

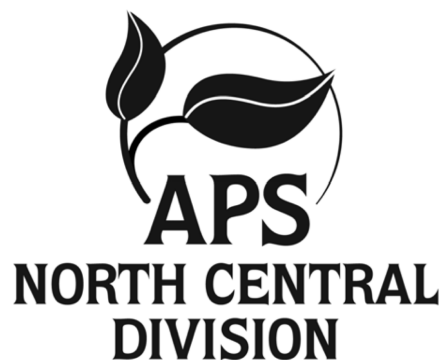
**Annual Meeting of the North Central Division
of the American Phytopathological Society**

**Hilton Garden Inn & Conference Center
410 S. Third St.
Manhattan, Kansas, 66502**

June 10 - 12, 2024

**Hosted by the Department of Plant Pathology,
Kansas State University**





North Central Division APS Meeting
June 10-12, 2024; Manhattan, Kansas

MONDAY, JUNE 10

- 8:00 - 5:00 pm Registration table open.
Conference Center Foyer, Hilton Garden Inn
- 3:00 pm + Poster set-up available.
McDowell/Tuttle/Alcove, Hilton Garden Inn
- 8:00 - 5:00 pm **Hands-on ELISA Wheat Viruses Workshop**
Instructors: Chandler Day, Judy O'Mara, Jessica Rupp Noller
Field, lab experience, & tours; Kansas State University
Lunch & transportation provided from Hilton Garden Inn
Registration required; Max capacity: 20 participants.
Summary: ELISA testing is a key test diagnostic labs around the country use regularly, but there are tips and tricks to ensure a successful test result. This workshop will provide instructors, students, and early career professionals with an overview of the process and hands-on experience from field sampling to interpretation of the result. Ensuring the use of industry best practices and optimization, you can provide stakeholders with timely results. This workshop will cover common wheat viruses and look at wheat diseases currently active in the field. The techniques and knowledge gained from this workshop will be broadly applicable to a wide variety of plant diseases. Agenda and details to follow.
- 1:00 - 5:00 pm **Plant Biosecurity Workshop**
Instructors: Giovanna Cruppe, James Stack
Pat Roberts Hall - Biosecurity Research Institute (BRI), 1900 Denison Avenue, Manhattan, Kansas
Transportation provided from Hilton Garden Inn
Registration required; Max capacity: 15 participants.
Summary: The Plant Biosecurity Workshop will convene at the *Biosecurity Research Institute (BRI)*, a Biosafety level-3-Ag biocontainment facility, located on the Kansas State University campus. The research program at the BRI includes plant, animal, and human health projects. The workshop will include an overview of Plant Biosecurity, in-depth discussions about BRI research with two high-consequence plant pathogens, a brief tour of the BRI, and a short biosecurity desktop exercise where participants will engage in decision-making to support

response to a potential plant health emergency.

6:00 - 9:00 pm

Welcome Reception

Flint Hills Discovery Center, 315 S. 3rd Street, Manhattan, Kansas

Across the street from the Hilton Garden Inn. Partially sponsored by Corteva.

Hors d'oeuvres and cash bar.

TUESDAY, JUNE 11

Venue: Hilton Garden Inn, 410 S. 3rd Street, Manhattan, Kansas

Kaw Nation/Big Basin Ballroom

8:00 - 8:15 am

Welcome and introductory remarks

Dr. Megan Kennelly, Head, KSU Department of Plant Pathology; **Dr. Jane Schuh**, Associate Dean of Research, KSU College of Agriculture & Life Sciences; **Dr. Karen Garrett**, APS President-Elect.

General Session

Moderator: Dr. Chris Little, Scientific Program Chair

8:15 - 8:45 am

"Plant health in an evolving One Health Concept: The nature and rationale for inclusion"

Dr. Jim Stack, Professor of Plant Pathology, Department of Plant Pathology, Kansas State University

Summary: There are multiple examples of, 1) plant pathogens causing diseases in humans and animals, 2) plants as reservoirs of human pathogens, and 3) plants and plant products that serve as vectors of human and animal pathogens. Those are the attributes that underpin One Health designation. The recent more prominent feature of plant health in the One Health Concept is long overdue; the nature and rationale for inclusion will be discussed.

8:45 - 9:15 am

"Advancing digital agriculture for improving agronomic decisions"

Dr. Ignacio Ciampitti, Professor of Farming Systems, Department of Agronomy, Kansas State University; Director of Research, Institute for Digital Agriculture and Advanced Analytics, Kansas State University

Summary: This presentation will focus on discussing the bottlenecks for translating research into actionable management within a context of digital agriculture. The use of more data-driven approaches, including probabilistic analysis, integration of new technologies such as satellite imagery, and development of new data visualization applications are all relevant options to assist producers on improving agronomic decision

9:15 - 9:45 am

"Team-based learning: A strategy to enhance student engagement in undergraduate courses"

Dr. Leonor Leandro, Professor of Plant Pathology, Department of Plant Pathology, Entomology, and Microbiology, Iowa State University

Summary: Student engagement is a key factor in creating an effective and enriching learning experience. In this presentation, I will describe the principles of

team-based learning (TBL), a collaborative learning and teaching strategy, and share how I used it to transform my approach to teaching mycology and plant pathology to undergraduate students.

9:45 - 10:00 am Break
Sponsored by BASF.

Student Presentations

Moderator: Dr. Jessica Rupp Noller, Student Oral & Poster Competition Chair

Student Session #1

Kaw Nation/Big Basin Ballroom

Abstracts available alphabetically by **presenting author*** last name. See pages 10-40.

(Abbreviations: ISU = Iowa State University; KSU = Kansas State University; MSU = Michigan State University; NDSU = North Dakota State University; SDSU = South Dakota State University; UNL = University of Nebraska - Lincoln; Utrecht = Utrecht University; WSU = Washington State University)

- 10:00 - 10:15 am "Identification of genetic factors involved in pathogenicity and virulence of *Xanthomonas translucens* pv. *undulosa* using EZ-Tn5 mutagenesis" **Fazal Manan*** (NDSU), Z. Liu (NDSU), I. Bankoke (NDSU), G. Shi (NDSU)
- 10:15 - 10:30 am "Effect of fertilization on soybean sudden death syndrome severity" **Leticia Camara Viera*** (KSU), T. Todd (KSU), R.B. Onofre (KSU), E. Adey (KSU)
- 10:30 - 10:45 am "Unraveling the wall-associated kinase immunity in the barley spot blotch pathosystem" **Meenu Sengar*** (SDSU), S. Solanki (SDSU), G. Ameen (SDSU), R. Brueggeman (WSU)
- 10:45 - 11:00 am "Molecular approaches to identify *Ascochyta rabiei* effectors" **Amian Arman*** (NDSU), M. Seidl (Utrecht), M. Ebert (NDSU)
- 11:00 - 11:15 am "Species diversity of soybean sudden death syndrome pathogens in Kansas" **Luiza Adami Montiero de Castro*** (KSU), R.B. Onofre (KSU), M. Chilvers (MSU), K. Andersen-Onofre (KSU), R. Debacker Moura (KSU), M. Kessler (KSU)
- 11:15 - 11:30 am "Fungicidal strategies for Septoria brown spot and frogeye leaf spot management in Iowa grown mungbean (*Vigna radiata*)" **Evelyn Platner-Heidt*** (ISU),
- 11:30 - 11:45 am "The role of irrigation in canopy microclimate and disease development" **Talon Mues*** (UNL), T. Jackson-Ziems (UNL), S. Taghvaeian (UNL), M. Shires (SDSU), R.B. Onofre (KSU), D. Mangel (UNL)
- 12:00 - 1:30 pm Lunch (on your own)
See attached map for downtown Manhattan restaurants within walking distance.

12:00 - 1:30 pm **Careers 101 Workshop**
Instructor: Horacio Lopez-Nicora, The Ohio State University
Kaw Nation/Big Basin Ballroom, Hilton Garden Inn
Registration required. Lunch provided for registered participants.

Student Session #2

Kaw Nation/Big Basin Ballroom

Abstracts available alphabetically by **presenting author*** last name. See pages 10-40.

(*Abbreviations:* Danforth = Donald Danforth Plant Science Center; KSU = Kansas State University; NDSU = North Dakota State University; Okla. St. = Oklahoma State University; UIUC = University of Illinois Urbana-Champaign; USDA, CCRU = USDA-ARS Cereal Crops Research Unit; USDA, HWWGRU = USDA-ARS Hard Winter Wheat Genetics Research Unit)

- 1:45 - 2:00 pm "Towards fine mapping and cloning of novel leaf and stem rust resistant genes in the Portuguese durum wheat landrace PI 192051" **Md Al Mamun*** (NDSU), G. Valentini (NDSU), U. Gill (NDSU), M. Aoun (Okla. St.),
- 2:00 - 2:15 pm "Genome-wide identification of Fusarium head blight resistance and susceptibility loci in USA wheat breeding programs" **Lawrence Tidbaki*** (KSU), G. Bai (USDA, HWWGRU), K.W. Jordan (USDA, HWWGRU), J. Rupp Noller (KSU)
- 2:15 - 2:30 pm "Haplotype and transcriptome analyses reveal genes and gene regions that promote FHB resistance in spring wheat" **Sittal Thapa*** (NDSU), J. Fiedler (USDA, CCRU), S. Xu (USDA, CCRU), G. Bai (USDA, HWWGRU), B. Poudel (Danforth)
- 2:30 - 2:45 pm "Screening diverse maize lines for resistance to Pythium root rot" **Harrison Hall*** (UIUC), M. Krone (UIUC), S.M. Mora (UIUC), T. Jamann (UIUC)
- 2:45 - 3:00 pm "Phenotyping and genotyping of near-isogenic lines harboring a major Fusarium head blight resistance gene (*Fhb1*) in spring wheat" **Shahed Safar*** (NDSU), R.S. Nandety (USDA, CCRU), M. Osenga (USDA, CCRU), J. Fiedler (USDA, CCRU), Y. Leng (NDSU), S. Zhong (NDSU)
- 3:00 - 3:15 pm Break
Sponsored by Syngenta.

Student Session #3

Kaw Nation/Big Basin Ballroom

Abstracts available alphabetically by **presenting author*** last name. See pages 10-40.

(*Abbreviations:* Missouri = University of Missouri; MSU = Michigan State University; NDSU = North Dakota State University; NWROC = Northwest Research Outreach Center; SDSU = South Dakota State University; UMinn = University of Minnesota)

- 3:15 - 3:30 pm "Occurrence of new *Sdh* gene mutations in *Alternaria solani* with potential impact in the management of early blight," **Sunil Shrestha*** (NDSU, J.

	Pasche (NDSU), N. Gudmestad (NDSU), I. Mallick (NDSU)
3:30 - 3:45 pm	"Microplate-based assessment of DMI fungicide sensitivity in <i>Cercospora beticola</i> ," <u>Austin Lien</u> * (UMinn), M. Bolton (USDA), N. Wyatt (USDA), A. Chanda (UMinn, NWROC)
3:45 - 4:00 pm	"Copy number variations at the <i>Rhg1</i> locus and their relationship with resistance to soybean cyst nematode (<i>Heterodera glycines</i>)" <u>Dinesh Poudel</u> * (NDSU), G. Yan (NDSU), G. Kreutz (NDSU), C. Miranda (NDSU)
4:00 - 4:15 pm	"Effectiveness of fungicides and biorationals controlling dis- appearing root rot on ginseng" <u>Jacqueline Metheny</u> * (MSU), M. Hausbeck (MSU)
4:15 - 4:30 pm	"Distribution and virulence phenotypes of soybean cyst nematode (<i>Heterodera glycines</i>) populations in South Dakota" <u>Nabina Karki</u> * (SDSU), M. Shires (SDSU), C. Tande (SDSU), C. Kotaska (SDSU)
4:30 - 4:45 pm	"Missouri on-farm strip trials: 5-year summary of soybean response to R3 foliar fungicide applications" <u>Feyisayo Akande</u> * (Missouri), A. Ohmes (Missouri Extension), C. Ellis (Missouri), R. Lee (Missouri). J. Lory (Missouri), M. Bish (Missouri), K. Bissonette (Cotton, Inc.), J. Calhoun (Missouri), W. Flanary (Missouri)
5:15 - 7:00 pm	Poster session & reception <i>McDowell/Tuttle/Alcove</i> <i>Drink ticket bar available.</i> <i>Partially sponsored by FMC.</i>
7:00 pm +	Banquet & Awards <i>Kaw Nation/Big Basin Ballroom</i> <i>Partially sponsored by Bayer.</i>

WEDNESDAY, JUNE 12

Morning	Poster take-down
8:30 - 10:00 am	NC-APS Division business meeting <i>Big Basin Ballroom</i>
10:15 - 12:00 pm	The Tactile Toolbox: 3D Printing for IPM Workshop Organizer: Horacio Lopez-Nicora, The Ohio State University <i>Registration required</i> <i>Kaw Nation Ballroom</i>

2024 NCAPS Sponsors



On behalf of the Organizing Committee -- Thanks for attending!

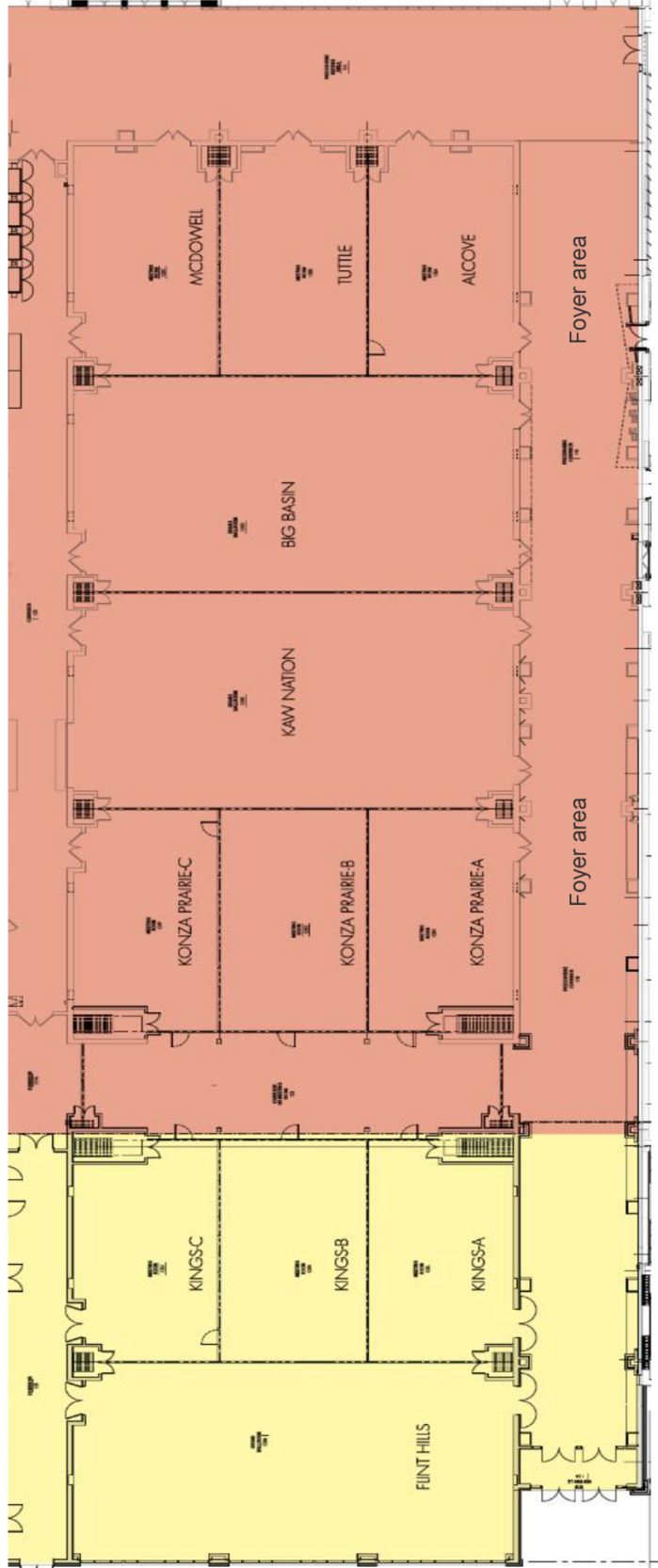
Megan Kennelly, NCAPS President

Chris Little, Scientific Program Chair

Jessica Rupp-Noller, Student Competition Chair

Kelsey Andersen-Onofre, Local Arrangements Chair

Rodrigo Borba Onofre, Sponsorship Chair



MANHATTAN CONFERENCE CENTER - FLOOR PLAN



ABSTRACTS

Ordered by presenting author's last name (underlined). Final abstracts will be published by APS.

A

Synthetic community approach for investigating microbiome assembly and interactions during Fusarium Head Blight (FHB) infection of barley

J. Acaso¹, B.R. Benz¹, E.L. Echartea¹, B. Whitaker², T. Baldwin¹, B. Geddes¹; ¹North Dakota State University, ²USDA-ARS; **Abstract:** Fusarium head blight (FHB) poses significant global challenges for food safety and security, leading to billions in economic losses and health risks due to the mycotoxin deoxynivalenol (DON). The complexity of resistance to FHB in small grains necessitates innovative solutions, with the plant microbiome emerging as a promising intervention. The plant microbiome, consisting of diverse microorganisms, is crucial in plant health and disease resistance. However, our understanding of phyllosphere microbiome assembly and interactions in the context of disease in barley is limited. We aim to define and assemble a synthetic community (*Hv* SynCom) from the barley head microbiome through culturing and to develop an experimentally tractable system to study the community members as they assemble into the head. This will enable investigations of microbiome assembly and interactions in the context of colonization by *F. graminearum*. From the barley phyllosphere, 16 microbial taxa were identified due to high abundance and occupancy rate. Growth experiments of barley inoculated with *Hv* SynCom demonstrated that early heading short barley varieties, sterile substrate, and media, and a close-controlled environment are essential features for a tractable system. This study could provide critical insights into sustainable crop production and disease control through the manipulation of the microbiome.

