**Multi-State S1083**

**Ecological and genetic diversity of soilborne pathogens and indigenous microflora**

**Report for project period: October 1, 2019 to September 30, 2020**

**Report date: October 19, 2020**

**Annual Meeting Date: September 8, 2020**

**Participants:**

Dr. Nathan Slaton (Administrative Advisor), University of Arkansas

Dr. Sydney Everhart, University of Nebraska

Dr. William Kingery, Mississippi State University

Dr. Shankar Ganapathi Shanmugam (Secretary), Mississippi State University

Dr. Alejandro Rojas, University of Arkansas

Dr. Terry Spurlock, University of Arkansas

Dr. Fulya Baysal-Gurel, Tennessee State University

Dr. Maria del Mar Jimenez-Gasco , Pennsylvania State University

Dr. Soledad Benitez Ponce (Chair), The Ohio State University

**Brief summary of annual meeting** (12,000 char) – can include link to minutes/video

The S1083 2019-2020 meeting was held September 08, 2020. In past years, the group had agreed to meet in conjunction with the annual meeting of the American Phytopathological Society (APS); however, due to the travel restrictions and changes in activities in 2020, from both APS and individual institutions, the group met virtually, through Zoom. During the meeting, which lasted 3 hours (10:00 am to 1 pm ET), attendees a) provided an update of their research, b) discussed about funding and writing opportunities, and c) began brainstorming on a potential review paper to be developed by the group. The meeting was led by Dr. Benitez Ponce, with Dr. Ganapathi Shanmugam as secretary. All attendees actively participated and contributed to the meeting. Common themes that were covered and discussed included the use of cover crops to manage soil borne pathogens and soil health, studies of plant endophytes and their potential role (or relationship with) as soilborne pathogens, and impact of management practices in microbial communities. Regarding funding opportunities, the team discussed about a proposal which was in early stages of development focused on rye as a cover crop. In addition, the focus of cover crop was highlighted as a common theme, and this could potentially be applied to management of soil borne diseases in specialty crops. A follow-up meeting was planned to discuss about development and writing of a review paper, as an output from this team. Finally, the group will aim to meet in person for the 2020-2021, during the 2021 APS annual meeting (to be held in Memphis, TN). Additional detail is provided in the meeting minutes, as well as the video recording, available in the following link: <https://osu.box.com/s/5es0a835zo6ynlkzzyd34rlqdfwtbrkv>.

**Accomplishments:**

**Short-term outcomes**

**1**. Isolation, characterization and screening of fungicide sensitivity of soilborne pathogens of row crops and woody ornamentals, including Sclerotinia, Rhizoctonia, Cercospora, Xylaria, and Phytopythium.

2. Evaluation of agricultural management practices, in particular the use of cover crop, for control of soil borne diseases. The use of cover crops for plant and soil health management has been evaluated across different production settings and states (OH, AR, TN, MS, NE). Cropping systems and diseases for which cover crop integration is being studied include: corn and soybean production, and ornamental nursery. In addition, cover crop contributions to disease development, as potential alternate host of soilborne pathogens is of relevance, as shown particularly for Xylaria and tap-root decline.

3. Characterization of microbial community profiles (bacteria and fungi) in soils, in response to management strategies aiming at building of suppressive soils. The effective implementation of cover crops or other management practices that contribute to disease suppression, is accompanied by our understanding of potential enrichment of beneficial microbial communities in soils. For instance, changes in microbial community profiles have been evidence in response to cover crops is currently on-going at multiple locations in this project.

**4.** Evaluation of the efficacy of chemical and biorational products for controlling soil-borne diseases in nursery (TN) and field crop production (OH, AR), with particular success in the woody ornamental industry.

5. Screening and characterization of available biorational products and novel isolates for biological control and plant growth promotion.

**Outputs**

1. Isolate collections and characterization of fungicide sensitivity and population variation for at least five different soil borne pathogens of row crops and ornamentals.

2. Establishment and/or continuing field experiments to evaluate cover crop and rotation management in five US states (OH, AR, TN, MS, NE), with emphasis on soilborne disease management.

3. Performed field trials to study tap root decline incidence, ecology and spatial distribution in two southern states (AR, MS)

4. Training of three postdoctoral researchers, 13 graduate students, five undergraduate students, and 11 research technicians (part and full-time).

5. Publication of 13 refereed journal articles and six extension reports and one patent.

6. Eighteen oral presentations in scientific meetings and eight workshops, extension presentations and training opportunities.

7. One doctoral and two master dissertations.

8. Description of a new disease in woody ornamentals: *Phytopythium vexans* as causal agent of crown and root rot of ginko and red maple.

**Activities**

1. Field experiments were established, or continued to be studied to understand cover crop and:

a) different soybean maturity groups (NE)

b) relationship with timing of cover crop termination (OH)

c) soil health and beneficial microbial communities (OH, MS, TN)

d) soilborne pathogen populations, including *Xylaria* species (Arkansas)

e) soilborne disease incidence and suppressive soils in nursery production (TE)

f) interactions with other management practices, such as rotational sequences (OH)

2. Isolation, characterization and screening of fungicide sensitivity of soilborne pathogens, including Sclerotinia and Rhizoctonia (NE), Cercospora (AR), Xylaria (AR). Phytophthora (-like) oomycetes in woody ornamentals (TN).

3. Survey fields to understand the ecology of taproot decline and its spatial distribution (AR)

4. Development, characterization and testing of biocontrol agents, including

a) isolate screening and genomic analysis of bacterial endophytes against rice fungal and oomycete pathogens (AR)

b) evaluation of commercial biorational products as alternatives for nursery ornamental production (TE)

c) efficacy of colonization and yield impact of arbuscular mychorrizal fungi in soybean (OH)

d) isolation and screening for fungi associated to soybean cyst nematodes (OH)

**Milestones**

1. Protocol development to evaluate fungal pathogen impact on seed quality (AR)

2. Isolation and PCR based characterization of *Xylaria*

3. Description of *(Phytopythium vexans*) as causal agent of crown and root rot of ginko and red maple

4. Tests of fungicide and biopesticide efficacy for developing guidelines for management, with results indicating variability in susceptibility to different fungicides, including insensitivity to currently used fungicides

5. Sampling and on-going characterization of microbial communities associated to multiple experiments across project participants

**Impact**

**Activities**

1. Test of different soybean varieties and fungicide sensitivities for control of tap root decline in soybean

2. Characterization of soilborne pathogens relevant in various cropping systems

3. Provided the woody ornamental nursery industry with improved, efficacious, cost-effective and sustainable recommendations for soil-borne disease management (TN).

4. Developed/continued research collaboration with industry partners for understanding of efficacy of microbial inoculants for soilborne disease management

5. Shared data of management practices, such as cover crop and rotation sequence with agronomist and extension specialist to be presented to growers

**Milestones**

1. As part of the various fungicide sensitivity assays, information was generated regarding specific effect of commercially available fungicides. Some of the results, could be applied to better manage specific soilborne diseases. For instance, for control of tap root decline, the β-tubulin inhibitors, thiophanate-methyl and thiabendazole, shown efficacy, and in-furrow applications might be more successful. Whereas for *Rhizoctonia zea*, evidence shows insensitivity to azoxystrobin fungicide.

2. Characterization of pathogenic populatins have resulted in understanding of variation in aggressiveness across different hosts, as well as new reports of pathogens in crops. For instance, in Nebraska, *Rhizoctonia solani* and *R*. *zeae* were most abundant in row crops collections, with R. zeae being more aggressive in soybean. In addition, a species of *Pythopythium* was identified as causal agent of root rot in ginko and red maple in nursery production.

