W_TEMP_5147: Managing Plant Microbe Interactions in Soil to Promote Sustainable Agriculture

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Statement of Issues and Justification

A. The Problem

Soilborne diseases continue to be a serious problem in US agriculture. They are caused primarily by fungi and nematodes, and attack the root systems, reducing the ability of plants to take up water and nutrients. They are difficult to diagnose, because above- ground symptoms may not be distinctive, and could be mistaken for nutrient deficiencies or drought. Unlike foliar diseases, they are difficult to manage with chemicals. They form survival structures in the soil that are resistant to chemicals, and it is near impossible to treat all the soil, except with fumigation, which can result in environmental degradation. Unlike with biotrophic diseases like rusts, there is little genetic resistance to soilborne diseases in available and industry accepted crop cultivars. Although some exceptions exist (i.e., Fusarium wilt diseases), in most cases host resistance to soilborne diseases is multigenic and difficult to breed for.

Economic Costs Due to Soilborne Plant Pathogens

According to the Food and Agriculture Organization (FAO) of the United Nations, plant diseases and pests reduce agricultural production by 20-40%, which represents a global cost of approximately 220 billion USD for diseases alone (www.fao.org). As such, 2020 was declared the International Year of Plant Health. For root diseases of mature crops, there are few effective and economical post-planting strategies for control. About 90% of the 2000 major diseases of the principal crops in the U.S. are caused by soilborne plant pathogens, most of which are fungi or fungus-like organisms (Panth et al., 2020). Many pathogens have a soil- or debris-borne over seasoning stage. In many cases, resistance to soilborne diseases is quantitative and specific pathogen race structures and population diversity parameters remain understudied.

Table 1. Crop production (million metric tons) for the top 10 crops (excluding vegetables) in the U.S. in 2021 (NASS, 2022).

Crop	Production	Rank	Crop	Production	Rank
Corn	383.9	1	Sorghum	13.7	6
Soybeans	120.7	2	Rice	9.7	7
Wheat	44.8	3	Cotton	3.8	8
Sugarbeets	33.3	4	Peanuts	2.9	9
Sugarcane	29.8	5	Barley	2.6	10

As examples, certain commodities quantify and report yield loss data to all categories of diseases including those caused by soilborne pathogens. Corn data from 2016 to 2019 showed an average yearly loss of 9.6 million metric tons (Mmt) (2.4% of the 2021 production value; Table 1) to soilborne diseases including Fusarium stalk rot (5.1 Mmt yr^{-1}), nematodes (1.7 Mmt yr^{-1}), Gibberella stalk rot (1.3 Mmt yr^{-1}), seedling blights (0.9 Mmt yr^{-1}), and root rots (0.7 Mmt yr^{-1}) (Mueller et al., 2020). Likewise, soybean data from 2015 to 2019 showed an average yearly loss of 3.9 Mmt (3.2% of the 2021 production value; Table 1) to soilborne diseases including seedling diseases (1.29 Mmt yr^{-1}), white mold (1.10 Mmt yr^{-1}), sudden death syndrome (1.01 Mmt yr^{-1}), Phytophthora root rot (0.66 Mmt yr^{-1}), and charcoal rot (0.04 Mmt yr^{-1}) throughout the northern and southern soybean-producing states. Detailed studies on the wheat crops in the Pacific Northwest had documented loss due to *Pythium*, *Fusarium*, and *Rhizoctonia*. For 2021, the cotton disease committee (Cotton, Inc.) estimated losses in 480 lb bales to Fusarium wilt (*Fusarium oxysporum* f.sp. *vasinfectum*), Verticillium wilt (*Verticillium dahliae*), cotton root rot (*Phymatotrichopsis omnivora*), and seedling diseases (several fungi) to be 64800 bales (0.35%), 67600 bales (0.37%), 26500 bales (0.14%), and 146100 bales (0.80%), respectively.

New or re-emerging soilborne diseases are a constant threat to crop production in the United States. Such issues arise through pathogen introductions, changes in host germplasm, and altered control measures. For example, wheat blast is a new disease in South America (Brazil) caused by a strain of the pathogen *Magnaporthe oryzae.* It has recently been detected in Bangladesh, but is not yet present in the United States. *Macrophomina* and Fusarium wilt in strawberries have become new problems, because of the loss of methyl bromide. In corn, tar spot (*Phyllachora maydis*) has emerged and caused disease losses in the U.S. since 2018 (Kleczewski et al., 2020). In soybean, *Xylaria necrophora* has emerged as a root rot and taproot decline pathogen in the southern U.S. (Garcia-Aroca et al., 2021) since ~2017. Several of the top 15 restricted, invasive quarantine pathogens listed by APHIS are soilborne and could represent biosecurity risks.

New invasive species have been discovered in North America in the last fifteen years, including*Phytophthora ramorum*, cause of sudden oak death, and *Phytophthora tenticulata*, have decimated native ecosystems in California. In natural ecosystems, once they become established, these pathogens cannot be easily managed. Laurel wilt, caused by *Raffaelea lauricola* and vectored by exotic ambrosia beetles, threatens the native laurels of the East Coast and the avocado industry in Florida and California. Boxwood blight, caused by *Cylindrocladium buxicola*, discovered in the US in 2011, has become endemic in 10 states.

Finally, the changing climate represents a challenge and an opportunity to address soilborne pathogen problems. Changing climate will result in more plant stress, drought conditions, salinity or in some cases a wetter climate, which will predispose plants to more disease. Models based on global field surveys have shown that increasing soil temperature may increase the abundance of soilborne fungal pathogens (Delgado- Baquerizo et al. 2020). Further, Chalconer et al. (2021) project that as climate changes, higher latitudes will see greater yields, but pathogen turnover is projected to increase in some of the most productive swaths of global agricultural land. This includes the central Great Plains of the United States. Charcoal rot, a drought and high-temperature disease, was in the top 10 soybean diseases for the southern United States every year from 2015-2019, but had appeared in the top 10 diseases of the northern U.S. more often (3 of 5 yrs) during this period compared to previous disease loss reports (≤ 2 of 5 yrs).

Cost to the environment

The cost of soilborne plant pathogens to society and the environment far exceeds the direct costs to growers and consumers. The use of chemical pesticides to control soilborne pathogens has caused significant changes in air and water quality, altered natural ecosystems resulting in direct and indirect effects on wildlife, and caused human health problems. For example, methyl bromide, a fumigant used to control soilborne diseases, has become notorious in recent years for contributing to the depletion of the ozone layer. The planned ban on production and importation of this product has been repeatedly delayed by a lack of cost-effective alternatives, and there remains an intensive search for replacement control methods. This fumigant was to be totally banned by 2005, but there are still a few critical use exemptions for the U.S. Larger buffers and restriction zones are needed for many pesticides. Soil fumigants are major contributors to volatile organic compounds affecting air quality, especially in the Central and Imperial Valley of California. Development of fungicide resistance continues to be a problem with the newer generation of reduced-risk fungicides with specific modes of action, such as the strobilurins. Additionally, plants evolved in the presence of microorganisms and are dependent on them in order to carry out many activities necessary for growth and reproduction. Thus, long-term chemical applications may permanently alter the microbial community structure sufficiently such that sustainable agriculture may be impossible.