Species diversity of soybean sudden death syndrome pathogens in Kansas

L. Adami Monteiro de Castro¹, R. Borba Onofre¹, M. Chilvers², K. Andersen-Onofre¹, R. Debacker Moura¹, M. Kessler¹; ¹Kansas State University, ²Michigan State University; **Abstract:** Sudden Death Syndrome (SDS) is one of the most important yield-limiting diseases of soybean in the United States. Five species of *Fusarium* have been identified as causal agents of this disease globally. *Fusarium virguliforme* was the only pathogen described as the causal agent in the United States for many years, however in 2019 *F. brasiliense* was confirmed in Michigan in 2019. The objective of this study was to identify the *Fusarium* species causing SDS in Kansas. During the 2022 season, symptomatic plants were sampled from 13 fields across five counties in Kansas resulting in 128 *Fusarium* spp. being isolated. Genetic analysis using PCR and sequencing of the translation elongation factor-1 α (EF-1 α) gene was conducted for all isolates. Additionally, isolates identified as part of the FSSC clade 2 were further sequenced for the nuclear rDNA intergenic spacer (IGS) region. Phylogenetic trees were constructed using the Maximum Likelihood method with bootstrap support from 1000 replicates. Of the 128 isolates, 113 isolates were confirmed *F. virguliforme*. Of the remaining isolates six were identified as FSSC clade 5, one as FSSC clade 3, two as FFSC, two as FOSSC and four as undescribed *Fusarium* spp. These data suggest that in the fields sampled, *F. virguliforme* is likely the dominant species causing SDS in Kansas.

Fungicide sensitivity of *Sclerotinia sclerotiorum* isolates associated with sugar beet in the United States

M. Aderoju¹, S. Bhandari¹, M.F.R. Khan¹, L. del Rio Mendoza¹, S. Kalika-Singh¹; ¹North Dakota State University; **Abstract:** *Sclerotinia sclerotiorum* is emerging as a threat to the sugar beet (*Beta vulgaris* L.) industry in the United States. This pathogen causes leaf blight, seedling damping-off and root necrosis. Fungicide use is the most effective method to manage this pathogen, but its efficacy varies depending on the geographic location and the pathogen's sensitivity to their active ingredients. In this study, we evaluated the sensitivity of *S. sclerotiorum* isolates from sugar beet fields from various U.S. states to multiple fungicides. The poison plate assay was used for evaluating *S. sclerotiorum* sensitivity to fungicides. EC50

values were calculated using the diameter of fungal colonies growing on agar plates containing 0.01, 0.1, 1, and 10 ppm of each fungicide measured after three days of incubation in dark at 23°C. The mean EC₅₀ for Priaxor+SHAM, Proline (DMI), Priaxor (SDHI+QoI), Topsin (MBC), Excalia (SDHI), Quadris (QoI), Quadris+SHAM were 0.10, 0.32, 0.54, 0.94, 4.43, 5.33 and 6.96 ppm, respectively. Isolates from Minnesota and North Dakota were the least sensitive compared to the baseline across all fungicides. Large differences in EC₅₀ to the same chemicals are an indication of potential resistance issues in the future. This information could be used as references to monitor future changes in sensitivity.

Genetic mapping of resistance to Goss's wilt of maize

O. Adesina¹, B. Li², J. Jaqueth², Y. Hao¹, E. Stier¹, S. Liu¹; ¹Kansas State University, ²Corteva Agriscience; **Abstract:** Goss's bacterial wilt and leaf blight, caused by the Gram-positive actinobacterium *Clavibacter nebraskensis* (Cn), is a significant bacterial disease of maize and affects the maize production in the US and Canada. Similar to many plant pathogens, genetic resistance remains one of the best control strategies. However, host responses to Gram-positive bacteria and the molecular and genetic resistance mechanisms associated with developing plant disease symptoms caused by Cn in maize are poorly understood. We analyzed gene expression upon Cn infection in both resistant (R) and susceptible (S) lines, identifying Cn-triggering expression of genes involved in multiple hormone pathways. The mutants of some of those genes are being collected from the public stock center or generated through CRISPR editing for genetic analysis. At the same time, mapping of the resistance gene has identified multiple genomic regions conferring Goss's wilt resistance. We have constructed mapping populations for further finer-scale mapping. In this poster, we will update our mapping result through using Bulk Segregant RNA-seq (BSR-seq). Our integrated approach aims to provide foundational data for mechanistic studies to mitigate the effects of this important maize disease.

Association mapping of bacterial leaf streak disease resistant loci in winter wheat

M. Ahmad¹, G. Ameen¹, C. Galvin¹, S. Solanki¹, A. Alhusays¹, M. Torres², H. Ashfaq³, P. St. Amand⁴, A. Bernardo⁴, G. Bai⁴, S. Seghal¹, K.D. Glover¹; ¹South Dakota State University, ²Southwestern College, ³University of Florida, ⁴USDA-ARS, HWWGRU; **Abstract:** Bacterial leaf streak (BLS) disease, caused by *Xanthomonas translucens* pv. *undulosa*, is a major threat to wheat production in the Northern Great Plains, leading to up to 40% yield losses. Despite lacking effective chemical control, genetic resistance remains the most reliable strategy for managing BLS. In pursuit of this goal, we assessed 480 hard winter wheat lines from the South Dakota winter wheat breeding program for BLS resistance. Using a modified disease scoring scale ranging from 1 to 9, we identified four highly resistant and 28 moderately resistant genotypes in the greenhouse experiment. Following genotyping with the RAD-GBS approach, we detected 11,012 polymorphic SNP markers. Through genome-wide association analysis (GWAS) using the FarmCPU model, we identified nine significant marker-trait associations (MTA) (p<0.005) across five wheat chromosomes (2A, 6B, 3A, 4A, and 3B). These QTLs collectively explain 13.8% of the observed phenotypic variance in BLS resistance among the tested genotypes. Notably, the resistance loci on chromosomes 2A, 6B, 3A, 3B, and 4A harbor a total of 22, 27, 3, 4, and 1 putative candidate gene(s) respectively, encoding proteins such as NB-LRR, PIK6-NB, WAK-2, and RGA1, known for their role in plant defense mechanisms. The four -highly resistant genotypes and the SNP markers linked to the identified QTLs are useful sources to accelerate BLS resistance breeding efforts and characterization of underlying resistance pathways.

Missouri on-farm strip trials: 5-year summary of soybean response to R3 foliar fungicide applications

F. Akande¹, A. Ohmes¹, C. Ellis¹, R. Lee¹, J. Lory¹, M. Bish¹, K. Bissonnette², J. Calhoun¹, W. Flanary¹; ¹University of Missouri, ²Cotton Inc.; **Abstract:** Soybean growers continue the practice of foliar fungicide applications as a management strategy to protect soybean crops from foliar diseases. The use of foliar fungicides in U.S. soybean production substantially grew in the early 2000's due to concerns about soybean rust. However, the use of foliar fungicides in Missouri soybean fields has doubled among

farmers. We completed 67 on-farm strip trials across Missouri fields between 2018 and 2023 to evaluate foliar fungicide applications on soybeans for efficacy, yield, and profitability. Producers chose the fungicide and agronomic practices at each site. The trials consisted of two treatments: 1) no fungicide or 2) fungicide applied to R3 soybean. Each treatment has a minimum of five replicates. The study showed a yield increase of 121 kg ha⁻¹ in fungicide-treated strips compared to non-treated strips. However, this yield increase is not profitable given current application costs and soybean prices. We observed *Septoria* brown spot (*Septoria glycines*) at each site with disease severity ratings that ranged from <0.1 to 10.7 prior to fungicide application. We speculate that fungicide applications are fostering fungicide resistance in *Septoria*. Preliminary analysis of 2023 strip trials indicate a larger yield difference between fungicide-treated and non-treated strips across locations. We are evaluating weather, agronomic, and environmental factors to explore the relationships of these variables with yield responses.

Emerging bacterial diseases in wheat due to climate change and host shifts

A. Alhusays¹, K.D. Glover¹, E. Fitzpatrick¹, M.S. Sengar¹, M. Torres², C. Galvin¹, S. Solanki¹, J. Gonzalez¹, G. Ameen¹; ¹South Dakota State University, ²Southwestern College; **Abstract:** The impact of climate change on agriculturally important crop plants is multidimensional. Pathogen virulence evolution and emerging new host-pathosystems are some of its undesirable outcomes. In wheat, Bacterial leaf streak (BLS) disease is predominantly caused by *Xanthomonas translucens* pv. *undulosa* (*Xtu*). Wheat is one of the most important cereal crops grown in the world and its health is paramount to global food security. However, our recent findings establish that at least three morphologically similar bacterial species, *Xanthomonas translucens* pv. *undulosa* (*Xtu*), *Pantoea ananatis*, and *P. agglomerans* are pathogenic on wheat. Because their symptoms are indistinguishable, it may be worthwhile to refer to this as bacterial complex leaf streak disease (BCLS). We hypothesize that climate change could be the major evolutionary driving force, either favoring the pathogenic *Pantoea* population in the host microbiome or inducing horizontal gene transfer resulting in a host range shift from corn to wheat. We are analyzing PromethION sequencing data on multiple strains of *Pantoea* spp. to find evidence of evolutionary mechanisms in these interacting bacterial communities. We suggest that resistance sources that work for *Xtu* may not be useful for *Pantoea* spp., and we need to address this issue immediately. The evolution of these deceptive pathogens in future climate scenarios should be taken as an urgent task in hand.

Towards fine mapping and cloning of novel leaf and stem rust resistant genes in the Portuguese durum wheat landrace PI 192051

Md. Al Mamun¹, G. Valentini¹, U. Gill¹, M. Aoun²; North Dakota State University, ²Oklahoma State University; **Abstract:** Leaf rust, caused by *Puccinia triticina* Erikss. (*Pt*), and stem rust, caused by *Puccinia graminis* f. sp. *tritici* Erikss. & E. Henn (*Pgt*), are significant diseases of durum wheat (*Triticum turgidum* L) worldwide. An underutilized landrace PI 192051 that originated from Portugal is resistant to multiple races of leaf and stem rust pathogens collected from East Africa and the United States. QTL for leaf rust and stem rust resistance were previously described on chromosomes 4A and 7A. The objectives of our current research include fine-mapping of novel leaf rust and stem rust resistance genes on chromosomes 4A and 7A and functional validation of candidate genes using mutagenesis, gene silencing, and over-expression. For leaf rust, an F8 RIL population derived from Rusty x PI 192051 was used to validate flanking markers of the 4A QTL by phenotyping the population with *Pt* race BBBQD and genotyping with molecular markers spanning the QTL. New recombinants from the F2 population from the same cross were also identified using the flanking markers. Our phenotyping and genotyping data suggest that the leaf rust resistance gene is present in a 34.8 Mb interval flanked by markers IWA5123 and IWA1720. Validation of flanking markers for stem rust resistance QTL on 7A and high-resolution genetic mapping of leaf and stem rust resistance genes is underway. Cloning these genes will facilitate their integration into durum and hexaploid wheat cultivars.

Fine mapping of a QTL conferring disease susceptibility to spot blotch in barley

O. Amusan¹, S. Zhong¹, S. Yang², Y. Leng¹; ¹North Dakota State University, ²Northern Crop Science Laboratory; **Abstract:** Spot blotch, caused by *Bipolaris sorokiniana*, is an important disease of barley. Use of host resistance is the most effective way to manage this disease. Our previous study identified two quantitative trait loci (QTL) (*QSbs-1H-P1* and *QSbs-7H-P1*) conditioning susceptibility to spot blotch caused by isolate ND85F in barley line ND5883 using a recombinant inbred line (RIL) population derived from the cross between Bowman (resistant) and ND5883 (susceptible). In this study, we aimed to finely map *QSbs-1H-P1* flanked by two SNP markers spanning an ~460 Mb region on chromosome 1H. A cross was made between Bowman and a susceptible RIL carrying only *QSbs-1H-P1* but not *QSbs-7H-P1* derived from the Bowman/ND5883 cross, and the F₂ population was evaluated for infection responses to isolate ND85F. A total of 826 resistant F₂ individuals were identified and genotyped with peak and flanking markers of *QSbs-1H-P1* through the Kompetitive Allele-Specific PCR (KASP) assay. Additional SNP markers were developed based on genome sequences of Bowman and ND5883, thereby delimiting the QTL between two markers 1H_23462390 and 1H_63553355 covering a physical distance of ~40Mb. More SNP markers will be developed for fine mapping of *QSbs-1H-P1* with the goal of identifying the candidate gene for the QTL.

Unravelling the Role of Oxalic Acid Tolerance in Sunflower Resistance to Sclerotinia Basal Stalk Rot

S. Angidi¹, W. Underwood², J. Pasche¹; ¹North Dakota State University, ²USDA-ARS; **Abstract:** Basal stalk rot (BSR) of cultivated sunflower is caused by a necrotrophic fungal pathogen, *Sclerotinia sclerotiorum*, causing significant yield losses worldwide. Typical symptoms of BSR include wilting in aerial tissues and basal stem lesions, which are mimicked by treatment with oxalic acid (OA). This study focuses on understanding the role of OA in BSR development and identifying genetic factors responsible for OA tolerance. In our prior investigation, OA tolerance was identified in the sunflower inbred line HA 61, which also exhibited partial resistance to BSR. Therefore, we developed a recombinant inbred line population to 1) map quantitative trait loci (QTL) contributing to BSR resistance, 2) map QTL contributing to OA tolerance, 3) assess the correlation between OA tolerance and BSR resistance. The results of this study will provide valuable insights into the genetic mechanisms governing resistance to BSR and the specific genes responsible for OA tolerance. Understanding the genetic basis of OA tolerance can potentially lead to the development of BSR-resistant crop varieties, offering a sustainable solution to mitigate the economic impact of this devastating disease.

Molecular approaches to identify *Ascochyta rabiei* effectors

A. Arman¹, M. Seidl², M. Ebert¹; ¹North Dakota State University, ²Utrecht University; **Abstract:** Chickpea is the third most important food legume in the world after dry bean and pea, and North Dakota is one of the top four chickpea producing states in the US. One of the major yield limiting factors affecting the chickpea production is Ascochyta Blight caused by the necrotrophic fungus *Ascochyta rabiei*. Ascochyta Blight is a seed borne disease of the above-ground parts of chickpea plants. Plant pathogens deploy a range of secreted molecules, collectively known as effectors, to colonize the host and establish their niche. These effectors aid the pathogen in virulence, competition among other microbes, and/or nutrient acquisition. To identify effectors of *A. rabiei*, we have sequenced the genome of our most virulent *A. rabiei* strain APNS4. The APNS4 genome assembly has 30 contigs that adds up to a genome size of about 40.4 Mb. Additionally, we are pairing our whole-genome sequencing data with an RNA sequencing experiment, where we use different time points (3DPI, 6DPI, 9DPI, and 14DPI) to study differential gene expression throughout the infection progress. By identifying fungal effectors, we are hoping to get a better understanding of the infection mechanism of this economically important pathogen that will ultimately help with the development of resistant chickpea varieties.

B

Comparing spray coverage of unmanned aerial applications at two carrier volumes in corn.

W. Barlow¹, L. Warner¹, K.A. Wise¹; ¹University of Kentucky; **Abstract:** The application of pesticides from unmanned aerial vehicles (UAV) in the United States has increased in recent years and questions remain about application parameters required to achieve adequate spray coverage, in a given crop. A trial was established in Princeton, KY in 2023 to compare spray coverage on the corn (*Zea mays* L.) ear leaf from two different spray carrier volumes applied by a UAV. Aerial applications occurred using a DJI T10, with carrier volumes of 9.35 or 18.71 liters per hectare. Both treatments contained 1% v/v Tracer Hot Pink marking foam colorant. Kromekote paper was cut into 8.9 cm x 43.2 cm strips and attached to the ear leaf of three plants, chosen arbitrarily, in the two center rows of each plot. The DJI T10 applied the treatment to the center of each plot at a height of 3 meters above the canopy. After application, the cards were collected and scanned. The images were analyzed in Image J and Cell Profiler to determine percentage of card area covered with dye. The spray card coverage data were analyzed in SAS 9.4 using Proc GLMMIX. The average spray coverage on cards was 1.62% and 2.24% (Image J) and 1.22% and 1.64% (Cell Profiler) for the 9.35 and 18.71 l/ha, respectively. Spray coverage did not differ between carrier volumes (Image J $P = 0.3611$; Cell Profiler $P = 0.3951$). This trial will be repeated in 2024 with the addition of a fungicide to determine the effect of carrier volume on efficacy against foliar disease severity in corn.

Reaction of wheat progenitor and related species to barley bacterial leaf streak pathogen *Xanthomonas translucens* pv. *translucens*

G. Belen¹, U. Gill¹, Z. Liu¹, T. Baldwin¹, J. Singh¹, S. Xu²; ¹North Dakota State University, ²USDA-ARS, Cereal Crops Research Unit; **Abstract:** Bacterial leaf streak (BLS) is an important disease of wheat and barley. *Xanthomonas translucens* pv. *undulosa* (Xtu) causes BLS on both wheat and barley while *X. translucens* pv. *translucens* (Xtt) causes BLS only on barley. Although Xtt cannot cause disease on wheat, it remains unknown if Xtt causes disease on wheat progenitor and related species. In this study, a total of 268 accessions of *Aegilops tauschii* (the D-subgenome donor of common wheat), 222 accessions of *Triticum urartu* (A-subgenome donor of common wheat and durum) and 18 accessions of *Aegilops umbellulata* were screened against a local Xtt strain by infiltration. Only a small percentage of *Aegilops tauschii* accessions produced some degree of water-soaking, an indication of a susceptible reaction, whereas the majority of accessions showed chlorosis in the infiltrated area, indicating a resistant reaction. However, it was found that a significantly higher percentage of *Aegilops umbellulata* and *Triticum urartu* accessions were susceptible to Xtt. To our knowledge, this is the first report of Xtt causing disease outside of barley. The lines showing different reactions will be crossed to create segregating populations in order to map the genes that confer resistance to Xtt. Resistant gene identification will not only be useful for the barley breeding program but will also advance our knowledge on the molecular mechanisms underlying host range determination in cereal BLS disease systems.

