ABSTRACTS FROM THE SOCIETY OF NEMATOLOGISTS ANNUAL MEETING 2020

POSTERS (S = synchronous; A = asynchronous; K = keynote; C = Cobb Foundation)

S-1
THE EFFECTIVENESS OF MYCORRHIZA AND MYCORRHIZA HELPER BACTERIA (MHB) CONSORTIUM ADDED WITH ORGANIC MATTER AGAINST PRATYLENCHUS COFFEAE ON COFFEE PLANTATIONS.

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The nematode Pratylenchus coffeae is a major threat in robusta and arabica coffee plants that potentially lower the yield to 78%. Until now there is no effective biological agents in the field because the researchers only focused on nematodes as a target and had less attention to the ecology of the rhizosphere. Therefore, in the biological control of nematodes involves factors affecting soil environment and the most important is the organic matter. Adding organic matter is believed to improve the effectiveness of mycorrhizal and Mycorrhiza Helper Bacteria (MHB) in controlling P. coffeae on coffee plantation.

Testing the doses of mycorrhizal and MHB combined with the dose of organic fertilizer on two years old coffee plants and productive coffee plants infected by nematodes P. coffeae (endemic) on random complete block design (RCBD) with factorial treatment 4 x 3. The observation on soil biology, plant growth, the population of P. coffeae, root and shoot damage assessment, the % colonization of mycorrhiza, and uptake of Phosphorus (P). For data analysis the SPSS software version 16 (including the ANOVA test) was used.

The results indicated that mycorrhizal and MHB combined with organic fertilizer can reduce significantly P. coffeae in two years old coffee plants and productive coffee plants. In two years old arabica coffee, the best treatment was M1O1 (250 spore of mycorrhiza+10 ml MHB 10⁸ combined with 5 kg organic fertilizer) reducing the nematode population up to 70%. In productive arabica coffee, the best treatment was M2O1 (1,000 spore of mycorrhiza+100 ml MHB 10⁸ combined with 15 kg organic fertilizer) decreasing the nematode population up to 68.7%. While in robusta coffee the best treatment was M3O2 (750 spore of mycorrhiza+10 ml MHB 10⁹ combined with 10 kg organic fertilizer) reducing the nematode population up to 55%.

Application of mycorrhizal and MHB combined with the dose of organic fertilizer proven to reduce P. coffeae population significantly, therefore it is recommended to be applied on coffee plantations infected by the nematode P. coffeae.

A-2
SEED-DELIVERED FLUOPYRAM MITIGATES ROOT-KNOT NEMATODE DAMAGE IN FRESH MARKET CARROT FIELD TRIALS.


Fresh market carrot production in California made up 87% of the 2019 US-grown harvest. Several root-knot nematode species (RKN), particularly Meloidogyne incognita and M. javanica, are major production challenges and are wide-spread in the lighter soil types throughout the central and southern part of the State. They cause severe root damage such as forking and galling that reduce marketability and revenues. Carrot cultivars resistant to the pathogens and registered effective contact nematicides or biocontrol agents are not available. Consequently, nearly 44% of California’s carrot growing areas were treated with a soil fumigant (1,3-dichloropropene, metam-sodium, or metam-potassium). Regulatory, environmental, and economic issues encourage the search for alternatives. Fluopyram was originally developed as a broad-spectrum succinate dehydrogenase-inhibiting fungicide. More recently, it received Federal EPA registration as a nematicidal seed treatment in soybeans and cotton. The goal of this project was to evaluate fluopyram as a carrot seed-delivered nematicide in a RKN-infested field. The trial was conducted from May 1 to July 16 during the 2019 season at the University of California South Coast Research and Extension Center, Irvine, CA. The soil at the trial site was a sandy loam infested with M. incognita (Pi 1.66 J2/100 cm³).

Carrots seeds cv. Choctaw were treated by Bayer CropScience with thiram (1.87 g/kg), metalaxyl (0.153 g/kg), and fluopyram at various rates (0.006, 0.01, or 0.035 mg/seed). Individual plots were 3-m long with 0.6-m wide beds. The experimental design was a randomized complete block with 5 replications. Trifluralin (0.85 kg product/ha) was incorporated into the seedbed as a pre-season herbicide. The soil-applied nematicide Salibro (2.2 L/ha, a.i. 50% fluazaindolizine) was used as a trial quality check. At harvest, ten randomly selected mature carrot roots from each plot were assessed for root-knot nematode disease symptoms (galling rating scale 0-10, 0= no galls, 10= completely galled, stunted, dying). Carrots rated 0-3 were considered marketable. All data were subjected to one-way ANOVA. Population counts and gall rating data were log (x +1) and arcsine (sqrt(x/10) transformed before statistical analysis. Means separation was performed by Fisher’s protected LSD (P < 0.05). The seeds sprouted evenly with no signs of phytotoxicity. At harvest, there was no difference in efficacy among the fluopyram rates. They reduced gall rating at harvest by 2 classes and forking by about 87%, which resulted in a marketable yield increase of 78% compared to the non-treated control. These results demonstrate fluopyram's excellent crop protection efficacy at a low seed-delivered application rate. It may be especially useful in low to moderately RKN-infested soils and in field areas where soil fumigants are not permitted.

S-3
VARIABILITY OF MELOIDOGYNE INCognITa INFECTION ON HEMP (CANNABIS SATIVA) CULTIVARS.

Cultivation of hemp is rapidly expanding, especially of medicinal varieties with high levels of THC or CBD. Susceptibility of hemp to plant-parasitic nematodes is poorly known due to lack of investigation for most of the past 100 years. Several greenhouse experiments were conducted to determine suitability of selected hemp cultivars as hosts for Meloidogyne incognita. In a small trial run, fiber cv. ‘Delores’ was heavily galled and yielded 0.5–1.2 million M. incognita eggs per plant (reproductive factor Rf = 36–81), whereas CBD cv. ‘Wife’ had little galling and no eggs. Fiber-type varieties were grown from seed, while CBD-rich varieties were propagated by means of terminal cuttings. Young plants were transplanted into a 3:1 mix of sand:peat in 15-cm-diam. pots, which were then infested with 5,000 eggs/pot of a stock M. incognita population. A pelleted fertilizer was added to the mix during planting and after one month a dilute solution of 20:20:20 soluble fertilizer was applied to each pot. In the first experiment, in summer, no supplemental light was provided. In the other experiments daylight was extended to 17 hours with overhead lights. All experiments were arranged in a randomized block design. ‘Rutgers’ tomato was used as a check on inoculum viability. Root systems were harvested 55–60 days after infestation and rated for galling, after which eggs were extracted and counted. Rf values were calculated: Rf = Pf/Pi; Rf-max (maximum) values are used for comparison. Galls were small, hard and white, and rarely consolidated into larger galls. In the first experiment, three fiber cultivars were all moderately galled, with maximum Rf-max values of 10.4–11.1. For CBD cultivars, ‘Wife’ again had very little galling and an Rf-max of 0.2. ‘Charlotte’s Web’ was an excellent host with an Rf-max of 39.6, followed by four other cultivars (Cherry, OG, Suver Haze, ‘Chalice’) with Rf-max values of 10.5–16.8. ‘ACDC’ and ‘Carolina’ were poor hosts (Rf-max 3.1 and 2.4, respectively). This experiment was repeated in part with cultivars Carolina, Charlotte’s Web and Wife, with results similar to the previous results. A similar experiment was conducted with ‘Charlotte’s Web’ and two additional cultivars (Siskiyou Gold, Special Sauce). ‘Charlotte’s Web’ had an Rf-max of 17.5, whereas ‘Siskiyou Gold’ was 4.8 and ‘Special Sauce’ 8.8. In all experiments, results for different cultivars were often highly variable; nevertheless, there is a wide range of reactions from highly susceptible to nearly immune among hemp cultivars, which could serve as a basis for developing resistant selections should M. incognita become a limiting factor in hemp production.

A-4 HOST SUITABILITY OF SUMMER COVER CROPS TO FOUR ROOT-KNOT NEMATODE SPECIES (MELOIDOGYNE SPP) IN FLORIDA. Bui, H.X., and J.A. Desaeger. Department of Entomology and Nematology, University of Florida Gulf Coast Research and Education Center, Wimauma, Florida, 33598, United States.

