research Scientist, AAFC, Saskatoon and President of Canada Phytopathological Society (CPS)  
“Development of resistance to *Sclerotinia sclerotiorum* in canola”

### **9:45 am - 10:15 am Session I: Technicians Oral Presentations**

Chair: Dr. Gary Peng, Research Scientist, AAFC, Saskatoon, Saskatchewan

9:45 am – 10:00 am: Chelsi Harvey (T1)  
“A genetic approach for improving wheat resilience to emerging stripe rust isolates using novel *R* gene sequences from *Thinopyrum intermedium*”

10:00 am - 10:15 am: Ryan Gourlie (S1/T2)  
“The pangenome of *Pyrenophora tritici-repentis* reveals high plasticity and the association of virulence factors with mobile elements”

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### **10:15 am - 10:30 am Break**

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### **Featured speaker:**

10:30 am - 10:45 am: Dr. Yoann Aigu, Postdoctoral Research Fellow, University of Alberta, Edmonton, Alberta  
“The occurrence and spread of clubroot in Alberta”

### **10:45 am – 12:00 pm Session II: Students Oral Presentations**

Chair: Dr. Reem Aboukhaddour, Research Scientist, AAFC, Lethbridge, Alberta

10:45 am - 11:00 am: Marla Roth (S2)  
“Identifying pH insensitive pathotypes of *Plasmodiophora brassicae*”



11:00 am – 11:15 am: Keisha Hollman (S3)

“The virulence of *Plasmodiophora brassicae* on canola with 2nd generation clubroot resistance”

11:15 am – 11:30 am: Zhiyu Yu (S4)

“Greenhouse and field evaluation of precipitated calcium carbonate for clubroot management”

11:30 am – 11:45 am: Emilee Storfie (S5)

“Identification and functional investigation into *Plasmodiophora brassicae* effector candidates during infection of *Brassica napus* genotypes”

11:45 am – 12:00 pm: Hui Liu (S6)

“Transcriptome analysis of rutabaga (*Brassica napus*) cultivars indicates glucosinolate-derived nitriles may play a role inducing plant defense against clubroot disease”

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**12:00 pm – 13:00 pm Lunch break**

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**13:00 pm – 13:45 pm Session III: Students Oral Presentations**

Chair: Autumn Barnes, Canola Council of Canada

13:00 pm – 13:15 pm: Ilakkiya Thirungnasambandam (S7)

“Validation of polyclonal antibodies against tan spot and fusarium head blight spore, fungal pathogens of wheat”

13:15 pm – 13:30 pm: Dilini Adihetty (S8)

“Screening of barley cultivars for resistance to *Bipolaris sorokiniana* isolates from the prairies”

13:30 pm – 13:45 pm: Yixiao Wang (S9)

“Understanding Verticillium stripe-blackleg interactions”



**13:45 pm – 3:00 pm Session IV: Student and Technician Poster presentations**

Chair: Dr. Kelly Turkington, Research Scientist, AAFC, Lacombe, Alberta

**Student Posters:**

1. Sijan Pandit (S12), R. Goyal, E. Schultz and S. Chatterton  
“Temporal analysis of *Aphanomyces* root rot development after zoospore inoculation of field pea”
2. Haitian Yu (S13), S.F. Hwang, R. Fredua-Agyeman and S.E. Strelkov  
“Effects of *Fusarium avenaceum* and *F. proliferatum* on seedling survival, growth, root rot severity and yield of canola”
3. Chunxiao Yang (S14), R. Fredua-Agyeman, S.F. Hwang, and S.E. Strelkov and L.Y. Gorim. “Determination of the optimum number of days to scan for root architectural traits”
4. Chunxiao Yang (S15), S.F. Hwang, R. Fredua-Agyeman and S.E. Strelkov  
“Optimization of *Fusarium oxysporum* inoculation methods for *Brassica napus*”
5. Claudia Escobar-Gil (S16), L. Galindo-Gonzalez, A. Akhavan and S.E. Strelkov  
“Transcriptomic profiling of the host-pathogen interaction in tan spot of wheat”

**Technician Posters:**

1. Albert J. Hannig (T3), S. Chatterton, M. Tetorya, Dilip Shah and Ravinder K. Goyal  
“A search for an effective control measure against root-rot in field pea: Effect of antimicrobial peptides on in vitro growth of *Aphanomyces euteiches*”
2. Mohamed Hafez (T4), T. Despins, K. Nakajima and R. Aboukhaddour  
“*Pyrenophora tritici-repentis* in Japan: first report on race structure and a novel ToxA haplotype”

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**3:00 pm – 4:00 pm**

**Judges meet to rank Nov. 4 student and technician oral presentations and posters**

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**NB: S = student**

**T = technician**



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**Friday, November 5<sup>th</sup> 2021**

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08:40 am – 08:45 am: Welcome by 2021 PPSA president Dr. Rudolph Fredua-Agyeman

**Featured speaker:**

8:45 am – 9:00 am: Dr. Kelly Turkington, Research Scientist, AAFC, Lacombe, Alberta  
“Fungicide timing for sclerotinia stem rot of canola”

**9:00 am – 9:30 am Session V: Students oral presentations**

Chair: Dr. Andrea Botero, Postdoctoral Fellow, University of Alberta, Edmonton

9:00 am – 9:15 am: Kyle Biscaglia-Horvat (S10)

“Effects of soil moisture and temperature on the pathogen dynamics of root rot complex in field pea”

9:15 am – 9:30 am: Longfei Wu (S11)

“Detection and validation of partial resistance to Aphanomyces root rot in field pea cultivar ‘00-2067’ via bulked segregant RNA-Seq (BSR-Seq)”

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**9:30 am – 10:30 am Break for participants**

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**9:30 am – 10:30 am Judges meet to select winning oral presentations and posters**

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**10:30 am – 11:00 am (Award Ceremony via zoom and group zoom pic)**

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**11:00 am – 12:30 pm Business meeting**