B. The Solution.

The future of sustainable agriculture in the U.S. will increasingly rely on the integration of biotechnology with traditional agricultural practices. Although genetic engineering promises enhanced yields and disease resistance, it is also important to recognize that plants exist in intimate associations with microorganisms, some of which cause plant disease while others protect against disease. Identifying, understanding and utilizing microorganisms or microbial products to control plant disease and enhance crop production are becoming more central parts of sustainable agriculture. Biological control or biologically-based pest management (BBPM) has the potential to control crop diseases while causing no or minimal detrimental environmental impact. For this proposal, we define biological control as the manipulation of microbial populations through cultural, physical or biological means including plant mechanisms. Some of the benefits of utilizing microorganisms include:

- Reduced dependence on chemical pesticides, which is important because of expanding demand for organic produce and increasing costs of such petroleum-based inputs
- Lack of development of pathogen resistance to biological control organisms
- Lower regulatory costs of registration
- Faster reentry times after application
- More selective action against pathogens and not against beneficial organisms
- Biodegradability of microbial pesticides and their by-products
- Reduced danger to humans or animals
- Improvement of soil quality and health
- Increased food safety
- Management of diseases in natural ecosystems
- Improve plant productivity via controlling abiotic stress
- Adaption to climate change, as pathogen distributions shift
- Increased N use efficiency and reduced N and P contamination of waterways and oceans

In the last five years, there has also been an increased awareness of soil health and the importance of microbes in providing benefits to plant health and productivity. Over the last 10 years, almost 30,000 papers have been published on the topic of soil health. There is an established non-profit (Soil Health Institute (https://soilhealthinstitute.org). States such as Washington have funded a soil health institute via legislative appropriations (Washington Soil Health Initiative https://agr.wa.gov/departments/land-and-water/natural-resources/soil-health). While, NRCS has developed on-line resources inform growers about soil health, there are no validated protocols for assessing soil health for all cropping systems and growers are clamoring for tests to help them determine their soil's health. Additionally, THERE IS A LACK OF UNDERSTANDING ABOUT HOW SPECIFIC MICROBIAL GROUPS DRIVE SOIL HEALTH. Research proposed BY THIS PROJECT addresses these knowledge gaps and is ADVANCING THE FIELD.

Objectives

To address this problem, this project will propose four objectives

1. **Objective 1**. Biocontrol via one or a small number of microbes and/or their products
2. **Objective 2**. Biocontrol via microbial communities and soil health
3. **Objective 3**. Creating and testing disease management and/or soil health strategies
4. **Objective 4**. Extension and outreach

How this project fits the goals of USDA and REE

This project fits REE Action Goal Framework Goal 1: Sustainable Intensification of Agricultural Production. In 2017, USDA also issued a set of goals, and this overlaps with USDA GOALS 2 & 3. GOAL 2: Maximize the Ability of American Agricultural Producers To Prosper by Feeding and Clothing the World and GOAL 3. Promote American Agricultural Products and Exports. This entire project is designed to help farmers and growers manage soilborne pathogens in an environmentally sustainable way, thus directly impact the large agricultural industry of the United States.

C. Addressing the Needs of the Stakeholders

1. **The Biopesticide Industry.**

Biopesticides are the fastest-growing crop protection market sector exhibiting substantial growth over the past three decades as the market has grown into a multibillion-dollar business. Demand for biopesticides has continued to expand dramatically in the last five to ten years. In the 5 years since our previous proposal, the global market for biopesticides has doubled from $1.5 billion to $3 billion US (Biological Pesticide Products Industry Alliance, 2017) and is expected to triple by 2027 (Batista and Singh, 2021; Sessitsch et al. 2018). Recent policy changes, such as the new Green Deal of the European Union, have expanded the biological product market. The Biological Pesticide Products Industry Alliance, established in 2001, had 31 member companies in 2006, 65 members in 2012 and now has over 120 members. The International Biocontrol Manufacturer's Association had 130 companies marketing microbial biocontrol agents in 2012. But now there are approximately 2530 biopesticide manufacturers (not including China and India) with about 98 of those in the Americas and 91 in Europe. This segment of the industry is expected to grow between 15% and 20% annually (http://www.ibma-global.org). This growth has been driven by expanding organic markets as well as increased public sensitivity to the risks and hazards of chemical pesticides. From 2008 to 2012, 15 microbial active ingredients have been registered by EPA. From 2012 to 2017, six *Bacillus* spp., two *Trichoderma* spp., one *Streptomyces* sp., one *Pseudomonas* sp. and one *Muscodor* sp. have entered the EPA regulatory process (EPA Biopesticide Workplans 2012-2017). In the last 5 years, there has been a concerted effort by larger companies, such as Syngenta, BASF, FMC, and Bayer, to develop inhouse expertise by partnering or acquiring smaller companies, to increase their investment in this field. For example, Bayer CropScience bought AgraQuest, acquiring Serenade and other *Bacillus* products.

Additionally, the recent development of next-generation sequencing technologies and capacity to analyze metagenomic data has led to a greater understanding of how soil microbial communities function and influence plant tolerance to biotic and abiotic stressors. This technology is presently being utilized by our members to gain insights into plant-microbe interactions and identify implications for biocontrol. In fact, there has been a significant increase in research publications on the use of soil inoculants and consortia of microbes for disease control (Canfora et al. 2021).

2. **The Microbiome Industry.**

This is an industry that did not exist 15 years ago. The total agricultural microbial industry is estimated to increase by 17% yearly, with a value of $12 billion by 2026 (Statistics Market Research Consulting. Agricultural Microbials - Global Market Outlook (2017–2026). https://www.researchandmarkets.com/research/vdth4t/global?w=4 (2020).

Much of this growth is in “microbiome companies” such as Indigo, which has developed seed endophytes, AgBiome, Robigo, Inviao, and BioConsortia. Monsanto (now part of Bayer) invested a significant amount of money in microbiome research in the last 10 years. In addition, there are dozens of startup companies getting into the field. This involves a major paradigm shift. Instead of just relying on developing biological control agents (BCAs) to apply to agriculture, the emphasis is to manage the rhizosphere and soil microbiome by management of the agricultural system, to promote disease suppression, nutrient uptake, and tolerance to abiotic stress. Nevertheless, many of these companies are using microbiome research to discover and market potential new inoculants. They are also marketing microbiome tests for farmers that would give them a diagnostic assessment of the soil health of fields, much in the same way companies have exploited this for the human gut microbiome. There is a large “probiotics” market for agriculture, but many of the claims made by developers of probiotics and soil health products and services are not based on sound research. Researchers in this project can provide sound, science-based evidence to help support adoption of products, services, and management practices that will be most valuable to growers.

3. **The Organic Industry**

As is readily apparent from reading the popular press, consumers are demanding plentiful low cost but safe food while simultaneously requiring reduced use of chemical pesticides. This has been evident by the rapid growth of the organic food industry. In 2016, there were 5.1 million acres in organic production (USDA Organic Survey), almost double the acreage from 2008. In 2019, this number increased to 5,495,270 acres. with 16,585 farms. The total farm gate value of organic products in 2016 was $7.5 billion, compared to 3.5 billion in 2011 (NASS). By 2019, sales increased to 9.9 billion dollars. Organic food is now available from large retailers such as Walmart, Whole Foods, Kroger, and others.

Organically-grown crops require non-synthetic methods for management of diseases, and organic growers are seeking scientifically-based disease management methods. A 2015 survey by the Organic Farming Research Foundation (OFRF) identified disease management and soil health as one of the top five research priorities. In 2020, The Organic Farming Research Foundation (OFRF) and Organic Seed Alliance (OSA) released an update survey in 2020 for California growers. Some of the production concerns that were identified include controlling disease pressure, minimizing adverse impacts of tillage on soil health, and adapting to climate change. One of the top research priorities is maintaining yield and soil health. Members of this project are doing this research. Many products that members of W-4147 have researched are certified as organic with the Organic Materials Review Institute (OMRI). During the last few years, more and more pesticides that control soilborne diseases have been taken off the market or regulated, including methyl bromide. Soilborne pathogens are well adapted to soil conditions, and once established are very difficult to eliminate. A classic example of a disease shift with the loss of methyl bromide has been the increased incidence of Fusarium wilt and charcoal rot of strawberries in California, which were not major problems 10 years ago. Even if chemical products are available, they are often too expensive to be economically practical and for many pathogens, chemical remedies have yet to be identified. Other approaches with great potential include the development of transgenic crops engineered with resistance genes to several pathogens. However, there is widespread public reluctance to accept these crops as evidenced by protests both here and in Europe. These concerns, combined with the natural ability of pathogens to overcome introduced resistance genes, has frustrated efforts to maximize this approach.