I**ndicators**

1. Understanding and application of management practices against taproot decline disease.

2. Adoption of cover crop practices by producers, including the ornamental nursery industry

3. Understanding of complexity of use and efficacy of biorational and biocontrol products

4. Availability of effective fungicide and resistance variety information to aid in grower’s decision making process

5. Training of scientists, growers and extension agents.

6. Delivery of scientific information to the scientific community (publications and presentations at scientific conferences), stakeholders and general public.

7. Presentation of alternatives for soilborne disease management.

**Publications**

1. **Baysal-Gurel, F.,** Liyanapathiranage, P., Panth, M., Avin, F.A., and Simmons, T. 2020. First report of *Phytopythium vexans* causing root and crown rot on flowering cherry in Tennessee. Plant Disease. https://doi.org/10.1094/PDIS-06-20-1166-PDN
2. Castroagudín, V. L., Weiland, J. E., **Baysal-Gurel, F.**, Cubeta, M. A., Daughtrey, M., Gauthier, N. W., LaMondia, J., Luster, D. G., Hand, F. P., Shishkoff, N., Williams-Woodward, J., Yang, X., LeBlanc, N., and Crouch, J. A. 2020. One clonal lineage of *Calonectria pseudonaviculata* is primarily responsible for the boxwood blight epidemic in the United States. Phytopathology. https://doi.org/10.1094/PHYTO-04-20-0130-R.
3. Dundore-Arias, E.A. Eloe-Fadrosh, L.M. Schriml, G.A. Beattie, F.P. Brennan, P.E. Busby, R.B. Calderon, S.C. Castle, J.B. Emerson, **S. Everhart**, K. Eversole, K. Frost, J. Herr, A.J. Huerta, A.S. Iyer-Pascuzzi, A. Kalil, J.E. Leach, J. Leonard, J.E. Maul, B. Prithiviraj, M. Potrykus, N.R. Redekar, J.A. Rojas, K.A.T. Silverstein, D. Tomso, S. Tringle, B. Vinatzer, and L. Kinkel. 2020. Community-driven Metadata Standards for Agricultural Microbiome Research. *Phytobiomes* 4: 115-121.
4. Hoeksema JD, Averill C, Bhatnagar JM, Brzostek E, Buscardo E, Chen K, Liao H, Nagy L, Policelli N, Ridgeway J, Rojas JA, Vilgalys R. 2020. Ectomycorrhizal plant-fungal co-invasions as natural experiments for connecting plant and fungal traits to their ecosystem consequences. Frontiers in Forests and Global Change 3:84.
5. Hudson, O., Walilullah, S. Hand, J. Gazis-Seregina, R., **Baysal-Gurel, F.**, and Ali, E. 2020. Novel method for detection of *Phytophthora capsici* in irrigation water using loop-mediated isothermal amplification. J. Vis. Exp., e61478, doi:10.3791/61478.
6. Koehler‐Cole, K., **S.E. Everhart**, Y. Gu, C.A. Proctor, M. Marroquin‐Guzman, D.D. Redfearn, and R.W. Elmore. 2020. Is allelopathy from winter cover crops affecting row crops?. *Agricultural & Environmental Letters*, *5*(1), e20015.
7. Ortega, L., Walker, K.A., Patrick, C., Wamishe, Y., Rojas, A. and Rojas, C.M., 2020. Harnessing Pseudomonas protegens to Control Bacterial Panicle Blight of Rice. Phytopathology, pp.PHYTO-02.
8. Panth, M., **Baysal-Gurel, F.**, Simmons, T., Addesso, K., and Witcher, A. 2020. Impact of winter cover crop usage in soilborne disease suppressiveness in woody ornamental production system. Agronomy 10(7), 995; https://doi.org/10.3390/agronomy10070995
9. Panth, M., Hassler, S., and **Baysal-Gurel, F.** 2020. Methods for management of soilborne diseases in crop production. Agriculture. Agriculture *10(*1), 16; <https://doi.org/10.3390/agriculture10010016>
10. Sciarresi, C., C. Proctor, E.R. Haramoto, L.E. Lindsey, G.I. Carmona, R. Elmore, **S. Everhart**, W. Looker, M. Marroquin-Guzman, J. McMechan, J. Wehrbein, R. Werle, and M. Salmeron. 2020. Evaluating short-season soybean management adaptations for cover crop rotations with a crop simulation model. *Field Crops Research* 250: 107734.
11. Tekin, N.T., Kaplanoglu, E., Erdemir, E., **Baysal-Gurel, F.,** Uyanik, C., and Hargrove, S. K. 2020. Modeling and Testing of Magnetic Speed Controlled Submersible Robot for Hydroponic Production. 2019 SoutheastCon. IEEE pp. 1-4. DOI:10.1109/SoutheastCon42311.2019.9020356.
12. Tolbert, A.C., T.N. Spurlock, and R. Hoyle. 2019. Cultivar responses to sudden death syndrome and harvest seed pathogens, 2018. doi:10.1094/PDMR14
13. Tolbert, A.C., T.N. Spurlock, and R. Hoyle. 2019. Efficacy of various seed treatments on taproot decline in Southeast Arkansas, 2018. doi:10.1094/PDMR14

**Extension Publications**

1. Baysal-Gurel, F. 2019. Control of Phytophthora on boxwood. Chase Digest August 2019 Issue Volume 7(8).
2. Baysal-Gurel, F. 2020. Phytophthora root and crown rot of boxwood. Tennessee Greentimes. Volume 21/No. 1 Spring 2020.
3. Chilvers, M., McCoy, A., Noel, Z., Rojas, A., Faske, T., Mueller, D., Smith, D., Tenuta, A., and Wise, K. Detection and Prevalence of Oomycete Seedling Diseases On Soybean. CPN 5003. DOI: doi.org/10.31274/20200918-1
4. Chilvers, M., McCoy, A., Noel, Z., Rojas, A., Faske, T., Mueller, D., Smith, D., Tenuta, A., and Wise, K. Detection and Prevalence of Oomycete Seedling Diseases on Soybean. CPN 5003. DOI: doi.org/10.31274/20200918-1
5. Faske, T., Smith, S., Spurlock, T., Wamishe, Y. MP154 Arkansas Plant Disease Control Products Guide. 2019.
6. T.N. Spurlock, Tolbert, A.C., and R. Hoyle. 2019. Taproot decline Trial Summaries 2018-2019. Arkansas Soybean Research Series.

**Patents**

U.S. Provisional Application No. 63/036,202. *Pseudomonas protegens* and products thereof to control bacterial panicle blight of rice. Filed June 8, 2020.

**Authorization:**

\*Limited to three pages or less exclusive of publications. Details may be appended.

**APPENDIX 1. Detailed activities by participating researchers**

**Dr. Sydney Everhart, University of Nebraska**

**Major Goals and Objectives**

Objective 1. Evaluate the biology and diversity of soilborne pathogens, associated and antagonistic microorganisms, and environmental conditions in the context of the whole-system phytobiome. This objective includes traditional, metagenomics, and spatial/temporal methodologies to understand microbial community dynamics that determine soilborne disease incidence and severity on economically important crops in the U.S.

Sub-objective (Everhart): characterize the effect of cover crops planted after different soybean maturity groups on chemical and physical properties of soil and soybean and corn yields.

Objective 2. Evaluate the efficacy of soil-borne disease management strategies (chemical, biorational/biological, cultural) and characterize the associations among microbial community profile, soil physicochemical properties, environmental factors and disease suppression.