C

Improving our ability to manage sorghum stalk rot through fungicide and spraying technologies practices in Kansas

L. Camara Vieira¹, R. Borba Onofre¹, T. Todd, K. Andersen-Onofre¹, J. Wisbey¹, C. Dinkel¹, M. Mariano¹, M. Kessler¹; ¹Kansas State University; **Abstract:** Stalk rot diseases threaten sorghum production worldwide

causing lodging, poor grain fill, and yield reduction. The objectives of this study were to evaluate fungicide timing and at-planting spray technologies to manage stalk rot diseases. The experiment was conducted in Russell, Kansas, planted on May 31, 2023. The treatments were arranged in a randomized complete block design with six replications and plots measuring 10 ft wide by 30 ft long. The target plant population was 35,000 plants/a, planted using a 4-row planter with 30-in row spacing. Stalk rot incidence was evaluated on 10 sorghum plants per plot prior harvest. Data were analyzed in SAS 9.4 using a mixed-effects analysis of variance ($\alpha = 0.05$). The treatments TRT3 (Xyway LFR fungicide 15.2 fl. oz./A, at-plant, 2x2), and TRT6 (Topguard EQ 5 fl. oz/A, at beginning of flowering), showed significant increases in productivity compared to the untreated control (TRT1), with respective average increments of 8.4 bu/a and 8.9 bu/a. Additionally, TRT7 (Adastrio 8 fl. oz/A, at the beginning of flowering) showed significantly higher yields compared to the untreated control (TRT1), TRT2 (Xyway LFR 15.2 fl. oz/A, at-plant, 2x2) and TRT5 (Xyway LFR 10.5 fl. oz/A, at-planting 2x2 + Topguard EQ 5 fl oz/A at beginning of flowering), with increases in yield of 11.35 bu/a, 8.69 bu/a, and 11.14 bu/a, respectively. Fungicide treatments had no significant impact on panicle size, greenness, or stalk rot incidence.

Effect of Fertilization on Soybean Sudden Death Syndrome Severity

L. Camara Vieira¹, T. Todd¹, R. Borba Onofre¹, E. Adele¹; ¹Kansas State University; **Abstract:** Sudden death syndrome (SDS), is one of the most yield-limiting soybean diseases in Kansas. In this study we evaluated the effect of different fertilizer and rates on the severity of SDS. The experiment was conducted at the Kansas River Valley Experiment Field, Topeka, Kansas in an annual corn-soybean rotation. Nitrogen (0, 120, 160 lbs/a), Phosphorus (0, 30, 60 lbs/a), and K (0, 150 lbs/a) treatments were applied before corn planting in a factorial complete block design with four replications. Plots were 15-ft wide and 30-ft long, consisted of six rows spaced at 30 inches. Asgrow 3833 soybeans were seeded on May 21, 2014, Asgrow 3731 on May 10, 2016, and Asgrow 38X6 on May 7, 2018, at 140,000 seeds per acre. At R6 maturity, SDS severity was assessed by rating leaf area with symptoms per plot, while soybean plant height was measured to the highest node with seed-bearing pods. The two center rows of each plot were harvested for yield. The data were analyzed in SAS 9.4, using a mixed-effects analysis of variance. According to the results, only phosphorus had a significant effect on SDS. As the phosphorus rate increased, the severity of SDS decreased. Additionally, phosphorus had a significant impact on yield and plant height, with higher doses resulting in a 20 bushel per acre increase in yield compared to the untreated control. Furthermore, we found a negative interaction between SDS and yield across all treatments: as SDS levels increase, yield decreases.

D

Statewide Evaluation of Fungicides for Frogeye Leaf Spot Control on Soybean in Iowa

N. Dangal¹, D. Mueller¹, S. Wiggs¹, J. González-Acuña¹, E. Nieto-Lopez¹; ¹Iowa State University; **Abstract:** Frogeye leaf spot (FLS) is a major yield-limiting disease of soybean in the United States, especially in the southern states. Uniform statewide field trials were conducted across seven geographic locations in Iowa from 2018 to 2023 to evaluate the effect of 29 different fungicides in controlling foliar diseases and yield response. Severity of any foliar disease was recorded throughout the growing seasons. However, final disease severity at the R6 stage and yield at maturity were used for analysis in this study. Out of 42 site-years, FLS was present at 33 locations with an average severity of 0.01 to 14.4% in the untreated control (UTC). Septoria brown leaf spot was also recorded in a few locations, but it was excluded from further analysis due to small sample size. A network meta-analysis was conducted separately to evaluate the treatment effect on FLS severity and yield. Results showed that the effect of fungicides on FLS severity and yield response varied significantly. Among the fungicides, several, including many premixes of active ingredients from different FRAC groups, had significantly less FLS and resulted in improved yield gains compared to the UTC. These findings suggest that various fungicides

effectively controlled FLS and safeguarded yield showcasing these options as significant tools for managing this important foliar disease.

A network meta-analysis of yield responses to timings of fungicide application for wheat stripe rust in the U.S.

C. Dias da Silva¹, E. De Wolfe¹, K. Anderson-Onofre¹, R.P. Lollato¹, N. Giordano¹; ¹Kansas State University; **Abstract:** Stripe rust (*Puccinia striiformis* f.sp. *tritici*) is one of the most important threats to wheat production worldwide. Fungicides are a critical tool for stripe rust management however there remain open questions about optimal timings for applications. The objectives of our study were to: 1) Compare the efficacy of five fungicide application timings for stripe rust control; and 2) Evaluate the effect of different modes of action. The application timings evaluated included T1 (Feekes 4 – 7); T2 (Feekes 8 – 10); T3 (Feekes 10.1 +); T4 (T1 + T2); and T5 (T1 + T3). We conducted a network meta-analysis of Plant Disease Management Reports. A systematic search resulted in 130 publications. Publications were removed from the dataset according to predetermined selection criteria resulting in 82 studies. We observed a significant difference between the application timings ($p < 0.0001$). For single applications, T2 resulted in an overall better yield improvement (895.66 kg/ha) than T1 (876.43 kg/ha) and T3 (873.16 kg/ha). For the dual applications, T5 resulted in an overall better yield improvement (978.16 kg/ha) than T4 (933.02 kg/ha). For the moderators, year ($p = 0.0010$) and state ($p = 0.0075$) had a significant effect on yield. Wheat growth habit (spring vs. winter) did not significantly impact yield response ($p = 0.1654$). These findings provide critical information that can be used by wheat producers to better time fungicide applications for the management of stripe rust.

Rotating DMI fungicides within spray programs to manage *Cercospora beticola* in sugar beet

J. Deleon¹, A. Chanda¹, A. Lien¹; ¹University of Minnesota NWROC; **Abstract:** Sugar beet growers rely upon demethylation inhibitor (DMI) fungicides to manage *Cercospora* leaf spot (CLS), caused by *Cercospora beticola*. In recent years, *C. beticola* isolates have developed patterns of cross-resistance to select DMI fungicides. In 2023, a field trial was conducted to evaluate the efficacy of 4-spray programs with two alternating DMI fungicides, beginning with either tetraconazole (TET) or mefentrifluconazole (MEF) in the first application and rotating with either a pre-mix of difenoconazole (DIF) + propiconazole or prothioconazole (PRO) in the third application. Weekly CLS severity ratings were used to calculate the sAUDPS. Additionally, 5 leaves were collected from each plot on Sept 18 to assess CLS severity and obtain four single-conidia isolates. The effective concentration of fungicide that inhibits 50% of growth compared to the control (EC50) was determined for TET, MEF, DIF, and PRO using a microplate-based assay. A multiplex probe-based qPCR assay was used to distinguish between five potential haplotypes (A-E) of *CbCyp51*, which encodes the DMI target enzyme. CLS severity was significantly lower in all fungicide treatments compared to the nontreated control. Haplotype E was predominant among the isolates which is known to be associated with decreased sensitivity to all four DMI fungicides. Overall, monitoring fungicide resistance is essential for ensuring the continued effectiveness of DMI fungicides and minimizing economic losses.

Map-based cloning of the Nec50 gene which regulates programmed cell death in barley

A. D'Eustachio¹, Z. Liu¹, S. Yang²; ¹North Dakota State University, ²Northern Crop Science Laboratory; **Abstract:** Biotrophic pathogens require living host cells, and the elicited programmed cell death deprives nutrients from them, suppressing their colonization process. In contrast, necrotrophic pathogens kill host cells and feed on dead tissues and PCD benefits them. Understanding the PCD mechanism is important to manipulate resistance and susceptibility pathways for the purpose of disease control. However, PCD signaling remains largely unknown. Lesion mimic mutants (LMM), exhibiting necrotic lesions spontaneously without pathogen infection, provide a powerful tool to identify genes or cellular events that regulate cell death in plants. We have cloned the Nec50 gene, which is a negative suppressor of PCD in barley. Loss-of-function mutation in Nec50 caused by EMS mutagenesis results in the formation of

tan to light brown lesions on the leaf at the jointing stage leads to premature leaf senescence. Genetic mapping delimited Nec50 to a 3M-bp region on chromosome 7H. RNA-seq analysis and mutseq technique identified a single point mutation of C→T transition at the 5'UTR of HORVU.MOREX.r3.7HG0668180, which encodes a putative O-fucosyltransferase 15-like protein. CRISPR-mediated gene mutagenesis of the nec50 mutant phenocopied, validating that HORVU.MOREX.r3.7HG0668180 is the Nec50 gene. Therefore, cloning of Nec50 sheds light on the molecular mechanisms of plant defenses.

A survey of foliar diseases affecting Cucurbitaceae in South Dakota.

J.P. Waduwarage Dona¹, S. Toporek¹; ¹South Dakota State University; **Abstract:** Cucurbit crops make up the largest vegetable acreage in South Dakota, however information on the prevalence and distribution of foliar diseases that affect them is unknown. In 2023, a total of 12 small, diversified vegetable farms were surveyed across eastern, central, and western South Dakota. Up to 40 symptomatic leaves were collected from bottle gourd, melon, cucumber, pumpkin, squash, watermelon or zucchini plants from each farm. Preliminary microscopic identification of sporulating leaves ($n = 422$) identified spores of *Alternaria* sp. (95% of leaves), *Colletotrichum* sp. (3.6%), *Podosphaera xanthii* (0.7%), and *Stagonosporopsis* sp. (0.5%). Symptomatic leaf tissues were surface sterilized with 5% bleach for 30 seconds and plated onto ¼ PDA amended with 40 mg/L streptomycin and 40 mg/L chloramphenicol. So far, isolates of nine species have been identified across 221 recovered isolates, of which 79.2 % were identified as *Alternaria* sp., *Fusarium* sp. (10.9%), *Stagonosporopsis* (4.5%), and other species (3.2%). *Alternaria* sp. are most commonly recovered from diseased cucurbit leaves, but also appear to be present as a common contaminant, possibly leading to false microscopic identification. A systemic characterization of fungal disease prevalence and causal pathogens will help direct better management strategies tailored to South Dakota growers.

F

Changes in phenylpropanoids in two sweet sorghum lines differing with contrasting responses to stalk pathogens

D. Funnell-Harris¹, G. Sarath¹, S. Sattler¹, E. Scully¹, N. Palmer¹, P. O'Neill¹, L. Bernhardson¹, M. Khasin¹; ¹USDA-ARS; **Abstract:** Sweet sorghum [*Sorghum bicolor* (L.) Moench] lines M81-E and Colman differed in responses to stalk rot pathogens *Fusarium thapsinum* and *Macrophomina phaseolina*: M81-E had shorter mean lesion lengths than Colman when inoculated with either fungus. The transcriptional and secondary metabolic responses to pathogen and mock inoculations were examined at 3 and 13 days after inoculation (DAI) in plants of these lines. Transcripts encoding monolignol biosynthetic and modification enzymes were associated with mock responses of both lines at 3 DAI. Monolignol biosynthetic genes were coexpressed with transcriptional activator SbMyb76 in all Colman inoculations, but only after *M. phaseolina* inoculation in M81-E. In mock-inoculations, defense-related genes were expressed at higher levels in M81-E than Colman. Line, treatment, and timepoint differences in phenolic metabolites did not account for lesion differences. Generalized additive models could relate metabolites to lesion length and quantitatively model disease progression. Only in M81-E, sinapic acid levels positively predicted lesions at 3 DAI when cell wall-bound syringic acid was low and soluble caffeic and lactic acids were high. These results provide potential gene targets for development of stalk rot resistant sweet sorghum varieties with high yields and quality of biomass and sugar.

G

Unraveling the Early Die Disease Complex in Potato

D. Gill¹, J. Pache¹, A. Sarwar¹; ¹North Dakota State University; **Abstract:** The potato early die disease complex (PED) causes premature senescence, yield, and quality loss in potatoes worldwide. *Verticillium* spp., *Colletotrichum coccodes*, and *Pratylenchus penetrans* all contribute to the development and severity

of the disease. Management options are limited due to pathogen biology and commercial-scale feasibility. Soil fumigation with metam sodium or chloropicrin is the most effective approach in suppressing the disease. Under high pathogen infestations, effects of fumigation may not be sufficient to economically reduce disease. Therefore, the disease remains a prominent factor in limiting yield each growing season. With absolute control of PED not achieved, further understanding of soil fumigation performance on high disease pressure production fields can improve the way we approach mitigating the disease. This research strives to contribute useful information on how *V. dahliae* and *C. coccodes* infect and colonize potatoes under commercial treatment of metam sodium or chloropicrin in grower fields. Insights are guided by data collected from in-season field observations and sampling along with molecular techniques for quantifying pathogen colonization of stem tissue over time. Providing molecular and physical observations of how PED persists in potatoes at production scale is valuable in assessing current management techniques and uncovers potential focus points for future research.

Effect of Cereal Rye Cover Crop Cultivars on Corn Seedling Disease

L. Gonzalez¹, A. Robertson¹; ¹Iowa State University; **Abstract:** Cover crops (CC) in agricultural systems provide numerous environmental services. Cereal rye (CR) is the most widely used CC in the Midwest, USA. However, losses in corn yield have been reported following a CR CC. Losses in yield may be due to low plant-available nitrogen (N) after CR termination, resulting in plants that may be more susceptible to seedling diseases caused by *Pythium* and *Fusarium* species. Greater CR biomass results in wider C:N, and consequently increased N immobilization. Cultivars of CR produce different amounts of biomass; thus, we hypothesized CR cultivar may affect the incidence and severity of corn seedling diseases. A growth chamber experiment was done in which corn was planted following four cultivars of CR (Serafino, Elbon, Aroostook, Rymin) and a control (No-CR CC). CR biomass, root rot severity, and identification of root pathogens were evaluated. Serafino produced the most biomass of all cultivars ($P > 0.05$). No differences in biomass occurred among the other cultivars. Corn seedlings planted after Serafino had greater root rot severity ($P < 0.05$) than the control and other treatments. *Fusarium oxysporum* was the most frequently recovered pathogen accounting for 75% of the total isolates. These results suggest CR CC biomass production affects corn seedling disease severity. Field trials are in progress. Data from these studies will inform best management strategies, specifically choice of CR cultivar, to mitigate corn yield loss after a CR CC.

The Frogspotter Tool: Repurposing Fungicide Trials into Predicting Frogeye Leaf Spot in Soybean in the US

J. González-Acuña¹, T. Allen², D. Mueller¹, S. Markell³, R. Webster³, D. Telenko⁴, D. Mangel⁵, D. Smith⁶, D. Malvick⁷, M. Bish⁸, C. Bradley⁹, T. Faske¹⁰, H.M. Kelly¹¹, P. Price¹², J. Scherer³, E. Sikora¹³; ¹Iowa State University, ²Mississippi State University, ³North Dakota State University, ⁴Purdue University, ⁵University of Nebraska - Lincoln, ⁶University of Wisconsin, ⁷University of Minnesota, ⁸University of Missouri, ⁹University of Kentucky, ¹⁰University of Arkansas, ¹¹University of Tennessee, ¹²Louisiana State University, ¹³Auburn University; **Abstract:** *Cercospora soja*, the causal agent of frogeye leaf spot (FLS) is a major pathogen of soybean. Plant pathology research groups in US universities have been setting up field trials and collecting data for several years, assessing the efficacy of different fungicides in controlling soybean diseases, including FLS. Fungicide trials from 2015-2022 were compiled from 7 US states ($n = 158$). Disease observations ($n = 188$) were classified as binary variables if FLS developed or not and used as response variables for developing logistic regression models. The dataset was separated using a 70:30 split to create the training/testing datasets. Hourly site-specific weather data were extracted, aggregated, and used to create 30-, 21-, 14-, and 10-d moving averages. From correlation analyses, the total number of hours with relative humidity $> 75\%$ was highly correlated with FLS development, and the highest correlation coefficient was 0.43 ($p < 0.001$) using the 21-day moving averages. Logistic regression models were created and evaluated against the testing dataset. The best-performing model was integrated into a decision support system (Frogspotter), currently being evaluated. In 2023, three risk thresholds were evaluated across eight states. The thresholds had an

effect on disease at three sites in 2023. The Frogspotter tool is a step toward a more precise approach to managing FLS and fungicide stewardship, which can help soybean farmers make more informed fungicide use decisions.