Summer cover crops can be a useful tool for managing plant-parasitic nematodes in Florida. A greenhouse experiment was done to determine the host suitability of four commonly used cover crops in Florida, sunnhemp (Crotalaria juncea), cowpea (Vigna unguiculata), sorghum sudangrass (Sorghum bicolor L. Moench) and sunflower (Helianthus annuus) to four tropical root-knot nematode (RKN) species Meloidogyne javanica, M. incognita, M. enterolobii and M. arenaria. Tomato (cv. HM1823) was included as the check. No visible root galls were observed for any of the RKN species on sunhemp and sorghum sudangrass at four and eight weeks after nematode inoculation (WAI). In contrast, tomato showed the highest gall damage for all tested RKN species at four and eight WAI. Sunflower and cowpea were infected by all four tested RKN species, but host suitability varied. On sunflower, M. incognita, M. javanica and M. arenaria caused more root galls than M. enterolobii. On cowpea, the opposite was noted with M. javanica and M. enterolobii inducing more root galls than M. arenaria and especially M. incognita both at four and eight WAI. Our preliminary observations showed that sunnhemp and sorghum sudangrass were poor hosts to all four tested RKN species. Sunflower and cowpea confirmed to be good hosts for root-knot nematodes, although sunflower showed less root galls with M. enterolobii, while cowpea showed less galls with M. arenaria and M. incognita. Nematode egg and juvenile counts are still pending and will further reveal the hosts status of these summer cover crops for four common RKN species in Florida. Our preliminary data confirm and stress the importance of RKN species identification when selecting cover crops as a nematode management strategy.

S-5 CAN THE EFFICACY OF ENTOMOPATHOGENIC NEMATODE AGAINST DIAMONDBACK MOTH AND IMPORTED CABBAGE WORM BE IMPROVED BY OTHER CULTURAL PRACTICES? Budathoki, Sabina, and K.-H. Wang. Department of Plant and Environmental Protection Sciences, University of Hawaii, Honolulu, HI 96822.

The success of foliar application of entomopathogenic nematodes (EPNs) to manage diamondback moth (DBM) and imported cabbageworm (ICW) in the field had been inconsistent and not promising due to its susceptibility to desiccation and UV radiation. Two field trials were conducted by integrating trap cropping (mustard green) with EPN (Steinernema feltiae MG14) application in a kale agroecosystem. It was hypothesized that mustard green planted at 1:1 ratio with the cash crop would lure DBM and ICW away from feeding on kale, and would reduce the pest pressure for EPN to be effective. A 2 × 2 (trap crop × EPN) factorial experiment i.e. trap crop vs no trap crop, and EPN (1.25 million IJs/ha, foliar spray) vs no EPN were established in a 2-week old kale farm. Treatments were replicated four times. Insects were monitored weekly over 6 weeks. Planting of mustard green suppressed abundance (45.8%) and damage (22.2%) of DBM on kale (≤ 0.05), whereas no significant effect was observed against ICW in Trial I. In Trial II, trap cropping did not affect DBM abundance and damage but did reduced ICW damage by 12.6% (P < 0.05). On the other hand, EPNs had no effect on DBM but reduced ICW numbers by 60% (P ≤ 0.05) in Trial I; whereas EPN significantly suppressed DBM numbers by 87.5% (P < 0.05) with no effects on ICW in Trial II. No interaction between trap crop and EPN was observed in all parameters measured. Thus, trap cropping did not enhance the efficacy of EPN. Another field study was conducted to examine the potential of using intermittent sprinkler irrigation to enhance the efficacy of EPNs against DBM and ICW in a head cabbage field. It was hypothesized that intermittent sprinkler irrigation could keep the leaf surface moist and would increase the longevity of EPNs applied as a foliar spray. A 3 × 2 (sprinkler irrigation regime × EPN) factorial designed experiment was installed where intermittent sprinkler irrigation regimes include 5-min sprinkler i) throughout day and dusk (SDd), ii) during dusk only (Sd), and iii) absent (NS), with and without EPN foliar spray (1.25 million IJs/ha) was conducted. Treatments were replicated 4 times. Abundance and damage of DBM and ICW were monitored...
weekly over 7 weeks. SDd and Sd both decreased DBM and ICW damage without effects on DBM and ICW abundance on the plant. EPN had no effect on reducing DBM and ICW abundance and damage. Unfortunately, no interaction occurred between sprinkler irrigation and EPN application, indicating that intermittent sprinkler irrigation also did not improve the performance of EPN on these pests. The only trend that EPN suppressed DBM or ICW successfully was when the pest pressure were below 0.5 per plant, the economic threshold of these pests on brassica crops.

A-6
UNRAVELING EARTHWORMS IMPACT OVER ENTOMOPATHOGENIC NEMATODE INFECTIVITY: GENERAL TREND OR SPECIES-SPECIFIC DEPENDENT? Chelkha, Maryam1,2, Blanco-Pérez, R.1, Vicente-Diez, I.1, González-Trujillo, M. M.1, Amghar, S.1, El Harti, A.1, and Campos-Herrera, R.2. 1 Research Team "Lombricidae, Improving Soil Productivity and Environment" (LAPSE), Ecole Normale Supérieure (E.N.S.), Centre d'Eau, Ressources Naturelles, Environnement et Développement Durable (CERN2D), Mohammed V University, Avenue Mohamed Bel Hassan El Ouazzani, BP: 5118. Takaddoum – Rabat, Morocco. 2 Instituto de Ciencias de la Vid y del Vino (CSC-Universidad de La Rioja-Gobierno de La Rioja), Finca La Grajera, Ctra. de Burgos Km. 6, 26007 Logroño, Spain.

Earthworms are beneficial to soil organisms that promote soil structure and enhance the decomposition of organic matter, but also contribute to the passive displacement in soils of biological control agents (BCAs) such as the entomopathogenic nematodes (EPNs) in the families Steinernematidae and Heterorhabditidae. However, a recent study showed that larval mortality and other fitness features of large (> 600 mm) steinernematid infective juveniles (IJ)s might decrease after being exposed to the earthworm species Eisenia fetida (Haplotaxida: Lumbricidae) or it's cutaneous excreta (CEx). Whether this phenomenon in the co-existence of EPNs and earthworms is the general trend or species-specific dependent is still unknown. We hypothesized that earthworms of different nature could affect EPN ability as BCAs differentially. This study aimed to investigate the interaction of three earthworm species (E. fetida, Lumbricus terrestris, and Perionyx excavatus) (Haplotaxida) and three EPN species (Steinernema feltiae, S. riojaense, and Heterorhabditis bacteriophora) (Rhabditida). In laboratory experiments, we evaluated the killing and reproductive ability of EPNs against Galleria mellonella (Lepidoptera: Pyralidae) larvae when inoculated in autoclaved soil, single applied or combined with earthworms or their CEx. In a subsequent study, we evaluated in multi-well plates the effect of the CEx in EPN activity at two concentrations (1.5 and 10 IJs/cm2). For the soil experiment, the presence of the earthworms E. fetida and L. terrestris caused a significant reduction (P < 0.05) in S. feltiae and H. bacteriophora infectivity. Similarly, the CEx produced by E. fetida and P. excavatus negatively affected S. riojaense and H. bacteriophora activity. However, the infectivity of H. bacteriophora and S. feltiae increased in the presence of the CEx produced by E. fetida and P. excavates. Moreover, in the multi-wells experiment, the CEx extracted from E. fetida and L. terrestris reduced the infectivity of S. feltiae and S. riojaense, and H. bacteriophora and S. riojaense, respectively, but only at specific timings. However, we observed the opposite trend for the interaction of P. excavatus and S. feltiae, even if depending on the initial IJ inoculum. Conversely, the EPN reproduction ability was not affected, except for a few cases, by any of the interactions under study. Overall, we concluded that the detrimental effect on EPN pathogenicity and reproductive capacity caused by the presence of earthworms, or their CEx, cannot be considered as a general trend. Thereby, there is no clear evidence of incompatibility for both beneficial soil organisms in the EPN use as BCAs.

S-7
EVALUATION OF SOYBEAN CULTIVARS FOR RESISTANCE TO A NEW PRATYLENCHUS SP. DETECTED IN NORTH DAKOTA. Chowdhury, Intiaz and G. P. Yan North Dakota State University, Department of Plant Pathology, Fargo, ND 58108.