Why a Multi-State, Multi-Disciplinary Approach? No single research institution has sufficient resources and diversity of expertise to solve the diverse disease problems that might be addressed through the use of biological controls. Many of these pathogens occur in multiple states and a coordinated research effort could provide more cost-effective outcomes. The more than 40 researchers in this multistate project also collaborate with other researchers in the U.S. and around the world, providing further impact and cross-fertilization of knowledge, as well as conducting the needed outreach activities for implementation of biocontrol options. This group is at the forefront of research in soil health and phytobiome research and in understanding the complex interactions among the soil microflora, which provide benefits to the plant. With the reduction in resources and human capital in agricultural research, it is more important than ever to gain synergy by leveraging resources with a multi-state group.

Goals and potential impact of the Project

The ultimate goals of this collaborative work of W-5147 are to:

- Provide society with a safe, low-cost food supply
- Reduce the environmental impact of soilborne disease control on ornamental, bioenergy, fiber and food crop production
- Protect natural and agroecosystems from invasive species
- Development of new industries and products for biologically based disease control

Related, Current and Previous Work

THE MICROBIOME

The newest area of biological control research is understanding and utilizing the communities of microorganisms found in plant-associated habitats such as soil, rhizosphere, and the endosphere. The microbial communities that inhabit these niches are called microbiomes. In this area of biological control research, instead of examining how one or a small number of microbes influences plant disease, large numbers of microorganisms, or an entire microbial community, are being examined to determine how they are affecting plant health and disease. Members of the W4147 have been leaders in this field. In the following, we describe several examples of this work.

Outreach to Stakeholders

One essential component of working in a new area of an applied science is to inform the stakeholders about the field and keep them updated on the newest advances and capabilities. It is also important to get stakeholder input concerning how fundamental results might be translated into practical solutions, and to form collaborations with the stakeholders to test putative solutions in the field. Members of our group regularly interact with growers to foster these relationships, and they have also given numerous presentations on such topics. These presentations have covered fundamental topics such as what is a microbiome (Marks et al. 2020a). They have also covered topics such as how scientists are elucidating the relationships between microbiomes and plant health as well as between microbiomes and plant disease (Marks et al. 2020b, Evin et al. 2021).

Microbiome Methods

Another essential component of working in new field is to create new methods. Since new fields are asking new questions, new methods are needed to answer them. Members of our group have been leaders in this area. The following are a few examples in chronological order.

Early on in the development of the field, one of our members published a new method that provided the simplest and fastest way to isolate DNA from soil. This method initiated a transformation in how DNA was isolated from environmental samples, contributing to the explosion of studies characterizing the vast and previously undescribed diversity of microbes inhabiting our planet. This soil DNA isolation method uses bead-beating-based cell lysis and column-based DNA purification (Borneman et al. 1996). Building on the commercial success of the kits that were based on these methods, a few of the researchers from BIO 101/MP Biomedicals started their own company, MoBio Laboratories, which became a leader in DNA and RNA isolation.

Another early advance was the development of a new method that provided a simple and rapid way to compare bacterial communities (Borneman and Triplett, 1997). Such analyses give researchers the ability to identify bacteria that are differentially abundant in two or more samples, which can be the key first step in identifying functionally important microbes (e.g. bacteria that cause or inhibit disease). Compared to the most commonly used method at the time (DGGE) (Muyzer et al. 1993), this new method (i) provided better taxonomic resolution, (ii) did not require specialized equipment, and (iii) it is still used today.

Another early advance was the development of a new method that provided a simple, fast, and culture-independent approach for identifying individual microbes that respond to specific chemical stimuli in environmental samples. This capability can be of considerable value in efforts to determine which microbes play key functional roles in complex ecosystems. By adapting a commonly used experimental strategy in mammalian biology, a culture-independent method was created that allowed the identification of microbes (bacteria, fungi, others) that actively divide in response to the addition of specific chemicals to an environmental sample (Borneman 1999). Here, a nucleotide analog (BrdU) and a specific chemical are added to an environmental sample such as soil and allowed to incubate. DNA is then isolated from that sample, the DNA that has incorporated the BrdU is captured using an antibody, and the resulting DNA is analyzed for its microbial community composition. An example experiment comparing uncaptured and antibody-captured DNA with a control soil and one amended with a solution of glucose and potassium phosphate, there were considerable differences between the treatment groups with the captured DNA but no differences with the uncaptured DNA.

A more recent advance was the development of a new method for bacterial community analyses that provided increased taxonomic resolution. This feature is crucial for differentiating similar bacteria, and for determining if samples or subjects harbor functionally important bacteria (e.g. bacteria that cause or inhibit diseases such as IBD as described below). This was the first, Illumina-based, amplicon sequencing method that frequently enabled species- and sometimes strain-level resolution of bacteria. This increased resolution was obtained by analyzing the rRNA ITS region (Ruegger et al. 2014).

In very recent work, one of our members created new methods to analyze modern omics datasets. For example, they created an inexpensive and quantitative reduced-representation sequencing (qRRS) strategy, omeSeq, for describing microbial communities (Olukolu 2021). They have also developed new bioinformatic software that automates metagenomic analyses (Kuster et al. 2021). These bioinformatic tools implement novel algorithms that provide strain-level and quantitative profiling of microbiomes, as well as the ability to handle new features derived from the qRRS protocol that delivers high-fidelity base calls and yields that exceed Illumina's maximum yield by as much as 50%. This new software is also backward compatibility with amplicon sequencing and shotgun sequencing of metagenomes and meta-transcriptomes. These tools are being used to understand how multi-way plant-pathogen-microbe interactions modulate plant diseases and defense response pathways.

Other members of our group are involved in creating standards for reporting omics metadata (Dundore-Arias et al. 2020). This critical work will allow investigators to compare studies across the globe, thereby amplifying the effects of any individual study.

Microbiome and Plant Health and Disease

The microbiome topics most relevant to this project involve determining the relationships between microbiomes and plant health as well as between microbiomes and plant disease.

An example of this involves replant disease. Replant disease often occurs when certain crops are ‘‘replanted’’ in a soil that had previously supported the same or similar plant species. This disease typically leads to reductions in plant growth, crop yields, and production duration, and its etiology remains ill-defined. A study of replant disease from one of our members examined the relationships among the microbiome, replant disease severity, plant genotype, and seed meal applications (Wang and Mazzola, 2019). Here, the authors determined that (i) the rate of seed meal application was linked to replant disease severity, (ii) such applications were more effective with a specific plant genotype, and (iii) these beneficial effects were linked to microorganisms that make more anti-bacterial and anti-fungal compounds. This work provides a blueprint for other such studies, where several components of the phytobiome are examined to obtain a much more complete understanding of how all of these complex variables enable effective disease control.

Another example involves survivor trees. The Survivor Tree Phenotype is a natural disease tolerance phenomenon – where a small number of healthy citrus trees are found in orchards that are severely affected by Huanglongbing. To characterize this potentially valuable phenomenon, members of our group initiated a multiyear study to identify the microbes in these trees. The same trees in orchards from several geographical locations in Florida were annually assessed for their HLB disease severity ratings and their composition of microbes. This work identified trees with little to no decline in their health, providing evidence that the Survivor Tree Phenotype is a reproducible phenomenon. A small number of microbes were positively associated with this phenotype, suggesting that host-microbe interactions may be causing this phenotype (Ginnan et al. 2020). Towards the goal of creating an Huanglongbing management strategy, future work will involve testing these microbes for their ability to control disease.