Mapping of Pathogen Aggressiveness in Populations of *Fusarium graminearum*

S. Gopisetty¹, C. Toomajian¹, S. Chowdury¹; ¹Kansas State University; **Abstract:** *Fusarium graminearum* (Fg) is the predominant cause of Fusarium Head Blight of wheat in the US, posing a risk to both food safety, due to mycotoxin contamination of grain, and crop yield. Understanding the genetic variability and population biology of Fg is important in developing sustainable disease management tools. Genotyping results from our Fg population genetics studies can be used to associate genetic variants with pathogen potential traits like disease severity and deoxynivalenol (DON) production. To identify genes with functional variation that contributes to variation in these traits within various Fg populations, we conducted greenhouse experiments to assess disease severity and DON production and tested for associations between these traits and thousands of genetic variants. Spring wheat varieties Wheaton (susceptible) and Rollag (moderately resistant) were grown in the greenhouse and single spikelets per head were inoculated with one of 150 Fg field isolates. Disease severity of each isolate on each variety was measured as the percentage of diseased spikelets per inoculated spike at 14 and 21 dpi. By testing for associations between genetic variants across the genome and disease severity and DON production using GWAS methods, we can detect genetic factors influencing isolate aggressiveness. Our results will suggest targets for pathogen control and will lead to the development of markers for tracking the spread of more pathogenic lineages.

Occurrence of five major viral pathogens of hemp in Wisconsin

D. Grunwald¹, R. Witherell¹, S. Stroschein¹, S. Ellison¹; ¹University of Wisconsin - Madison; **Abstract:** Viruses limit hemp yield and profitability. A survey of hemp (*Cannabis sativa* L.) was conducted from 2022-2023 for viruses in Wisconsin research plots. 166 samples from three genetically distinct classes (37 Feral, 72 Grain/Fiber, and 57 high cannabinoid) were collected at two time points, representing approximately 1 month and 4 months post-planting. Samples were tested for tobacco streak virus (TSV) and alfalfa mosaic virus (AMV) by ELISA, and for beet curly top virus (BCTV), citrus yellow vein associated virus (CYVaV), and *Cannabis sativa* mitovirus 1 (CasaMV1) by RT-PCR. All viruses were represented by at least one sample collected. Significant effects of timepoint and classification were observed by using generalized mixed linear models. Greater incidence of CasaMV1 was observed in grain/fiber and high cannabinoid varieties relative to feral varieties. TSV incidence was significantly greater at the later timepoint. There was an interaction between classification and timepoint for AMV, with greater incidence occurring in grain/fiber varieties at later timepoints. CYVaV and BCTV were infrequently detected. Co-infection of viruses occurred in around 33% of samples. To our knowledge, this is the first viral survey of hemp in Wisconsin. These results will serve to inform local growers as to the viral reservoir capacity of hemp, which may inform integrated pest management schemes to mitigate the spread of these viruses to other row crops.

H

Screening diverse maize lines for resistance to *Pythium* root rot

H. Hall¹, M. Krone¹, S.X. Mideros Mora¹, T. Jamann¹; ¹UIUC; **Abstract:** *Pythium* root rot in maize is caused by many oomycete species, including *Globisporangium ultimum* var. *ultimum* (Guu). There are no reports of resistance genes effective against *Pythium* root rot. Our research aims to discover resistance by screening a diverse set of maize inbred lines. We hypothesize that resistance to *Globisporangium* species varies among maize lines and is heritable. The seeds of thirty diverse inbred maize lines were plated on water agar and inoculated with Guu. Root lengths were measured after four days. Nine maize lines were selected for greenhouse screening. Guu inoculum was made using sand and cornmeal mixed into containers with soil. After fourteen days, the plants were rated for disease severity and washed and dried to measure root mass and length. There was a wide range of resistance across lines in in vitro experiments.

Many of the lines susceptible in the in vitro experiments remained susceptible in the greenhouse, but the moderately resistant tropical lines CML103 and CML333 had increased root weight and length when inoculated with Guu. There was no correlation between in vitro and greenhouse root lengths, but there was a high correlation ($r = 0.71$) between replicates of the greenhouse screen. Future research will include screening additional *Globisporangium* species and diverse environmental conditions. Our results suggest multiple resistance mechanisms to Pythium root rot, offering novel disease management and crop improvement strategies.

Adapting Techniques for Evaluating Cercospora Leaf Spot in a Field Nursery for Varied Climates

L. Hanson¹, H. Corder², T. Goodwill¹; ¹USDA-ARS- SBRU, ²Michigan State University; **Abstract:** Cercospora leaf spot (CLS) disease nurseries were conducted in Colorado (CO) since the 1950's. CO was a suitable place to evaluate plant response to the pathogen, *Cercospora beticola*, since that region had few other leaf spot issues. Because of a housing development boom and irrigation water limits in CO, the nursery needed to be relocated. In 2006, the CLS nursery was moved to Michigan (MI) where the climate is humid and CLS is a perennial issue. The MI climate, however, also is conducive to the development of other foliar diseases, which necessitated modification of techniques used to create a successful CLS nursery. The Ruppel and Gaskill methods were initially used in Michigan, but key changes resulted in improved consistency. A change recommended for any production area the use of conidia counts instead of leaf:water ratios to determine inoculum levels. In addition, in MI, irrigation in the nursery is applied minimally, susceptible spreader rows are planted perpendicular in the field, and leaves collected for inoculum are selected to avoid other common leaf spots. In 2009 and 2010, the Ruppel and Gaskill methods and the MI methods were compared. In both years, the peak of the epidemic was at least 10 days earlier using the MI methods. The nursery has continued to show effective disease development under varied summer conditions. A closer look at how changes to the Ruppel and Gaskill methods have improved the performance of the CLS nursery will be discussed.

Quantifying Soybean Cyst Nematode Resistance Breakdown: A Real-time Assessment of 'Peking' and PI 88788

C. Hicks¹, P. Karki¹, D. Mangel¹; ¹University of Nebraska - Lincoln; **Abstract:** Soybean cyst nematode (SCN; *Heterodera glycines* Ichinohe) is the most economically significant soybean pathogen in Nebraska. SCN can cause significant yield loss but is often unmanaged due to a lack of noticeable above-ground symptoms. The primary management practice for SCN is the use of genetic resistance sources. Although there are many known plant introductions that can serve as genetic resistance sources, only two are widely available for growers: PI 88788 and PI 548402 (Peking). Currently, PI 88788 is used in approximately 95% of SCN-resistant varieties and this has led to reduced efficacy or resistance breakdown. As 'Peking' use increases in more varieties, there are concerns about its overuse and potential breakdown. The goal of this project is to demonstrate the resistance source breakdown in real-time by quantifying the change in nematode reproduction across several 28-day generations of the pest. Based on the effectiveness of 'Peking' resistance, results are likely to show that it degrades quicker than PI 88788. However, a rotation of the two resistance sources could slow resistance breakdown in both varieties. This project aims to encourage Nebraska soybean producers to use rotation strategies and extend the life of resistance sources.

I

***Alternaria alternata* internal rot of *Capsicum* species, an understudied pathosystem causing losses in South Dakota peppers.**

K. Imai¹, S. Toporek¹; ¹South Dakota State University; **Abstract:** *Alternaria alternata* causes an internal rot of *Capsicum* spp. by infecting flowers during anthesis, but this disease has never been reported in the United States. The objective was to identify associated fungi during the 2023 growing season from one

pepper farm in South Dakota with varietal internal rot disease incidence ranging from 3 to 50%. One symptomatic pepper each from ten plants representing ten varieties of *Capsicum chinense*, one asymptomatic pepper each from four plants representing four varieties, and three flowers each from six plants representing six varieties were sampled. *Alternaria alternata* was recovered from all ten symptomatic peppers, two and one of which also contained *Epicoccum* sp. and *Mucor* sp., respectively. *Cladosporium* sp., *Fusarium* sp., *Mucor* sp. and *Stemphylium* sp. were recovered from non-symptomatic peppers. In flower samples, *Alternaria alternata* and *Epicoccum* sp. were recovered from all six varieties and *Mucor* sp. was recovered from flowers of only one variety. Overall, *Alternaria alternata* was strongly associated with internal rot disease incidence and was present in all flowers. This preliminary information will guide future work on pathogenicity, intervening management strategies, and non-destructive detection.

Reaction of Hard Red Spring Wheat Cultivars and Breeding Lines to Multiple Leaf Diseases

Md. M. Islam¹, A. Friskop¹, J.P. Cook², A. Green¹, A. Friskop¹, Z. Liu¹; ¹North Dakota State University, ²Montana State University; **Abstract:** Hard red spring wheat production in the Northern Great Plains can be affected by several leaf diseases including tan spot (TS), Septoria nodorum blotch (SNB), and bacterial leaf streak (BLS). In this study, we evaluated 51 hard red spring wheat cultivars or elite lines from variety trail in North Dakota and 137 breeding lines from Montana State University (MSU) for their reaction to these diseases and the necrotrophic effector ToxA. The goal was to identify the sources of resistance and understand the role of ToxA in disease development. Approximately, 21% and 40% of cultivars or elite lines were resistant to TS and SNB, respectively. A relative low percentage (18%) of ToxA sensitivity was identified in the MSU breeding lines, and we observed a higher percentage of lines in it with resistance to the two fungal diseases. Overall, ToxA sensitivity was highly correlated ($r = 0.80$) to SNB susceptibility compared to that ($r = 0.27$) for TS susceptibility. The majority of cultivars and breeding lines were susceptible to BLS and only one breeding line showed a good level of resistance as the resistant check Boost. The work provides useful information for wheat growers and breeders in cultivar/line selections and confirms the important role of ToxA playing in both fungal diseases.

J

Assessment of Soil Microbiome Adding to Nitrogen Use Efficiency in Soil to Optimize Nitrogen Fertilizer Application in Winter Wheat

R. Jain¹, C. Graham¹, S. Solanki¹, G. Ameen¹, S. Sharma¹; ¹South Dakota State University; **Abstract:** Efficient utilization of available soil nutrients by plants heavily relies on ubiquitous yet specialized interacting microbiome components. Specific to nitrogen (N), free-living soil bacterial communities harboring mineralization pathway genes make N available to plants while modulating losses to environment, enhancing nitrogen use efficiency (NUE) and soil health. This research aims to quantify genes involved in N mineralization, microbial community diversity analysis, and to create an improved soil health index for increasing NUE in winter wheat. Winter wheat was planted at two South Dakota research locations i.e. Quinn and Sturgis treated with four N application rates: 0, 50, 100, and 150 lbs. N/acre with two soil depths 0 to 3 and 3 to 6 inches to evaluate microbial activity and gene expression at the start of maximum N uptake in the plant. Extracted soil DNA from these samples, after normalization was used for qPCR using *AmoA*, *Urec*, *ChiA*, and *Apr* genes and subsequent metagenomics analysis based on 16s amplicon sequencing. Our initial qPCR results on *AmoA* and *Urec* suggest that higher microbial activity for N-cycle genes occur at 50 and 100 lbs N/acre at 0-3 inches depth. Currently, we are analyzing the full-length 16s data on the EMU pipeline to identify community modulation due to added nitrogen. This research would reduce the environmental footprint of chemical-based fertilizers, further contributing to improved soil health and environmental sustainability.

K

Sensitivity of Sugar Beet-Derived *Cercospora beticola* Isolates to Different Fungicide Classes

S. Kalika-Singh¹, M.F.R. Khan¹, L. del Rio Mendoza¹, M. Aderoju¹, S. Bhandari¹; ¹North Dakota State University; **Abstract:** *Cercospora* leaf spot (CLS) caused by *Cercospora beticola*, is a severe foliar disease affecting sugar beet in the Red River Valley. The disease can reduce yields and sucrose content by up to 50%. Managing CLS effectively often involves using resistant cultivars and timely fungicide application, but *C. beticola* has developed resistance to various fungicide classes, e.g., Organotin, Demethylation Inhibitors (DMI), and Quinone Outside Inhibitor (QoI). This study investigated the response of 40 *C. beticola* isolates obtained from a sugar beet field in Foxhome, Minnesota, during the 2022 growing season. The isolates were grown on CV-8 media amended with concentrations of Pyraclostrobin (QoI), Prothioconazole (DMI), Copper hydroxide and Mancozeb (Multisite), and Triphenyltin hydroxide (Organotin) ranging from 0.01 to 10ppm. After 14 days of incubation in the dark at 23°C, colony diameters were measured at two perpendicular directions. This study was repeated twice, each time with two replications. The results demonstrated that only Mancozeb and Triphenyltin hydroxide suppressed isolate growth at the range of concentrations tested. All other fungicides proved ineffective. Ongoing research aims to find the optimal concentrations of ineffective fungicides needed to inhibit isolate growth effectively. Future research will involve conducting greenhouse experiments and mutation analysis to better understand how these isolates respond to different fungicides.

Distribution and virulence phenotypes of Soybean cyst nematode (*Heterodera glycines*) populations in South Dakota

N. Karki¹, M. Shires¹, C. Tande¹, C. Kotaska¹; ¹South Dakota State University; **Abstract:** Soybean cyst nematode (*Heterodera glycines* Ichinohe; SCN) is the most economically important soybean pathogen in the United States. The basis of effective management of nematodes lies in the precise and rapid diagnosis of the nematode population in the field. HG typing documents the diversity of SCN populations and sources of resistance for management. To determine the distribution of HG types in South Dakota, 219 samples were collected and processed for extraction of eggs and cysts with counts ranging from 250-56,000 eggs/100 cc of soil. 14 samples with counts above 10,000 eggs/100 cc of soil were HG typed on seven indicator lines and a susceptible check, Williams 82. Preliminary data suggests all tested samples were virulent on indicator line PI 88788, the major source of resistance for soybean cultivars. Nearly 57%, 79%, and 100% of tested populations could reproduce on indicator lines PI 548402 (Peking), PI 209332, and PI 548316 (Cloud) respectively. HG type 1.2.5.7 is the most prevalent virulent phenotype in the state. Multiple samples were HG typed from Brookings and Moody counties, which revealed variation in the HG type within counties, leading to the need for more detailed testing to understand the true variation in HG types across the state. Future steps for this project are to sample and HG type SCN from all major soybean producing counties in the state to track the development of nematode resistance.

Soybean Cyst Nematode (*Heterodera glycines* Ichinohe) Virulence Assay Against Resistance Sources in Nebraska

P. Karki¹, C. Hicks¹, D. Mangel¹; ¹University of Nebraska - Lincoln; **Abstract:** Soybean cyst nematode (SCN), one of the major yield-limiting pathogens of soybean, is currently found in 59 counties of Nebraska that produce more than 93% of the state's soybean. Often, yield loss occurs without any above-ground symptoms, which makes it difficult to manage. The primary management option is the use of SCN-resistant varieties. Several soybean plant introductions serve as resistance sources against SCN. Unfortunately, the single source, PI 88788, is used in approximately 95% of resistant varieties. Past studies have found that the prolonged use of PI 88788 has increased the virulence status of SCN. The objective of our study is to assess the virulence status of Nebraska SCN populations on resistant varieties and assign SCN HG type. To complete this objective, we randomly selected soil samples submitted by

the Nebraska soybean producers in 2021, 2022, and 2023 through the Nebraska Soybean Board's free SCN test program. So far, the results have indicated the dominance of HG type 2.5.7, suggesting increasing reproduction of SCN in PI 88788, PI 209332 and PI 548316 resistance source. As the study continues, we hypothesize that the resistance from PI 88788 is breaking down. The results from this study aim to increase the number of Nebraska producers who use effective management practices against SCN.

Spontaneous chlorate resistance mutations in *Fusarium verticillioides*

M. Kaur¹, C. Toomajian¹, J. Leslie¹, T. Krska¹; ¹Kansas State University; **Abstract:** Mutations drive the adaptation of pathogenic species, and understanding mutation patterns allows our understanding of their adaptability to environmental changes. We studied spontaneous mutations inactivating the nitrate reductase (*nit1*) gene in *Fusarium verticillioides* when grown in the presence of chlorate. Spontaneous *nit1* mutants (116) were selected following growth on media containing 1.5% chlorate and the mutant *nit1* genes were sequenced. In total, 99 sites were mutated in the gene, with no more than 4 mutants per nucleotide site. The gene showed 69% single nucleotide polymorphisms (SNPs), 13% insertions, and 18% deletions. Insertions and deletions led to frame-shifted translation and presumptive protein biosynthesis termination. Of the 81 SNPs, 39% generated premature stop codons, 23% altered amino acids near essential sites, and 38% changed amino acids not previously known to be essential for enzyme activity. We also sequenced 1 kb of the *fum1* gene, which was not under selection pressure in this experiment, in 92 mutants as a control. No mutations were detected in the *fum1* sequences. Although chlorate is not mutagenic per se, *F. verticillioides* responds to its toxicity by mutating proteins involved in its metabolism. The distribution and limited size of mutations in the *nit1* genes are consistent with errors in DNA replication and/or repair systems. Such errors could enable *F. verticillioides* to rapidly adapt and survive environmental stresses.

Genetic mapping of a recessive resistance to stripe rust in barley

S. Kaur¹, Z. Liu¹, S. Yang²; ¹North Dakota State University, ²Northern Crop Science Laboratory, Edward T. Schafer Agricultural Research Center; **Abstract:** Barley (*Hordeum vulgare* L.) is one of the most important cereal crops grown worldwide. Stripe rust caused by *Puccinia striiformis* f. sp. *hordei* (Psh) is one of the major barley diseases in the United States. This disease can cause seed quality reductions and yield losses reaching up to 70% and is managed via targeted breeding efforts which aim to stack resistance quantitative trait loci (QTL). One recessive gene, named rps.gz which was reported in the Ethiopia cultivar Grannelose Zweizeilige (GZ), confers broad-spectrum resistance to stripe rust. Here, we developed a Steptoe x Grannelose Zweizeilige (GZ) biparental population (F2) and performed QTL analysis. A selected panel of 196 F2 progenies were phenotyped with Psh race 33 at the seedling stage in the greenhouse and were genotyped with the 20K barley SNP markers. The obtained segregation ratio for resistance/susceptible fits 1:3, indicating that the disease phenotype is controlled by a single dominant susceptibility gene. Interval mapping revealed a single QTL located on chromosome 4H with a logarithm of the odds (LOD) score of 8.9. This QTL was flanked by two markers, AVRIG07556 and AVRIG07785, which delimited the gene to a 32-Mb region. Molecular markers were designed to narrow down the gene region. We aim to pinpoint this QTL, design gene-specific/diagnostic markers, and clone the gene, thus providing a valuable tool for breeding programs to control stripe rust in barley production.