12:30 pm- Meeting Adjourns



## ORAL PAPER ABSTRACTS

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- T1 A genetic approach to improve wheat resilience to emerging stripe rust isolates using novel R gene sequences from *Thinopyrum intermedium*.** CHELSI HARVEY, G.T. ARAUJO, M. FRICK, A. LAROCHE, J.D. LAURIE, R.J. GRAF, & A. BILICHAK, *Lethbridge Research & Development Centre, Agriculture and Agri-Food Canada, 5403 1st Avenue South, Lethbridge, AB, T1J 4B1; (A.B.) Morden Research and Development Centre, Agriculture and Agri-Food Canada, 101 Rte 100 #100, Morden, MB R6M 1Y5*

Stripe rust disease caused by the pathogen *Puccinia striiformis* f.sp. *tritici* remains a major biotic stress to wheat crops worldwide. Susceptibility to this disease can lead to a vast reduction of grain quality and yield. The main source of resistance to this disease is the presence of expressed resistance (R) genes in wheat lines. With the continued emergence of new virulent stripe rust isolates, many R genes are already or will likely become ineffective. Modifying known R gene sequences offers the potential for use of previously defeated genes. This modification focuses primarily on the coiled-coil (CC) and leucine-rich repeat (LRR) domains characteristic of most known R genes in monocots, including wheat. An alternative to altering the DNA sequences of previously characterized R genes is introducing new R genes into wheat, thereby increasing the available genetic repertoire for infection response. Historically, wild species have provided disease R genes to transfer to wheat thus prompting us to explore the wild relative intermediate wheatgrass (*Thinopyrum intermedium*) as a source of these genes. We observed stripe rust response under field conditions and narrowed our selection to 15 target wheatgrass lines displaying effective resistance. These lines were analyzed to discover a set of putative R genes for further investigation. Here, we report the identification of novel R genes discovered by resistance gene enrichment sequencing of *Thinopyrum intermedium* lines. Additionally, we will describe two approaches to employ these sequences in wheat with the ultimate goal of increasing resistance to virulent stripe rust races.

- S1/T2 The pangenome of *Pyrenophora tritici-repentis* reveals high plasticity and the association of virulence factors with mobile elements.** RYAN GOURLIE<sup>1</sup>, M. MCDONALD<sup>2</sup>, M. HAFEZ<sup>1</sup>, R. ORTEGA-POLO<sup>1</sup>, S. STRELKOV<sup>3</sup>, AND R. ABOUKHADDOUR<sup>1\*</sup> *Agriculture and Agri-food Canada, LRDC, Cereal Pathology, 5403 1st Ave South, Lethbridge, AB, Canada, T1J 4B1, <sup>2</sup>School of Biosciences, University of Birmingham, Edgbaston, Birmingham, United Kingdom, B15 2TT, <sup>3</sup>Department of Agricultural, Food and Nutritional Science, 410 Agriculture/Forestry Centre, University of Alberta, Edmonton, AB T6G 2P5, Canada*

Pangenome analysis of 41 global *Pyrenophora tritici-repentis* (Died.) Drechs. (tan spot of wheat) isolates revealed a high accessory gene count with 57% of all genes were accessory genes (not present in all isolates). A clear distinction between pathogenic and non-pathogenic genomes was observed in size, gene contents and phylogenetic relationships. Over 69% of the core genes had known functional domains, while only 28% of accessory genes had known functions. The pangenome is open, with the total set of genes continually increasing as genomes are added. Comparison of long-read assemblies with reference genomes revealed major structural organizations with the virulence genes *ToxA* and *ToxB* being nested in large transposons. *ToxA* and its associated transposon *ToxhAt*, are nested within what seems to be a 143 kb crypton. Whereas *ToxB* is present as multi-copy within a separate 294 kb transposon. Our study highlights the role of mobile elements in driving the evolution of virulence in this pathogen.



**S2 Evaluation of the pH sensitivity of *Plasmodiophora brassicae* strains.** MARLA ROTH, VICTOR P. MANOLII, SHEAU-FANG HWANG AND STEPHEN E. STRELKOV. *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB T6G 2P5*

Clubroot, caused by *Plasmodiophora brassicae*, is a serious disease of canola (*Brassica napus*). Although disease development is favored in acidic soils, it is not clear whether there is differential sensitivity to pH among *P. brassicae* pathotypes. Clubroot-susceptible canola was grown in a potting medium/field soil mix inoculated with isolates representing pathotypes 3A, 3H, 3D, 5G or 5X of *P. brassicae* at pH 6.1 (no lime control) or pH 7.2 and pH 8.0 in replicated greenhouse trials. Clubroot severity was assessed after 6 weeks. No symptoms of clubroot were observed in non-inoculated controls, while severe clubroot developed at pH 6.1 regardless of pathotype. Preliminary results indicated that there was a significant decrease in clubroot severity for pathotype 5X at pH 7.2. In contrast, there was a moderate but significant increase in clubroot severity for pathotype 5G at this pH; the isolate representing this pathotype originated from a high pH (7.8) field. At pH 8.0, symptom development was very limited for most pathotypes, including pathotype 5G. In a subsequent experiment, pathotypes 3A, 3D and 5G were evaluated further at pH values of 7.0, 7.3, 7.6 and 7.9 (and in a no lime control). Severe clubroot developed in the control and pH 7.0 and 7.3 treatments with all pathotypes, while disease severity was significantly lower at pH 7.6 and 7.9. Pathotype 3A caused greater disease severity at pH 7.9 than did pathotypes 3D and 5G. These results suggest some variation in the sensitivity of *P. brassicae* stains to soil pH.