Root-lesion nematodes, Pratylenchus spp., are one of important groups of plant-parasitic nematodes that has a worldwide distribution and a broad host range including soybeans. One of the key management strategies against root-lesion nematodes is host resistance. Hence, two greenhouse experiments were conducted in this study to ascertain resistance levels of soybean cultivars to the new species of root-lesion nematodes detected in ND (Experiment 1) and the habitat preference of this new root-lesion nematode species (Experiment 2). A total of twenty soybean cultivars used in the region were evaluated in both experiments. Additionally, the local soybean cultivar Barnes and a non-planted control were used as positive and negative controls, respectively. The experiments were conducted in five replicates with naturally infested soil. Experiment 1 was harvested at 15 weeks after planting while Experiment 2 was harvested at 9 weeks to aid in separation of roots and soil, and both experiments were repeated once. The ratio of the postharvest population density of a test cultivar relative to the postharvest population density of a susceptible check was used to scale resistance rating. Among the 20 soybean cultivars evaluated, a soybean cultivar with the ID# M consistently produced the greatest postharvest population densities in both trials of Experiment 1 and Experiment 2 and therefore it was selected as the susceptible check. Additionally, cultivar ID# O also showed susceptible reaction in all the trials. In contrast, four cultivars were the least susceptible across all the trials. Combined results of all the trials indicated that seven of the cultivars were moderately resistant, nine of the cultivars and the local cultivar Barnes were moderately susceptible, and four of the cultivars were susceptible. However, none of the cultivars evaluated were classified as resistant. Analysis of the habitat preference of the new species in roots versus soil revealed that above or close to 50% of the root-lesion nematodes resided in the root habitat and the remaining of the nematodes were recovered from soil at nine weeks after planting for a majority of the cultivars tested. These results provide an insight into the virulence of the new root-lesion nematode species identified in ND on commercial soybean cultivars.

A-8
KILLER POOP: THE STORY OF HOW ANIMAL BY-PRODUCTS DEFEATED PRATYLENCHUS PENETRANS. Cole, Emilie1 and M. Quintanilla1. Michigan State University, Department of Entomology, 288 Farm Ln. East Lansing MI 48823.

The root lesion nematode, Pratylenchus penetrans, is a significant pest of potatoes especially when in conjunction with the soil borne fungus Verticillium dahliae. Together these pests form the disease complex known as Potato Early Die (PED) which can result in yield losses up to 50% in severely infected fields. Although management has typically relied on broad spectrum fumigants and nematicides, growers are becoming increasingly conscientious of their soil health, and therefore are interested in alternative management strategies.
Utilizing composts and manures for pest control is ideal in the potato system since they improve soil quality and health, are easy to apply and are affordable for the grower. Hence, the objectives for this project were to: 1) identify promising compost blends and manures to control *P. penetrans* and 2) determine the effect of the promising products on *P. penetrans*, *V. dahliae* and yields under field conditions. In 2018, a selection of five compost blends provided by Morgan Composting, Inc. (LAB, Dairy Doo, Poultry Compost, Worm Castings and Dairy Doo with Spelt) and poultry manure were tested against a pure sand control for their ability to reduce *P. penetrans* survivorship under laboratory conditions. For each product, five arenas were created, and each arena was inoculated with 200 *P. penetrans*. After a week, survivorship was assessed. The poultry manure and LAB resulted in the lowest survivorship with 0% each. These products were then tested at variable rates of (5, 15, 30, 50, 75 or 100%) using the same design as described previously. At 5% LAB, survivorship was 0% of control while the poultry manure was 3% of control. As rate increased, survivorship decreased. In the field, Dairy Doo, Poultry Manure, and LAB Compost were all tested at high (5 ton/ac) and low (1.25 ton/ac) rates against a negative (untreated) control and positive (Vydate [oxamyl]) control in a completely randomized block design. Over the course of the season, soil samples were collected to monitor *P. penetrans* and *V. dahliae* populations and yields were collected at harvest. The Poultry Manure high treatment resulted in a significant reduction in *P. penetrans* populations while Poultry Manure low resulted in significantly higher marketable yields. *V. dahliae* incidence was not significantly impacted by treatment, however the number of viable microsclerotia were significantly reduced two weeks after application in the LAB Low treatments. Overall, this study suggests that Poultry Manure and LAB Compost have significant potential for root lesion nematode control. Reliable control of *V. dahliae* using composts and manures, however, has not been elucidated and will likely require a multifaceted approach.

S-9

THE LIFE CYCLE DEVELOPMENT AND REPRODUCTION OF MELOIDOGYNE ENTEROLOBII, *M. INCNIGITA*, AND *M. JAVANICA*: A COMPARATIVE STUDY. Collett, Raymond¹, M.S. Dance¹, M. Marais¹, and H. Foure¹. ¹Unit for Environmental Sciences and Management, North-West University, Potchefstroom, South Africa, ²Agricultural Research Council-Tropical and Subtropical Crops (ARC-TSC), Mbombela, South Africa. ³Nematology Unit, Biosystematics, Agricultural Research Council-Plant Health and Protection (ARC-PHP), Queenswood, Gauteng, South Africa.

Research pertaining to *Meloidogyne enterolobii* is limited to 317 scientific documents being published since its first identification in 1983. Collectively, only 3% of these sources pertains explicitly to the life cycle of *M. enterolobii*. It was thus the aim of this study to determine the life cycle and reproduction of *M. enterolobii*, in comparison to that of *M. incognita* and *M. javanica*, under greenhouse conditions over a period of for 25 days. Ten mature females of each species were removed from existing in vivo reared populations for SCAR-PCR and perineal pattern investigations to verify the identity of the species. Three susceptible crop host genotypes were used, namely maize (‘P-2432-R’), soybean (‘DM-5953-RSF’), and tomato (‘Moneymaker’). Each crop seedling was inoculated with 24-h old motile second-stage juveniles (J2) of the respective *Meloidogyne* species. The glasshouse temperature ranges were 20 to 28 °C. At time intervals of 3, 5, 10, 15, 20, and 25 days after inoculation (DAI) five root replicates of each crop host and root-knot nematode species were removed and staining using the sodium-hypochlorite-acid-fuchsin method. The life-stage development of each *Meloidogyne* species was determined by randomly selecting 20 nematode individuals from each root. At 20 and 25 DAI egg masses were observed and stained using the eosin-Y staining protocol; 10 egg-masses of each *Meloidogyne* species were then isolated and the number of eggs per egg-mass counted to determine their reproduction properties. Statistical analyses were done using Factorial ANOVAs (life stage development) and Student’s t-Tests (reproduction). Degree days (DD) were also calculated for each species. Significant (*P* ≤ 0.05) differences existed for the number of each of the life stages of each species among some of the time intervals. *Meloidogyne enterolobii* developed more rapidly from one life-stage to the other compared to its two counterpart species. Although females were observed for all three species 15 DAI, single eggs were observed for *M. enterolobii* only. Egg masses were, however, produced by females of all three species 20 and 25 DAI. The presence of J2 generations of *M. enterolobii* and *M. javanica* from 20 DAI compared to those of *M. incognita* (recorded from 25 DAI only) confirmed the quicker development of *M. enterolobii* as well as of *M. javanica*. Ultimately, the shorter DD needed by *M. enterolobii* to complete its life cycle compared to those of *M. javanica* and *M. incognita* represents novel information, both fundamentally and for its applicability. An improved advisory approach to farmers can now, for example, focus on rather using crop genotypes with shorter growing periods. Other management strategies can also be streamlined to focus on combating *M. enterolobii* by, for example, interfering with its rapid life-cycle duration.

A-10

DEVELOPMENT OF AN EFFECTIVE SCREENING METHOD FOR DETECTING ROOT-KNOT NEMATODE IN SWEETPOTATOES. Culbreath, Julianna¹, W. Rutter², and C. Khanal¹. ¹Department of Plant and Environment Science, Clemson University, Clemson, SC 29634, ²USDA, ARS, Charleston, SC, 29414.