Influence of Row Spacing, Plant Population, and Variety Selection on Sudden Death Syndrome of Soybean in Kansas and Iowa

M. Kessler¹, R. Borba Onofre¹, D. Mueller², N. Dangal²; ¹Kansas State University, ²Iowa State University; **Abstract:** Sudden death syndrome (SDS), most commonly caused by the fungal pathogen *Fusarium virguliforme*, poses a significant threat to soybean production. The disease is favored when planting occurs in cool, wet soils, and is exacerbated by post-planting rainfall. Growers rely on pre-planting management strategies, including resistant cultivars and seed treatments, with no control options after planting for SDS. We assessed the impact of agronomic practices on SDS through on-farm trials in Kansas and Iowa, which included evaluating row spacing, plant population, and variety selection.

Currently, the influence of these practices on disease development and suppression is not fully understood. Preliminary results indicate that row spacing, plant population, and soybean varieties play an important role in managing this disease. Higher disease incidence was observed with higher plant populations and narrower row spacing, while lower disease incidence was associated with resistant varieties. Further analysis of our findings will aid in making optimal SDS management decisions.

Genetic Study of Tar Spot Outbreak in the Great Plains

C. Kotaska¹, S. Enter¹, N. Karki¹, T. Mues², M. Shires¹, M. Bish³, R. Borba Onofre⁴, T. Jackson-Ziems², C.L. Strunk¹; ¹South Dakota State University, ²University of Nebraska - Lincoln, ³University of Missouri, ⁴Kansas State University; **Abstract:** *Phyllachora maydis*, the causal agent of tar spot on corn, is estimated to have caused a loss of 38.1 million bushels from 18 corn growing states in the United States and 3.9 million bushels in Ontario, Canada in the 2023. Tar spot was first reported in Indiana in 2015, Canada in 2020, and the Great Plains region in 2021. After widespread outbreaks of the disease were noted in 2022 and 2023, the question of genetic diversity and adaptation to the Great Plains region was raised. Symptomatic corn samples from Kansas, Missouri, Nebraska, and South Dakota were collected in the 2022 and 2023 growing seasons. Total nucleic acids were extracted utilizing a modified CTAB method and PCR performed with ITS 4 and ITS 5 primers. The positive PCR products were Sanger sequenced; sequences were aligned in Geneious Prime 2023.1 and analyzed for single nucleotide polymorphisms (SNP). There were no SNP's present in any sequences and most samples were at least 99% match to *P. maydis* reference sequences in NCBI GenBank. For further examination, we are currently performing genetic analysis of gene regions for partial small subunit, partial large subunit, 5.8S subunit, internal transcribed spacer 1, internal transcribed spacer 2, partial glyceraldehyde dehydrogenase, and partial actin. It is hoped that these additional genomic regions will reveal if there are any differences in tar spot within the Great Plains region, indicating adaptation of the fungus to the unique growing area.

L

Microplate-based assessment of DMI fungicide sensitivity in *Cercospora beticola*.

A. Lien¹, M. Bolton², N. Wyatt², A. Chanda¹; ¹University of Minnesota; ²Edward T. Shafer Agricultural Research Center, USDA-ARS; **Abstract:** *Cercospora beticola*, the causal pathogen of Cercospora leaf spot (CLS), is responsible for significant economic losses in sugar beet. CLS is primarily managed by alternating two to three demethylation inhibitor (DMI) fungicides in a four to six fungicide spray program. Monitoring *C. beticola* for reduced sensitivity to DMI fungicides is a common practice to aid effective fungicide spray programs. Here, we developed a high-throughput microplate-based assay to estimate the sensitivity of *C. beticola* to four DMI compounds. Additionally, isolates collected after repeated exposure to DMI fungicides in a field trial were subjected to a multiplex probe-based qPCR assay to distinguish between five potential haplotypes (A to E) of *CbCyp51*, which encodes the DMI target enzyme. The determination of *CbCyp51* haplotypes, previously associated with sensitivity or insensitivity, corroborated the accuracy of the microplate-based assay. *CbCyp51* haplotype D was associated with insensitivity to tetraconazole (TET) and prothioconazole (PRO) and was prevalent among isolates exposed to TET and PRO in the field. *CbCyp51* haplotype B was associated with reduced sensitivity to difenoconazole (DIF) and mefentrifluconazole (MEF) and was prevalent among isolates exposed to DIF and MEF in the field. This microplate assay provides a valuable tool for rapid and efficient assessment of DMI insensitivity in *C. beticola* to provide effective disease management strategies in sugar beet.

Unveiling genetic diversity of *Verticillium* spp. from potato in central Minnesota

K. Lerohl¹, A. Ranjan¹; ¹University of Minnesota; **Abstract:** Verticillium wilt (VW) of potatoes is primarily caused by soilborne fungal pathogen *Verticillium dahliae* (VD), though other species can also infect. VW can result in crop losses ranging from 10% to 70% depending on disease severity. VD

produces microsclerotia that can persist in soil for over a decade, posing challenges for control. VD has been classified into three different races previously based on resistance profiles in tomato cultivars. Recently races have been redefined by the possession of newly characterized effectors. Resistance to races 1 and 2 of VD is attributed to Ve genes, but their role in resistance to VW in potato remains understudied. Thus, understanding pathogen load in soil, its variability and genetics is essential for managing the disease. This study assesses pathogen load in soil samples collected from a naturally infected disease nursery in central Minnesota, alongside isolation of VW causative agents from soil and infected potato plants. Our study found that the field soil contained 6-19 propagules per gram, with no isolates obtained directly from soil. Using morphological and PCR-based techniques, 14 isolates isolated from the infected stem were identified as VD or *V. longisporum*, all belonging to race 3. The identification of prevalent *Verticillium* spp. and races yields novel insights into pathogen dynamics and paves the way for developing race-specific resistant potato varieties and future surveys across the state.

Creating a Preliminary Cell Atlas of ‘Fusarium-Wheat’ Host Pathogen Interaction

N. Lnu¹, S. Solanki¹, K. Glover¹, G. Ameen¹; ¹South Dakota State University; **Abstract:** Fusarium head blight (FHB) is a destructive fungal disease caused by *Fusarium graminearum* (Fg), causes huge crop loss. The Fg-host interaction is highly dynamic, posing a challenge to study the gene expression at seed's sub-tissue levels. Qualitative FHB resistance gene *Fhb1* from wheat has recently been cloned, yet there is still debate on gene pinpointing and have an unclear understanding of conserved downstream cascade of resistance. Thus, we are leveraging Visium's spatial transcriptomic approach to combine microscopic observation with gene expression profiling to accomplish unbiased mapping of global host-fungal gene expression. Currently we are optimizing the molecular and data analysis pipeline and successfully achieved expertise on inoculation, sectioning, and RNA isolation protocols required for Visium spatial assay from both immature heads and rachis of the *Fhb1* containing variety ‘Brick’ and a more susceptible check ‘Boost’. We have successfully obtained 10-30 ng RNA from 8µm tissue sections after staining and visualization by using Alexa Fluor 488 WGA conjugate. Our preliminary analysis suggests sample collection at 4-6 DAI is suitable for fungal visualization by confocal microscopy and collecting RNA for library preparation. We are also confirming RNA QL values using Bioanalyzer and optimizing tissue permeabilization time for library preparation. This will be the first ever cell atlas of gene expression in the tissue context during Fg-*Fhb1* interaction.

The Tactile Toolbox: A 3D Printing Library for Crop Scouts and Researchers

H. Lopez-Nicora¹, A. Sisson²; ¹The Ohio State University, ²Iowa State University; **Abstract:** A key issue in integrated pest management (IPM) practice and field crop research is accurate quantification of disease and insect injury severity. Severity assessment is crucial when evaluating fungicide efficacy or insect leaf defoliation, but visual assessment can lead to inconsistent results. Training tools to aid assessment include standard area diagrams and interactive computer software, but they have limitations that can be overcome with 3D printing tools to improve accuracy in data collection. By providing a tactile experience, individuals can increase understanding as they interact with physical 3D printed models, which can offer customization, flexibility, and an immersive IPM learning experience. The Tactile Toolbox working group was developed through the North Central IPM Center to better understand 3D printing needs and devise innovative 3D printing solutions for IPM. Working group activities include establishment of a library for 3D printing materials for sharing and accessing models related to crop diseases, insect damage, and assessment techniques; and organization of a 3D printing workshop for academia, industry, and government agencies to exchange ideas, showcase advancements, and promote adoption of 3D printing in IPM research and practices. This workshop is designed to demystify essentials of 3D printing. The Tactile Toolbox working group aims to improve accuracy and efficiency in diagnostics, assessment, and overall decision-making.

M

Identification of genetic factors involved in pathogenicity and virulence of *Xanthomonas translucens* pv. *undulosa* using EZ-Tn5 mutagenesis

F. Manan¹, Z. Liu¹, I. Bankole¹, G. Shi¹, Z. Liu¹; ¹North Dakota State University; **Abstract:** *Xanthomonas translucens* pv. *undulosa* is the causal agent of bacterial leaf streak of wheat, an economically important disease worldwide. The molecular mechanisms of pathogen virulence and host pathogen interaction remain largely unknown. In this study, we conducted a genome-wide mutagenesis through an EZ-Tn5 system to identify bacterial genes that are involved in Xtu pathogenicity or virulence/avirulence. A total of 2,688 random mutants of the local strain LB10 were generated and are being tested in the greenhouse for their pathogenicity/virulence on resistant and susceptible wheat and triticale lines. Among the 1,200 mutants tested, 14 mutants were less virulent on susceptible lines compared to wild type and 3 mutants lost virulence. However, no mutants were found to become virulent on resistant lines. Whole genome sequencing of those mutants revealed the insertion mutations occurred in a variety of bacterial genes, including type III secretion system, biofilm production, transcription factor and primary metabolism. Targeted gene disruption was used to validate a mutation in the argininosuccinate synthase gene and similar level of reduced virulence was observed. This work will help us understanding molecular mechanisms of bacterial virulence/pathogenicity for Xtu and may provide knowledge to develop management strategies.

Effect of spatial and temporal separation of cereal rye from corn on soil infiltration rates, corn seedling disease, and yield in Iowa

R. Matthiesen¹, G. Simmons², P. O'Brien², A. Robertson¹; ¹Iowa State University, ²USDA-National Laboratory for Agriculture and the Environment; **Abstract:** Winter cereal rye (*Secale cereale*) (CR) is the most common cover crop (CC) grown in Iowa. Corn yield reduction may occur when corn is planted following a CR CC. Spatially or temporally separating corn from CR mitigates reduced corn yield. The effect on ecosystem services is unknown. We investigated the effect of corn row proximity to CR and CR termination timing on soil infiltration rates, corn seedling disease, and yield. There were five treatments: corn separated from CR by (i) 38 cm, CR terminated 10 days before corn planting (DBP); (ii) 38 cm, CR terminated at planting; (iii) 19 cm, CR terminated 10 DBP; (iv) 19 cm, CR terminated at planting; and (v) 0-9.5 cm terminated at planting. A no CR control was included. These data were collected: CR biomass at termination; infiltration rates 10 d after corn emergence; percent root rot severity at V3; yield adjusted to 15.5% moisture. CR biomass was greatest when CR was planted 0-19 cm from corn and terminated at corn planting (iv, v) compared to all other treatments. Infiltration rates were greater when termination occurred 10 DBP (i, iii) compared to at planting (ii, iv, v) with the greatest rate in the no CR (vi). Root rot severity was greater when CR was 0-19 cm from corn and terminated at planting (iv, v) compared to CR planted in 38 cm rows (i, ii). These data will inform improved best management practices for incorporating a CR cover crop into corn production to mitigate corn yield reduction.

Identification and Evaluation of Biological Control Agents from North Dakota Soybean Fields for the Control of *Sclerotinia sclerotiorum*

M. Matloob¹, H. Renfro-Becton¹, R. Webster¹; ¹North Dakota State University; **Abstract:** *Sclerotinia sclerotiorum* causes crop diseases, resulting in significant yield and economic losses globally. In 2023, it caused a 32.3-million-bushel loss in North Dakota soybean due to *Sclerotinia* stem rot. To improve disease management, we have been exploring biological control agents (BCAs) derived from bulk soil of North Dakota soybean fields as an alternative to chemical control. This study aims to isolate and characterize BCAs in soil samples across North Dakota. So far, a total of 218 fungal and 148 bacterial isolates from six soybean fields in 2023 have been screened for biological control activity against *S. sclerotiorum*. Preliminary results indicate nine fungal and 15 bacterial isolates with potential against *S. sclerotiorum*. Based on molecular identification, majority of bacterial BCAs are *Pseudomonas*, *Rhodococcus*, and *Bacillus* species, with a small percentage of uncultured bacteria. Among fungi, eight

belong to *Aspergillus*, *Clonostachys*, *Penicillium*, and *Fusarium* genera and one is still unknown. Some identified BCAs exude unknown compounds that hinder fungal growth while others compete for resources, suppressing *S. sclerotiorum* development in dual culture and detached leaf assays. Liquid chromatography-mass spectrometry and other molecular analyses will uncover the identity of these antifungal compounds. This ongoing study shows progress in discovering and characterizing bioagents from soil ecosystems as efficient antagonists against *S. sclerotiorum*.

Effectiveness of fungicides and biorationals controlling disappearing root rot on ginseng

J. Metheny¹, M. Hausbeck¹; ¹Michigan State University; **Abstract:** Ginseng (*Panax quinquefolius*) is a perennial herb known for the root's medicinal properties. Disappearing root rot is a highly destructive disease caused by *Ilyonectria mors-panacis*. Cultural strategies and fungicides are used to control disappearing root rot. Fungicides and biorationals were tested for their efficacy against the disease in four greenhouse trials. Ginseng seedlings, approximately 6 weeks old, were inoculated with *I. mors-panacis* by dipping the roots into a slurry containing mycelial fragments and spores. Within 1 to 4 hours of inoculation, seedlings were planted and drenched with a fungicide or biorational product or not treated. Seven fungicides and three biorationals were tested each year and applied as a soil drench; an additional fungicide was added in 2023. Not all tested products are currently registered for use on ginseng. Disease severity was visually assessed four times over the course of each study using a 0-5 scale (0 = healthy; 5 = plant death). In each trial, the control received a final disease severity rating > 4.2. Penthiopyrad, fludioxonil, thiophanate-methyl, prothioconazole + tebuconazole, and triticonazole + pyraclostrobin were effective compared to the control in all trials, isofetamid in 3 of 4 trials. The biorational products included in our trials were similar to the control for disease severity at the last assessment date. Our results can support disease management programs and efforts to register new fungicides for ginseng.

Dual fungicide applications for Fusarium head blight management in winter wheat in Kansas

R. Debacker Moura¹, K. Andersen-Onofre¹, R. Borba Onofre¹, J. Rupp Noller¹, E. De Wolf¹, A. de Trinidad¹, C. Dias da Silva¹; ¹Kansas State University; **Abstract:** Fusarium head blight (FHB), caused by *Fusarium graminearum*, is a globally important wheat disease. A single fungicide application made at early flowering (Feekes 10.5.1) can result in approximately 50% disease control. We hypothesize that control can be improved by making an application at Feekes 10.5.1 followed by a second application 4 days later. Field experiments to evaluate the efficacy of dual applications were conducted in 2022 and 2023 in two locations in Kansas. Plots were inoculated with *F. graminearum* and received supplemental irrigation to promote disease establishment. Treatments consisted of 1) an untreated control, 2) Prosaro – 6.5 fl oz/A at flowering; 3) Caramba 13.5 fl oz/A at flowering; 4) Miravis Ace 13.7 fl oz/A at flowering; 5) Prosaro Pro 10.3 fl oz/A at flowering; 6) Sphaerex 7.3 fl oz/A at flowering, 6) Miravis Ace 13.7 fl oz/A fb Prosaro Pro 10.3 fl oz/A; 7) Miravis Ace 13.7 fl oz/A fb Sphaerex 7.3 fl oz/A; and 8) Miravis Ace 13.7 fl oz/A fb Tebuconazole 4 fl oz/A). FHB incidence, severity, DON, and grain yield were evaluated. The data collected were subjected to mixed model analysis of variance. Results from Manhattan, KS in 2023 demonstrated that treatments with Miravis Ace alone or in combination with another fungicide application had higher yields (79.5-92.0 bu/A) and lower indices (25.2-32.9%) than the untreated control (58.08 bu/A; 59.2%, respectively). Additional results will be presented.

The role of irrigation in canopy microclimate and disease development

T. Mues¹, T. Jackson-Ziems¹, S. Taghvaeian, M. Shires², R. Borba Onofre³, D. Mangel¹; ¹University of Nebraska - Lincoln, ²South Dakota State University, ³Kansas State University; **Abstract:** Spread of tar spot into the Great Plains region has created concern about the impact of irrigation on disease development and potential for yield loss. Research was initiated on four producer fields across eastern Nebraska and South Dakota to evaluate the role of center-pivot irrigation on canopy conditions and tar spot development. The tar spot pathogen, *Phyllachora maydis*, is favored by cool (optimal 17-22°C) and humid (>75%) environments. Irrigated fields may provide conditions favorable for severe levels of

disease. Weather stations collected data every five minutes on air temperature, relative humidity, precipitation, and leaf wetness at three different regions of the field: under the pivot's inner span, outer span, and the rainfed corner. Plants in these zones were also assessed for disease. Results from the first year of research showed greater disease incidence and severity under irrigation than in the rainfed environment. Incidence under irrigated conditions was 100% by the final rating, while rainfed incidence reached a high of 55%. Severity was greater in the irrigated treatments but was not significant except for the initial rating. Compared to the rainfed corner, air temperature decreased (up to 7.75 hrs) along with vapor pressure deficit (up to 6.25 hrs). Relative humidity and leaf wetness duration increased, for up to 5.75 and 10.5 hrs, respectively. This research will be continued during the 2024 growing season with modification to the trial design.