**S3 The virulence of *Plasmodiophora brassicae* on canola with ‘2<sup>nd</sup> generation’ clubroot resistance.** KEISHA .B. HOLLMAN, V.P. MANOLII, S.F. HWANG AND S.E. STRELKOV *Department of Agricultural, Food and Nutritional Sciences, University of Alberta, Edmonton, AB T6G2P5, Canada*

Clubroot, caused by *Plasmodiophora brassicae*, is a damaging soilborne disease of canola (*Brassica napus*) first identified on the Canadian Prairies in 2003. Clubroot resistant (CR) canola cultivars, carrying what is now known as ‘1<sup>st</sup> generation’ resistance, were introduced in 2009-10 and soon became the most effective and widely used clubroot management tool. Unfortunately, new pathotypes of *P. brassicae* have emerged that can overcome 1<sup>st</sup> generation resistance and are found in an increasing number of fields. In response, canola breeders have developed a new set of cultivars with so-called ‘2<sup>nd</sup> generation’ resistance. While the nature of this resistance is not in the public domain, and may differ among cultivars from different companies, it is believed to be distinct from 1<sup>st</sup> generation resistance. Studies are underway to characterize the virulence of *P. brassicae* populations on different hosts with 2<sup>nd</sup> generation resistance. Isolations of the pathogen were made from symptomatic 2<sup>nd</sup> generation CR canola crops identified in the field and tested for their virulence on a suite of seven commercial canola cultivars carrying 2<sup>nd</sup> generation resistance. These cultivars were also tested with selected *P. brassicae* pathotypes that break 1<sup>st</sup> generation resistance. Preliminary results indicate that about half of these cultivars developed moderate to severe clubroot when challenged with pathotypes that can overcome 1<sup>st</sup> generation resistance. This suggests that resistance-breaking strains of *P. brassicae* may be difficult to control solely via the deployment of resistant cultivars, and that an integrated strategy, including cultural and other approaches, is warranted.



- S4 Greenhouse and field evaluation of precipitated calcium carbonate for clubroot management**  
**ZHIYU YU, SHEAU-FANG HWANG AND STEPHEN E. STRELKOV.** *Department of Agricultural, Food and Nutritional Sciences, University of Alberta, Edmonton, AB, T6 G2P5.*

Clubroot, caused by *Plasmodiophora brassicae*, is a threat to canola (*Brassica napus* L.) production in western Canada. Pathotypes of *P. brassicae* have emerged in recent years that can overcome the resistance in most canola cultivars. The application of lime has long been used to manage clubroot in cruciferous vegetables and could be another tool for disease control in canola. Precipitated calcium carbonate (PCC), an environmental-friendly lime product derived from the beet, corn and sugarcane industries, was evaluated for its efficacy in clubroot management. Two canola hybrids, '45H31' (clubroot susceptible) and 'CS2000' (moderately resistant), were seeded 1 week after the application of PCC at 3, 6 or 9 t/ha at three field sites and in a greenhouse trial with plastic tubs. Untreated control (UTC) treatments did not receive any PCC. Significant reductions ( $p < 0.05$ ) in clubroot severity relative to the UTC were observed in all of the PCC-treated plots or tubs. In the greenhouse tub trial, an increasing rate of PCC resulted in decreasing disease severity in both cultivars, while plants in the PCC-treated tubs grew significantly taller and produced more biomass than the UTC. In the field, a higher PCC rate also resulted in lower disease severity and higher yields; at all three field sites, the lowest disease severities and highest yields were observed at the highest rate (9 t/ha). The results suggest that PCC is an effective lime product for clubroot control and could be included as part of an integrated management strategy for this disease in canola.

- S5 Identification and functional investigation of *Plasmodiophora brassicae* effector candidates during infection of *Brassica napus* by pathotype 3A.** **EMILEE STORFIE, L. GALINDO-GONZÁLEZ, Q. ZHOU, S.F. HWANG, AND S.E. STRELKOV.** *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, T6G 2P5, Canada*

Canola (*Brassica napus*) contributes \$26.7 billion annually to the national economy, with production concentrated in the prairies. Clubroot disease, caused by the obligate parasite *Plasmodiophora brassicae*, poses a significant threat to this crop. To mitigate this threat, clubroot resistant (CR) canola cultivars with strong resistance to many pathotypes are widely grown. In recent years, however, this resistance has been overcome in an increasing number of fields due to pathotype shifts in *P. brassicae* populations. Pathotype 3A is one of the predominant resistance-breaking pathotypes. Transcriptomic analyses were conducted on pathotype 3A infections of *B. napus* rutabaga cultivars 'Laurentian' (susceptible) and 'Wilhemsburger' (resistant) at 7, 14, and 21 days after infection. To validate these analyses, the abundance of 28 putative effector transcripts was assessed using NanoString technology and a positive correlation was found between the two datasets. Non-redundant transcripts of the predicted effectors were analyzed across timepoints and cultivars, with transcripts displaying low covariance being compared with a previous study evaluating another resistance-breaking pathotype, 5X. Two highly expressed putative effectors, *SPR01261.1* and *SPQ99289.1*, which were identified in both studies, were selected for functional analyses. *SPR01261.1* is predicted to encode a serine carboxypeptidase, whereas *SPQ99289.1* encodes an unknown protein with a kinase domain. Currently, investigations are underway to functionally assess each putative effector's signal peptide, and to determine each effector's protein function. The identification and characterization of these and other effectors will contribute to an improved understanding of the molecular interactions between *P. brassicae* and its *B. napus* host.



- S6 Transcriptome analysis of rutabaga (*Brassica napus*) cultivars indicates glucosinolate-derived nitriles may play a role in inducing plant defense against clubroot disease.** HUI LIU, Q.Q. ZHOU, S.E. STRELKOV, S.F. HWANG, AND J.A. OZGA. *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB T6G 2P5, Canada*

Clubroot, caused by the obligate biotrophic protist *Plasmodiophora brassicae*, is one of the most damaging diseases of the Brassicaceae. Glucosinolates (GSL) are a group of defense-related secondary metabolites found in species of this family, and many of their hydrolysis products, including isothiocyanates, thiocyanate, and nitriles, are implicated in plant defense processes against many pathogens and herbivores. We analyzed the GSL pathway in a database from a recently published study (Zhou et al., 2020 *Int J Mol Sci* 21, 8381) where the authors compared transcriptomic profiles of two rutabaga (*Brassica napus* subsp. *rapifera*) cultivars which showed resistant ('Wilhelmsburger') and susceptible ('Laurentian') responses to *P. brassicae* inoculation. The results indicated that several genes that lead to production of nitriles along the indolic GSL degradation pathway are more highly upregulated 7 days after pathogen inoculation in 'Wilhelmsburger' than in 'Laurentian'. Nitriles serve as defensive compounds against plant pathogens because of their toxic nature and are also proven to elicit defense response pathways in plants. These data suggests that GSL-derived nitriles may play a role in inducing enhanced plant defense against the clubroot pathogen in the rutabaga cultivar 'Wilhelmsburger'. Further research to understand the role of GSLs in defense against *P. brassicae* in cruciferous plants is warranted.