The guava root-knot nematode (*Meloidogyne enterolobii*) is an aggressive plant pathogen that attacks a diverse range of crops, leading to substantial economic losses for growers globally. In recent years, *M. enterolobii* has been identified as an emerging pathogen of sweetpotatoes (*Ipomoea batatas*). Generally, detecting a plant-parasitic nematode infection is a challenging task that requires a trained eye, as most symptoms of plant parasitic nematodes are generic to a wide scope of plant pathogens. Here, a sweetpotato storage root survey method has been developed and implemented to detect and identify *M. enterolobii* and possibly assist in monitoring the root-knot nematode (RKN) population. The sweetpotato skins are removed, using an industrial vegetable peeler, and two 10 ml samples are collected and processed for DNA analysis. Additionally, *M. enterolobii* eggs are extracted from the extra skins, to be used for inoculation of susceptible and RKN resistant solanaceous crops. This method allows for high-throughput screening as up to 9 kg of roots can be tested at once, providing growers and breeders with a manageable and practical option to check fields for this RKN. This approach is also sensitive enough to detect a miniscule amount of nematode eggs in the storage roots, which would permit growers to employ management practices before significant yield loss occurs. Compared with other RKN detection methods, this method is highly efficient, as large samples can
be processed quickly while retaining high sensitivity and accuracy. To date, this procedure has detected and recovered 100% of the *M. enterolobii* spiked controls and RKN infection has been successfully recovered from all of the symptomatic samples. Further experiments need to be conducted, but this sweetpotato storage survey method presents a promising and novel approach at screening, monitoring and ultimately managing the guava root-knot nematode population for growers and breeders worldwide.

A-11
IMPACT OF MANURE-BASED SOIL AMENDMENTS ON PLANT-PARASITIC NEMATODE TAXA ASSOCIATED WITH HOP (CV. CENTENNIAL). Darling, Elisabeth1, E. Cole,1 H. Chung2 and M. Quintanilla1, 1Michigan State University, Department of Entomology, 288 Farm Ln. East Lansing, MI; 2Michigan State University, Department of Entomology, 293 Farm Ln. East Lansing, MI.

Plant-parasitic nematodes commonly encountered in Michigan are damaging to an abundance of crops, yet impact to regional hop production remains unclear. For this reason, our objectives were (1) to document the nematode genera in a well-established hopyard with no history of compost or nematicide applications, (2) to conduct preliminary greenhouse trials to investigate the impact of the northern root knot nematode (*Meloidogyne hapla*) and the hop cyst nematode (*Heterodera humuli*) on young plants, and (3) to establish a field trial to observe the potential control of hop nematode populations with various soil amendments. In June 2020, 75 soil samples were collected in Greenville prior to treatment to determine nematode taxa present. Preliminary greenhouse trials were conducted using pooled Centennial hop rhizomes with four replicates. Finally, a RCBD field trial was established to determine if manure-based composts were effective at controlling different types of nematode pests.

Plant-parasitic nematodes encountered in the field trial included: *Helicotylenchus* spp., *Tylenchorhynchus* spp., *Pratylenchus* spp., *Xiphinema* spp., and *H. hapla*. *H. humuli* was not present in the field trial. Based on nematode pressure in field trial, preliminary greenhouse trials and past literature, we hypothesize that five nematodes have the potential to impact Michigan hop growth: *M. hapla*, *H. humuli*, *Pratylenchus* spp., *Tylenchorhynchus* spp., and *Xiphinema* spp. In preliminary greenhouse trials, *M. hapla* and *H. humuli* infested plants developed reduced bine heights of 69.4% and 40.4% respectively, compared to healthy controls. We found two compost applications (pelletized poultry manure and LAB) and one nematicide (Velum [fluopyram]) to be suitable options for controlling *Pratylenchus* populations on hop. Across all treatments, *Pratylenchus* nematodes and *Xiphinema* populations decreased when soil amendments were applied, while *Helicotylenchus* and *Tylenchorhynchus* nematodes were not impacted by any of the treatments. Hop cone yield was negatively correlated with root lesion populations ($R^2 = 0.5571$, $P < 0.05$), but further studies need to be done to determine thresholds and document symptoms.

C-12
UPDATE ON THE PROGRESS OF PROJECT NEMATODA. Eisenback, J. D., School of Plant and Environmental Sciences, Virginia Tech, Blacksburg, VA 24061.

Project Nematoda is a collection of every description of nematodes, both parasitic and free-living. The first round is nearly complete with just three letters, R, S, and T remaining to be inputted into the Zotero database. The collection is a bibliography that is linked to the original descriptions in the form of PDFs. More than 18 thousand species have been entered and new species are being entered as they are published or found on the internet from various journals and libraries. Taxonomists are encouraged to submit PDFs of their published descriptions for incorporation into the Project. A few volunteers regularly submit descriptions from their personal collections and two taxonomists place them directly into the Zotero database. Additional resources are welcomed and more users of the Project are invited. Since the database has limited access, participants and users are directed to ask permission to use the database from the administrator of the Project via email at jon@vt.edu.

S-13
GRAFTING OF BELL PEPPERS TO CONTROL THE SOUTHERN ROOT-KNOT NEMATODE DAMAGE. Franco-Navarro, Francisco, and A. T. Ploeg. Department of Nematology, University of California, Riverside, CA 92521.

The Southern Root-Knot Nematode (SRKN), *Meloidogyne incognita*, is a serious pest in the southern United States on many solanaceous crops including pepper, tomato and eggplant. Grafting of susceptible varieties onto nematode-resistant rootstocks is a method to limit nematode infestation and damage in solanaceous crops and even can help to eliminate the use of nematicides. In the present study, the goal was to evaluate the resistance of some pepper varieties to *M. incognita* race 3 and their usefulness as rootstocks for a susceptible commercial pepper cultivar under greenhouse conditions. A greenhouse trial was conducted to evaluate the effect of eight Capsicum materials as rootstocks on plant growth, infestation of the SRKN, and resulting disease symptoms. Susceptible bell pepper cv. 'Baron' was used as the scion for all rootstocks. Non-grafted and self-grafted 'Baron' and non-grafted rootstocks were included as controls. At the start of the trial, all plants were inoculated with 1500 SRKN J2s. After 70 days, the number of egg masses, root galling, number of J2s per gram root and the reproduction factor were highest in non-grafted and self-grafted 'Baron'. On most of the rootstocks, galling was not observed. Furthermore, they had fewer egg masses, J2s per gram of root, and a lower reproduction factor. Grafting was successful with most rootstocks, but incompatibility between scion and rootstock was observed in a few combinations. Although resistance to SRKN was maintained after grafting of the scion variety onto most of the resistant rootstocks, the biomass and yield were not significantly different among non-grafted and grafted plants. We conclude that grafting susceptible bell pepper on to resistant rootstocks can prevent nematode infestation and build-up and does not negatively affect growth of the susceptible scion.

A-14
NEMATODE COMMUNITY DIVERSITY AND COMPOSITION ACROSS TWO ENVIRONMENTAL GRADIENTS IN THE SANDHILLS OF WESTERN NEBRASKA. Gattoni, Kaitlin1, A. Borgmeier2, E. Gendron3, J. P. McQueen1, P. Mullin1, K. Powers2, T. Powers3, and D. L. Porazinska1. 1Dept. of Entomology and Nematology, University of Florida, FL 32611. 2Department of Plant Pathology, 406 Plant Science, University of Nebraska-Lincoln, Lincoln NE 68583.
Lakes at the Crescent Lake National Wildlife Refuge of the Western Nebraska Sandhills are unique ecosystems existing along a natural pH gradient amid grass-covered sand dunes. The distinctive chemistry of the lakes is driven by potassium (K) and sodium (Na) resulting in their extreme alkalinity (pH > 10). There is no documentation of macroorganisms living in these highly alkaline lakes, but there are complex communities of microorganisms. The goal of this research is to describe nematode diversity along two environmental gradients: 1. alkalinity gradient spanning a pH from 7 to 10.4, and 2. habitats including lakes, their shorelines, and surrounding prairies.

In October of 2019, we collected a total of 45 sediment/soil samples from and around 5 lakes (Island, Gimlet, Bean, Kokjohn and Border). Each sample was a mix of 12-16 cores from an area ~ 20 m². Nematodes were extracted from 50 mL subsamples, counted and identified with microscopy, and then processed for 18S rDNA metabcoding. We hypothesized that: 1. The most alkaline sediments and soils would support the most depauperate nematode communities, and 2. all three habitats would support distinct nematode communities. To test the hypotheses, alpha diversity was measured by the Shannon index and beta diversity was measured by the Bray-Curtis distance dissimilarity matrix. The presence of significant (P<0.05) differences in alpha diversity were tested with a one-way ANOVA followed by Tukey’s Honest Significant Differences (HSD) and differences in beta diversity were visualized with PC plots and tested with PERMANOVA.