Another example is the use of anaerobic soil disinfestation. Management of soilborne diseases in strawberries and other diseases used to be accomplished via methyl bromide fumigation. Because of environmental concerns, methyl bromide and other fumigants have been or are being phased out, creating a need for alternative disease management strategies. One of our members has performed a series of studies examining the use of anaerobic soil disinfestation as an alternative strategy to manage these diseases. In one of these studies, the efficacy of such treatments was linked to changes in the soil microbiomes (Mazzola et al. 2018). This work provides another important example of how the microbiome is important for disease management of soilborne plant pathogens. Additional research on this topic determined how soil metabolites were influenced by anaerobic soil disinfestation (Hewavitharana et al. 2019), providing key clues about the putative mechanisms driving this beneficial effect, and identifying molecules that may be used in future disease management strategies.

Cultural and Chemical Inputs and the Microbiome

Members of our group have also published several landmark papers on how cultural and chemical inputs affect the wheat microbiome. We were the first to document, with next-generation sequencing, that glyphosate applied at rates used by farmers, has minimal impact on both bacterial and fungal communities in the rhizosphere and bulk soil. (Schlatter et al. 2017, 2018). Biosolids from treated sewage are applied to fields in eastern Washington as a source of N. We showed that biosolids can shift both bacterial and fungal communities, even four years after the application (Schlatter et al. 2017, 2019). We also documented the bacterial and fungal microbiome of dust emitted from these biosolid treated fields (Schlatter et al. 2018).

Many of the soils in eastern Washington are becoming acidified because of use of ammonia fertilizers over 60 years, and growers are adding lime to increase soil pH. In a series of papers, we examined how liming affects bacterial and fungal microbiomes (Schroeder et al. 2018, Yin et al. 2021). In a series of papers comparing long-term no-till sites to conventionally-tilled sites, we found that tillage had a stronger effect on fungal communities than bacterial communities (Yin et al. 2017, Poudyal-Sharma et al. 2017). Earthworms also play a major role in no-till systems, by moving carbon into deep soil profiles. We identified specific bacterial communities present in the drilosphere, lining the burrows of the earthworms, and also unique communities present in the gut and casts (Schlatter et al. 2019, a and b). Soils in the hilly Palouse region of Eastern Washington are very deep wind deposited loess soils, with topsoil up to 10 ft deep. In our Mediterranean climate with little summer precipitation, wheat plants rely on moisture stored in this deep profile. We showed very distinct communities at each depth in a no-till system (Schlatter et al. 2020). The top layer was enriched with copiotrophic bacteria that colonized roots and degraded above ground biomass. At 10 cm, there was an acidified layer with a distinct community of acid tolerant bacteria, with reduced richness and tolerance. In the lower layers were oligotrophic slow growing bacteria, and many more unknown taxa. Finally, we defined the core microbiome of wheat grown across 4 locations in distinct precipitation zones of Eastern Washington (Schlatter et al. 2018, 2020). There was tremendous variation in communities in the bulk soil of each location, but the rhizosphere selected for a narrower group of common taxa.

We have described how several different management practices associated with potato production in the Pacific Northwest influence the soil and potato microbiome. For example, we have described the short-term response of the soil microbial community to 1,3-dichlropropene application and, using amplicon sequencing to assess the microbial community, found minimal detectable response (Zeng et al. 2019). Additionally, we documented legacy effects of metam sodium use on the soil microbiome and how previous exposure to metam sodium changes the response of the microbial community if re- exposed to the fumigant (Li et al. 2022). We characterized the microbiome in soils closely associated with seed potato (tare soils) and found that the microbiome varies as a function of seed source and that a large number of potentially plant pathogenic taxa were detected by sequencing tare soil, including pathogens of plants other than potato (Delventhal et al. 2022). Additionally, we found that the potato rhizosphere microbiome establishment appears to be heavily influenced by the microbial community of the soil in which the seed tuber is planted (Delventhal et al. 2022). This suggests that on farm management practices that influence the soil microbiome are an important contributing factor to rhizosphere microbiome establishment that could influence plant productivity.

Identification and Testing of New Biocontrol Agents

As mentioned in previous sections, the field of biological control of soilborne pathogens continues to expand (Lahlali et al. 2022). As of 2022, there are 29 *Trichoderma* products registered with EPA, over 30 with *Pseudomonas*, and 20 with *Bacillus* (http://npic.orst.edu/NPRO/). There are 13 bacterial based BCAs registered in the EU (Bonterra et al. 2022). But this list is expanding by discoveries in the microbiome, which is becoming a tool for finding biocontrol agents (Malacrino et al. 2022, Berg et al. 2021). For the first time, we can see all of the fungal and bacterial community members. In fact, although these BCAs have been extensively researched the last 40 years, they are often just one minor component of the total community. This is why the component of microbiome and phytobiome are of increasing importance in this project.

W-4147 members have been instrumental in discovering and testing many of these new biocontrol agents in the last five years (states doing the research in parentheses). These include

Burkholderia ambifara, Bacillus simplex, *B. safensis, Paenibacillus graminis* and *Lysobacter enzymogenes* (NE) to control *Pythium* and *Rhizoctonia*

Trichoderma atroviride- (AK)

Hyalorbilia aff. *oviparasitica* (basionym: *Dactylella* aff. *oviparasitica*) CA- to control *Heterodera schactii*

Bacillus subtilis- against alfalfa pathogens (DE)

Trichoderma to suppress greenhouse diseases (*Rhizoctonia* and *Pythium* (NH)

Burkholderia and *Pseudomonas protegens*- boxwood blight (VA)

Another exciting finding from the group (CA) is the discovery, characterization and application of bacteria belonging to the genus *Collimonas*, their antifungal properties, and their ability to work synergistically with *Bacillus* bacteria to protect plants from soilborne fungal pathogens (Doan et al, 2019). They discovered a novel *Collimonas*-produced antifungal metabolite (carenaemin) (Akum et al 2021), and found that fungi respond to and adapt after repeated exposure to *Collimonas* biocontrol bacteria (Mosquera et al, 2021). They are collaborating with UC Davis engineers to encapsulate and stabilize *Collimonas* bacteria for field application (Kawakita et al 2021a, Kawakita et al 2021b).

Members continue to look at the mode of action of BCAs, to gain insight on how they may work and be optimized.

This includes:

Pseudomonas spp, which produce antifungal compounds such as phenazine and 2,4-diacetylphloroglucol (WA) (Kwak et al. 2013)

The role of biofilms and electron transport (redox potential and Fe availability) in colonization of roots by phenazine producers (WA) (LeTourneau et al. 2019)

The role of ribosomal proteins surfactin and plipastatin in biofilm formation (DE)

Bradyrhizobium japonicum, a PGPR, increases root biomass via regulation of auxin efflux transporters PIN2, PIN3, PIN7 and ABCB 19 (CA).

The role of signal molecules in zoosporic Phytophthora (*Phytophthora erythroseptica)* and how this may influence disease (ME) (Jiang et al. 2019)

The role of antifungal compounds in *Lysobacter enzymogenes* (NE) (Wang et al. 2015).

Disease Suppressive Soils and Plant Protecting Microorganisms

Over the last 20 years there have been surprising and exciting discoveries for natural methods to suppress or eliminate pathogens, and/or protect plants. One of these are naturally suppressive soils, different paradigm than inoculating with BCAs. Disease-suppressive soils are where indigenous microbes protect the plant against pathogens. Often the disease becomes established in a field, and cause disease for a few years, but then declines, even though the pathogen is still present. Understanding suppression is key to managing the agroecosystem to promote this phenomenon, which is sustainable long-term, and does not require any inputs. This is especially valuable for low-input systems or for organic agriculture.