- S7 Validation of polyclonal antibodies against fungal pathogens of wheat: tan spot and fusarium head blight.** IIAKKIYA THIRUGNANASAMBANDAM<sup>1,2</sup>, T. VUCUREVICH<sup>2</sup>, T. SHELTON<sup>2</sup>, C. WATT<sup>2</sup>, N. KAV<sup>1</sup>, AND A. LAROCHE<sup>2</sup>, <sup>1</sup>*Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, T6G 2P5*, <sup>2</sup>*Agriculture and Agri-Food Canada, Lethbridge Research and Development Centre, Lethbridge, AB, T1J 4B1*

Wheat is a staple food crop across the world with 760 million tonnes consumed annually. In 2020, The Canadian Prairie provinces produced 35 million tonnes of it. Airborne fungal pathogens pose a serious threat to wheat growers all over the world including Canada. Tan spot (TS) caused by *Pyrenophora tritici-repentis* (Died.) Drechs and fusarium head blight (FHB) caused by *Fusarium graminearum* (Schwein) Petch are important wheat diseases that impact the quality of crops and their yields. We describe the validation of polyclonal antibodies against TS and FHB spores through indirect enzyme linked immunosorbent assay (ELISA). An indirect ELISA involves a series of additions that bind to one another: first an antigen/spore coated onto a plate; then primary antibodies; secondary antibodies that are conjugated to enzymes, and lastly colorimetric reagent that is measured by spectrophotometry. An indirect checkerboard ELISA is used to determine the effective specificity of polyclonal antibodies against fungal spores by using different concentrations of both antigens and antibodies. Alterations in the concentrations can be adapted to measure either spores or the antibody. The experiment was repeated twice with three replicates in each. The linear range of detection was between 1300 and 115,000 spores for FHB while being smaller for TS spores. Cross-reactivity of the TS antibodies will be verified against FHB spores and vice-versa FHB antibodies will be challenged with TS spores. These results suggest that these new antibodies will be very useful for antigenic detection of TS and FHB spores.



**S8** **Screening of barley cultivars for resistance to *Bipolaris sorokiniana* isolates from the Prairies**  
DILINI D. ADIHETTY, K. XI, H. KLEIN-GEBBINCK, R. ABOUKHADDOUR, H.R. KUTCHER, J.R. TUCKER, X. WANG, W. XU, A.D. BEATTIE, A. BADEA, T.K. TURKINGTON, AND S.E. STRELKOV. *Department of Agricultural, Food and Nutritional Science, 410 Agriculture/Forestry Center, University of Alberta, Edmonton, AB T6G 2P5, Canada; (K.X.) Alberta Agriculture and Forestry, Field Crop Development Centre, Lacombe, AB, T4L 1W1, Canada; (H.K.-G.) Beaverlodge Research Farm, Agriculture and Agri-Food Canada, Beaverlodge, AB T0H 0C0, Canada; (R.A.) Lethbridge Research and Development Center, Agriculture and Agri-Food Canada, Lethbridge, AB T1J 4B1, Canada; (H.R.K.) College of Agriculture and Bioresources, Department of Plant Sciences, University of Saskatchewan, Saskatoon, SK S7N 5A8, Canada; (J.R.T.) Brandon Research and Development Center, Agriculture and Agri-Food Canada, Brandon, MB R7A 5Y3, Canada; (X.W.) Morden Research and Development Center, Agriculture and Agri-Food Canada, Morden, MB R6M 1Y5, Canada; (W.X.) Morden Research and Development Center, Agriculture and Agri-Food Canada, Morden, MB R6M 1Y5, Canada; (A.D.B.) Department of Plant Sciences, College of Agriculture and Bioresources, University of Saskatchewan, Saskatoon, SK S7N 5A8, Canada; (A.B.) Brandon Research and Development Center, Agriculture and Agri-Food Canada, Brandon, MB R7A 5Y3, (T.K.T.) Lacombe Research and Development Center, Agriculture and Agri-Food Canada, Lacombe, AB T4L 1W1, Canada; and (S.E.S.) Department of Agricultural, Food and Nutritional Science, 410 Agriculture/Forestry Center, University of Alberta, Edmonton, AB T6G 2P5, Canada*

Spot blotch, caused by *Bipolaris sorokiniana* (Sacc.) Shoemaker, is an important foliar disease of barley found with increasing frequency on the Canadian Prairies. Yield losses of 16%-33% can occur under favorable conditions, and as such, the deployment of spot blotch-resistant cultivars can help to mitigate the impact of this disease. It is critical to evaluate the virulence of *B. sorokiniana* populations to identify any shifts that could reduce the effectiveness of resistance. Eleven *B. sorokiniana* isolates collected in 2018-2020 from Manitoba (5 isolates), Saskatchewan (3 isolates) and Alberta (3 isolates) were tested for their virulence on three barley cultivars, 'AAC Synergy', 'CDC Fraser', and 'Revanche'. Host reactions were rated on a 0 (no disease) to 9 (severe symptoms) infection response (IR) scale, 8-10 days after inoculation at the seedling stage. Four of the five isolates from Manitoba were highly virulent (7.5 to 9) on all three cultivars, while the fifth was highly virulent on 'AAC Synergy' and 'Revanche', but caused an IR of 4.6 on 'CDC Fraser'. One isolate from Saskatchewan and another isolate from Alberta caused an IR of 4.5 and 4.8, respectively, on 'AAC Synergy'. These data suggest that current *B. sorokiniana* populations on the Prairies can either overcome or at least show some adaptation to the resistance in cultivars such as 'AAC Synergy' and 'CDC Fraser'. The identification of isolates virulent on "resistant" cultivars highlights the importance of more improved resistant cultivars combined with integrated disease management practices for the sustainable management of spot blotch of barley.