The alpha diversity of the shoreline and prairie nematode communities was significantly higher than that of lake sediment communities. As predicted, the most alkaline lakes supported more depauperate nematode communities than the neutral lakes, however, the shoreline and prairie nematode diversity was less constrained by pH. Beta diversity generally supported our second hypothesis and indicated distinct communities among the three habitats. Specifically, the 1st PC clearly separated the prairie from the lake and shoreline communities while the 2nd PC further separated the lake from the shoreline communities, although the more alkaline shoreline communities were most like those of the alkaline lake sediments. Predatory nematodes, specifically Tobrilidae and Tripylidae, were dominant within the lake sediment and shoreline soils. The prairie nematode communities were dominated by bacterivores, such as Cephalobidae, as well as plant parasites (i.e. Criconematidae), predators (i.e. Actinolaimidae) and omnivores (i.e. Dorylaimidae). Our results indicate that both habitat and alkalinity gradients impact the nematode community diversity and composition. Future investigation will determine the contribution of specific factors (i.e., abiotic and biotic) in driving the assemblages of these distinct nematode communities.

S-15
DOES SUPPLEMENTING FUMIGATION WITH NON-FUMIGANT NEMATICIDES IMPROVE NEMATODE MANAGEMENT IN POTATOES? Grubau, Zane J.1 and C. Liu1.1Entomology and Nematology Dept., University of Florida, Gainesville, FL 32611.

Nematicide application is the primary option for controlling nematodes in Florida potato production. Sting nematode (Belonolaimus longicaudatus) and stubby-root nematodes (楠idorus spp. and Trichodorus spp.) are the most predominant nematode pests in Northeast Florida, where Florida potato production is most prevalent. Preplant fumigation is commonly used, but many growers also use at-plant and post-plant non-fumigant nematicides, in part for stubby-root nematode management, since that pest is typically not managed well by fumigation. Older chemistries, particularly oxamyl, are commonly used, but new products such as Majestene (formulation of dead Burkholderia bacteria and fermentation media) are also available. Demand for new, non-fumigant nematicides is driven in part by interest in minimizing non-target effects, such as potential effects on free-living nematodes. The objectives of this research were to (1) determine if supplementing fumigation with at-plant and post-plant non-fumigant nematicide application improves management of stubby-root nematodes and sting nematode and (2) determine the effect of these nematicides on free-living nematodes. To investigate this, a replicated (n=6) field trial was conducted in Hastings, FL. Treatments were (1) untreated control; (2) 1,3-dichloropropene (1,3-D) preplant fumigation; and 1,3-D preplant fumigation in conjunction with non-fumigant treatments including (3) oxamyl at planting; (4) oxamyl and Majestene at planting, (5) oxamyl and Majestene at planting plus Majestene at 22 days after planting, and (6) Majestene at planting and 22 days after planting. Any of the nematicide treatments significantly (P<0.05, Fisher’s protected LSD) reduced sting or stubby-root nematode soil abundances compared with untreated control, but there were no differences among combinations of fumigant and non-fumigant products. Similarly, any nematicide increased marketable yield compared with untreated control. None of the nematicides significantly affected free-living nematode abundances. In summary, fumigation with 1,3-D was effective for nematode management, and applying at-plant or post-plant non-fumigant nematicides in addition to 1,3-D fumigation did not provide any additional benefit.

A-16
ADVANCED CONTROL OF CODLING MOTH (CYDIA POMONELLA) USING THE BENEFICIAL NEMATODE STEINERNEMA CARPOCAPSAE IN APPLE AND PEAR ORCHARDS IN THE PACIFIC NORTHWEST: Haines, Stephanie, Diana K. Londoño, Tye Shauck, and Shaun D. Berry. BASF Corporation, Agricultural Solutions, Research Triangle Park, NC.

The most significant insect pest in apple and pear trees in the Pacific Northwest (PNW), USA is the codling moth (Cydia pomonella). In the PNW, codling moths generally produce 2-3 generations per year, but have been observed to produce up to 5 generations in some hot spots. During early larval stages, codling moth larvae burrow into apples and feed on the fruit, resulting in lost crops and lost income for growers. Growers have limited options for controlling this pest at the larval stage. This challenge is compounded for growers maintaining an organic orchard. Few biological control options are available, and the ones currently on the market, (e.g. viruses, plant extracts, and pheromones) tend to target 1st instar larvae and adults. A promising means of decreasing codling moth populations is entomopathogenic nematodes (EPNs), specifically the species Steinernema carpocapsae (Sc), which was originally isolated from codling moth larvae. These beneficial nematodes can fill in the gap left by other biocontrol agents, by attacking the mature and overwintering larvae in the soil and the base of tree trunks. A few years ago, successful trials were conducted in apple orchards in Germany and the United Kingdom, where beneficial nematodes reduced fruit damage by nearly 75%. In 2019, field trials in the PNW during spring, summer, and fall resulted in codling moth larval mortality of 89%, 74%, and 90% respectively. Results demonstrated that Sc nematodes are a powerful tool for apple growers to incorporate into an integrated pest management system.
A FUNCTIONAL ANALYSIS OF HORIZONTALLY TRANSFERRED CELLULASE IN THE NEMATODE PRISTIONCHUS PACIFICUS. **Han, Ziduan (Paul)**1 and **R.J. Sommer**, 1Max Planck Institute for Developmental Biology, Department for Integrative Evolutionary Biology, Max-Planck-Ring 9, Tübingen, Germany 72076.

The gain of genetic information through a non-parental relationship is termed horizontal gene transfer (HGT). While this phenomenon has been demonstrated as an important evolutionary force in prokaryotes, HGT is not broadly recognized in eukaryotes due to its rare detection. However, one exception is in the phylum Nematoda, where multiple HGT events have occurred. Groups of plant-parasitic nematodes are well known for their acquisitions of genes from bacteria which are utilized to invade plants and digest plant materials. Pristionchus nematodes, although omnivorous free-living bacterial-feeders, have also acquired cellulase genes horizontally. Eight genes containing a cellulase domain have been detected in the *P. pacificus* genome that strongly suggests they are under positive selection. RNA-seq data revealed these genes are expressed at all life stages. In addition, a transcriptional reporter driven by the promoter of the major component cellulase-2 shows expression in the pharyngeal gland cells. Strikingly, upon starvation an additional robust secretory system expression is also induced. These cellulases are likely secreted to the environment where robust enzymatic activity can be detected. To further investigate the ecological meaning of cellulase in *P. pacificus*, we generated a cellulase null mutant by knocking out all eight cellulase genes using CRISPR/Cas9. The *P. pacificus* cellulase mutant behaved similarly compared to the wild type when grown on the standard food source *E. coli* OP50. However, when grown on bacteria capable of producing biofilm, a protective structure containing cellulose, the *P. pacificus* cellulase mutant developed significantly slower and had a potentially smaller brood size. Thus, our study demonstrates that the horizontally acquired cellulase can increase the fitness of *Pristionchus* and provided new insight into this evolutionary phenomenon in eukaryotes.

ALTERNATIVE MANAGEMENT PRACTICES FOR THE NORTHERN ROOT-KNOT NEMATODE (MELOIDOGYNE HAPLA) IN DAYLILY (HEMEROCALLIS SPP.) PRODUCTION. **Howland, Amanda,**1 E. Cole1, K. Poley2, and M. Quintanilla1. 1Michigan State University, Department of Entomology, East Lansing, MI. 2Michigan Corn Research Manager, Lansing, MI.

In the United States, the floriculture industry was valued at $4.77 billion in 2018. Daylily (*Hemerocallis* spp.) production is a major component of the floriculture industry, but one of its main pathogens is plant-parasitic nematodes. In Michigan, the northern root-knot nematode, *Meloidogyne hapla*, can cause over 20% yield loss in daylily production and reduce marketability due to their presence on the roots. Previous, ongoing field trials testing different management strategies showed four treatments with promising control of *M. hapla* that also increased plant growth. Therefore, a three-year field trial was established in 2019 in Zeeland, MI with the objective to test these top treatments to determine which would work best for managing *M. hapla* in daylily production fields. The treatments included in the trial are Indemnify Root Dip (Dip) + TerraClean 5.0, Dip + Indemnify, Dip + AzaGuard, Dip + Compost, and an untreated control. In the field, the five treatments were arranged in a randomized block design with fivereplications. Daylilies were dipped in Indemnify and immediately planted; the additional treatments were then applied to each respective plot. To monitor *M. hapla* population levels, soil samples were taken yearly at treatment (spring), mid-season, and end-season. Mid-season plant height data and root samples were also taken; root samples were stained with acid fuchsin to determine presence of *M. hapla* inside the roots. Preliminary results from the first two years showed the Dip + AzaGuard treatment has the best *M. hapla* control, and the Dip + Indemnify treatment has the highest plant growth. The field trial will end in the fall of 2021. In addition to final nematode analysis and plant measurements, a cost benefit analysis will be performed to provide daylily producers the economically best management strategy to reduce root-knot nematode populations in their field.