Intensive studies of disease suppressive soils have led to the development of new methods of analysis (Gross et al. 2007, Borneman and Becker. 2007, Bolwerk et al. 2005, Benitez et al. 2007) and new insights into the nature of soilborne disease suppression (Weller et al. 2002, Hoitink and Boehm 1999). The most interesting direction has been the use of microbiome research to describe these complex communities. Members of W-4147 are recognized as leaders in this area, as evidenced by an invited review article titled “Disease Suppressive Soils: New Insights from the Soil Microbiome” which was published in *Phytopathology* in 2017 (Schlatter et al. 2017). In this paper, Schlatter, Weller, Thomashow and Kinkel, all members of W-4147, speculate on the future of research in this area, show three case studies (take-all, *Rhizoctonia*, and *Streptomycetes*) and construct a series of testable hypotheses. **Many of the advances in the study of suppressive soils have been made by members of W-4147.**

These include:

Rhizoctonia root rot and bare patch (Yin et al. 2015, 2021)

Heterodera schachtii cyst nematode of sugar beet (Chen et al. 2021 and Witte et al. 2021)

Take-all of wheat (Yang et al, 2022, submitted)

Apple replant (Cohen and Mazzola, 2005)

Methods that transform resident microbial communities in a manner which induces natural soil disease suppression have potential as components of environmentally sustainable systems for management of soilborne plant pathogens to reduce the need for pesticides.

Differentiation from other regional workgroups in the area of plant pathology

At the present time, there is no Multistate Project that completely overlaps with this group. The closest is S1083: Ecological and genetic diversity of soilborne pathogens and indigenous microflora. This project mainly focuses on crops in the Southern US, while ours focuses on cropping systems in the Western US. There are several important differences between the two regions, the main one being that water is more limited in the West, and there are more irrigated crops. Crops in the South are primarily centered on corn, soybean and peanuts, whereas there is a greater diversity of crops in the West, especially in California and Washington.

The rest of the projects in plant disease focus on specific pathogens or cropping systems.

Objectives

1. Biocontrol via one or a small number of microbes and/or their products
2. Biocontrol via microbial communities and soil health
3. Creating and testing disease management and/or soil health strategies
4. Extension and outreach

Methods

We have a wide range of research interests and programs in W-5157. But many of these are collaborative and cut across different states that focus on some important cropping systems. We present them according to objectives. Some of the collaborative themes include: potato soil health (WA, OR, ME), plant pathogenic nematodes (CA, WA, ME, MS), and soybean health (especially charcoal rot) (KS, MS). The phytobiome and testing of biocontrol agents are other topics that are being researched by most of the members.

Objective 1. Biocontrol via one or a small number of microbes and/or their products

Bacteria to Promote Growth of Camelina. Members in WA and MT are collaborating on a large Department of Energy CAP grant to increase the nitrogen use efficiency of camelina, a potential biofuels crop. They are isolating bacteria from the rhizosphere, and testing for plant growth promotion. Collaborators in WA have developed a collection of over 3000 bacterial isolates and are screening with organic and inorganic N sources.

Biocontrol agents in potato production in Maine Biological control agents will be identified and tested for their efficacy for potential commercialization. Dilution plating on selective media will be applied for screening beneficial microbes. Agar plate tests, greenhouse assays and field trials will be performed over the time course. The goal is to obtain next-generation microbes that are effective in the control of major potato pathogens.

Biocontrol in soilless production (NH), In this objective, we will characterize effects on disease suppression efficacy of introduced beneficial microbes to help growers use biofungicides in soilless substrates containing wood. Results from the previous project provide preliminary evidence that addition of wood components to peat may enhance efficacy and plant growth promotion effects of *Trichoderma* biocontrol strains. Objectives of this proposed project are to (1) Characterize the effects of wood substrate blends on disease suppression efficacy of fungal and bacterial based bioproducts and (2) Investigate effects of processed wood amendments on activity of *Trichoderma* biocontrol agents.

Objective 2. Biocontrol via microbial communities and soil health

Soil Health in Potato Production. Over the last four years, SCIR has funded a project to look at the potato soil health across the US (https://potatosoilhealth.cfans.umn.edu). Members of W-5147 from Maine, Oregon and Washington have participated in this project by providing samples for microbiome analysis. In addition, OR and WA have initiated a project, funded by the Potato Consortium, that takes a more detailed look at the role of the soil microbiome. Growers have observed greater production in fields on virgin soil that has never been cropped. A three-year project is comparing the microbiome of paired fields from virgin and cropped soils in WA, both in the irrigated production area of the Columbia Basin and the west side of the Cascades. The populations of soilborne fungal pathogens and nematodes will be assessed. Amplicon sequencing will be performed on bulk soils. Soils from all locations will be grown in a common garden in Pullman, WA to assess yield and agronomic aspects. In addition, a more complete microbiome analysis will be done on plants in the common garden, including rhizosphere, root and tuberosphere. Another cooperative project between OR and WA involves developing artificial intelligence (AI) to identify general of pathogenic potato nematodes (*Meloidogyne*, *Pratylenchus*, and *Trichodorus*) from microscope pictures. We are obtaining thousands of photos and training and developing an algorithm with convolution neural network (CNN) methods.

In addition, specific research from Maine will focus on the following. 1) Impacts of organic matter on microbiomes of soil and plants. Living organic materials such as rotation crops, cover crops, and varieties of potatoes will be applied. Meanwhile, non-living organic matters such as compost, lobster shell meal, manures, and other plants by-products will be added to soil as amendments. 2) Impacts of chemical and non-chemical materials on the microbiomes of plants and soil. While applying chemical and biological products to control plant diseases, soil and plant microbiomes will be examined in diversity, abundance, and functions to determine the effects of various applications. Illumina sequencing-based metagenomics will be applied to conduct the studies. 3) Metabolisms of plants related to their responses to plant diseases under various soil conditions as described in 1) and 2). Liquid or gas chromatography coupled to high-resolution mass spectrometry (HRMS) will be used to detect and identify simultaneously biomarkers of effect and exposure accumulating in samples. By examining microbe metabolism, an optimized pattern of soil microbiome will be determined in the suppression of plant pathogens.

In Oregon, research in the potato cropping system will seek to characterize the soil microbiome as a function of crop rotation and use of cover crops, soil amendments, and fumigation. Different crop management practices (i.e., rotation lengths, cover crops, organic amendments, fumigation) will be used to experimentally manipulate soil microbial composition and chemical and physical properties. Data will be collected to determine how these management practices affect the soil biotic and abiotic properties, soil microbiomes and crop health and yield. On-farm studies will also be conducted to survey and collect soils and relevant management histories from fields with different yield potentials in the PNW. Soil biotic and abiotic properties will be measured to determine links between management practices, the soil microbiome, and crop productivity. Additionally, the population dynamics of *Spongospora subterranea* in soils will be studied in multiple grower fields. Pathogen inoculum density in soil will be quantified monthly and hourly soil water and temperature data will be collected throughout the growing season. We will examine the correlation between disease expression (i.e., root galling and powdery scab) and pathogen inoculum density in soils among the field locations. Soil moisture and temperature data will be used to model risk for powdery scab and PMTV infection. We will also characterize the bulk and rhizosphere soil microbiome for plants from each field location to determine if there are microbial taxa antagonistic to *S. subterranea* present in the soils. The long-term goal of the project is to develop diagnostic tools that would inform growers of their risk for losses due to powdery scab and PMTV transmission.

Effect of previous rotation crops on wheat microbiome. Plants are strong drivers of microbiomes and can select a core microbiome. We hypothesize that the previous rotation crop can shape the microbiome of the following wheat crop. Researchers in WA and OR will examine how rotation crops such as winter pea, canola, and other cereals can affect the microbiome in the following season. We will use amplicon sequencing of bacteria and fungi. We will especially emphasize arbuscular mycorrhizal fungi and *Rhizobium.* Canola, in the family Brassicaceae, is one of the few non-mycorrhizal families, can reduce AMF in wheat. These studies will be conducted in long-term rotation studies in WA.