- S9** **Understanding Verticillium stripe-blackleg interactions.** YIXIAO WANG, S.E. SRELKOV, AND S.F. HWANG *Department of Agricultural, Food and Nutritional Sciences, University of Alberta, Edmonton, AB T6G2P5, Canada*

Verticillium stripe, caused by *Verticillium longisporum*, is a relatively new disease of canola (*Brassica napus*) in Canada. In contrast, blackleg of canola (*Leptosphaeria maculans*) is ubiquitous and has been recorded in Canada for more than three decades. Both *V. longisporum* and *L. maculans* cause a similar discoloration of the stem in cross-section, and both pathogens may occur in the same field. Therefore, an improved understanding of Verticillium stripe - blackleg interactions could help growers to distinguish between these diseases and facilitate the selection of best management strategies. Field experiments were conducted over 2 years near Edmonton, Alberta, using the canola hybrids '45H31' and 'CS2000'. The hybrids were inoculated with *V. longisporum* and *L. maculans* individually and together at five different rates. Control treatments did not receive inoculum of either pathogen. Disease assessments were conducted at plant maturity on a 0 to 5 scale for blackleg (0 = a completely healthy plant and 5 = plant death) and 0 to 4 scale for Verticillium stripe (0 = a completely healthy plant and 4 = peeling of the stem epidermis and large numbers of microsclerotia). Both canola hybrids were susceptible to Verticillium stripe, which caused greater yield losses than blackleg. When both pathogens were inoculated together, yield losses ranged from 4.4% to 24.2%. Therefore, integrated disease management strategies should consider the possible occurrence of both *V. longisporum* and *L. maculans*.

- S10** **Effects of Soil Moisture and Temperature on the Pathogen Dynamics of Root Rot Complex in Field Pea.** KYLE BISCAGLIA-HORVATH, M. HARDING AND S. CHATTERTON. (K.B. & S.C.) *Lethbridge Research and Development Centre, Agriculture and Agri-Food Canada, 5403 1 Ave S, Lethbridge, AB, Canada;* (K.B.) *Department of Biology, University of Lethbridge, 4401 University Dr W, Lethbridge, AB, Canada;* (M.H.) *Crop Diversification Centre South, South 301 Horticultural Station Road East, Brooks, AB, Canada.*

Pea root rot is a disease complex comprised of multiple biotic and abiotic stressors resulting in decay of the root system. Pathogens contributing to root rot in field peas include *Aphanomyces euteiches* and various *Fusarium* spp. However, little is understood of the interaction between biotic and abiotic stressors within the disease complex. The objective of our study was to study the effects of soil moisture and temperature on disease development and host colonization in artificially inoculated multiple infection scenarios. CDC Meadow was selected as a susceptible cultivar and grown for the period of one month in sterilized soil inoculated with varying concentrations of two isolates of *Fusarium avenaceum* and one isolate of *Aphanomyces euteiches*, either singly or in combination. Environmental effects studied were (i) soil moisture (drought stressed, field capacity and saturated) and (ii) temperature (day: 16 hrs/night: 8 hrs; 28°C/20°C, 23°C/15°C and 18°C/10°C). At one-month plants were processed for biomass and roots were indexed for disease severity against a visual rating scale. Replicates were subsampled for DNA extraction from the tap and lateral root regions; and pathogen biomass will be quantified to determine host colonization and pathogen localization. Results to date have shown a positive correlation between volumetric water content and disease severity and a negative correlation between disease severity and temperature in samples containing *A. euteiches*. Samples containing a mixed population of *F. avenaceum* and *A. euteiches* displayed the lowest disease severity when watered to field capacity. By improving our understanding of these interactions, we hope to more accurately predict risk of severe root rot based on environmental conditions and knowledge of the predominant pathogen composition in field soils.



- S11 Detection and validation of partial resistance to *Aphanomyces* root rot in field pea via Bulk Segregant RNA-Seq.** LONGFEI WU, RUDOLPH FREDUA-AGYEMAN, STEPHEN E. STRELKOV, KAN-FA CHANG AND SHEAU-FANG HWANG. *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, T6G 2P5, Canada*

*Aphanomyces* root rot, caused by *Aphanomyces euteiches*, can cause severe yield loss in field pea (*Pisum sativum* L.). Polygenic resistance to this disease has been reported in the field pea cultivar '00-2067'. Bulk segregant analysis-RNA-Seq (BSR-Seq) was conducted with an F<sub>8</sub> RIL population derived from the cross 'Carman' × '00-2067' to detect and validate the basis of this resistance. Disease reactions were assessed under controlled conditions in three independent experiments, indicating a significant genotypic effect ( $P < 0.05$ ) and significant correlation coefficient ( $0.51 < r < 0.58$ ,  $P < 0.001$ ) among the three experiments. Resistant (R) and susceptible (S) bulks were generated that showed extreme resistance or susceptibility, respectively, to *A. euteiches*. RNA-seq analysis of the R bulks generated 44,595,510 ~ 51,658,688 reads and 98.08% ~ 99.44% were aligned, of which intragenic rates were 93.40% ~ 93.72% and exonic rates were 84.50% ~ 85.13%. For analysis of the S bulks, 43,848,192 ~ 45,664,302 reads were generated with alignment rates of 99.04% ~ 99.53%, intragenic rates of 92.86% ~ 93.43% and exonic rates of 83.37% ~ 84.50%. The aligned sequences in this study were linked to 44,757 genes in a reference genome, of which 42.60% ~ 43.12% and 42.66% ~ 43.13% were expressed in the R and S bulks, respectively. Two-hundred ninety-seven candidate genes that showed differential expression between the R and S bulks were selected for further study of the plant defense mechanisms against *A. euteiches* and for SNP development in pea.