MORPHOLOGICAL AND MOLECULAR CHARACTERIZATION OF CACTODERA MILLERI GRANEY & BIRD, 1990 FROM COLORADO AND MINNESOTA. **Kantor Mihail**1, Z. A. Handoo 1, A. M. Skantar1, M. N. Hult1, S. L. Hafez 2, K. Kromroy1, K. Sigurdson1 and M. Grabowski1. 1Myology and Nematology Genetic Diversity and Biology Laboratory, USDA, ARS, Northeast Area, Beltsville, MD 20705, USA; 2University of Idaho, Parma, ID 83600, USA; 3Minnesota Department of Agriculture, Plant Protection Division, Saint Paul, MN 55155, US.

The genus *Cactodera,* has fifteen known species. *Cactodera milleri* was originally described infecting the roots of common lambsquarter (*Chenopodium album*) in Michigan. *Chenopodium quinoa* is a known host for *C. milleri* since 1990 when Graney and Bird included it in the host range study of this species. In 2019, *C. milleri* cysts were recovered from soil samples collected from a *Chenopodium quinoa* field, located in Mosca, Alamosa, county, Colorado, USA. A moderate number of lemon shaped cysts and juveniles were recovered from the affected quinoa plants. Same species was also recovered and identified from samples submitted over the years by the Minnesota Department of Agriculture as part of the Animal and Plant Health Inspection Service (APHIS) efforts to survey states for the presence of *Pale Potato Cyst Nematode*. The cysts and juveniles (J2) were recovered from soil samples through sieving and Baermann funnel extraction. The nematode species was identified by both morphological and molecular means as *Cactodera milleri*. To our knowledge this represents the first report of *Cactodera milleri* from Colorado and Minnesota.

DYNAMICS OF ENTOMOPATHOGENIC NEMATODE FORAGING AND INFECTIVITY IN MICROGRAVITY. **Kaplan, Fatma**1, **D. Shapiro-Ilan**2, and **K. C. Schiller**1. 1Pheronym, Inc., Davis, CA, 95618, 2US Department of Agriculture, Agricultural Research Service, Byron, GA, 31008.

Microgravity is a unique environment to elucidate host–parasite biology. Entomopathogenic nematodes (EPNs), model parasites, kill host insects with mutualistic bacteria and provide environmentally friendly pest control. It is unknown how microgravity affects a multistep
insect invasion by parasites with mutalistic bacteria. EPNs respond directionally to electromagnetic cues and their sinusoidal locomotion is affected by various physical factors. Therefore, we expected microgravity to impact EPN functionality. Microgravity experiments during space flight on the International Space Station (ISS) indicated that EPNs successfully emerged from consumed insect host cadavers, moved through soil, found and infected bait insects in a manner equivalent to Earth controls. However, nematodes that developed entirely in space, from the egg stage, died upon return to Earth, unlike controls in microgravity and on Earth. This agricultural biocontrol experiment in space gives insight to long-term space flight for symbiotic organisms, parasite biology, and the potential for sustainable crop protection in space.

S-21
SCREENING OF SUMMER COVER CROPS AGAINST MELOIDOGYNE ENTEROLOBII. Khanal, Churamani. Department of Plant and Environmental Sciences, Clemson University, Clemson, SC 29634.

Greenhouse studies were conducted to evaluate ten commercially available summer cover crops against the guava root-knot nematode (Meloidogyne enterolobii). The experiments were established on 10-cm diameter plastic pots as a randomized block design with five replications. Each pot filled with sterilized soil received five seeds per pot and were inoculated with 10,000 eggs of M. enterolobii a week after planting. The number of eggs each crop supported at the end of the experiments (45 days after inoculation) varied greatly and ranged from 10 to 106,513. Sunflower and cowpea were good hosts for M. enterolobii and supported up to 158% more eggs per plant compared to that of the control (tomato cv. Rutgers). Although not significantly different, sesame and buckwheat suppressed egg reproduction by, respectively, 65% and 75% to that of the control. Sunn hemp, grain sorghum, sorghum-sudangrass, and millets (brown top-, pearl-, and Japanese-millet) were poor hosts and significantly suppressed the nematode egg reproduction, the suppression being 96% to 99% in relation to the control. While grain sorghum, sorghum-sudangrass, and the three millets supported minimal nematode reproduction, any juveniles and males were not found in the soil. Plant dry weights were also measured at the end of the experiments and the biomass of each crop was greater than that of the control. Grain sorghum, sorghum-sudangrass, cowpea, pearl millet, sunflower, buckwheat, and sesame had significantly higher plant biomass than the control.

A-22
ASSESSMENT OF COMBINED EFFECT OF SECONDARY METABOLITES PRODUCED BY THE ENTOMOPATHOGENIC BACTERIUM PHOTORHABDUS L. SONORENSIS (ENTEROBACTERIACEAE) ON THE ROOT KNOT NEMATODE, MELOIDOGYNE INCognita. Kusakabe, Ayako1 and Stock, S. Patricia1,2. 1Department of Entomology, University of Arizona, Tucson, Arizona, 85721, 2School of Animal and Comparative Biomedical Sciences, University of Arizona, Tucson, Arizona, 85721.

The combined effect of Photorhabdus l. sonorensis-derived SMs was tested against the root-knot nematode, Meloidogyne incognita. The effects of these SM on the host plant were also measured. Three SMs, two phenylpropanoids (AK1, AK2) and one alkaloid (AK3), previously isolated by our team and with shown nematicidal activity were considered. Results from these assays showed activities that ranged from antagonistic, additive, to synergistic. The LC50 of two SM combinations tested, AK1+AK2 (119 µg/mL) and AK1+AK2+AK3 (19.9 µg/mL), were significantly lower than that observed for each single compound. Both SM mixtures had a synergistic effect against M. incognita infective juveniles with a calculated index of 0.17 and 0.24, respectively. The AK2+AK3 mixture had an additive effect (index = −0.03); whereas AK1 + AK3 showed an antagonistic effect (index = −0.31). Cow pea seedlings treated with AK1 + AK2 showed a reduction in the number of galls and the number of egg masses after six weeks post M. incognita inoculation. These results were dependent on the concentrations tested. Root infection was suppressed with increasing concentration of AK1+AK2 however, number of egg masses/gall was not affected. Growth parameters (total plant dry weight, root and stem length, leaf mass/leaf area, and root to shoot ratio), chlorosis, and leaf chlorophyll content (chlorophyll a, chlorophyll b, the ratio of chlorophyll a and b, and total chlorophyll) were not affected by all tested combinations between the AK1 + AK2 concentrations tested. Additionally, no phytotoxic effects were observed on the cowpea seedlings.

S-23
GREENHOUSE EFFICACY TESTS OF CONTACT NEMATICIDES WITH COTTON GROWN IN SOIL INFESTED WITH LESION NEMATODES (PRATYLENCHUS BRACHYURUS). Loffredo, Angelo and J.O. Becker. Dept. Nematology, University of California, Riverside, CA 92521.