Plant Parasitic Nematode Population Densities Associated with Crown Rot Symptoms of Corn in the U.S. Corn Belt

C. Mukuma¹, T. Jackson-Ziems¹, C. Termunde¹; ¹University of Nebraska; **Abstracts:** Corn is the most important cereal crop produced in the U.S. In recent years, the emergence of crown rot (CR) disease has become a potential production concern in certain states. Literature indicates that most CR diseases involve interactions of fungal complexes and other microbes. However, the primary causal organism(s) of CR in corn or their interaction with other microbes is unknown in the U.S. Corn plant samples were collected by Bayer Crop Science in 2022 from 8 Midwest states. Adjacent plant pairs with and without aboveground symptoms were collected by digging around each plant to include an intact root ball and about 8 inches of the plant stalk. Nematodes were extracted from 45 plant pairs. Population densities (nematodes/gram root or 100 cm³ soil) of root-lesion (*Pratylenchus* spp), spiral (*Helicotylenchus* spp.) and stunt (*Tylenchorhynchus* spp.) nematodes were estimated. Differences in population densities between symptomatic and asymptomatic plants were statistically analyzed with a paired t-test. The population density of root-lesion nematodes from roots of symptomatic plants was greater (p -value = 0.02019) than that of asymptomatic plants. However, no differences were observed among the population densities of other nematode genera from the roots and root ball soil. Identifying interactions of microbes, such as root-lesion nematodes, with crown rot disease could provide more information towards understanding the disease and developing management strategies.

Exploring Interactions of Root-lesion Nematode (*Pratylenchus scribneri*) and Fungal Pathogen (*Verticillium dahliae*) in the Potato Early Die Complex

B. Mumia¹, J. Pasche¹, G. Yan¹; ¹North Dakota State University; **Abstract:** Root-lesion nematodes (*Pratylenchus* spp.) are migratory endoparasites that can cause high yield losses in potato. Synergistic interactions between root-lesion nematodes, *Verticillium dahliae*, and other fungal pathogens constitute the potato early die (PED) disease complex, with up to 50% yield loss. Previous studies have focused mainly on the interaction of *P. penetrans* with *V. dahliae*. However, *P. scribneri* is an important root-lesion nematode that may interact with *V. dahliae*. Infective juveniles (J2s) of *Pratylenchus* spp. induce wounds on feeder roots that may serve as pathogen entry points and/or promote production of root exudates leading to faster germination of *V. dahliae* microsclerotia. *Pratylenchus scribneri* was collected from a potato field in North Dakota and multiplied through a sterile carrot culture technique. Preliminary results confirmed the identity of *P. scribneri* with a single melt peak of 81.5°C and 136 bp amplicon size using qPCR. Four isolates of *V. dahliae* from North Dakota and Minnesota, identified through qPCR in an earlier study, were raised on potato dextrose agar. Microplot experiments with *P. scribneri* and *V. dahliae* infested separately and co-infested will be conducted. The effect of co-infestations will be determined by PED symptoms, severity, growth and yield of Russet Burbank and Umatilla Russet potato cultivars. This study will provide critical information for management of PED, *P. scribneri* and *V. dahliae* in potato.

N

Safeguarding Canola Yield: Monitoring the Breakdown of Resistance to Clubroot in Commercial Canola Cultivars

N. Narra¹, L. Jennings¹, A. Arens¹, L. del Rio Mendoza¹, V. Chapara¹; ¹North Dakota State University;

Abstract: In the United States, North Dakota is the top canola producer, with a record of 3.4 billion pounds produced in 2023. Of these 23% came from Cavalier County. However, diseases like clubroot, caused by *Plasmodiophora brassicae*, limit its production. Many strategies have been proposed to manage clubroot. The use of genetically resistant cultivars is one of the most effective strategies. However, there are many clubroot pathotypes, and genetic resistance to clubroot may be pathotype-specific. A preliminary study of pathotype determination revealed the presence of pathotype 3D (a mutant) in Cavalier County, which has the potential to overcome first-generation clubroot resistance in canola. Field studies conducted at NDSU REC Langdon, in Cavalier Co. in 2022, 2023 were aimed to assess the breakdown of resistance by pathotype 3D in nine frequently grown commercial canola cultivars. Each trial had four replications and was set up in Randomized complete block design with susceptible varieties InVigor L233P and CP9978TF planted as the checks. Clubroot severity was rated 60 days after planting. All the resistant cultivars performed as expected and are highly resistant ($P < 0.0001$) to clubroot. The results of this study suggest that the genetic resistance to pathotype 3D present in these cultivars is holding. Nevertheless, continuous monitoring of the reaction of cultivars to clubroot is critical to ensure sustainable canola production in North Dakota.

Sugarbeet Tare Soil and its Role in Storage

E. Nelson¹, S. Kandel¹, K. Fugate¹, G. Secor²; ¹USDA-ARS, ²North Dakota State University; **Abstract:**

Following harvest, sugarbeet (*Beta vulgaris* L.) roots are agitated over rollers, chains, and belts to dislodge as much soil as possible from roots prior to storage. Despite such efforts to free roots of soil, some soil remains adhered to roots, and is integrated into storage piles, yet the effects of this soil, referred to by the industry as tare soil, on storage are largely unknown. The study was conducted to characterize the bacterial microbiome of tare soil and its effect on storage. Tare soil samples were collected from sugarbeet roots obtained from five storage piles in the Red River Valley of Minnesota and North Dakota. Total genomic DNA was extracted from tare soil samples and bacterial communities were characterized by sequencing of the full-length 16S rRNA gene. Firmicutes, Proteobacteria, and Actinobacteria were the dominant bacterial phyla in the tare soil. The most abundant bacterial genera identified in the tare soil were *Clostridium*, *Leuconostoc*, and *Pantoea*. In addition, freshly harvested sugarbeet roots were collected from the storage pile and performed storage experiments using clean or surface disinfected roots prior storage. Our data showed that respiration rate was significantly decreased in both clean and surface disinfected sugarbeet roots before storage as compared to unwashed roots with tare soil. The preliminary findings of the study suggest that the tare soil microbiome can influence the storage quality in sugarbeet roots.

P

Validation of the real-time PCR assay for rapid detection and identification of the pin nematode, *Paratylenchus nanus* type B

S. Paudel¹, G. Yan¹; ¹North Dakota State University; **Abstract:** This study validates the real-time PCR

assay developed for identification of *Paratylenchus nanus* type B, a prevalent plant-parasitic nematode in field peas in North Dakota. Species-specific primers were designed targeting the internal transcribed spacer region of ribosomal DNA. The specificity of the primers was evaluated by doing in-silico analysis and was confirmed by PCR tests. The real-time PCR assay was optimized for amplification temperature and primer concentration. Detection sensitivity was determined using DNA extracts from 20 nematodes followed by two-fold serial dilutions. The assay could detect an equivalent to 1/51 of a single nematode. Standard curve was generated by plotting quantification cycle (Cq) values against corresponding log₁₀

numbers of nematodes with amplification efficiency of 92.6%. Subsequently, the assay was applied to 21 field soil samples collected from 10 counties in North Dakota and Minnesota. Nematodes were extracted using sugar centrifugal-floatation method and DNA was extracted from single nematodes by proteinase K method. Real-time PCR was done using the designed species-specific primers. Amplifications observed in the assay showed presence of the target *P. nanus* type B DNA in all the soil samples with pin nematodes. These results validated the developed real-time PCR assay to be specific, sensitive and efficient, providing a useful tool for rapid detection of *P. nanus* type B from infested fields.

Genomic Regions Associated with Spot Blotch (SB) Resistance in Barley in the Interspecific Cytonuclear Multi-Parent Population (CMPP)

T. Pawar¹, J. Tilstra¹, M. Ahmad¹, A. Beery², S. Bodenheimer^{2,3}, S. Yang⁴, S. Solanki¹, E. Fridman², G. Ameen¹; ¹South Dakota State University, ²Plant Sciences Institute, Volcani Agricultural Research Organization (Israel), ³The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture (Israel), ⁴Cereals Crops Research Lab; **Abstract:** Spot blotch is one of the major foliar diseases of barley (*Hordeum vulgare*), caused by the fungal pathogen *Bipolaris sorokiniana* in the upper Midwest of the USA. It also causes root rot, black point, and reduces kernel weight which deteriorates the malting quality. Identifying genetic loci associated with resistance to spot blotch is crucial for future marker-assistant selection or introgression breeding. In this study, 352 doubled haploid lines belonging to the interspecific Cytonuclear Multi-Parent Population (CMPP) were evaluated by using a virulent isolate ND85F of pathotype 1 of *B. sorokiniana* at the seedling stage under greenhouse conditions. Three genotypes showed resistance and 44 genotypes showed moderately resistance reactions on a disease rating scale of 1 to 9. The CMPP population was genotyped with 50K single nucleotide polymorphism (SNP) markers using Illumina Infinium iSelect genotyping array. After quality control filtering of the genotyping dataset, 19,741 SNPs were selected for GWAS. The genome-wide association analysis identified four significant marker-trait associations (MTA). The identified MTAs were found at Chromosomes 1H, 5H, 6H and 7H. This study provided markers that can be utilized to genetically improve resistance to spot blotch disease in barley. Further testing of these genotypes in field conditions and different environments is needed to confirm the gene effects for these identified loci.

Exploring Pathogenic Potential: Isolation and Analysis of *Streptomyces* spp. From Soil and Scab-Infected Potatoes

G. Petzold¹, K. Tyagi¹, A. Ranjan¹; ¹University of Minnesota; **Abstract:** *Streptomyces* is a genus of gram-positive bacteria that thrives in soil. The majority of *Streptomyces* spp. are helpful to humans and plants, producing antibiotics and anticancer drugs for human use and acting as biocontrol agents against plant pathogens. However, a few *Streptomyces* spp., including *Streptomyces scabies*, can cause common scab of potato disease. This disease causes raised textured and deep pitted lesions on tuber surfaces, affecting their quality and marketability. In this research, we used aseptic microbiological techniques to isolate 76 potential *Streptomyces* spp. strains from soil and common scab-infected potatoes using semi-selective STR media. Of the 76 strains, 51 were confirmed to be *Streptomyces* spp. with *Streptomyces* specific 16S rRNA PCR followed by sequencing. These isolates were further characterized by a morphological study and pathogenicity-associated *TxtAB* gene analysis. Furthermore, a pathogenicity radish seedling assay was performed to estimate their pathogenicity. Results from the 16S rRNA phylogenetic analysis and these studies indicate that *Streptomyces* spp. have wide genetic variability that can contribute to pathogenicity. Overall, we successfully isolated *Streptomyces* spp. from both tuber and soil samples and have analyzed them for various characteristics common to *Streptomyces* spp. Further work should support identifying novel pathogenicity-related genes and mechanisms within these confirmed pathogenic *Streptomyces* spp.

Fungicidal Strategies for Septoria Brown Spot and Frogeye Leaf Spot Management in Iowa Grown Mungbean (*Vigna radiata*)

E. Platner-Heidt¹, M.E. O'Neal¹, S. Arti¹, J. Batzer¹, D. Mueller¹; ¹Iowa State University; **Abstract:** The expansion of mungbean (*Vigna radiata*) cultivation in the U.S. is driven by consumers, adaptability to

climates, and compatibility with existing soybean-production infrastructure. Mungbean's short growing season (60-90 days) makes it suitable for various cropping systems as a row or cover crop. However, because of its similarity to soybean as a legume, concerns arise regarding potential overlap with common soybean diseases. We surveyed diseases pathogenic to mungbean in Iowa and developed a fungal library to assist breeders in resistance programs. This assessment confirms that diseases overlap from soybean to mungbean, including *Septoria glycines*, and *Cercospora sojina*. We then explored if applying fungicide can protect mungbean from foliar disease and yield loss. To generate information for mungbean cultivation, we assessed the impacts of fungicide applications on two mungbean varieties (Berken and OK2000) in Story County, Iowa, during the summers of 2022 and 2023. We evaluated the efficacy of Revytek and Warrior II tank mixture in reducing the impact of major pathogens, *Septoria glycines*, and *Cercospora sojina*, when applied at various growth stages (R1 through R5). An application of pesticides during the early R stages slowed the progression of foliar diseases (p -value = 0.03). Treated mungbean yielded around 420 lbs/ac more than the control. These data show that farmers can manage foliar mungbean diseases with commercially available fungicides.

Summary of 5 Years of Illinois Soybean Cyst Nematode Egg Count Samples submitted to the University of Illinois Plant Clinic

D. Plewa¹, A. Colgrove¹, E. Mahecha¹; ¹University of Illinois Plant Clinic; **Abstract:** Soybean cyst nematode (*Heterodera glycines*, SCN) is the greatest yield limiting soybean pathogen in the United States and has been confirmed in over 80% of Midwest fields. Identifying SCN in the field can be challenging due to non-specific symptoms that may resemble those caused by other biotic and abiotic issues. The fact that SCN is widespread, may be overlooked or misidentified in the field, and the potential for yield loss make SCN one of the most important soybean pathogens in the US. Hence, regular sampling, early detection, and previous SCN field data can assist farmers in future integrated pest management (IPM) decisions. The data presented is of historical SCN egg counts classified by risk levels representing 2674 soil samples from 74 Illinois counties submitted to the U of IL Plant Clinic from 2019 through 2023. This work highlights the persistence of SCN in fields across Illinois. The number of samples submitted varied across the years. The average risk levels for all samples were high (29.33% of samples), followed by low (26.37%), moderate (18.13%), none detected (14.4%), and extreme (8.7%). Despite variations in sample numbers and egg counts from year to year, SCN risk levels remain consistently "high" in the state. These findings underscore the importance of continued vigilance for this pathogen. The analysis of past SCN data provides valuable insights for growers and researchers to develop IPM strategies for future seasons.

Identification and Assessment of Pathogenic *Fusarium oxysporum* species complex isolates on Sugar Beet

K. Pollok¹, L. Hanson²; ¹Michigan State University, ²USDA-ARS; **Abstract:** The sugar beet industry in Michigan is a valued source of revenue for the state, producing \$2 billion in 2022. *Fusarium* yellows, caused by strains in the *F. oxysporum* species complex (FOSC), was first reported in MI in 2007. Since then, it has been an increasing concern for growers. The main strategy to reduce this disease is by using resistant varieties. The varieties used in MI are screened in Minnesota using MN isolates, but distinct FOSC strains vary across production regions. As such, resistance in one region is not guaranteed in others. The objectives of this study were to identify *Fusarium* spp. isolates collected in MI that were pathogenic on sugar beet and assign virulence levels. Isolates of *Fusarium* spp. were collected from symptomatic MI sugar beet and identified to the species level. FOSC isolates were greenhouse screened on a pair of USDA sugar beet cultivars with varying susceptibility. Pathogenicity and virulence were assessed weekly for six weeks post inoculation via foliar ratings with a root rating and re-isolation taken at the end. Of the isolates screened, about 75 percent of them were pathogenic on sugar beet but the majority had low virulence. While a small portion of isolates were moderately virulent, no highly virulent isolates have been identified yet. The moderately virulent isolates are promising for future applications in screening for broad spectrum resistance to *Fusarium* spp. and developing a resistance screening system in MI.

Copy number variations at the *Rhg1* locus and their relationship with resistance to soybean cyst nematode (*Heterodera glycines*)

D. Poudel¹, G. Yan¹, G. Kreutz¹, C. Miranda¹; ¹North Dakota State University; **Abstract:** Soybean cyst nematode (SCN, *Heterodera glycines*) poses a significant threat to soybean production. The *Rhg1* locus of soybean, characterized by both copy number variations (CNVs) and sequence variations, confers robust resistance to SCN, with CNVs having a dominant role. To determine CNVs at *Rhg1* among 100 soybean breeding lines, a SYBR Green-based quantitative PCR (qPCR) assay was adapted and validated using 12 soybean accessions with known copy numbers. The copy number of each line was determined using the 2- $\Delta\Delta C_q$ method relative to the reference Williams 82 (single copy) and compared with its resistance responses to two SCN populations, HG type 7 and 2.5.7, with differential reproduction rates on the major SCN-resistant source, PI 88788. Copy numbers among the breeding lines ranged from 1 to 11. Only one breeding line was resistant (Female Index, FI < 10%) to HG type 7 and had 11 copies, while all the susceptible lines (FI > 60%) had a single copy. 42 breeding lines with ≥ 6 copies were either resistant or moderately resistant (FI = 10-30%) to HG type 7, and most of those lines were moderately resistant to HG type 2.5.7, with few being moderately susceptible (FI = 30-60%). Significant negative correlations between copy numbers and FI values were observed. Higher copy numbers were consistently associated with increased resistance to HG type 7. These results show the importance of *Rhg1* CNVs in determining levels of SCN resistance and selecting resistant soybean lines.

Assessing Sclerotinia Stem Rot Resistance Across Early Maturity Soybean PI Lines

S. Poudel¹, H. Renfroe-Becton¹, R. Webster¹; ¹North Dakota State University; **Abstract:** *Sclerotinia sclerotiorum*, the causal agent of Sclerotinia stem rot (SSR), is a devastating fungal pathogen that led to the loss of over 11 million bushels in northern US soybeans in 2022. While utilizing resistant cultivars can effectively limit SSR, no commercial varieties have been found to offer complete resistance. In this study, we screened 23 soybean accessions from the USDA germplasm collection, with different maturity groups (ranging from 000 to V). Four previously established check lines (Dwight, 51–23, SSR51 and 52-82B) were used, as suggested by Webster et al. (2021). The greenhouse experiment was conducted in a randomized complete block design. The plants were inoculated at the R1 growth stage (beginning of flowering) with mycelium plugs from a highly aggressive isolate of *S. sclerotiorum* (WI-20) using the cut-petiole method. Lesion length was monitored, and data were collected at 7-, 10-, and 14-days post-inoculation. Area under the disease progress curve (AUDPC) values were generated from this data. While all PI lines showed disease symptoms, the level of resistance varied significantly among them ($P < 0.001$). PI 423989A exhibited the highest resistance level with an AUDPC value of 79.36, while the most susceptible line was PI 458535 with an AUDPC of 732.7. Additional lines are being screened. This research can help in identifying highly resistant lines for utilization in breeding programs to develop resistant cultivars in the Northern Great Plains.