Sub-objective (Everhart): Conduct surveys for *R. solani* and *Sclerotinia* sclerotiorum from fields in Nebraska and from other North Central states to determine sensitivity of isolates to commercial fungicides with different modes of action and to characterize the structure of pathogen populations within and between fields.

**Accomplishments:**

Objective 1: Our aim was to characterize soil fungal community diversity in a soybean- oat/rye system in three experimental trials in Nebraska. A fallow control treatment was utilized at each location and cover crops consisted of a 50:50 mixture of oats and cereal rye. The soil was sampled at planting and at harvest. Bulk DNA was purified and the ITS region sequenced. A community sequencing analysis is underway and will be used to compare fungal diversity within and across locations. Results will contribute to understanding how cover crops impact fungal communities in Nebraskan soils and identify fungi that may be potential indicators of disease incidence and yields. Other aspects of our research contributed to understanding the impact of cover crop rotations, understanding of allelopathy, and methods of data curation.

Objective 2: We have determined the “baseline sensitivity” of *Sclerotinia sclerotiorum* isolates never exposed to fungicides and identified a single discriminatory dose for future assays. We estimated sensitivity to all four fungicides for 154 isolates. Results thus far show no association of fungicide exposure and fungicide sensitivity within field populations, however, individual isolates with less sensitivity have been observed. A total of 139 isolates of *Rhizoctonia* were characterized according to their morphological and sequences to their subgroup level. Results from our study indicated that the *R. solani* AG-4 and *R*. *zeae* were most abundant in Nebraska from row crops collection. Interestingly, *R. zeae* were most aggressive to soybean when temperature at which *in planta* evaluations were conducted was higher than for *R. solani*. This was a surprising finding of this research because *R. zeae* was previously reported mostly as a pathogen of significance for turf grasses but was not considered to be an aggressive pathogen on row crops. We also characterized the diversity and pathogenicity of *Rhizoctonia* spp. from Sandhills. Isolates were pathogenic on native grasses (sand bluestem and needle-and-thread) and soybean. The Sandhills grasslands harbor a unique composition of *Rhizoctonia* spp. that have potential to cause disease on soybean when this area is converted to cultivated land. Fungicide sensitivity was determined for all *Rhizoctonia zeae* isolated. A crucial finding was that Rhizoctonia zeae was completely insensitive to azoxystrobin fungicide, which is currently one of most common fungicides used owing to its expected high specificity of action. Population structure of *Rhizoctonia zeae* is being characterized, which will enable a deeper insight into the biology and mode of spread of this pathogen.

**Impacts**

Training

Objective 1: Postdoc (Margarita Marroquin-Guzman) provided training in computational analyses and completed undergraduate and graduate student mentoring. This postdoc has now moved to a permanent position elsewhere.

Objective 2: Training continued for 3 PhD students, all of whom presented their research at various scientific meetings and one student completed their dissertation. In the past year, one undergraduate students from UNL was involved in this project and received training in microbiology and molecular genetic techniques.

Dissemination

Results have been disseminated via presentation of posters and oral presentations at local, regional, and national scientific meetings, which are listed below.

Target Audience

The primary target for dissemination includes the broader scientific community concerned with pest evolution and pesticide resistance management. Targets also include postdocs, graduate students, and undergraduate students who receive training in microbiology, molecular genetics, genomics, and scientific communication.

**Products**

Publications:

1. Koehler‐Cole, K., S.E. Everhart, Y. Gu, C.A. Proctor, M. Marroquin‐Guzman, D.D. Redfearn, and R.W. Elmore. 2020. Is allelopathy from winter cover crops affecting row crops?. *Agricultural & Environmental Letters*, *5*(1), e20015.
2. Sciarresi, C., C. Proctor, E.R. Haramoto, L.E. Lindsey, G.I. Carmona, R. Elmore, S. Everhart, W. Looker, M. Marroquin-Guzman, J. McMechan, J. Wehrbein, R. Werle, and M. Salmeron. 2020. Evaluating short-season soybean management adaptations for cover crop rotations with a crop simulation model. *Field Crops Research* 250: 107734.
3. Dundore-Arias, E.A. Eloe-Fadrosh, L.M. Schriml, G.A. Beattie, F.P. Brennan, P.E. Busby, R.B. Calderon, S.C. Castle, J.B. Emerson, S. Everhart, K. Eversole, K. Frost, J. Herr, A.J. Huerta, A.S. Iyer-Pascuzzi, A. Kalil, J.E. Leach, J. Leonard, J.E. Maul, B. Prithiviraj, M. Potrykus, N.R. Redekar, J.A. Rojas, K.A.T. Silverstein, D. Tomso, S. Tringle, B. Vinatzer, and L. Kinkel. 2020. Community-driven Metadata Standards for Agricultural Microbiome Research. *Phytobiomes* 4: 115-121.

Oral and poster presentations:

1. Gambhir, N., Kodati, S., Adesemoye, A.O., and Everhart, S.E. 2020. Fungicide resistance: Screening and risk-assessment of *Rhizoctonia zeae* populations in Nebraska. APS Plant Health 2020 Meeting oral presentation on Aug. 14, 2020.
2. Gambhir, N., Kodati, S., Adesemoye, A.O., Olutoyosi, A.O., Bissonnette, K., Bradley, C.A., Chilvers, M., Fakhoury, A.M., Jackson-Ziems, T.A., Leandro, L.F.S., Little, C.R., Malvick, D.K., Mathew, F.M., Nelson, B.D., Sassenrath, G., Smith, D.L., Telenko, D.E.P., Wise, K.A., and Everhart, S.E. 2020. Distribution and population structure of *Rhizoctonia zeae* in the North Central United States. APS Plant Health 2020 Meeting poster presentation on Aug. 3, 2020.
3. Nieto-Lopez, E.H., Miorini, T.J.J., Chilvers, M., Giesler, L.J., Jackson-Ziems, T.A., Kabbage, M., Mueller, D.S., Smith, D.L., Tovar-Pedraza, J.M., Willbur, J.F., and Everhart, S.E. 2020. Fungicide sensitivity of *Sclerotinia sclerotiorum* from dry bean and soybean in the U.S. APS Plant Health 2020 Meeting poster presentation on Aug. 3, 2020.
4. Higgins, R., C. Wulkop, E.H. Nieto-Lopez, and S. Everhart. 2020. Sources of white mold resistance derived from wide crosses in common bean and fungicide sensitivity of *Sclerotinia sclerotiorum* from multi-site locations. National Sclerotinia Initiative Meeting poster and oral presentation on January 25, 2020.

Dissertation:

1. Kodati, S., 2019. "Diversity and Pathogenicity of *Rhizoctonia* Spp. from Different Plant Hosts in Nebraska”, (Doctoral dissertation, The University of Nebraska-Lincoln). AAI27667794.
<https://digitalcommons.unl.edu/dissertations/AAI27667794>

**Problems**

Covid-19 response has limited our ability to disseminate results in scientific meetings that were scheduled but cancelled in 2020, including the American Phytopathological Society North Central Division meeting.

**Plans for next year**

Research towards both Objectives 1 and 2 are currently in preparation for publication, with two dissertations currently in preparation. Development of molecular markers will enable deeper insight into the mode of spread and biology of *Rhizoctonia zeae*. By understanding where and why soilborne diseases occur and possible problems with fungicide resistance, we will develop recommendations on how farmers can achieve optimal soil health.