The root-lesion nematode P. brachyurus is a migratory endoparasite with a broad host range and occurs widely in the world’s warm climates. It causes necrosis in the root cortex and predisposes cotton to secondary microbial infections. In recent years, several new contact nematicides have been developed with excellent efficacy against root-knot nematodes. However, efficacy against other plant-parasitic nematodes, including Pratylenchus spp., has not been widely reported. Two greenhouse experiments were performed to evaluate the nematicidal efficacy and crop safety of three products on upland cotton (Gossypium hirsutum cv. GC510) grown in a pasteurized sand with about 2% organic matter. The soil was infested with greenhouse culture raised Meloidogyne incognita and supported up to 158% more eggs per plant compared to that of the control. Sunn hemp, grain sorghum, sorghum-sudangrass, and millets (brown top-, pearl-, and Japanese-millet) were poor hosts and significantly suppressed the nematode egg reproduction, the suppression being 96% to 99% in relation to the control. While grain sorghum, sorghum-sudangrass, and the three millets supported minimal nematode reproduction, any juveniles and males were not found in the soil. Plant dry weights were also measured at the end of the experiments and the biomass of each crop was greater than that of the control. Grain sorghum, sorghum-sudangrass, cowpea, pearl millet, sunflower, buckwheat, and sesame had significantly higher plant biomass than the control.
strongly reduced the lesion nematode population. After one week, the number of P. brachyurus recovered from the soil samples was about 3 to 13% of the non-treated control. Two weeks later, the soil populations in the nematicide treatments increased to about 7 to 27% of the control. Nimitz was the most effective nematicide and reduced the number of nematodes recovered from the roots to less than 10% of the control. A second trial was performed with the same setup but with a three-week-old cotton seedling transplanted into each pot just before treatment application. At trial termination after 60 days, the nematicides Nimitz, Vydate, Velum One, and Salibro reduced the total extracted nematode population (soil and roots) to about 12, 17, 33, and 39%, respectively of the non-treated control. The nematicides had no apparent influence on plant growth.

A-24
MORE INSIGHT TO THE GENOME OF THE POTATO AND TOBACCO CYST NEMATODES GLOBODERA SPECIES. Madani, Mehrdad1, Andy Vierstraete6, Solke H. De Boer2, and Maurice Moens.3 1 Former researcher at Canadian Food Inspection Agency, 93 Mount Edward Road, Charlottetown Laboratory, Charlottetown, PE, Canada. 2 Biology Department, Gent University, K.L. Ledeganckstraat, 35 9000. Gent Belgium. 3 Research Institute for Agriculture, Fisheries and Food (ILVO), 9280 Merelbeke, Belgium and Department of Plants and Crops, Ghent University, Coupure Links 653, Ghent Belgium.

Ribosomal DNA has been shown usefulness for diagnostics, polymorphisms analysis, and for evolutionary research in many species of plant-parasitic nematodes. This study presenting data on the structure and organization a rarely studied part of the rDNA cluster genes, i.e. intergenic spacer (IGS) in the genome of the Potato and Tobacco Cyst Nematode (Globodera pallida and G. rostochiensis, PCN, G. tabacum, TCN). PCNs have been estimated to cause losses of 9% of total potato production worldwide. The presence of a 5 S gene, repeats and sub-repeat elements with interspecies sequence divergence and heterogeneity, and various lengths within IGS, has been reported from different eukaryotic organisms. Also, diagnostic PCR using IGS derived primers have been developed for Root Knot Nematodes of the Meloidogyne and Pinewood nematodes of the Bursaphelenchus. In this study, the intergenic spacer of the rDNA from these species was amplified, sequenced, and analyzed. Genomic DNA was prepared using the lysis buffer method from a single cyst from populations collected in Europe, the USA, and Canada. Several putative primers were designed based on the sequence data of the lower and upper streams of each 28 S, and 18 S genes of these species, and consequently internal primer to PCR amplify the whole IGS for each studied species. Prior to sequencing cloning was performed using PGEM-T easy vector and transformed to the Escherichia coli strain JM109. To analyses retraction pattern of each PCR product, four restriction enzymes, HinfI, SacII, HincII, and Drai, were separately used in Restriction Fragment Length Polymorphism (RFLP) test.

Gel electrophoresis of the PCR reaction, showed the PCR amplicon consisted of a single 2.8 Kb fragment for G. tabacum, while for G. pallida it consisted of a 2.7 kb fragment accompanied by a faint band of 2.6 kb. G. rostochiensis amplification resulted in two bands, sized 2.7 and 1.7 Kb. Sequence analyses of the IGS-PCR products using ‘Tandem Repeats Finder’ revealed the presence of long and short repetitive sequences as well as sub-repeats with different sizes. An approximately 400 bp long region without any internal repetitive elements, were identified in a position between the two repetitive regions suggesting that there is a 5 S gene in the IGS of these species. To the best of our knowledge this is the first research on the IGS region of these species.

S-25
EFFECTS OF A TURMERIC (Curcuma longa) SOLUTION ON THIRD-STAGE JUVENILES OF THE RAT LUNGWORM (Angiostrongylus cantonensis). Marahatta, Sharadchandra P. College of Agriculture, Forestry and Natural Resource Management, University of Hawai‘i at Hilo, Hilo, HI 96720, USA.

Rat lungworm, Angiostrongylus cantonensis, (RLW) is a challenging zoonotic, parasitic nematode of the tropics. Curcumin, a bioactive substance in turmeric (Curcuma longa), has been used as a supplement to treat the RLW infected patients diagnosed with eosinophilic meningitis in Hawai‘i. Semi-slugs (Parmarion martensi) are often infected with RLW in the east Hawai‘i region and are commonly found in vegetable fields, lawns, compost making areas, and greenhouses. RLW infected semi-slugs are considered as threats to Hawaiian agriculture and to human health. Research was carried out to: (i) determine the percent of semi-slugs infected with RLW in Hilo, Hawai‘i, and (ii) evaluate the effects of a turmeric suspension on third-stage juveniles of the RLWs. Semi-slugs were collected from the University of Hawai‘i at Hilo (UH-Hilo) compound and immersed in 50 ml tap water in 50 ml Falcon tubes. After 72 hours of incubation at room temperature, RLWs were isolated and observed under an inverted microscope. RLWs were placed in petri dishes and incubated in 20 drops of 0%, 0.5%, 1.0% or 2.0% turmeric solution or 66.66% isopropyl alcohol. All treatments were replicated 3 times. Treated RLWs were observed and their body shapes were compared with heat killed RLWs at 22 minutes after initiation of the test. RLWs were found in 83.33% of the semi-slugs surveyed (n = 12). On average, each semi-slug was loaded with 61 ± 28 (mean ± SEM) RLWs. Within 22 minutes of incubation, at least 50% of the RLWs treated with 1% turmeric and 66.66% isopropyl alcohol showed partial semi-coiled position, whereas 50% or more RLWs treated with 0%, 0.5%, 2% turmeric were in coiled position. None of the treatments resulted in RLWs turned into C-shaped. In contrast to this, all heat-killed RLWs were in a C-shaped position. Thus, turmeric solution tested or isopropyl alcohol incubated for 22 minutes did not appear to be a viable method to kill RLW. More research is needed to further explore the potential of turmeric solutions with higher concentrations or longer incubation time against RLW.

A-26
THE ASSEMBLY OF MICROBIAL GUT COMMUNITIES OF NEMATODES FROM TAYLOR VALLEY STREAMS IN ANTARCTICA. McQueen, J. Parr1, K. Gattoni1, E. Gendron1, P. Sommers1, J. Darling1, S. Schmidt1, and D. L. Porazinska1. 1 Dept. of Entomology and Nematology, University of Florida, FL 32611 2University of Colorado-Boulder, Boulder, CO 80303, USA.

The knowledge of nematode gut microbial communities holds the potential to better understand basic nematode ecology, including a further refinement of nematode feeding habits. Recent work examining the gut microbiome of the bacterivorous C. elegans has indicated the presence of a species-specific gut microbiome that is conserved among individuals collected from a wide range of
habitats and is distinct from the surrounding environment. However, the knowledge of gut microbiomes of other nematode species and how these microbial communities assemble is extremely limited. Microbial mats found within streams of the Dry Valleys in Antarctica provide a relatively simple and tractable natural ecosystem with only two morphologically distinct nematode species representing two distinct feeding habits (i.e., bacterivorous *Plectus murrayi* and omnivorous *Eudorylaimus antarcticus*). The main objectives of this study were to: 1. characterize nematode gut microbiomes, and 2. examine potential factors that could drive the assembly of these communities.