Chemotype diversity, population structure and pathogenicity of a *Fusarium graminearum* population sampled from North Dakota in 2021 and 2022

A. Poursafar¹, S. Zhong¹, C. Monson¹, Y. Leng¹, O. Amusan¹, S. Safar¹, A. Riasat¹, J. Mullins¹; ¹North Dakota State University; **Abstract:** *Fusarium graminearum* is the major causal agent of Fusarium Head Blight (FHB) of wheat and barley in North America. The fungal pathogen significantly affects the yield and quality of the crops. Isolates of *F. graminearum* were traditionally categorized as one of the three chemotype groups: 3-acetyl-deoxynivalenol (3ADON), 15-acetyl-deoxynivalenol (15ADON), and nivalenol (NIV). More recently, a novel type A trichothecene (NX-2) was found for the fungus. In this study, we aimed to characterize *F. graminearum* isolates collected in 2021 and 2022 from North Dakota for possible changes in chemotypes, population structure and pathogenicity in wheat. Analysis of 261 isolates identified 3ADON and 15ADON chemotypes, while no NIV and NX-2 chemotypes were detected. The isolates with 15ADON chemotype were predominant accounting for 69% of the isolates while 31% of the isolates were of 3ADON chemotype. A total of 150 randomly selected 3ADON and

15ADON isolates were inoculated on two spring wheat genotypes, Alsen (resistant to FHB) and Wheaton (susceptible to FHB), for pathogenicity tests in greenhouse. The results indicated that significant differences in virulence exist among the isolates tested. The analysis of trichothecenes produced by the fungal isolates in grains are in progress. This study provides updated information on the existing *F. graminearum* populations in North Dakota and may facilitate a better management of the disease.

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Validation of GWAS candidate genes associated with *Fusarium virguliforme* as a root rot pathogen in soybean (*Glycine max* L.)

N. Rafi¹, T. Mukaila¹, F. Mathew¹; ¹North Dakota State University; **Abstract:** Sudden death syndrome, caused by *Fusarium virguliforme* in soybean (*Glycine max* L.), resulted in an estimated total yield loss of 527,109 MT in the United States in 2023. There are commercially available soybean cultivars that have partial resistance to *F. virguliforme*. However, the exact defense mechanisms against this fungus are unclear. This study aimed to compare the expression levels of GWAS candidate genes known to be associated with foliar resistance to *F. virguliforme*. Specifically, we focused on gene expression levels in the presence of the fungus as a root rot pathogen. The experiment was conducted twice using a completely randomized design with one susceptible ('Spencer') and one resistant ('Ripley') variety. Plants were collected at 0-, 1-, 3-, 10-, 14-, and 21 days post-inoculation (dpi) and flash-frozen for RNA extraction. Gene expression levels were estimated through reverse transcription-quantitative PCR and analyzed using the 2- $\Delta\Delta C_t$ method. For Ripley, four genes were significantly upregulated, SIK1 (LRR-RLK) at 10 dpi ($P < 0.001$), *Glyma19g109300* (NBS-LRR) at 3, 10, 14, and 21 dpi ($P < 0.001$), *Glyma18g231000* (LRR-containing resistance) at 3 dpi ($P < 0.001$), and *Glyma02g211000* (integral membrane protein) at 10 dpi ($P = 0.001$), respectively. These genes were not significantly upregulated in Spencer at $\alpha = 0.05$. This study lays the foundation for future research on resistance genes against *F. virguliforme* and the development of cultivars with improved resistance

A Systematic Analysis of Genome and Phenome Associations in Soybean White Mold Pathogen for Fungicide Resistance and Evolving Virulence

K. Reese¹, S. Sharma¹, J. Kleinjan¹, C. Graham¹, G. Ameen¹, S. Solanki¹; ¹South Dakota State University; **Abstract:** White mold, caused by the fungal pathogen *Sclerotinia sclerotiorum*, is a notable concern for soybean growers in the Midwestern US, including South Dakota (SD). Limited resistance against white mold in the commercially available soybean varieties necessitates the use of fungicides as a primary management tool, however, it also acts as selection pressure and favors the evolution of fungicide-resistant populations. Thus, we initiated a systematic approach to survey the prevalence and genetic diversity of *Sclerotinia sclerotiorum* in SD for fungicide resistance and virulence. During a 2023 statewide disease survey, more than 30 representative isolates were retrieved from multiple locations. These isolates are being evaluated in a greenhouse for their virulence diversity on commercial varieties. Presently, we are assessing these isolates using DNA marker-based MBC and SDHI fungicide resistance mutations in corresponding genes. Initial analysis identified no mutation in the β -tubulin gene that has previously been associated with high MBC resistance. However, there could be novel mutations that may not be detected using a partial-length marker-based approach. To detect novel mutations quickly, we are optimizing full-length gene marker analysis using a MinION sequencer. Fungicide plate assays will validate DNA-based results of 2023 and 2024 isolates. Our goal is to develop a logical approach to understanding white mold pathogen diversity and support SD farming communities.

Molecular mapping of Type I Resistance to Fusarium head blight in a Spring Wheat Population.

A. Riasat¹, A. Poursafar¹, S. Safar¹, A. Karmacharya¹, Y. Leng¹, S. Xu², S. Zhong¹; North Dakota State University, ²USDA-ARS Cereal Crops Research Unit; **Abstract:** Fusarium head blight (FHB) is a devastating disease affecting worldwide wheat production. Host resistance is the most effective way to manage the disease. Type I resistance for restriction of initial infection by the pathogen is one of the major components of FHB resistance. However, it is not well characterized, and not many QTLs for this type of resistance have been identified so far. This study aimed to identify and map QTL associated with Type I resistance to FHB in a spring wheat mapping population consisting of 130 doubled haploid (DH) lines derived from the cross Grandin × PI277012. This population was previously genotyped with SSR markers and were phenotype for FHB resistance, with two QTLs for Type II resistance detected on chromosome 5A. We re-phenotype the DH population for Type I resistance using the dip inoculation method in three greenhouse experiments in 2023 and 2024 and constructed a new genetic map using previously identified SSR markers combined with SNP markers from genotyping with the 9k SNP chips. QTL analysis identified one QTL for Type I resistance located on chromosome 4B in the repeated greenhouse experiments. We also identified three QTLs for the Plant height (PH) on chromosomes 4B, and 5A, one QTL for the spike length on chromosome 5A, and two QTLs for the flowering date (FD) on chromosomes 2B and 5D. Further greenhouse and field experiments will be conducted to validate the QTL for Type I resistance.

Dissection of Nonhost Resistance in Barley to the Heterologous Leaf Rust Pathogen *Puccinia triticina*

M. Ritzinger¹, S. Zhong¹, S. Yang²; ¹North Dakota State University, ²Northern Crop Science Laboratory - Edward T. Schafer Agricultural Research Center; **Abstract:** Barley and wheat leaf rusts are important diseases caused by the biotrophic fungi, *Puccinia hordei* and *P. tritici*, respectively. Annual losses to leaf rust range from 1 to 20% but can reach 60% under epidemic conditions. These fungi possess a suite of effectors, have high evolutionary and dispersal capabilities, and are highly host-specific. Infection compatibility is typically determined by the gene-for-gene interactions of the pathogens' avirulence (*Avr*) genes and host resistance (*R*) genes. However, several studies have reported host plant accessions which exhibit significant levels of susceptibility to a typically incompatible pathogen — termed “heterologous infections”. Recent technological advances have greatly expanded our ability to dissect the genetic basis underlying these heterologous infections. Our group has identified two wild barley accessions, WBDC016 and WBDC103, which are susceptible to multiple common races of *P. tritici*. To dissect the genetic basis of the observed susceptibility, crosses with each were made with the resistant cultivar Rasmusson, and linkage mapping was performed with the derived F₂ progeny. In the former population, significant quantitative trait loci (QTL) were identified on chromosomes 4H and 7H; in the latter, QTL were detected on chromosome 6H (2) and 7H. The results of this research could ultimately provide valuable insights into the basis of nonhost resistance and inform new strategies for controlling leaf rusts of cereals.

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Assessing microbial contaminants in the raw sugarbeet juice: A microbiome-driven analysis

K.C. Sabina¹, Z. Liu¹, E. Branch¹, M. Bill¹, S. Kandel²; ¹North Dakota State University, ²USDA-ARS; **Abstract:** Microbial contaminants in raw sugarbeet juice cause substantial sucrose loss in the factory and create technical challenges during juice purification and crystallization. However, limited information is available regarding the status of microbial contaminants in raw sugarbeet juice and its quality in the United States. In this study, we aim to profile fungi, especially yeast communities present in raw sugarbeet juice during the sugar extraction process in the factory. Genomic DNA was extracted from 49 raw juice samples and fungal ITS rRNA gene was amplified. Sequencing library preparation, sequencing

libraries, and data analysis are underway. The findings of the study will help to identify fungal contaminants present in sugar processing streams and contribute towards the development of control measures to minimize microbial contamination and reduce sucrose loss during sugar processing.

Phenotyping and Genotyping of Near-Isogenic Lines Harboring a Major Fusarium Head Blight Resistance Gene (*Fhb1*) in Spring Wheat

S. Safar¹, R.S. Nandety², M. Osenga², J. Fiedler², Y. Leng¹, S. Zhong¹, A. Poursafar¹, A. Riasat¹, O. Amusan¹; ¹North Dakota State University, ²USDA-ARS Cereal Crops Research Unit; **Abstract:** Fusarium head blight (FHB) caused by *Fusarium graminearum* is a highly destructive disease in cereal crops including wheat. Use of host resistance is one of the major approaches in the management of the disease. Among the quantitative trait loci (QTL) previously identified for FHB resistance, *Fhb1* derived from Sumai 3 is one of the most effective ones commonly used in wheat breeding programs. In this study, we introduced *Fhb1* into six spring wheat varieties (WA8283, Linkert, ND VitPro, Surpass, Glenn, Dayn) and one durum variety (ND Riveland) via crosses and backcrosses using Alsen as the FHB resistance donor parent. The resulting near-isogenic lines (NILs) (BC₇F₂) with homozygous *Fhb1* and their recipient parents were phenotyped for FHB in the greenhouse and field, and also genotyped by 90K and 3K single nucleotide polymorphism (SNP) arrays. The results showed that only Linkert-NILs and WA8283-NILs carrying *Fhb1* demonstrated significant improvement in FHB resistance compared to their respective recurrent parents, and they carried < 5% and < 3% of resistant donor alleles from Alsen, respectively, based on genotyping with the 90K and 3K SNP arrays. Our study indicated that FHB resistance conferred by *Fhb1* is significantly influenced by the genetic background of the recipient parents. The NILs developed with *Fhb1* can be used as elite germplasm for wheat breeding programs to develop new varieties with improved FHB resistance.

Managing Potato Common Scab Under North Dakota Field Conditions

A. Sarwar¹, C. Clarke², J. Pasche¹; ¹North Dakota State University, ²USDA-ARS Genetic Improvement for Fruits and Vegetables Laboratory; **Abstract:** Potato common scab, caused by at least 10 species of *Streptomyces*, is a major potato disease impacting tuber quality and marketability. While low dose applications of the herbicide 2,4-D show some promise for common scab management, results are inconsistent. We evaluated the efficacy of low dose foliar 2,4-D applications, 1-naphtheneacetic acid-potassium salt (NAA-PS) (Rejuvenate®) seed treatment, and pre-plant broadcast of manganese sulfate for common scab management. Four field trials were conducted during 2022 and 2023, each comprising eight treatments of these products alone and in combination. Significant differences in common scab were observed in one trial under lower disease incidence (14% to 34.5%) and severity (3.2% to 8.1%). In this trial all treatments significantly reduced common scab incidence and severity compared to the nontreated control. The greatest common scab reduction was observed with 2,4-D (1.6 fl. oz/a) applied 24 days after emergence followed by a second application 14 days later. This translates to a 60% reduction in common scab disease incidence and severity. In one of four trials, the application of NAA-PS as a seed treatment alone significantly reduced emergence compared to all other treatments and yield compared to some treatments. These results support previous research that the application of 2,4-D, NAA-PS, or manganese sulfate provide inconsistent results for the management of common scab under field conditions

Crown Rot of Corn

C. Schmidt¹, A. Robertson¹; ¹Iowa State University; **Abstract:** Crown rot of corn has become more prevalent in the Midwest. The etiology of this disease is not well understood. We hypothesized that crown rot is like stalk rot and develops when conditions are stressful during grain fill. Since foliar fungicide applications may reduce stalk rot severity, we investigated if fungicide applications could also mitigate crown rot. Experimental plot trials were established at three locations in Iowa in 2023. A foliar fungicide was applied at growth stages V6, R1, or both growth stages. A non-sprayed treatment was included as a check. Foliar disease severity and the incidence of crown rot were assessed at R5.5. No effect of treatments on foliar diseases or crown rot was detected ($P < 0.1$). Dry conditions during the 2023 growing

season were not conducive to disease development. Despite these inconclusive results, the trials will be repeated in 2024.

Unraveling the wall-associated kinase immunity in the barley spot blotch pathosystem

M. Sengar¹, S. Solanki¹, G. Ameen¹, R. Brueggeman²; ¹South Dakota State University, ²Washington State University; **Abstract:** Plant immunity relies on immune signaling receptors that activate upon detecting pathogens, often triggering programmed cell death to combat infection. However, necrotrophic fungal pathogens like *Bipolaris sorokiniana* can exploit these responses, promoting disease and reducing crop yield. Previously, we identified two wall-associated kinase (WAK) receptors, Sbs1 and Sbs2, which are manipulated by the pathogen in barley-spot blotch interactions. The goal of this study is to explore the function of these atypical WAK receptors, particularly whether Sbs1 and Sbs2 interact as protein complexes, similar to other receptor-like kinases. We hypothesize that both Sbs1 and Sbs2 co-localize at the plasma membrane and interact with each other. To validate our hypothesis, we are using complementary assays of co-localization, and bimolecular fluorescence complementation (BiFC). For co-localization, we constructed 35S:*Sbs1-GFP-his* and 35S:*Sbs2-CFP-HA* expression clones and for the BiFC, we constructed Sbs1-YN and Sbs2-YC fusion proteins and transiently expressed them in tobacco leaves using *Agrobacterium tumefaciens* strain GV3101. We conducted confocal laser scanning microscopy after 72 hours post-infiltration and observed that the fusion of Sbs1-YN and Sbs2-YC exhibited YFP fluorescence at the edges of the plasma membrane, indicating a potential interaction between the two proteins. The results from this study will elucidate the WAK-mediated immunity mechanism in plants.

Targeting Organic Acid Responsive Genetic Pathways in *Sclerotinia sclerotiorum* for Disease Suppression in Organic Soybeans Cultivation

S. Sharma¹, S. Solanki¹, G. Ameen¹; ¹South Dakota State University; **Abstract:** The major bottleneck in organic soybean production with limitations on fungicidal usage is white mold disease caused by fungus *Sclerotinia sclerotiorum* (S.c.). To address this issue, we have started developing a sustainable alternative to chemicals by leveraging pathogen genetics as a tool to identify and target the organic acid responsive genes in S.c. for disease suppression. Major fractions of organic molecules (OM) present in manure includes humic acid (HA) and fulvic acid (FA). We have tested 33 diverse isolates for their responsiveness to a concentration gradient of HA and FA using 20, 30, 40, 50 and 60 mgL⁻¹ concentration in vitro plate assay using both mycelia and sclerotia. ImageJ data of growth, sclerotia number and size identified that isolates significantly differ (p<0.05) in their OM concentration responsiveness for above characteristics and association could be either positive, negative, or null. We have selected two isolates with contrasting responsiveness at 20-30 mgL⁻¹ OM concentration and are currently performing long read transcriptomics on DNA degraded RNA libraries on Oxford PromethION sequencer to identify responsive genetic pathways. The identified gene sets will be used in multitarget bio-nanoparticle stabilized complex bio-fungicide development leveraging information available from previous white mold studies. Thus, our research goal is to develop next generation management practices suitable for both organic and manure applied soybeans.

***Fusarium luffae*: Potential concern for soybean growers in US Midwest**

S. Sharma¹, K. Reese¹, J. Kleinjan¹, C. Graham¹, J. Gonzalez¹, S. Solanki¹, G. Ameen¹, S. Ali¹; ¹South Dakota State University; **Abstract:** Soybean (*Glycine max* (L.) Merr.) is the leading oilseed crop in the United States contributing 90% to the total oilseed production. During 2023 scouting in South Dakota, we identified soybean fields with plants having unusual wilted and blighted leaves. After isolation of the pathogen, microscopic studies along with DNA isolation were performed for identification using multiloci primers. Based on multiloci primers, the pathogen was identified as *F. luffae* on NCBI and Fusarioid polyphasic identification database. The pathogenicity test was confirmed using leaf and whole plant inoculations. The symptoms include blighted leaves and within 96 hours all inoculated plants were defoliated, and the emerging leaves also showed disease after emergence. Our initial pathogenicity data

suggests that it can also cause disease in roots and pods. The new pathogen showed more aggressiveness in comparison to already reported *F. equiseti*. We are currently assembling its first whole genome for accelerated identification of potential virulence effectors and as a resource for the scientific community. We are also screening commercial varieties and diverse germplasm to identify the diversity in pathogen virulence and for genome wide association study (GWAS) leveraging SoySNP50K BeadChip genotypic data. Our research aims to explore the pathogenomics of *F. luffae* as well as potential sources of resistance in soybean germplasm for this newly identified fungal pathogen species in USA.