**Dr. Alejandro Rojas and Dr. Terry Spurlock, University of Arkansas**

**Objective 1.** Evaluate the biology and diversity of soil-borne pathogens, associated antagonistic microorganisms, and environmental conditions in the context of the whole-system phytobiome. This objective includes traditional, metagenomics, and spatial/temporal methodologies to understand microbial community dynamics that determine soil-borne disease incidence and severity on economically important crops in the U.S.

**Objective 2.** Evaluate the efficacy of soil-borne disease management strategies (chemical, biorational/biological, cultural) and characterize the associations among microbial community profile, soil physicochemical properties, environmental factors and disease suppression.

**Accomplishments**

The research has been focused on three main areas: development of biocontrol agents for control of soilborne pathogens, soybean seed quality issues and their management, and ecology of soilborne pathogens. The development of biocontrol agents is a project in collaboration with Dr. Clemencia Rojas, and the aim is to characterize the potential biocontrol range of bacteria against rice fungal and oomycete pathogens. These bacteria were previously identified as potential biocontrol agents of rice panicle blight. The project is funded by the Arkansas Rice Promotion Board and we identified *Burkholderia* and *Pseudomonas* as a potential biocontrol agent of fungal pathogens that affect mainly rice, but also caused an impact on other field crops. The genomes of these bacteria have been sequenced and we are focused on the characterization of these bacteria in vitro and as seed treatments. We are using a seed plate assay and germination test to discriminate those strains that have a higher potential for biocontrol.

With respect to seed quality, we have different lines of investigation that converge on a similar goal: determining the factors that drive seed quality and efficient and effective use of fungicides to control foliar pathogens. The first line is establishing a baseline sensitivity of *Cercospora* against several foliar fungicides, which is a proposal with Dr. Travis Faske funded by the Soybean Promotion Board. The goal is to determine the extent of resistance of *Cercospora* in the field to different chemistries now being used by growers, mainly triazoles (FRAC 3 - DMI) and Azoxystrobin (FRAC 11 – QoI). A collection of Cercospora isolates was established from at least 3-5 counties in Arkansas, and those isolates were initially tested for resistance against Tetraconazole (DMI). We are testing the same set of isolates against Azoxystrobin. Part of this project is also contributing to ongoing collaborations with Dr. John Rupe on understanding the role of foliar fungicides on impacting seed infection. Reduced seed quality is associated with Cercospora and Phomopsis. Using plots with different fungicide applications were used and developing pods and mature seeds were collected for the detection of fungal pathogens using DNA based tools, especially those species associated with reduced seed quality.

In the area of the ecology of soilborne pathogens, we are actively working on research of Taproot decline (TRD), *Xylaria* sp. (*X. arbuscula* aggregate) in collaboration with Dr. Spurlock. We aim to characterize how cover crops could improve or cause issues with soilborne diseases. In this area, we have been working on testing cover crops as alternative hosts for this pathogen and we are working on developing molecular diagnostic methods to track this pathogen. A field trial was established at Milo Shult Research & Extension center at Fayetteville. The aim is to study the relation between plant infection and severity of symptoms using isolation and PCR. The trial is arranged in a randomized block design with four cultivars inoculated or non-inoculated and each treatment combination has four replications. Each plot is 4-rows, 36 ft long and the planting rate was 6 seed/ft. Inoculum was grown on millet and corn cob for 4 weeks, colonized material was air dried and placed with seed at 0.2 g/row-ft. The trial was planted in June 19th. Soil samples were collected, and roots were also collected to determine infection. After harvest, half of the treatments will be planted with cereal rye to evaluate the effect of cover crop on pathogen survival.

In collaboration with Dr. Spurlock, fields in southeast Arkansas will be sampled based on plant health, divided in quadrants of ‘healthy’ and ‘symptomatic’. Soil samples will be collected in the different points and used to determine inoculum density of TRD and the associated soil microbial community.

Dr. Spurlock’s laboratory continues to understand the regional distribution of TRD and efficacy of commercially available in-furrow and seed treatment fungicides as well as susceptibility of commonly planted soybean varieties to disease. Because the spatial aggregation of TRD can be relatively small (compared to other ‘patchy’ soilborne diseases) gaining consistency among variety responses in randomized plot evaluations has been difficult. Still, fungicide efficacy has been clearer. The β-tubulin inhibitors, thiophanate-methyl and thiabendazole, have shown efficacy. However, because the causal fungus can infect and colonize for a period extending from planting through the growing season, seed treatment fungicides‘ efficacy has not demonstrated longevity suitable for control in all situations. In-furrow application has been better, but the number of farmers willing to use in-furrow products on soybean are much smaller than those that would like to use a seed treatment package only. This points to the importance of finding moderately resistant and resistant varieties. This project is funded by the Arkansas Soybean Promotion Board.

**Impact**

Student and staff training and involvement

|  |  |  |  |
| --- | --- | --- | --- |
| **Role**  | **Non-Students or faculty**  | **Students with Staffing Roles**  | **Computed Total by Role**  |
| **Undergraduate**  | **Graduate**  | **Post-Doctorate**  |
| Scientist  | 2 | 2 | 2  | 0  | 6  |
| Professional  | 0  | 0  | 0  | 0  | 0  |
| Technical  | 4 | 0  | 0  | 0  | 4 |
| Administrative  | 0  | 0  | 0  | 0  | 0  |
| Other  | 0  | 2 | 0  | 0  | 2  |
| Computed Total  | 6 | 4 | 2 | 0  | 12 |

Student Count by Classification of Instructional Programs (CIP) Code

|  |  |  |  |
| --- | --- | --- | --- |
| Undergraduate  | Graduate  | Post-Doctorate  | CIP Code  |
| 2 | 0  | 0  | 01.00 Agriculture, General.  |
| 0  | 2 | 0  | 01.11 Plant Sciences.  |

Target Audience

Growers, crop consultants, private industry persons, and those interested in further education and outreach.

**Products**Publications

Ortega, L., Walker, K.A., Patrick, C., Wamishe, Y., Rojas, A. and Rojas, C.M., 2020. Harnessing Pseudomonas protegens to Control Bacterial Panicle Blight of Rice. Phytopathology, pp.PHYTO-02.

Hoeksema JD, Averill C, Bhatnagar JM, Brzostek E, Buscardo E, Chen K, Liao H, Nagy L, Policelli N, Ridgeway J, Rojas JA, Vilgalys R. 2020. Ectomycorrhizal plant-fungal co-invasions as natural experiments for connecting plant and fungal traits to their ecosystem consequences. Frontiers in Forests and Global Change 3:84.

Tolbert, A.C., T.N. Spurlock, and R. Hoyle. 2019. Cultivar responses to sudden death syndrome and harvest seed pathogens, 2018. doi:10.1094/PDMR14

Tolbert, A.C., T.N. Spurlock, and R. Hoyle. 2019. Efficacy of various seed treatments on taproot decline in Southeast Arkansas, 2018. doi:10.1094/PDMR14

T.N. Spurlock, Tolbert, A.C., and R. Hoyle. 2019. Taproot decline Trial Summaries 2018-2019. Arkansas Soybean Research Series.

Extension Publications

Chilvers, M., McCoy, A., Noel, Z., Rojas, A., Faske, T., Mueller, D., Smith, D., Tenuta, A., and Wise, K. Detection and Prevalence of Oomycete Seedling Diseases On Soybean. CPN 5003. DOI: doi.org/10.31274/20200918-1

Chilvers, M., McCoy, A., Noel, Z., Rojas, A., Faske, T., Mueller, D., Smith, D., Tenuta, A., and Wise, K. Detection and Prevalence of Oomycete Seedling Diseases on Soybean. CPN 5003. DOI: doi.org/10.31274/20200918-1

Faske, T., Smith, S., Spurlock, T., Wamishe, Y. MP154 Arkansas Plant Disease Control Products Guide. 2019.