## POSTER ABSTRACTS

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- S12 Temporal analysis of *Aphanomyces* root rot development after zoospore inoculation of field pea.** SIJAN PANDIT, R. GOYAL, E. SCHULTZ AND S. CHATTERTON. *University of Lethbridge, 4401 University Drive, Lethbridge, AB, T1K 3M4, Canada; (S.C.) Lethbridge Research and Development Centre, Agriculture and Agri-Food Canada, 5403-1 Avenue South Lethbridge, AB, T1J 4B1, Canada; (R.G.) Lacombe Research and development center, Agriculture and Agri-Food Canada, 6000 C and E trail, Lacombe, AB, T4L 1W1*

*Aphanomyces* root rot (ARR), caused by the oomycete *Aphanomyces euteiches*, is the most devastating soil borne disease in field pea (*Pisum sativum*). ARR can cause complete yield loss and efficient disease management options are unknown. Symptoms include honey brown lesions on lateral roots which later progress to tap root and epicotyl resulting in stunting, chlorosis and premature plant death. Symptoms might not be evident right after pathogen colonization of the roots. Understanding disease development in relation to pathogen inoculum is necessary for plant-pathogen interaction studies. The objective of this research was to develop consistent system for inoculating plants to achieve uniform disease and to determine time point of pathogen colonization, along with symptom manifestation, progression and pathogen biomass in roots with time after *A. euteiches* zoospores (asexual spores) inoculation. Two independent greenhouse trials with pea cultivars 'CDC Meadow' and 'CDC Dakota' were conducted. Two weeks old plants were inoculated with zoospores ( $10^3$ /ml) using a root soak method. Mock-inoculated plants were soaked in mineral-



salt solution. Roots were sampled for disease severity rating at 2, 6, 12, and 24 hours post inoculation (hpi), then 2 days post inoculation (dpi) followed by alternate days until 20 dpi. Symptoms were evident on inoculated plants at 6 dpi. However, qPCR of samples within first two days detected pathogen as early as 12 hpi on CDC Meadow, although more results on pathogen biomass are pending. This time period between pathogen entry and symptoms manifestation is crucial for the study of early plant defense responses to ARR.

- S13 Effects of *Fusarium avenaceum* and *F. proliferatum* on seedling survival, growth, root rot severity and yield of canola.** HAITIAN YU, S.F. HWANG, R. FREDUA-AGYEMAN AND S.E. STRELKOV. *Department of Agricultural, Food and Nutritional Sciences, University of Alberta, Edmonton, AB T6G2P5, Canada*

Seedling blight and root rot can have a substantial impact on plant stand uniformity and canola (*Brassica napus* L.) production in western Canada. Species of *Fusarium* are among the most common casual agents of seedling blight and root rot, with *F. avenaceum* reported to be predominant. In contrast, while *Fusarium proliferatum* can infect numerous crop plants and cause rots, blights, wilts and diebacks, this fungus has not been evaluated extensively on canola. In this study, we conducted field and greenhouse experiments to determine the effects of *F. avenaceum* and *F. proliferatum* on the growth of canola. Generally, for both *F. avenaceum* and *F. proliferatum*, as the inoculum concentration increased, seedling emergence, shoot weight, and yield decreased under field conditions. Disease severity in all treatments was significantly higher than in the controls. In the case of *F. proliferatum*, disease severity in both cultivars examined increased as the inoculum concentration increased, and emergence significantly declined as the inoculum concentration increased from zero (control) to low to medium. Canola yield was significantly lower than in the control at the high inoculum concentration. Under greenhouse conditions, plant counts, plant height, shoot weight, and disease severity were negatively affected by *F. proliferatum* inoculum, and disease severity increased and plant height decreased with increasing inoculum concentration. In conclusion, while both *F. avenaceum* and *F. proliferatum* reduced emergence and increased disease severity in canola, *F. proliferatum* showed a more marked effect.

- S14 Determination of the optimum number of days to scan for root architectural traits.** CHUNXIAO YANG, R. FREDUA-AGYEMAN, S.F. HWANG, S.E. STRELKOV AND L.Y. GORIM. *Department of Agricultural, Food and Nutritional Sciences, University of Alberta, Edmonton, AB T6G 2P5, Canada*

Canola (*Brassica napus* L.) is an important cash crop in Canada. Root architectural traits can vary among *B. napus* genotypes and could influence the response to different root pathogens, the efficiency of nutrient and water uptake from the soil, as well as other important biological and ecological processes. However, comparative studies on root architecture in *B. napus* are limited. The objectives of this study were to determine the optimal number of days at which root architectural traits of *Brassica* accessions could be measured accurately, and the sampling day at which significant differences could be observed among four *B. napus* cultivars ‘L255PC’, ‘Westar’, ‘L150’ and ‘Mendel’. Eleven parameters related to root architecture, including total root length (TRL), total



surface area of roots (TRSA), average root diameter (RAD), Number of tips (NTP), total primary root length (TPRL), total lateral root length (TLRT), total tertiary root length (TTRL), basal link length (BLL) and the proportion of TTRL, TRSA and Volume in 7 diameter classes (0-3.5 mm with 0.5 mm gap) were compared among the four *B. napus* cultivars at 7, 14 and 21 days after 1-week pre-germination. Roots were examined using an EPSON Perfection V800 scanner and WinRHIZO software, with the data analyzed using a mixed-effect linear model. The preliminary data suggested that 14-day-old seedlings were optimal for the accurate measurement of root architectural traits under hydroponic conditions in the growth chamber. This serves as a reference point for additional studies on root architecture and its influence on disease resistance and other processes.