Occurrence of new *Sdh* gene mutations in *Alternaria solani* with potential impact in the management of early blight

S. Shrestha¹, J. Pasche¹, N. Gudmestad¹, I. Mallik¹; ¹North Dakota State University; **Abstract:** Reductions in efficacy of single-site mode of action (MoA) fungicides has been an ongoing challenge in the management of potato early blight caused by *Alternaria solani*. Single-site MoA fungicides, including succinate dehydrogenase inhibitors (SDHI), play a substantial role in successful early blight management; however, reduced-sensitivity or resistance has developed in *A. solani* to SDHI fungicides registered for use on potatoes. We detected four new mutations, three in *SdhB* and one in *SdhD*, along with the co-occurrence of two mutations in a single isolate, associated with SDHI fungicide resistance. In vitro sensitivity assays conducted on the *A. solani* isolates with one or two of the novel mutations displayed reduced sensitivity to resistance to fluopyram and pydiflumetofen, with resistance factors ranging from 2 to >500 and 1.5 to >100, respectively. Significantly reduced disease control was observed by fluopyram and pydiflumetofen in 12 and 9 of 12 *Sdh* mutant *A. solani* isolates, respectively, in in vivo greenhouse assays. Loss in disease control in the greenhouse ranged from 20.4% to 65.8% for fluopyram and 2.7% to 84.9% for pydiflumetofen compared to the baseline isolates. Results from in vitro ($r = 0.877$; $P < 0.0001$) and in vivo assays ($r = 0.859$; $P < 0.0001$) indicated the cross sensitivity in most of the *A. solani* isolates between the fluopyram and pydiflumetofen. Grower recommendations for early blight management will be improved by integrating these findings.

Enter to Get Trapped: Stem Rust Infection Mechanism in Cereal Crops as a Driving Force Against *Rpg5* Immunity Receptor Evolution

S. Solanki¹, G. Ameen¹, R. Brueggeman²; ¹South Dakota State University, ²Washington State University; **Abstract:** Immunity activation relies on the perception of pathogens. The host molecules manipulated by pathogens are susceptibility targets of the genome that are under selective pressure and may or may not have direct function in immunity. However, pathogen selection pressure could lead to the fusion of these targets into host immune receptors as an atypical integrated domain (ID) serving as ‘pathogen sensors.’ Stem rust resistance gene *Rpg5* in barley represents a model for studying the aforementioned evolutionary trajectory of plant NLR-ID proteins. Our genome mining identified a putative kinase in the barley genome that could be the ancestral paralog of the atypical kinase-ID domain of *Rpg5*. We named it ‘PRK1’ and characterize it as an ortholog of the Arabidopsis guard cell kinase *AtApk1b*. We hypothesize that pathogen effectors can manipulate PRK1 to induce stomatal opening in the dark as an entry route, thus forcing the host to evolve the *Rpg5*NLR-ID_{Kinase} which acts as an immunity sensor to recognizes the pathogen’s attempt to manipulate PRK1. We have previously shown that PRK1 is expressed in barley stomata as well as in mesophyll cells. We are transforming Arabidopsis *apk1a-b* mutants with PRK1 driven by Arabidopsis promoter to validate if barley PRK1 can rescue stomatal guard cell opening phenotype and assess any phenotypic effects on non-host Arabidopsis. Our overarching goal is to dissect NLR-ID evolution mechanism in plant immunity using *Rpg5*-stem rust model.

T

Haplotype and transcriptome analyses reveal genes and gene regions that promote FHB resistance in spring wheat

S. Thapa¹, J. Fiedler², S. Xu², G. Bai³, B. Poudel⁴; ¹North Dakota State University, ²USDA-ARS Cereal Crops Research Unit, ³USDA-ARS Hard Winter Wheat Genetics Research Unit, ⁴Donald Danforth Plant Science Center; **Abstract:** Several quantitative trait loci (QTLs) associated with Fusarium head blight (FHB) resistance have been identified, of which seven of them are well-characterized (*Fhb1...Fhb7*). Chinese spring wheat, Sumai3 is the source for three major QTLs for FHB viz., *Fhb1*, *Fhb2* and *Fhb5*. Interestingly, this FHB-resistant cultivar was developed by crossing two moderately susceptible parents, “Funo” and “Taiwanxiaomai”, which suggests that the resistance was inherited by appropriate combinations for favorable haplotype blocks. This study aims to decipher those specific combinations of haplotype blocks in Sumai3 and trace the movement of resistance-associated gene regions across various derivatives. The whole genome sequencing of the Sumai3 along with resequencing of thirty lines harboring Sumai3 background was carried out and the haplotype blocks were characterized. Coupled with RNASeq information, we aim to explore expression patterns of resistance-associated genes and their conjugation to these defined haplotype blocks. Preliminary analysis has shown that the entire 3B chromosome in Sumai3 harboring *Fhb1* was inherited by a single parent, Taiwanxiaomai. But contradictorily, Taiwanxiaomai possessing this chromosome is moderately susceptible, suggesting Funo-derived genes are necessary for strong resistance. This comprehensive analysis of the genomes of these important sources of FHB-resistance will expand our knowledge on what is required to combat disease.

Genome-Wide Identification of Fusarium Head Blight Resistance and Susceptibility Loci in USA Wheat Breeding Programs

L. Tidakbi¹, G. Bai², K.W. Jordan², J. Rupp Noller¹; ¹Kansas State University, ²USDA-ARS; **Abstract:** Fusarium Head Blight (FHB) is a major pathogenic fungal disease in wheat caused by the *Fusarium* species complex such as *Fusarium graminearum* (*Gibberella zeae*) with a major economic impact on wheat production and human health due to the production of mycotoxins. Breeding for resistance in adapted wheat varieties is not trivial due to the complexity of the wheat genome. We are identifying FHB resistance quantitative traits loci (QTLs) and developing a customized Practical Haplotype Graph (PHG) database coupling phenotypic and genotypic data from a diversity panel of 300 wheat cultivars. The objective of identifying novel QTLs for wheat resistance to FHB and making a haplotype-informed prediction is underway with our customized wheat PHG database and two years of field and greenhouse data evaluated. Results show significant variability in phenotypic FHB severity, fusarium-damaged kernels (FDK), and deoxynivalenol (DON) across breeding lines. Additionally, ~15x sequencing data from these lines produced nearly 600,000 variants across the wheat genome for association analyses and model building. Using best linear unbiased estimates of our traits, we mapped novel QTLs conferring resistance and susceptibility alleles for FHB using this panel of wheat lines using mixed linear model genome-wide association studies (GWAS). Markers will be validated and developed into diagnostic markers for marker-assisted selection of FHB-resistant lines in breeding programs across the USA.

Discovering Bacterial Leaf Streak Disease Resistance Loci in Barley in the Interspecific Cytonuclear Multi-Parent Population (CMPP)

J. Tilstra¹, ¹M. Ahmad¹, T. Pawar¹, A. Beery², S. Bodenheimer^{2,3}, S. Yang⁴, S. Solanki¹, E. Fridman², G. Ameen¹; ¹South Dakota State University, ²Plant Sciences Institute, Volcani Agricultural Research Organization (Israel), ³Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture (Israel), ⁴Cereals Crops Research Unit, Edward T. Schafer Agricultural Research Center; **Abstract:** A noticeable impact of climate change is the outbreak of emerging diseases. One such disease that was not new to the US but has become predominant in the Northern Great Plains is Bacterial leaf streak or BLS. BLS is a bacterial disease of barley, wheat, and oats caused by *Xanthomonas translucens*. The initial disease symptoms are water-soaking and progressing into large necrotic streaks on leaves. BLS can also infect barley spikes causing black chaff symptoms reducing yield and rendering inferior-quality barley grains that cannot be used for malting. Due to the absence of resistant varieties and limited chemical or biological control, it is essential to find and deploy host resistance in the current, high-yielding varieties.

In this study, we screened 692 doubled haploid lines belonging to the interspecific Cytonuclear Multi-Parent Population (CMPP) to discover marker-trait association (MTAs) for BLS resistance. We used spray inoculations and recorded the disease phenotypic data using our 1 to 9 BLS disease rating scale. The CMPP population was genotyped using the 50K Illumina Infinium chip. After quality control, we used 19,988 SNP markers for a genome-wide association study and used MLM, GLM, Blink, and farmCPU models to find MTAs linked to BLS resistance. Our results found significant MTAs on chromosomes 1H, 5H, and 7H. Ongoing field testing will validate our results to confirm the BLS genetic resistance in these identified genomic regions.

“N” Field Observations: Extension Programing in the 21st Century

A. Timmerman¹, T. Jackson-Ziems¹, D. Mangel¹, J. Thomas¹, S. Melvin¹, C. Burr¹, A. Jhala¹, J. Peterson¹;

¹University of Nebraska; **Abstract:** The way Extension across the country engages with clientele has dramatically changed in the last 10 years, but especially during/following the pandemic. One of the driving factors affecting engagement is the variation of attention span among generational groups. The attention span of Millennials is 12 seconds vs. Gen Z is 8 seconds before they move onto the next item of interest. Thus, how do we “catch” their attention in that short amount of time? In addition, where clientele receive their information has also changed. According to survey data from 2022, 77% of Gen Z adults and 67% of millennial rely on social media for news of current events. In response to these changes, the Nebraska Extension Crop and Water team created “N” Field Observations. The three main goals of which are to provide timely agronomic information to Nebraska farmers and agronomists, to strengthen the engagement of extension educators and specialists, and to provide real time cropping updates to empower stakeholders to make management decisions. The program is formatted to run at least one video every week on social media platforms; X, Instagram, YouTube, and Tik Tok. In 2023, “N” Field Observations shared 36 videos that were viewed 40,447 times, with 133 shared and 635 engagements via X; 2,170 reach and 40 shares via Facebook; 810 reach and 74 likes via Instagram; and 3,878 views via YouTube. Overall, the use of social media is a positive way to interact with clientele in the 21st Century.

V

Predictive models for wheat stripe rust infection in Kansas

E. Vicentin¹, K. Andersen-Onofre¹, E. De Wolf¹; ¹Kansas State University; **Abstract:** Wheat stripe rust, *Puccinia striiformis* f.sp. *tritici* (Pst), causes severe yield loss in Kansas with statewide yield losses resulting often exceeding \$170 million. The objective of this study was to develop predictive models for stripe rust infection-based bioassays conducted in wheat field environments over 3 years. These bioassays involved inoculating susceptible wheat seedlings with Pst and placing plants in a field environment or a dew chamber at 13°C for 16 hours. The plants placed in the dew chamber helped verify the viability of spores used in the daily inoculations (control). The environment at the field site was monitored with a weather station equipped with temperature, humidity, rain, and leaf wetness sensors. Following their treatment, the plants were placed in a growth chamber and incubated at 12-20°C with 16 hours of light. Disease incidence and severity were visually evaluated 14 days after inoculation. Logistic regression was used to model the probability of infection based on environment observed during the field exposure. Preliminary models correctly classified 79-84% of 96 cases correctly, and k-fold cross-validation estimates of model accuracy were 72-73%. These models indicate that stripe rust infection is favored by temperature 9-23°C and at least 6 hours with humidity >83%. The presence of rain (>0 mm) also favored infection. These models are being adapted for use as part of a forecasting system for stripe rust in Kansas.

W

Surveying plant-parasitic nematodes in Kentucky corn fields

L. Warner¹, W. Barlow¹, K.A. Wise¹; ¹University of Kentucky; **Abstract:** Plant-parasitic nematodes can devastate corn (*Zea mays* L.) yield depending on type and population. A survey was conducted in 2023 to better understand plant-parasitic nematode distribution in Kentucky corn fields. Samples were collected from 40 fields across 13 counties and sent to the University of Illinois Plant Clinic for corn vermiform nematode quantification. Nematode populations were reported as frequency per 100cc of soil and severity levels were based on density as determined by Niblack et al., 2005. Results confirmed that samples from all counties had at least one field with lance (*Hopolaimus* spp.), lesion (*Pratylenchus* spp.), and spiral (*Helicotylenchus* spp.) nematodes present. Sixty-one percent of counties sampled had at least one field with stunt (*Tylenchorhynchus* spp.) nematodes, 15 percent of counties had at least one field with dagger (*Xiphenema* spp.) nematodes, and almost 8 percent of counties had at least one field with ring (*Criconeimella* spp.), pin (*Paratylenchus* spp.), and root knot (*Meloidogyne* spp.) nematodes present, respectively. Overall, fields sampled generally had non-significant to minor nematode risk levels. Stunt nematodes were detected at a moderate risk level in 8% of fields, and very high levels of lesion and spiral nematodes were detected in 3.3% and 2.5% of fields, respectively. This survey is ongoing and will help determine corn nematode distribution and future management considerations in Kentucky.

Effect of Adjuvants on Fungicide Performance Evaluated by Visual Rating and Multispectral Imaging in Oats

A. Weber¹, J. Skelton², C. Hoerning², B. Berghuis³; ¹WinField United, ²Land O'Lakes, ³University of Wisconsin River Falls; **Abstract:** Foliar fungicide applications are used to manage rust pathogens in agricultural production. Adjuvants containing non-ionic surfactants (NIS) can enhance pathogen management with fungicides due to improved leaf coverage and canopy penetration. Qualitative visual ratings have been traditionally used to quantify disease severity. In contrast, multispectral imaging can provide quantitative data by detecting spectral aberrations associated with disease symptoms. We assessed Oat Crown Rust (*Puccinia coronata*) severity through visual ratings and multispectral imaging following a fungicide application. Fungicide treatments included Azoxystrobin (AZOXY) alone and AZOXY + Prothioconazole (PROT). Both fungicide treatments were applied alone or with a NIS. Visual ratings were collected at seven and fourteen days after pathogen inoculation. Multispectral imaging data was collected daily with multispectral scanners (Phenospex PlantEye F500) to quantify disease pressure. Assessing oat crown rust through visual qualitative ratings and multispectral imaging were both effective methods to evaluate products to manage of oat crown rust on oat plants. Statistically less disease was observed with an application of AZOXY + PROT and AZOXY + PROT + NIS compared to untreated control.

Survey of Rose Rosette Disease in the Hamann Rose Garden of Lincoln, NE

L. Wilson¹, K. Broderick¹; ¹University of Nebraska - Lincoln; **Abstract:** The loss of cultivated rose perennials has greatly increased since the introduction of the rose rosette disease (RRD) to the US. RRD is caused by the rose rosette Emaravirus, which is primarily vectored by grafting and feeding of eriophyid mites (*Phyllocoptes fructiphilus*). Symptoms of RRD include witches' broom, phyllody, and stunting, impacting the blooms and reducing the longevity of perennial roses. RRD was first reported east of the Rocky Mountains in the 1940s, and soon found its way to Nebraska in 1968, marking the first Midwest state to be affected by RRD. Recently, RRD has become more prevalent in Nebraska, affecting numerous rose varieties. In 2022, RRD was visually diagnosed in the Hamann Rose Garden, a historic garden in Lincoln, NE. The Hamann Rose Garden was Nebraska's first test garden, established in the 1940's. The garden currently contains over 100 varieties of both cutting and landscape roses. This survey used both conventional and quantitative reverse transcription-PCR to confirm RRD. Our observations will contribute to the education of visitors to this public garden, direct management of RRD, and the understanding of RRD susceptibility in Hybrid Tea, Floribunda, and Grandiflora rose lineages in Lincoln, Nebraska.

Y

Validation of the real-time PCR assay for rapid detection and identification of the pin nematode, *Paratylenchus nanus* type B

S. Paudel¹, G. Yan¹; ¹North Dakota State University; **Abstract:** This study validates the real-time PCR assay developed for identification of *Paratylenchus nanus* type B, a prevalent plant-parasitic nematode in field peas in North Dakota. Species-specific primers were designed targeting the internal transcribed spacer region of ribosomal DNA. The specificity of the primers was evaluated by doing in-silico analysis and was confirmed by PCR tests. The real-time PCR assay was optimized for amplification temperature and primer concentration. Detection sensitivity was determined using DNA extracts from 20 nematodes followed by two-fold serial dilutions. The assay could detect an equivalent to 1/51 of a single nematode. Standard curve was generated by plotting quantification cycle (Cq) values against corresponding log10 numbers of nematodes with amplification efficiency of 92.6%. Subsequently, the assay was applied to 21 field soil samples collected from 10 counties in North Dakota and Minnesota. Nematodes were extracted using sugar centrifugal-floatation method and DNA was extracted from single nematodes by proteinase K method. Real-time PCR was done using the designed species-specific primers. Amplifications observed in the assay showed presence of the target *P. nanus* type B DNA in all the soil samples with pin nematodes. These results validated the developed real-time PCR assay to be specific, sensitive and efficient, providing a useful tool for rapid detection of *P. nanus* type B from infested fields.

Z

Whole-genome nucleotide polymorphism applied to a *Colletotrichum coccodes* global population

M. Zaccaron¹, C. Johnson¹, B. Pandey², F. Shahoveisi³, J. Pache¹, N. Gudmestad¹, I. Mallick¹, J. Pasche¹; ¹North Dakota State University; ²Oregon Department of Agriculture Plant Health Program, ³University of Maryland; **Abstract:** *Colletotrichum coccodes* causes black dot and plays a role in the potato early die complex. For the first time, SNPs from whole-genome sequencing were used in *C. coccodes* phylogeny. A group of 42 isolates collected from potatoes grown in Australia, Chile, South Africa, Europe, and the US were sequenced using an Ion torrent platform. The resulting WGS-SNP based phylogenetic tree consisted of two major clades, which were further divided into four subclades. This clustering was supported by analysis of principal components, principal coordinates, and molecular variance. Isolates belonging to different subclades were separated from each other by substantial nucleotide polymorphisms. Location of origin explain 48% of genetic variation among isolates. Geographic source of isolates did not completely explain subclade clustering. However, most isolates from a given region tended to belong to the same subclade. One of the subclades was composed exclusively by isolates from South Africa. However, US isolates clustered in three subclades, grouping with isolates from every other region. Results suggest some geographic restriction of *C. coccodes* genotypes; however, it appears global dissemination of this pathogen is present. Therefore, resistance breeding efforts would benefit from being informed of *C. coccodes* global pathogenicity profile. WGS will be an important tool to mechanistically understand *C. coccodes* pathogenesis and how plants can overcome it.