Patents

U.S. Provisional Application No. 63/036,202. *Pseudomonas protegens* and products thereof to control bacterial panicle blight of rice. Filed June 8, 2020.

Conferences and presentations

Rupe, J.C., Rojas, J.A., Holland, R., “Impact of cultivar on soybean foliar and seed diseases in Arkansas”. Southern Soybean Disease Workers Meeting 2020.

Delgado L, Winters S, Rojas C. and Rojas A. Bacterial biocontrol agents against fungal and oomycete pathogens of rice and other field crops. South Division APS 2020 Meeting.

Taproot decline of soybean is caused by a novel *Xylaria* sp. that produces phytotoxins associated with foliar symptoms. **T. Garcia-Aroca**1, P. Price1, M. Tomaso-Peterson2, T. Wilkerson2, T. Spurlock3, T. Faske3, B. Bluhm3, K. Conner4, E. Sikora4, R. Guyer5, H. Kelly5, T. Allen2, and V. Doyle1; 1Louisiana State University, 2Mississippi State University, 3University of Arkansas, 4Auburn University, 5University of Tennessee. Southern Division APS 2020 Meeting.

Moscoso, J., Winters S., Rojas A. Potential host range and effects of taproot decline on rotation cover crops. Annual Meeting APS 2020.

Pate S., Kelly H., Guyer R., Lawrence K., Rojas A., Kemerait R., Colyer P., Price P., Wilkerson T., Allen T., Thiessen L., Isakeit T., and Mehl H. Analyzing Populations of Cotton Seedling Disease and Evaluating Seed Treatment Efficacy for 2018 and 2019. Annual Meeting APS 2020.

Rupe J., Rojas A., Mozzoni L., Holland R., and Gbur E. Effect of *Phomopsis longicolla* seed infection on soybean seed composition. Annual Meeting APS 2020.

Sharfadine, S., Delgado L, Winters S, Rojas C. and Rojas A. Bacterial biocontrol agents against fungal and oomycete pathogens of rice and other field crops. Annual Meeting APS 2020.

Gil J., Colle, M., Miles T., and Rojas A. Genome assembly and comparative genomics of plant associated *Rhizoctonia* spp. Annual Meeting APS 2020.

Spurlock, T. N. Ashley/Chicot County Arkansas Soybean Update. 2020 (regional production meeting)

Spurlock, T. N. Tri-County Arkansas Soybean Update. 2020 (regional production meeting)

**Plans for next year**

The overall aim is to mitigate the impact of plant diseases in Arkansas field crops and reduce unnecessary fungicide inputs. Hopefully, combining variety selection based on disease resistance, followed by well-judged use of proper fungicides or biologicals, we could minimize the impact of pathogens. However, pathogens are constantly evolving or being introduced, so there is a continued need to screen crop varieties for resistance and fungicides/biologicals for their activity or simply understand the factors that drive their biology and epidemiology.

For the biocontrol of fungal pathogens in rice, in collaboration with Dr. Rojas, we are working on the development of the biocontrol agents and its characterization for use in greenhouse and eventually testing in the field. We are in the process of characterizing the genomes of the selected agents and understand the potential compounds involved in the biocontrol mechanisms. We are also using microbiome approaches to study soilborne fungi and beneficial fungal communities in field and horticultural crops. In this aspect, we have funding from the Arkansas Department of Agriculture to look at strawberry production systems in Arkansas and cover crop as a mechanism to increase soil health in field crops with funding from the Soybean promotion board. Regarding taproot decline, we will continue characterizing the host range of the pathogen. In collaboration with Dr. Spurlock, we will continue to study the ecology and epidemiology of this pathogen in the field using traditional and DNA based detection methods.

**Dr. Fulya Baysal-Gurel, Tenesse State University
Reporting Period: 2020**

**Major Goals and Objectives**

Overall Goal: Identify effective soil-borne disease management strategies for field nursery production of woody ornamentals to manage soil-borne pathogens that can be easily and readily adopted by field nursery growers.

Objective 1. Evaluate the efficacy of chemical and biorational products for controlling soil-borne diseases with different application methods, intervals and reduced-rate applications in woody ornamentals.

Objective 2. Develop improved soil-borne disease management strategies based on cultural approaches for suppression of Rhizoctonia and (or) Phytophthora spp. and other soil-borne pathogens.

Objective 3. Characterize the associations between microbial community profile and soil-borne disease suppression expressed in different soil-borne disease management strategies.

Objective 4. Engage in outreach and technology transfer with woody ornamental nursery growers.

**Accomplishments:**

**Evaluate the efficacy of chemical and biorational products for controlling soil-borne diseases with different application methods, intervals and reduced-rate applications in ornamentals.**

Soilborne diseases caused by pathogens such as *Phytophthora*, *Rhizoctonia*, *Fusarium*, *Verticillium*, and *Pythium* species are the most important diseases of woody ornamentals. Gingko (*Gingko biloba*) and red maple (*Acer rubrum* L.) ‘October Glory’ plants grown in containers and fields in Tennessee have shown root and crown rot symptoms with dark brown to black lesions in 2017 and 2018. The objective of this research was to isolate and identify pathogens affecting gingko and red maple plants in nurseries of Tennessee and develop fungicide/biofungicide management recommendations for nursery producers. Isolations were made from the infected roots. Several *Phytophthora*-like colonies with spherical zoospores, filamentous to globose oogoni, and whitish mycelium, were isolated on V8-PARPH medium. For confirming identity, total genomic DNA was extracted followed by the sequence analysis of the internal transcribed spacer (ITS) regions, and large subunit (LSU) of the nuclear ribosomal RNA (rRNA) as well as cytochrome c oxidase subunit I (Cox I) and cytochrome c oxidase subunit II (Cox II) of mitochondrial DNA (mtDNA). Based on morphological and molecular analysis, *Phytopythium vexans* was described as a causal agent of crown and root rot from the infected gingko and red maple plants. To complete Koch’s postulates, a pathogenicity test was performed by drenching 100 ml V8 agar medium slurry of *P. vexans* inoculum on 1-year-old potted ginkgo plant root systems as well as red maple ‘October Glory’. Necrotic lesion development was observed in the root system 45 days after inoculation and *P. vexans* was re-isolated from the roots of both gingko and red maple. All control gingko and red maple plants remained disease-free and no pathogen was re-isolated. In addition, the efficacy of fungicides, biofungicides, fertilizer and host-plant defense inducers (traditionally recommended for management of oomycete diseases) for control of Phytopythium crown and root rot was evaluated on gingko and red maple ‘October Glory’ seedlings in greenhouse and field trials. In both greenhouse and field trials, the fungicides such as Empress Intrinsic, Pageant Intrinsic, Segovis and Subdue MAXX were effective, and biofungicide such as Stargus was promising to reduce the disease severity caused by pathogen *P. vexans* on gingko and red maple plants. This comparative study will help nursery producers to make proper management decisions for newly reported Phytopythium crown and root rot disease of gingko and red maple plants.

**Characterize the associations between microbial community profile and soil-borne disease suppression expressed in different soil-borne disease management strategies.**

*Phytophthora nicotianae* is a soilborne plant pathogen which can infect 255 genera in 90 families and is one of the most devastating soilborne pathogens in the south-eastern United States. This pathogen can affect a diverse group of plants, including woody ornamentals, causing wilting and chlorosis of leaves, stem and crown necrosis, while below ground symptoms comprise root necrosis.