- S15 Optimization of *Fusarium oxysporum* inoculation methods for *Brassica napus*.** CHUNXIAO YANG, S.F. HWANG, R. FREDUA-AGYEMAN AND S.E. STRELKOV. *Department of Agricultural, Food and Nutritional Sciences, University of Alberta, Edmonton, AB T6G 2P5, Canada*

Fusarium wilt, caused by *Fusarium oxysporum*, was first reported on canola (*Brassica napus* L.) in Alberta about 20 years ago. Due to climate change and other factors, the disease has high potential to become an issue in Canadian canola production. The objective of this study was to evaluate the effect of seedling age at the time of inoculation, length of time roots are exposed to inoculum, and the concentration of inoculum on fusarium wilt severity. Seven, 14 and 21-day-old seedlings of the canola 'Westar' were inoculated with *F. oxysporum* by incubating the roots in one of three conidial suspensions ( $10^5$ ,  $10^6$  or  $10^7$  conidia/mL) for 5 min, 2 h, 6 h or 12 h. The seedlings were then transplanted into a potting mixture and maintained under greenhouse conditions. Twenty-one days after inoculation, the seedlings were assessed for fusarium wilt severity. Seedlings inoculated at 21 days developed the greatest disease severity, while those inoculated at 7 days showed the lowest severity. Incubation of the roots in the inoculum suspension for 12 h resulted in significantly greater disease severity than incubation for 5 min, 2 h or 6 h. Inoculation with a concentration of  $10^6$  or  $10^7$  conidia/mL resulted in significantly greater disease than inoculation with  $10^5$  conidia/mL. Based on these results, fusarium wilt development appears strongest when the roots of older seedlings are incubated with higher concentrations of *F. oxysporum* for longer periods.

- S16 Transcriptomic profiling of the host-pathogen interaction in tan spot of wheat.** CLAUDIA ESCOBAR-GIL, L. GALINDO-GONZALEZ, A. AKHAVAN AND S. STRELKOV. *Department of Agricultural, Food and Nutritional Science, 410 Agriculture/Forestry Centre, University of Alberta, Edmonton, Alberta, T6G 2P5. Canada*

Tan spot, caused by the necrotrophic fungus *Pyrenophora tritici-repentis*, is a foliar disease that affects wheat (*Triticum aestivum* and *Triticum turgidum*) by decreasing the kernel weight and number of kernels per spike. In recent years, tan spot has become one of the most detrimental diseases of wheat in Canada. Research on this pathosystem has focused on understanding the genetic basis of host resistance, yet important aspects of the plant-pathogen interaction remain unanswered. Using high-throughput RNA-sequencing, we have identified differences between isolates of *P.*



*tritici-repentis* during saprophytic vs. parasitic growth. Differences were also identified *in planta*, at 12, 36 and 72 h after inoculation, between isolates producing the necrotrophic effectors ToxA and ToxB. The number of reads recovered indicated an increase in the ToxA-producer, while minor changes in read numbers were found for the ToxB producer. Preliminary analysis showed that the expression patterns correctly clustered the biological replicates according to each treatment, validating the experiment. For the ToxA-producing isolate, the number of differentially expressed genes decreased over time, in contrast with the ToxB-producing isolate, which presented a constant pattern of expression. This study will provide genomic-based resources to improve understanding of virulence mechanisms in *P. tritici-repentis* and identify patterns of molecular interactions between the host and pathogen. The knowledge can be applied to the development of improved tan spot management programs, by identifying genes involved in virulence and potential targets in the host. Additionally, through comparative genomics with other necrotrophic fungal pathogens, these genes can provide insights into pathogenicity-related processes.

**T3** **A search for an effective control measure against root-rot in field pea: Effect of antimicrobial peptides on *in vitro* growth of *Aphanomyces euteiches*.** ALBERT J. HANNIG<sup>1</sup>, S. CHATTERTON<sup>2</sup>, M. TETORYA<sup>3</sup>, D. SHAH<sup>3</sup> AND R. K. GOYAL<sup>1</sup>, <sup>1</sup>AAFC, Lacombe Research and Development Centre, Lacombe, AB, T4L 1W1, Canada, <sup>2</sup>AAFC, Lethbridge Research and Development Centre, Lethbridge, AB, T1J 4B1, Canada, <sup>3</sup>Donald Danforth Plant Science Center, St. Louis, MO, 63132, USA

Dry field pea (*Pisum sativum* L.) is an important export crop grown in the Canadian prairies. With the recognition of benefits of plant-based proteins in human diet, and the fact that field pea has a high protein content in seeds, the demand of this crop is expected to grow significantly. In addition, because of its nitrogen fixing ability which enables it to keep a low carbon footprint, field pea has gained further importance in crop cultivation choices. However, a growing presence of root-rot pathogens in the Prairies fields and lack of effective control measures have seriously threatened the pea production system. *Aphanomyces euteiches* is one of the pathogens responsible for root-rot that infects seedlings resulting in severely reduced plant growth and seed yield, thus causing substantial economic losses. Long-lived dormant oospores of the pathogen in soil make it difficult to manage the disease. Here in this study, we are exploring to assess the efficacy of antimicrobial peptides (AMPs), which are a part of plant innate immunity but have a different mechanism of action than classical R-genes, against *A. euteiches*. Thirteen AMPs were tested for their ability to inhibit the growth of *A. euteiches* under *in vitro* conditions. Using a resazurin stain assay the inhibitory concentration (IC<sub>50</sub>) values of the AMPs were determined. The most effective AMPs were PN60 and PN51 with average IC<sub>50</sub> values of 3.03 and 0.79  $\mu$ M, respectively. A highly potent activity of these peptides was very promising which can be further developed into a novel approach to manage *A. euteiches*-induced root rot in field pea.



- T4** *Pyrenophora tritici-repentis* in Japan: first report on race structure and a novel *ToxA* haplotype. MOHAMED HAFEZ, T. DESPINS, K. NAKAJIMA & R. ABOUKHADDOUR. *Agriculture and Agri-Food Canada, Lethbridge Research and Development Center, Lethbridge, T1J 4B1, AB, Canada. (K.N) Mie Prefectural Agricultural Research Institute, Matusaka, Japan.*

Tan spot, caused by *Pyrenophora tritici-repentis* (*Ptr*), is a destructive foliar wheat disease reported worldwide. The fungus was first described as a pathogen on wheat in Japan in the 1920s, but since then there are no reports on the race structure or the dominant effectors secreted by *Ptr* in Japan. In this study, ten single-spore isolates of *Ptr* were collected from bread wheat and from four locations within the Mie prefecture in Japan. These isolates were evaluated for their virulence on four differential wheat genotypes, and tested for the presence/absence of the effector genes, *ToxA* and *ToxB* and its homolog *tox b*, in multiplex PCR assays. Eight isolates were designated as race 2 (*ToxA*-producers), and two isolates were classified as race 1 (*ToxA* and *ToxC*-producers), based on their virulence patterns. The necrosis-inducing *ToxA* gene was present in all tested isolates, whereas *ToxB*, the chlorosis-inducing gene, and its homolog *tox b* were totally absent. Sequence analysis for *ToxA* amplicons from these isolates indicated the presence of a novel *ToxA* haplotype. Comparative sequence analysis and re-sequencing of *ToxA* from reference *Ptr* isolates showed that all the previously published *Ptr ToxA* haplotypes (3 haplotypes) were identical. This is the first report correctly confirming the presence of an additional new *ToxA* haplotype in *Ptr*, which has two non-synonymous mutations at the DNA level that alter the amino acid sequence of the *ToxA* effector. The overall results highlight the value of exploring variability in *Ptr* and its *ToxA* haplotypes around the world to gain a better understanding of the evolution of this important fungus.