Core microbiome of camelina, a biofuels crop (WA and MT). As part of a DOE grant with researchers in Montana and WA, we will determine the core microbiome of camelina. This will be done by growing camelina in soil from a range of locations and using amplicon sequencing of DNA from the rhizosphere and root. We are especially interested in microbes that may increase nitrogen use efficiency (NUE). We will grow lines with high and low NUE in soil with high and low N levels and use shotgun sequencing and metatgenomics to determine what groups are correlated N level. In collaboration with JGI and Lawrence Livermore Labs, we will also do metabolomics on representative genera from the culture collection, to see what metabolites are consumed and produced.

Utilization of a Novel Bioinformatic Pipeline. This project is to identify novel host-microbe interactions that lead to the development of new biologically-based pest management strategies. Bode Olukolu created several novel bioinformatics tools which he assembled into a new automated pipeline called Qmatey. This pipeline can be used to examine multipartite host-microbe interactions at the strain level, among other features, including the analysis of viruses. Several different types of data can be fed into this analysis pipeline including metagenome, amplicon, genome, and others. **We expect that a considerable number of our project's members will be contributing to this project**, because many of them perform host-microbe interaction research.

Utilization of Spatial Transcriptomics. This project is to identify novel host-microbe interactions that lead to the development of new biologically-based pest management strategies. The Borneman lab is adapting novel spatial transcriptomic methodologies to examine host-microbe interactions. The key experimental design principle employed by this study is determining gene expression patterns that are localized with respect to the processes under investigation. This targeting of gene expression will enable the identification of a small number of genes potentially involved in the processes under investigation. Standard approaches, in contrast, typically use samples and analytical methods that are not highly localized to these processes. Such analyses can produce muddled gene expression data confounded by a much more complex set of functional processes taking place in the larger samples. **Spatial transcriptomics** enables gene expression analyses to be performed at a microscopic scale on thin sections of tissue, facilitating the discovery of new and important molecular mechanisms at a cellular level. **We expect that a considerable number of our project's members will be contributing to this project**, because many of them perform host-microbe interaction research.

Natural disease suppression in soilless substrates (NH). Our previous research indicates that the blending of processed pine wood-derived components into peat may enhance the natural suppression of damping-off disease of radish. Wood processed in different ways can result in components with unique physical, chemical, hydrological, and biological properties which in turn, may influence microbial driven natural disease-suppressiveness. Objectives of this project are to (1) Evaluate the effects of wood preconditioning processes on soilborne disease and to (2) Characterize bulk and rhizosphere microbiomes of peat-wood substrate blends. Research has shown that wood fiber substrates have a unique microbial community composition (Montagne *et al*., 2017) compared to peat and coconut coir but little is known about how these differences translate to disease suppressiveness. As a result, inclusion of wood products into soilless substrates is expected to affect substrate microbiome structure and function. We propose to build on the knowledge in the literature by characterizing soil microbes in peat-wood blends. This research will provide a foundation for future research to investigate correlations between microbial activity and disease suppression.

Endophytes and N-transferring endosymbiosis (NJ). We hope to develop a better understanding of how plants extract nutrients from microbes within their tissues. This is mostly focused on nitrogen transfer mechanisms. We will also be working with the plant breeder Walter Goldstien to help him develop nitrogen-fixing corn by using microbes extracted from highly nitrogen efficient landrace corn. We want a better understanding of when and how these endosymbioses evolved. In examining bryophytes we are finding nitrogen-transfer endosymbioses in non-photosynthetic filaments (caulonemata). We will further develop this information to better understand how early plants evolved these nitrogen-transfer endosymbioses. This is important to understand how important and fundamental these intracellular nutritional symbioses are in plants. In addition we are working on an organic bioherbicide that is focused on killing plants by stimulating endophytic bacteria in weeds to become pathogenic and thus leading to death of the weed host. We have previously submitted a patent application on this technology.

Objective 3. Creating and testing disease management and/or soil health strategies

Measuring the impact of soybean health on soybean diseases (KS and MS).

Management practices to alter the soil community structure will be implemented in replicated research plots at the Southeast Research and Extension Center fields in Parsons, KS. Treatments will include two treatments that are likely to increase disease (soybeans and corn stubble, both hosts of disease organisms), three treatments that are likely to decrease disease (brassica cover crop, animal manure, and solarization), and a fallow control.

Soils will be sampled at three times during the season to determine: soil nutrients, soil community structure, and pathogen populations. Soils will be sampled prior to implementation of treatments (early March), mid-season (late June) and after harvesting soybeans (October). Final soybean yield will be collected. Standard soil nutrient analysis will be performed at the K-State Soil Testing Lab. Soil community structure will be determined with PLFA. Because of the cost of this measurement, we will use funds from additional sources to complete this analysis. Disease presence will be measured using standard plating analysis of soils in the Department of Plant Pathology on campus.

5. *phaseolina* populations will be determined by counting the number of colony-forming units (CFUs). Charcoal rot disease severity will be measured by randomly selecting ten plants per plot at the R7-R8 growth stage for root and stem severity ratings. The plants will be scored by splitting the stem and taproot of each plant and rating the degree of gray discoloration and microsclerotia in the vascular and cortical tissues on a scale of 1-5. *M. phaseolina* root populations will be estimated by grinding the split roots after the severity evaluation. The ground plant tissue and soil samples will be plated on microbiological media and incubated. CFUs of *M. phaseolina* will be counted and transformed to CFUs per gram of root tissue or gram of soil.

We will also look at sudden death and soybean cyst nematode, which are major problems in soybean production.

Resistance to cereal cyst nematode (*Heterodera* spp.) and Fusarium crown rot. This nematode is a problem in the inland Pacific Northwest. Researchers in WA and OR will continue to work with plant breeders to screen for resistance in the greenhouse and evaluate adapted varieties. In addition, in collaboration with nematologists and researchers at CA (Borneman) we will examine the microbiome of cysts and look for suppressive fungi. We will continue to screen synthetic wheat lines and exotic sources for resistance to Fusarium crown rot.

Managing Cyst Nematodes by Predicting Suppressive Soils. This project is to create a new biologically-based strategy to manage crop damage caused by cyst nematodes. Cyst nematodes of the family *Heteroderidae* are one of the most damaging groups of obligate pathogens of economically important crops worldwide (Miller 1986, Jones, Haegeman et al. 2013). The idea for this project came from the following important finding. We performed a study examining indigenous populations of a fungus in field soils cropped to sugar beets in the Imperial Valley of California. This fungus was formerly called *Dactylella oviparasitica*, but it now belongs to a group of nematophagous fungi called the *Hyalorbilia oviparasitica* clade (Yang et al. 2012, Chen et al. 2021, Witte et al. 2021, Smith Becker et al. 2022). We determined that soils with detectable levels of these fungi all produced similar and substantial reductions in the population densities of the sugarbeet cyst nematode (*Heterodera schachtii*) after two nematode generations. In addition, the degree of nematode suppression was not correlated to the initial population densities of these fungi (Smith Becker et al. 2022). **To our knowledge, this is the first study (Smith Becker et al. 2022) showing that the presence of an indigenous biological control microorganism could be used to accurately predict whether the population densities of a plant pathogen would be suppressed.**

One logical new direction is to determine whether we will obtain similar results when some key variables are changed, such as geographic region, crop species, soil type, and/or the cyst nematode population. If we obtain the expected results, we anticipate that we will be able to translate this knowledge into cyst nematode management strategies that enable us to predict which soils can be safely planted with cyst-nematode susceptible crops. **Several members of our multistate project will contribute to this work** – to develop strategies to manage the sugarbeet cyst nematode, the soybean cyst nematode, and the cereal cyst nematode. Project members will include Ole Becker, James, Borneman, Tim Paulitz, Gretchen Sassenrath, and Tessie Wilkerson.