The objective of this research experiment was to evaluate the impact of cover crops on the soilborne diseases in field nursery production.

Winter cover crops (triticale or crimson clover) were seeded at the manufacturer recommended rates in September - October (optimal timing for each cover crop species) in 2.4 x 14.6 m field plots with four replicates per treatment at Pleasant cove nursery, Rock Island, TN USA (Warren Co.). Plots were prepared by disk harrow and cover crop seeds broadcast, followed by a cultipacker to incorporate seed into the soil. Plots with no cover crop (bare soil) were used as control. A preemergent herbicide (Sureguard (Valent BioSciences LLC., Libertyville, IL USA)) was applied post-transplant within tree rows to prevent weed/cover crop competition at the base of the trees. Each plot was sampled randomly at four locations each within rows and within middles, mixed in situ with a spade, and placed in a plastic bucket. The soil was stored for one week, at an ambient temperature in a greenhouse before use. The greenhouse bioassays were conducted at the Tennessee State University Otis L. Floyd Nursery Research Center (TSUNRC) in McMinnville, TN, USA. The soil sample from each field treatment -1) cover crop- triticale, 2) cover crop- crimson clover, and 3) bare soil (control); and replication was divided into round plastic containers (top diameter-16 cm, bottom diameter-13.5 cm and height-16 cm) with 3 kg of soil per container. Those soils were then used as either inoculated (with *P. nicotianae*, the rice grain method [6]) or non-inoculated. Isolate FBG201507 of *P. nicotianae* was obtained from the culture collection of Dr. Fulya Baysal-Gurel at the TSUNRC. For each bioassay, ten single-pot replications per treatment were arranged in a randomized complete block design. Rooted cuttings of red maple were transplanted into the containerized field soil, and disease severity was assessed 2 months later. Drip irrigation system was used once per day for 1 min during the experiment. The severity of root rot was assessed using a scale of 0-100% at the end of the experiment. Plant width and height were recorded at the beginning and end of the experiment to be able to calculate the difference. Total plant fresh weight and root weight were recorded at the end of the experiment. The presence of *Phytophthora* was confirmed by plating root samples on PARPH-V8 selective medium. Disease severity, pathogen recovery, total plant weight, root weight and increase in plant height were analyzed with a one-way analysis of variance (ANOVA) using Proc GLM in SAS, and means were separated using Tukey test (α=0.05).

In the greenhouse bioassay without the addition of pathogen inoculum, *Phytophthora* disease severity was significantly lower in soil collected from the cover crop treatments compared to the bare soil treatment. But there were no significant differences between triticale and crimson clover cover crops in disease severity. The pathogen recovery was significantly lower in triticale cover crop treatment compared to the bare soil treatment. There were no significant differences between crimson clover cover crop treatment and the bare soil treatment in pathogen recovery. There were no significant differences between the cover crop treatment and the bare soil treatment in total plant weight and height increase with no pathogen inoculum introduction.

In the greenhouse bioassay with the addition of *P. nicotianae* inoculum, *Phytophthora* root rot severity and pathogen recovery was significantly lower in soil collected from the cover crop treatments compared to the bare soil treatment. Disease severity was lower in triticale cover crop treatment compared to crimson clover cover crop treatment. There were no significant differences between triticale cover crop treatment and crimson clover cover crop treatment in *Phytophthora* pathogen recovery. Total plant weight was significantly greater in the cover crop used soil compared to the bare soil. Increase in maple plant height was significantly greater when the crimson clover cover crop was used compared to the bare soil.

Overall, the cover crops were effective in reducing *Phytophthora* pressure in maple production system, however, longer period of cover cropping might be required to see the prolonged effect of cover crops. Growers can get benefit of incorporating these cover crops into production by reducing the need for synthetic crop protection materials.

**Impact**

1. Evaluate the efficacy of chemical and biorational products for controlling soil-borne diseases with different application methods, intervals and reduced-rate applications in woody ornamentals.

Impact: Provided the woody ornamental nursery industry with improved, efficacious, cost-effective and sustainable recommendations for soil-borne disease management.

2. Develop improved soil-borne disease management strategies based on cultural approaches for suppression of Rhizoctonia and (or) Phytophthora spp. and other soil-borne pathogens.

Impact: Provided the woody ornamental nursery industry with an environmentally friendly disease management strategy that suppresses soil-borne disease population densities comparable to chemical products.

3. Characterize the associations between microbial community profile and soil-borne disease suppression expressed in different soil-borne disease management strategies.

Impact: Provided information regarding how specific management strategies, such as green manure or compost amendments, can suppress plant pathogens and increase the abundance of antagonist populations.

4. Engage in outreach and technology transfer with woody ornamental nursery growers.

Impact: We were able to provide significant outputs to the woody ornamental nursery industry, researchers, extension specialists and agents on the effectiveness of different application methods, intervals and reduced fungicides rates, efficacy of soil-borne disease management strategies such as organic matter amendment, cover crop and solarization for reducing soil-borne pathogen populations. This will lead to outcomes of growers changing their behavior to include more sustainable production practices and reduction in the amount of fungicides applied in field-grown nurseries of woody ornamental.

Personnel and training

The lead researcher is faculty advisor for Milan Panth M.S, Ravi Bika M.S and Krishna Neupane M.S; Sandhya Neupane Ph.D. She is serving as a member at the committee of Victoria Deren and Vivek Ojha since fall 2018, M.S. This project provided also research and Extension experience for Dr. Farhat Avin as post-doctoral researcher.

**Products**

Publications

**Baysal-Gurel, F.,** Liyanapathiranage, P., Panth, M., Avin, F.A., and Simmons, T. 2020. First report of *Phytopythium vexans* causing root and crown rot on flowering cherry in Tennessee. Plant Disease. https://doi.org/10.1094/PDIS-06-20-1166-PDN

Panth, M., **Baysal-Gurel, F.**, Simmons, T., Addesso, K., and Witcher, A. 2020. Impact of winter cover crop usage in soilborne disease suppressiveness in woody ornamental production system. Agronomy 10(7), 995; https://doi.org/10.3390/agronomy10070995

Castroagudín, V. L., Weiland, J. E., **Baysal-Gurel, F.**, Cubeta, M. A., Daughtrey, M., Gauthier, N. W., LaMondia, J., Luster, D. G., Hand, F. P., Shishkoff, N., Williams-Woodward, J., Yang, X., LeBlanc, N., and Crouch, J. A. 2020. One clonal lineage of *Calonectria pseudonaviculata* is primarily responsible for the boxwood blight epidemic in the United States. Phytopathology. https://doi.org/10.1094/PHYTO-04-20-0130-R.

Hudson, O., Walilullah, S. Hand, J. Gazis-Seregina, R., **Baysal-Gurel, F.**, and Ali, E. 2020. Novel method for detection of *Phytophthora capsici* in irrigation water using loop-mediated isothermal amplification. J. Vis. Exp., e61478, doi:10.3791/61478.

Panth, M., Hassler, S., and **Baysal-Gurel, F.** 2020. Methods for management of soilborne diseases in crop production. Agriculture. Agriculture *10(*1), 16; <https://doi.org/10.3390/agriculture10010016>

Tekin, N.T., Kaplanoglu, E., Erdemir, E., **Baysal-Gurel, F.,** Uyanik, C., and Hargrove, S. K. 2020. Modeling and Testing of Magnetic Speed Controlled Submersible Robot for Hydroponic Production. 2019 SoutheastCon. IEEE pp. 1-4. DOI:10.1109/SoutheastCon42311.2019.9020356.