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#### GUEST SPEAKER ABSTRACTS

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**The occurrence and spread of clubroot in Alberta (2005-2020).** YOANN AIGU, V.P. MANOLI AND S.E. STRELKOV. *Department of Agricultural, Food and Nutritional Science, 410 Agriculture/Forestry Center, University of Alberta, Edmonton, AB T6G 2P5, Canada*

Clubroot, caused by the obligate parasite *Plasmodiophora brassicae*, is a soilborne disease of *Brassica napus* and other cruciferous hosts. Infection by *P. brassicae* is associated with the formation of large galls on the roots of susceptible plants, leading to yield losses estimated at 10%-15% globally. In Alberta, clubroot was first identified on canola in 2003 and targeted surveys have been conducted annually since 2005, generating a large data set. By 2020, clubroot had been detected in almost 3400 fields. At present, the disease is managed mainly by planting clubroot resistant canola cultivars, which initially became available in 2009 and 2010. However, in 2013, resistance-breaking (RB) genotypes of *P. brassicae* were detected for the first time in Alberta. By 2019, RB genotypes had been confirmed in 239 clubroot-infested fields vs. 2928 fields infested by the wild-type (WT) genotype. In this presentation, the patterns and rates of spread will be compared for RB and WT genotypes of *P. brassicae*, and epidemiological approaches, including mathematical modeling and the application of the minimum convex polygon method, will be applied to improve understanding of pathogen dissemination and evaluate the efficiency of efforts to limit disease spread.



**Fungicide timing and assessment of risk factors for sclerotinia stem rot of canola.** THOMAS KELLY TURKINGTON, H. KLEIN-GEBBINCK, H. KUBOTA, B. TIDEMANN, G. SEMACH, C. GEDDES, S. CHATTERTON, M. HARDING, PRABHATH LOKURUGE, A. MULENGA, E. KARPPINEN, P. MOOLEKI, D. TOMASIEWICZ, G. PENG, W. MAY, R. MOHR, G. TELMOSSE, D. PAGEAU, J. FENG, E. MCBAIN, AND S.E. STRELKOV. (T.K.T., H.K., B.T.) Lacombe Research and Development Center, Agriculture and Agri-Food Canada, Lacombe, AB T4L 1W1, Canada; (H.K.-G., G.S.) Beaverlodge Research Farm, Agriculture and Agri-Food Canada, Beaverlodge, AB T0H 0C0, Canada; (C.G., S.C.) Lethbridge Research and Development Center, Agriculture and Agri-Food Canada, Lethbridge, AB T1J 4B1, Canada; (M.H.) Alberta Agriculture and Forestry, Crop Diversification Centre South, Brooks, AB T1R 1E6, Canada; (P.L., A.M.) Scott Research Farm, Agriculture and Agri-Food Canada, Scott, SK S0K 4A0, Canada; (E.K., P.M., D.T.) Canada-Saskatchewan Irrigation Diversification Centre, Agriculture and Agri-Food Canada, Outlook, SK S0L 2N0, Canada; (G.P.) Saskatoon Research and Development Centre, Agriculture and Agri-Food Canada, Saskatoon, SK S7N 0X2, Canada; (W.M.) Indian Head Research Farm, Agriculture and Agri-Food Canada, Indian Head, SK S0G 2K0, Canada; (R.M.) Brandon Research and Development Center, Agriculture and Agri-Food Canada, Brandon, MB R7A 5Y3, Canada; (G.T., D.P.) Normandin Experimental Farm, Agriculture and Agri-Food Canada, Normandin, QC G8M 4K3, Canada; (J.F.) Alberta Agriculture and Forestry, Crop Diversification Centre North, Edmonton, AB T5Y 6H3, Canada; and (E.B., S.E.S) Department of Agricultural, Food and Nutritional Science, 410 Agriculture/Forestry Center, University of Alberta, Edmonton, AB T6G 2P5, Canada

Fungicide use decisions can be challenging for producers trying to manage sclerotinia stem rot (SSR) of canola caused by *Sclerotinia sclerotiorum*. The objective of this work was to determine the appropriate times in terms of crop growth stage, weather conditions and inoculum load for fungicide applications to improve SSR management. An experiment was conducted in 2019 at 10 locations across Canada and included two seeding rates (60 and 120 seeds m<sup>-2</sup>) and nine fungicide applications arranged in a four-replicate factorial design. Applications included a check and single or dual fungicide applications (prothioconazole) starting at the yellow bud (YB) stage and then 1-4 weeks after YB. Rainfall, and in-canopy and ambient temperature and relative humidity were monitored along with assessment of aerial inoculum load. Weather conditions and reduced inoculum load at most sites limited SSR, except Beaverlodge, AB and Outlook, SK. The greatest reduction in SSR at Beaverlodge tended to occur for the single fungicide application three weeks after YB and the dual application at YB and again three weeks later. At Outlook, the one week after YB tended to have the lowest SSR. No yield data were available at Beaverlodge due to early onset of winter and subsequent deer damage, while at Outlook no yield differences occurred which was likely due to SSR levels being low (<4% incidence in the check treatments). Preliminary results suggest that optimal timing for reducing sclerotinia stem rot will vary based on crop growth stage, weather conditions, and inoculum loads prior to and during flowering.



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