Effects of wood substrate chemistry on plant disease and pathogen activity (NH). As an explanation for suppression of damping-off observed in the previous project, it is possible that certain physiochemical properties of the wood interfere with growth of *R. solani* but this has not been studied for processed wood substrates. To address the knowledge gap, two assays will be performed to investigate the effects of wood components on *R. solani* growth *in-vitro* and disease severity *in-vivo*.

Characterizing and predicting powdery scab suppressive activity in soils (OR).A greenhouse experiment will be set up to compare disease suppression from different soils collected in Oregon. Soils with diverse management histories will be collected from four locations. Soils will be collected from potato and non-potato agricultural systems in Oregon (e.g., forage crops, long-term pasture, etc.). For each soil, the chemical and physical properties (i.e., fertility, soil particle sizes, etc.), microbial biomass, and basal respiration will be determined. Three different potting mixtures of soil will be used to determine the relative importance of soil edaphic and soil biological factors leading to powdery scab suppressive activity. The powdery scab susceptible cultivar 'Shepody' will be used in the disease suppressive activity assay. Bulk and rhizosphere soils will be sampled before, during, and after flowering by destructive sampling of plants at each time point. Root galling and powdery scab will be assessed during destructive sampling. Bulk soils will be sampled directly from each pot and plant rhizoplane soils will be extracted. The microbiome of the resulting soil samples (i.e., bulk, rhizoplane) will be characterized. The suppressive activity of the field soils will be compared and relationships between soil suppression and soil chemical, physical and biological properties will be identified. An analysis of differential abundance will be performed to determine if specific taxa are associated with disease suppression, or disease incidence and severity. By identifying key soil factors associated powdery scab suppression, we can learn which management practices may promote suppressive activity. Similarly, microbial indicator species of disease suppressive activity may eventually serve as a diagnostic tool that could be used to assess a field's risk for disease development. Both outcomes represent novel ways these data could be applied to improve powdery scab management.

Objective 4. Extension and outreach. SEE SECTION ON OUTREACH PLAN IN FOLLOWING SECTION

Measurement of Progress and Results

Outputs

Outcomes or Projected Impacts

- Outcome 1. Knowledge and understanding of the phytobiome of agricultural systems and the benefits and services they provide to crop productivity
- Outcome 2. Understanding of not only the identity of the key drivers providing ecosystem services, but also their function through the use of metagenomics, transcriptomics and metabolomics.
- Outcome 3. Identification of communities and consortia involved in natural suppression of soilborne plant pathogens
- Outcome 4. Reduced use of chemical pesticides and increased use of biologically based products.
- Outcome 5. Reduced damage by soilborne pathogens and increased crop productivity and profitability.
- Outcome 6. Safe, low-cost agricultural products
- Outcome 7. Benefits to growers, consumers, and the environment by making significant progress in producing low cost safe agricultural products.
- Outcome 8. A greater understanding of the basic molecular and biochemical mechanisms will allow a better selection and improvement of existing new strains, and a more rational implementation of these organisms.
- Outcome 9. Knowledge of the genomic and biochemical diversity of microbial communities and biocontrol agents, and how they function in agroecosystems.
- Outcome 10. Understanding how the biocontrol agents interact with the plant and the environment, to predict their limitations and inconsistency in the field.
- Outcome 11. Expanded tool kit of disease management options for both organic and conventional growers, leading to improved agricultural productivity and sustainability.

Milestones

(**1**):(2024): (Year 1): Objective 1: Isolate and identify potential biocontrol agents. Test in vitro and in vivo pot experiments. Objective 2. Survey soils for suppressive activity against soilborne pathogens. Define and characterize phytobiomes of agriculture systems. Objective 3. Establish and maintain research and demonstration sites. Test potential management strategies (biological, biorational, cultural, organic amendments, genetic) in field trials. Objective 4. Transmit knowledge and technology to growers and stakeholders. Develop and implement disease management guidelines for organic and conventional producers.

(**2**):(2025): (Year 2): Objective 1. Test potential biocontrol agents in greenhouse and pot assays. Objective 2. Survey soils for suppressive activity against soilborne pathogens. Identify causal antagonists or suppressive microbial communities (including fungi) with culture-dependent and/or independent (DNA based) techniques. Define and characterize phytobiomes of agriculture systems. Objective 3. Test potential management strategies (biological, biorational, cultural, organic amendments, genetic) in field trials. Objective 4. Transmit knowledge and technology to growers and stakeholders. Develop and implement disease management guidelines for organic and conventional producers.

(**3**):(2026): (Year 3): Objective 1. Test biocontrol agents under commercial conditions, greenhouse or field. Objective 2. Survey soils for suppressive activity against soilborne pathogens. Identify causal antagonists or suppressive microbial communities (including fungi) with culture-dependent and/or independent (DNA based) techniques. Define and characterize phytobiomes of agriculture systems. Objective 3. Test potential management strategies (biological, biorational, cultural, organic amendments, genetic) in field trials. Develop application and management practices. Objective 4. Transmit knowledge and technology to growers and stakeholders. Develop and implement disease management guidelines for organic and conventional producers.

(**4**):(2027): (Year 4): Objective 1. Test biocontrol agents under commercial conditions, greenhouse or field. Objective 2. Characterize how agriculture practices (tillage, rotation, tillage) and beneficial practices organic amendments) drive and shift the phytobiome. Define communities associated with soil health. Objective 3. Test potential management strategies (biological, biorational, cultural, organic amendments, genetic) in field trials. Develop application and management practices, determine cost-benefits, and foster stakeholder involvement. Objective 4. Transmit knowledge and technology to growers and stakeholders. Develop and implement disease management guidelines for organic and conventional producers.

(**5**):(2028): (Year 5): Objective 1. Commercialize potential biocontrol agents. Objective 2. Characterize how agriculture practices (tillage, rotation, tillage) and beneficial practices organic amendments) drive and shift the phytobiome. Define communities associated with soil health. Objective 3. Develop application and management practices, determine cost-benefits, and foster stakeholder involvement. Objective 4. Transmit knowledge and technology to growers and stakeholders. Develop and implement disease management guidelines for organic and conventional producers.

Outreach Plan

The goal of the Outreach Plan for the W-5147 Project will be to deliver fundamental and applied information about biological control of soilborne pathogens. We will target growers, other members of the agricultural community, scientists, and the general public using several mechanisms.

1. **Outreach to Growers**. Over 50% of our members have extension appointments and regularly meet, consult, and teach growers, pest control advisors, and industry representatives via field days, workshops, 344 talks and published extension bulletins.

2. **Online Outreach**. Members of group engage in numerous forms of online media - e.g., our members created the Cornell Soil Health Website (http://soilhealth.cals.cornell.edu/). Growers can send in samples to have soil health status evaluated and to learn of major soil quality constraints that need to be addressed.

3. **Outreach to Industry**. Many of the products developed by our group are being marketed by the biocontrol industry including products such as Root Shield and Plant Shield.

4. **Training the Next Generation of Biocontrol Scientists and Practitioners**. Almost all of our members have teaching responsibilities. They teach courses in biological control and plant pathology, and train graduate and undergraduate student researchers.

5. **Public education to general community and grades K-12**. Our members participate in science fair judging, Master Gardener Training, 4-H and others.

6. **Providing Information to Policy Makers**. Our members will continue to be engaged with policy makers at all levels - e.g., see Subcommittee in Governance section.

7. **Publishing Results in Peer-Reviewed Scientific Journals.** Our group has an excellent publication record, which we plan to continue. In the last 5 years, we have published 362 peer reviewed publications, 23 book chapters, 3 theses and 66 conference proceedings/abstracts by our members, among many other activities.