Oral and poster presentations:

Ojha, V., Oliver, J., Addesso, K., **Baysal-Gurel, F.,** Youssef, N., and Simmons, T. 2020. Optimization of *Phytophthora* effective systemic fungicides for ambrosia beetle management. SNA Research Conference Vol; 64 2020. Entomology section.

Neupane, S., Simmons, T., and **Baysal-Gurel, F.** 2020. Management of Phytophthora root and crown rot using biofumigation on field grown boxwood.SNA Research Conference Vol; 64 2020. Pathology and Nematology Section (S. Neupane received the SNA Student Travel Grant).

Panth, M. and **Baysal-Gurel, F.** 2020. Impact of cover crop usage on soilborne disease suppressiveness in field nursery production. SNA Research Conference Vol; 64 2020. Pathology and Nematology Section (2020 Bryson L. James Student Research Competition 3rd place and M. Panth received the SNA Student Travel Grant).

**Baysal-Gurel, F.** Brown, M.S., Oliver, J., Addesso, K. 2019. Evaluation of fungicides and biofungicide to control Phytophthora root rot and ambrosia beetles on flood-stressed flowering dogwoods. Ambrosia beetle working group meeting proceeding. October 15, 2019. Griffin, GA.

 Oliver, J., Addesso, K., **Baysal-Gurel, F.**, Ojha, V., Brown, M., Youssef, N., O’Neal, P., Ranger, C., Reding, M., Bray, A., Schultz, P., Werle, C., Sampson, B., Saroli, J., Mafra-Neto, A. 2019. Ambrosia beetle ecology, pest status and management in tree ornamental systems. Ambrosia beetle working group meeting proceeding. October 15, 2019. Griffin, GA.

Extension publications:

**Baysal-Gurel, F**. 2020. Phytophthora root and crown rot of boxwood. Tennessee Greentimes. Volume 21/No. 1 Spring 2020.

**Baysal-Gurel, F**. 2019. Control of Phytophthora on boxwood. Chase Digest August 2019 Issue Volume 7(8).

Other Products:

**Baysal-Gurel, F.** 2020. Impact of climate change on plant diseases and their management. Climate change workshop. Jackson, TN. January 22, 2020.

**Baysal-Gurel, F.** 2020.Boxwood blight in Tennessee. Boxwood health workshop. George Washington Carver Center, USDA, Beltsville, MD. February 20, 2020.

**Baysal-Gurel, F.** 2020. Pepper diseases and their management. 2020 in-service training. Murfreesboro, TN. February 25, 2020.

**Baysal-Gurel, F.** Plant Diseases. 2020 Warren County Master Gardener Class. McMinnville, TN. March 3, 2020.

**Baysal-Gurel, F.** 2020. Impact of climate change on plant diseases and their management. Climate change workshop. Jackson, TN. March 11, 2020.

**Plans for next year**

1. Evaluate the efficacy of chemical and biorational products for controlling soil-borne diseases with different application methods, intervals and reduced-rate applications in woody ornamentals. We will continue to evaluate chemicals and biorational products for use in soil-borne disease management at greenhouse and field conditions.

2. Develop improved soil-borne disease management strategies based on cultural approaches for suppression of soil-borne pathogens. We will continue to evaluate cultural approaches for use in soil-borne disease management at on-farms and on-station.

3. Characterize the associations between microbial community profile and soil-borne disease suppression expressed in different soil-borne disease management strategies. Microbial community analyses will be performed on field experiment root and soil samples.

**Dr. Shankar Ganapathi Shanmugam and Dr. William Kingery - Mississippi State University**

**Major Goals and Objectives**

**Research objective:** Determining Management-related Factors that Impact the Severity and Incidence of Soybean Taproot Decline (TRD)

Our aim with this project is to understand how management practices in Mississippi soybean production systems (e.g., early-planting, precision seed placement, irrigation systems, residue management) relate to the occurrence of Soybean taproot decline (TRD) in order to verify the effects of altering management practices, and to determine the potential for enhancing the disease suppressive ability of these endemic soils. The aim is to broaden the range of cost-effective practices for TRD suppression that are amenable to incorporation into Mississippi soybean production systems.

**OBJECTIVE 1**: Survey of fields for TRD incidence and severity as well as distribution within agricultural fields that differ in farming system classification.

**OBJECTIVE 2:** Evaluation of the ecology of Xylaria spp. based on pathogen population in soil and microbiome structure and function.

**Accomplishments**

Objective 1:

A total of 20 soybean fields across six Delta counties were evaluated for the field-scale characteristics of symptomology of Soybean Taproot Decline. Evaluations were made at the R5 stage of crop development. A two-tier rating system for characterization was utilized. Field-scale distribution of symptoms were rated with the use of a 7-category system extending from no symptoms visible to symptomology present throughout. The categories included descriptors for symptoms appearing in concentration, i.e., clusters of infected plants, through symptoms distributed throughout the field. Fields were divided into four quarters with an overall rating given as the average of the four. Infection intensity percentage was measured by counting the number of infected plants per 100 plants in a linear section of row. This was done at two randomly chosen positions within each of the field quarters mentioned above.158 survey transects across twenty soybean fields were conducted to rate TRD severity and distribution. Plant growth stage and residue ratings were also documented at each survey transect.

Objective 2:

Two fields in the Mississippi Delta were sampled at locations within the fields that represented three ranges of Soybean Taproot Decline intensity (0%, 1-50%, and > 50% of plants infected along one linear meter of row, i.e., infection intensity grouping of low, medium an high). From each site, soil core plugs were collected along the one meter of row, to a 10-cm depth and composited. Two additional Delta fields, each with nine geo- referenced location per field were measured for percentage infection along one linear meter of row and soil sampled as described above at three dates (June, July and August). At the final sampling, all plants at each of the nine locations were collected and returned to the lab to make a final determination of Soybean Taproot Decline infection and to determine grain yield.

**Preliminary Results:**

Amplicons targeting bacterial 16S and fungal 18S rRNA genes (ITS2) were sequenced using Illumina MiSeq sequencing platform. The distribution of the 500 most abundant OTUs (operational taxonomic units) in the soil samples from the two locations indicated two primary patterns of soil bacterial community structure. The location has a significant effect on the bacterial community structure. The important part of the structural variability was related to differences associated with the sampling dates. There was no significant difference in the pattern of bacterial community structure with mid and late sampling dates (p=0.618 and 0.529, respectively) indicating that the disease severity had a significant effect on the soil bacterial community composition.

**Impacts**

Target Audience

Outcomes and information from this project will help soybean producers in both understanding and managing taproot decline disease. The increasing distribution and occurrence of the disease throughout the state suggests that project benefits may extend to all soybean acres in Mississippi. However, the monetary impact is difficult to project as the economics of TRD are somewhat unknown given the highly variable in-field distribution and infection intensity. Indirect benefits are associated with an increased understanding of taproot decline ecology, particularly in how the disease interacts and responds to crop and residue management practices. The survey and classification of farming systems in relation to TRD occurrence and severity is anticipated to provide direct benefits to soybean growers by identifying practical management decisions that may reduce the impact of TRD.

Training

A Post-Doctoral associate (Nisarga Narayana), an Undergraduate Research Scholar (Rachel Hill) and a Student Worker (Abigail Allison) are being training in soil microbial ecology lab (Mississippi State University).

Funded Projects

Shanmugam, S.G (Principal), W.L. Kingery (Co-Principal), Tom Allen, Teresa Wilkerson and Mark Shankle .“Determining Management-related Factors that Impact the Incidence and Severity of Soybean Taproot Decline (TRD)” . Mississippi Soybean Promotion Board- $67,139 (April 1, 2020 - March 21, 2021).

**Dr. M. Soledad Benitez Ponce – The Ohio State University**

**Major Goals and Objectives**

*Objective 1. Evaluate the biology and diversity of soil-borne pathogens, associated antagonistic microorganisms, and environmental conditions in the context of the whole-system phytobiome. This objective includes traditional, metagenomics, and spatial/temporal methodologies to understand microbial community dynamics that determine soil-borne disease incidence and severity on economically important crops in the U.S.*

Sub-objectives (Benitez):

- Characterize potential antagonistic and/or synergistic populations contributing to SCN incidence in Ohio

*Objective 2. Evaluate the efficacy of soil-borne disease management strategies (chemical, biorational/biological, cultural) and characterize the associations among microbial community profile, soil physicochemical properties, environmental factors and disease suppression.*

*Sub-objectives (Benitez):*

- Determine consistent responses of soil and plant associated microbiomes to cropping practices across different locations and years

- Evaluate the efficacy of microbial-based products in management of soil borne diseases

- Determine conditions that contribute to consistent and successful application of biological control products, and interactions with the soil and plant microbiome

**Accomplishments:**

Objective 1: In 2019-2020, we are evaluating the communities of fungi associated to soils with known incidence of soybean cyst nematodes, across Ohio. This survey will allow us to ask questions about a) potential fungi to be used in SCN disease management, as well as b) fungi and SCN interactions that might be relevant for increased disease incidence. This work is funded through the Ohio Soybean Council, and is also possible due to access to samples through with OSU Plant’s Disease Clinic and OSU’s Soybean Pathology lab (Dr. Anne Dorrance). We are currently processing a total of 76 samples representing production farms across 15 counties in Ohio. From these, 43% of the samples did not report presence of SCN; 42% of the samples have less than 1000 eggs per 100 cc of soil; and 14% of samples have greater than 1000 eggs/cc of soil. In addition, all samples were collected in 2019 and have a history of soybean production, with 50% reporting only soybean in their rotation.

In addition, we are also studying the communities of fungi associated to the nematode cyst. Following a baiting technique, to enrich for SCN-associated fungi in soils, we were able to determine that the most prevalent fungal species isolated from nematode cyst is *Fusarium solani*; future work will focus on characterizing the fungi associated to SCN cysts, and determine cyst-associated fungal community dynamics in cyst age.

As part of this project we are training two graduate students in the Department of Plant Pathology.

Objective 2: The successful application of biological control products and other microbial inoculants is often dependent on environmental variables and management practices, as complex interactions often occur in the plant-soil interface. The Benitez Lab is focusing on three aspects of management practices that can contribute to crop productivity and plant health a) evidence that can increase implementation of diversified cropping practices; b) evaluation of beneficial fungi in management of abiotic and biotic stress; and c) determining conditions or interactions that contribute to microbial inoculant efficacy in field production.

*Chicken-grazing and diversified vegetable production.* In 2020, a greenhouse experiment was completed to evaluate the impact of different concentrations of soils exposed to chicken manure in tomato perfmonance and associated endophytes. The greater percent of soils with history of chicken grazing resulted in stunting of tomato seedlings. In addition, a collection of bacterial endophytes was generated, and is currently being analyzed to determine if endophyte community structure is influenced by history of chicken grazing. This work was part of a MS student in the Department of Plant Pathology, OSU.

*Crop diversification through cover crops and rotations.* During 2020 we continued with our research in corn-soybean rotational systems, specifically focused on the corn-soybean-wheat rotational system, and the use of a rye cover crop. This project is performed with support of the Ohio Soybean Council, and OSU’ SEEDS grants, and in collaboration with Dr. Laura Lindsey, OSU. A MS student recently completed her program, studying the impact of corn-soybean-wheat rotation in soybean productivity, soil health and bacterial communities. Her results indicate the beneficial effect of the 3-year rotation system on years and sites with conditions less ideal for soybean production. Futhermore, she identified bacterial taxa, of the genus Pseudomonas, to be consistently enriched in the three-year rotation system. As part of this experiment, in fall 2020 we have incorporated additional treatments into the rotation experiment, to evaluate the interactions the three-year rotation and the use of cover-crops. As the project moves forward, we will use the information from the 2020 growing season as a baseline to determine if additive effects of cover crop treatment in crop health and productivity and microbial communities are observed. In a parallel experiment, we are studying the impact of timing of rye cover crop termination in subsequent soybean, across multiple years. However, due to university restrictions, our 2020 cover crop termination trial was discontinued.

*Arbuscular mychorrizal fungi applications in soybean production.* We are also continuing research on beneficial fungi, with emphasis on the arbuscular mychorrizal fungi (AM). This research is funded through OSU Industry Partnership SEEDS grants and is focused on evaluating the impact of AM inoculants on soybean production and impact of SCN. For the third year, the impact of AM inoculation in soybean productivity in fields with high incidence of soybean cyst nematodes was evaluated. Even though, no significant responses to commercial AM seed treatments on soybean performance in field trials has been observed, across the three-years there is a consistent trend indicative of a potential benefit when both AM and conventional seed treatment (nematicide and fungicide mix) is used. We will monitor not only for AM colonization in soybean roots, but also determine specific AM species colonizing, as a proxy to evaluate colonization success of inocula.

**Impacts**

Training

Three MSc students have been (or are being) trained as part of this experiments; as well as two undergraduate summer interns, and two research associates who contribute to this work.

Dissemination

 Results have been presented through multiple venues, with special focus on virtual meetings and presentation during 2020.

Target Audience

The target audience for this work is the scientific community in general. In addition, some of the material and examples of methods applied in our research are presented to undergraduate and graduate students in the two courses I teach/co-teach: Phytobacteriology and Current topics in Plant Microbiome and its application in agriculture. Finally, through our collaborators, in particular Dr. Laura Lindsey, and funding source, the Ohio Soybean Council, our research results regarding rotational systems and cover crop in particular, with corn and soybean producers.

**Products**

Publications: In preparation.

Oral and poster presentations:

1. Vazquez-Catoni AM and Benitez-Ponce MS. The impact of chicken-grazed soils on the tomato plant microbiome. Poster presentation - Plant Health 2020.
2. Willman M, Taylor L, Levy D, Isack Y, Benitez MS. Transcriptional responses to interactions among arbuscular mycorrhizal fungi, soybean cyst nematode, and soybean. Poster presentation Plant Health 2020.
3. Vazquez-Catoni AM and Benitez-Ponce MS. The impact of chicken grazed soils on the tomato plant microbiome. Abstract accepted for presentation at ASM Microbe 2020 – Conference Canceled.
4. Huo D., Lindsey LE,Benitez-Ponce MS.Three-year soybean-wheat-corn rotation benefits on soybean production, soil health and soil bacterial community are site and year dependent. Oral - presentation Plant Health 2020.

Dissertations:

Ana M. Vasquez-Catoni. 2020. The Ohio State University, Plant Pathology. MS Thesis: impact of chicken-grazed soils on the bacterial microbiome of tomato plants.

Daowen Huo. 2020. The Ohio State University, Plant Pathology. MS Thesis: Three-year soybean-wheat-corn rotation benefits on soybean production, soil health and soil bacterial community are site and year dependent.