Organization/Governance

Governance of the W-5147 Project will follow recommended guidelines with the Technical Committee being comprised of research scientists, extension specialists, an Administrative Advisor, and a NIFA Representative. Our Administrative Advisor will be Scot Hulbert. Officers of the Technical Committee will be the Chair and Secretary. The Secretary will be elected each year and will advance to Chair the following year. The location and organizers of the Annual Meetings will be determined by an election. At the Annual Meetings, presentations will be made describing research and extension accomplishments by the members of the Technical Committee. The Secretary will be responsible for taking the minutes at the Annual Meetings, and will work with the Chair to create the Annual Report. Consistent with the sustainability goals of the USDA and the Objectives of the W-5147 Project, we will form a subcommittee that will endeavor to increase funding for soilborne biological control research. We note that USDA used to have a program that only funded biological control research. One of the goals of the W-5147 Project will be to resurrect that program or one that has similar goals. These goals will include increasing the safety and sustainability of US Agriculture through fundamental and applied research and extension projects in biological control. Finally, several new members have been recruited into the project since the last renewal including Johan Leveau, Rachael Vanette, Harsh Bais, Chris Little, Gretchen Sassenrath, Anissa Poleatewich, Bode Olukolu, Kiran Mysore, Chuanxue Hong, Ken Frost, Jed Eberly, Ryan Graebner and Mike Kolomiets, all who have participated in meetings and reports.

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Land Grant Participating States/Institutions

NH,NC,MS,DE,CA,TX,ME,KS,NJ,OR,MT,MN,NY,NM,TN,SC

Non Land Grant Participating States/Institutions

ARS-WA

Participation

Participant	Is Head	Station	Objective	Research						Extension	
				KA	SOI	FOS	SY	PY	TY	FTE	KA
Adhikari, Tika		North Carolina - North Carolina State University	1,2,3	202 212 215 216	1122 1122 1122 1122	1081 1081 1081 1081	1.00	0.40	0.40	0.1	216
Bais, Harsh	Yes	Delaware - University of Delaware	1,2	212	1530	1080	0.10	0.00	0.00	0	0
Becker, Ole		California -Riverside : University of California, Riverside	1,4	216	2410	1120	0.20	0.20	0.00	0.2	216
Borneman, James		California -Riverside : University of California, Riverside	1,2,3,4	212	1440	1040	0.10	0.00	0.00	0	0

Participant	Is Head	Station	Objective	Research						Extension	
				KA	SOI	FOS	SY	PY	TY	FTE	KA
Eberly, Jed		Montana - Montana State University	2	102	110	1100	0.10	0.00	0.00	0	0
Farrar, James J		California Cooperative Extension	4	0	0	0	0.00	0.00	0.00	0.02	216
Frost, Kenneth	Yes	Oregon - Oregon State University	1,2,3,4	212	2110	1060	0.30	0.00	0.00	0	0
Fuhrmann, Jeffry		Delaware - University of Delaware	1,2	215 215	1820 1820	1100 1101	0.10	0.00	0.00	0	0
Garcia, Kevin		North Carolina - North Carolina State University	1	102 102 103 205	2420 611 611 1411	1010 1010 1010 1010	0.10	0.00	0.00	0	0
Hao, Jianjun		Maine - University of Maine	1,2,3,4	212	1310	1017	1.00	1.00	1.00	0.1	212
Kerrigan, Julia	Yes	South Carolina - Clemson University	2,3	212 203	4020 4020	1160 1060	0.10	0.00	0.00	0	0
Kinkel, Linda L.	Yes	Minnesota - University of Minnesota	1,2,3,4	215	4010	1070	0.10	0.00	0.00	0	0
Kolomiets, Mike		Texas AgriLife Research	1	212	1510	1160	1.00	0.00	0.00	0	0
Leveau, Johan		California -Davis : University of California, Davis	1	215	2410	1100	0.10	0.00	0.00	0	0
Little, Chris	Yes	Kansas - Kansas State University	2,3	212	2410	1102	0.10	0.00	0.00	0	0
Olukolu, Bode	Yes	Tennessee - University of Tennessee	1,3	212	1510	1080	0.20	0.00	0.00	0	0
Paulitz, Tim		ARS-WA	1,2,4	212	1549	1160	0.10	0.00	0.00	0	0
Pethybridge, Sarah		New York -Geneva : Cornell University	3,4	212	1410	1160	0.30	0.20	0.50	0.2	212
Poleatewich, Anissa		New Hampshire - University of New Hampshire	1,2,3,4	216 215	2499 2123	1160 1103	0.20	0.50	0.00	0	0
Sanogo, Soum		New Mexico - New Mexico State University	1,2,3,4	212	2410	1160	0.20	0.00	0.10	0	212
Sassenrath, Gretchen F		Kansas - Kansas State University	2,3,4	215 215 203	1820 1540 2499	1060 1060 1060	0.10	0.00	0.00	0	0
Somenahally, Anil		Texas AgriLife Research	1,2,3	102	110	1070	0.20	0.00	0.20	0	0

Participant	Is Head	Station	Objective	Research						Extension	
				KA	SOI	FOS	SY	PY	TY	FTE	KA
Subbarao, Krishna V.		California -Davis : University of California, Davis	1,2,4	212 212	1430 1440	1160 1160	0.10	0.00	0.00	0	0
Vannette, Rachel		California -Davis : University of California, Davis	2	205 212	2160 3110	1070 1130	0.10	0.00	0.00	0	0
white, james f.	Yes	New Jersey - Rutgers University	1,2,3	212	2499	1160	0.70	0.00	0.00	0	0
Wilkerson, Tessie		Mississippi - Mississippi State University	1,2,3,4	212	2410	1160	0.10	0.00	0.00	0	0

Combined Participation

Combination of KA, SOI and FOS	Total SY	Total PY	Total TY
215-2123-1103	0.1	0.5	0
216-2499-1160	0.1	0.5	0
102-611-1010	0.03	0	0
102-2420-1010	0.03	0	0
103-611-1010	0.03	0	0
205-1411-1010	0.03	0	0
212-2410-1160	0.1	0	0
212-1530-1080	0.1	0	0
212-1549-1160	0.1	0	0
215-2410-1100	0.1	0	0
0-0-0	0	0	0
215-1820-1100	0.05	0	0
215-1820-1101	0.05	0	0
212-1510-1160	1	0	0
205-2160-1070	0.05	0	0
212-3110-1130	0.05	0	0
212-1430-1160	0.05	0	0
212-1440-1160	0.05	0	0
Grand Total:	**6.70**	**2.30**	**2.20**

Combination of KA, SOI and FOS	Total SY	Total PY	Total TY
212-1310-1017	1	1	1
203-2499-1060	0.03	0	0
215-1540-1060	0.03	0	0
215-1820-1060	0.03	0	0
212-2499-1160	0.7	0	0
212-2110-1060	0.3	0	0
212-1440-1040	0.1	0	0
102-110-1100	0.1	0	0
215-4010-1070	0.1	0	0
216-2410-1120	0.2	0.2	0
212-1410-1160	0.3	0.2	0.5
102-110-1070	0.2	0	0.2
212-2410-1160	0.2	0	0.1
212-1510-1080	0.2	0	0
203-4020-1060	0.05	0	0
212-4020-1160	0.05	0	0
212-2410-1102	0.1	0	0
202-1122-1081	0.25	0.4	0.4
212-1122-1081	0.25	0.4	0.4
215-1122-1081	0.25	0.4	0.4
216-1122-1081	0.25	0.4	0.4
Grand Total:	**6.70**	**2.30**	**2.20**

Program/KA	Total FTE
0	0
0	0
0	0
0	0
0	0
0	0
216	0.01
0	0
0	0
0	0
0	0
212	0.03
0	0
0	0
0	0
0	0
0	0
0	0
216	0.07
212	0.07
0	0
212	0
0	0
0	0
0	0
216	0.03
Grand FTE Total:	**0.62**