In January 2019, replicate samples of two types of cyanobacterial mats (i.e., Black – dominated by *Nostoc* and Orange – characterized by *Phormidium*) were collected from four streams across a geographical and biogeochemical gradient (i.e., Canada Stream, Bowles Creek, Delta Stream, and Von Guerard) in Taylor Valley, Antarctica. Samples of mats as well as individual specimens of hand-picked nematodes were processed for 16S metabarcoding to characterize bacterial communities. We hypothesized that: 1. nematode species would be characterized by species-specific gut microbiomes, and 2. gut microbiomes would be explained more by nematode species and/or feeding habits than their biogeography and environmental characteristics. To test these hypotheses, metrics of alpha diversity (e.g., OTU richness, Shannon diversity, and Simpson’s diversity) and metrics of community composition (e.g., Bray-Curtis) were compared across the microbial communities examined and tested using ANOVA and PERMANOVA, respectively.

Sequencing data from all 24 mat replicates confirmed that both mat types were dominated by cyanobacteria (contributing 45% and 48% of bacterial communities within Black and Orange mats, respectively) and that the composition of cyanobacterial genera differed among the two mat types. Preliminary data from 12 individuals extra cited from a single mat supported our hypotheses by indicating smaller cyanobacterial contribution (<25%) to bacterial communities within nematode guts than the mat they were sampled from, as well as differences in relative abundance of specific bacterial taxa (e.g., Firmicutes, Proteobacteria) between *P. murrayi* and *E. antarcticus*. Other bacterial taxa (e.g., Verrucomicrobia, Planctomycetes) show enrichment in nematodes compared to the mat, indicating the possibility of these being selected for within the gut by nematode ecology or feeding habits. The in-progress analysis of an additional ~500 nematode individuals will elucidate specific details of how these microbiomes vary between nematode species and what specific factors play a role in their assemblies as well as allowing for a more robust statistical approach.

S-27

**FUNGAL FEEDING PREFERENCES OF TWO Aphelenchoides spp. FROM FLORIDA STRAWBERRY FIELDS, Aphelenchoides besseyi, A FACULTATIVE PLANT-PARASITIC NEMATODE, AND, A. pseudogoodeyi, AN OBLIGATE FUNGAL-FEEDING NEMATODE. Oliveira, Clemen1, J. Desaeger2, J. A. Brito3, N. Peres4, T. Seijo5, R. Inserro6**

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*Aphelenchoides besseyi* and *A. pseudogoodeyi* are two foliar nematodes associated with strawberry in Florida. In 2016, *A. besseyi* was found on several strawberry farms in Florida, causing severe stunting and considerable damage. Recent surveys in Florida strawberry fields have revealed the presence of many other *Aphelenchoides* spp. One of those is a newly discovered species, *A. pseudogoodeyi*, which appears to be a strictly fungal feeding nematode (Oliveira et al., 2019). It is hypothesized that both nematode species could feed and reproduced on fungi present in the soil, which could serve as a reservoir of theses nematodes and contribute for an increase of their populations at the end of the crop season. Feeding and reproductive abilities of these two nematodes were assessed on Florida isolates of four fungi pathogenic to strawberry; *Colletotrichum gloeosporioides* *Macrophomina phaseolina*, *Pestalotiopsis clavispora*, and *Botrytis cinerea*, one non-pathogenic; *Fusarium oxysporum* and one fungus pathogenic to peach; *Monilinia fructicola*. *Aphelenchoides besseyi*’s reproduction were significantly higher (p-value<2e-16) on strawberry pathogenic isolates of *B. cinerea*, *C. gloeosporioides*, and *P. clavispora* than on the non-pathogenic isolates of *F. oxysporum*, and *M. fructicola*. On the contrary, *A. pseudogoodeyi*’s reproductive rates did not differ across the fungus cultures. *Macrophomina phaseolina* was an exception because it did not produce mycelium in the medium used being a poor host for both nematodes. Furthermore, initial stages (Egg and Juveniles) of *A. pseudogoodeyi* were significantly higher than adults. On the other hand, *A. besseyi* dwelled in adult stages (Females and males). Our findings indicate that *A. besseyi*, which adults if more often found, is more selective in its feeding preference on fungi than *A. pseudogoodeyi*. Fungi play an important role in maintaining the populations of the two nematodes in the soil. Furthermore, the strawberry pathogenic fungi on residues of strawberry plants perpetuate *A. besseyi* infestation in Florida strawberry fields. The removal of strawberry plants residues infected by fungi is a desirable and effective management practice to suppress *A. besseyi* in commercial strawberry field in Central Florida.

S-28

**INTEGRATED MANAGEMENT OF POTATO EARLY DIE: DISEASE COMPLEX DYNAMIC AND TREATMENT EFFECTIVENESS. Parrado, Luisa M.**

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In 2018 and 2020 trials were conducted both in the lab and in the field to control *Pratylenchus penetrans*, a plant-parasitic nematode, and *Verticillium dahliae*, a fungal plant pathogen. These two organisms can interact and together cause the Potato Early Die complex (PED) which, when present, can reduce potato quality and yield by 30-50%. Our 2018 field and lab trials indicated that chicken manure and Layer Ash Blend (LAB), which is composed of chicken manure + cattle manure + wood ash, are most effective in controlling *P. penetrans* and producing highest yields. As for the biocontrol products commercially available, in 2018, MeloCon (*Paecilomyces lilacinum*) did not control *P. penetrans* populations under a lab and field settings. However, literature suggests that when integrated with organic soil amendments or another nematode antagonist like *Bacillus* spp., nematicide effect is maximized. In 2020, field trials were conducted at Three Rivers, MI on a commercial farm. We evaluated the effectiveness of biocontrol agents (*P. lilacinum* and *B. amylophiliquefaciens*) and organic soil amendments (chicken manure, LAB and a high carbon compost), alone and combined, on managing the PED complex. One month after the treatments were applied, *V. dahliae* propagule density reduction in soil was of 66%
with chicken manure + MeloCon. Verticillium dahliae infection in potato stems significantly increased from July 13 to August 11 (p < 0.001), where the greatest infection was under the Vydate treatment (70%). Although there were no significant differences between treatments, P. penetrans root infection was the lowest in potato plants treated with chicken manure + Vydate (p = 0.0237). Regarding disease severity, it was highest in the untreated plants (65.112) and the most effective treatment overtime was LAB + MeloCon (% control = 63.6%). Lastly, tuber yield was the highest under LAB treatment (403 CWT/ac) and the lowest vascular discoloration presence was on tubers from LAB + MeloCon plots (11.6%). Thus far, our results allow us to conclude that in combination with chicken manure or LAB, P. lilacinum is effective on managing PED.

A-29 MANAGEMENT OF PLANT-PARASITIC NEMATODES AND SOIL HEALTH USING SORGHUM/SORGHUM-SUDANGRASS HYBRIDS AS A COVER CROP. Paudel, Roshan, K. -H Wang, and P. Waisen. Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822.

Sorghum and sorghum-sudangrass hybrids (SSgH) are used as cover crops to increase soil organic matter and suppress root-knot nematodes in various cropping systems. A wide range of SSgH varieties that include forage sorghum, energy sorghum, sudangrass, and sorghum-sudangrass hybrids were examined to identify the most efficient varieties in tropical climate for 1) plant-parasitic nematode suppression, 2) soil water conservation, and 3) soil microbiome enhancement. It is hypothesized that different age of SSgH will have different allelopathic performance. Greenhouse tube bioassays were conducted to compare the amendments of 10 SSgH varieties to sunn hemp and an unamended control against Meloidogyne incognita infection on mustard green 'Hirayama' (Brassica juncea) using sterile sand: soil mix. Soil was amended with chopped tissues at 1% (w/w, dry weight basis) collected from 1- and 2-month old plant tissues grown in open field. Each tube was inoculated with 220 second-stage juveniles (J2) of M. incognita on the same day. Results showed that the one-month-old tissue in Trial I was most suppressive to M. incognita female formation by ‘NX2’, ‘LA’ and ‘CV’, but only ‘NX2’ and ‘LA’ remained suppressive along with ‘512’ to root-knot when using the 2-month old tissues. Bioassay results from Trial II identified ‘LA’ and ‘CV’ to be highly suppressive to root-knot females, followed by ‘NX2’, ‘512’, and ‘MII’, all of which were significantly more suppressive than the unamended control (P < 0.05). Using the greenhouse results, a field trial was initiated at Poaohomo Experiment Station on May 28, 2020 to compare 7 SSgH varieties to a fallow control in 3.6 × 1.2 m² plots in a randomized complete block design with four replications. Cover crop was terminated at 2.5 months after planting using a flail mower, and eggplant seedlings were transplanted 2 weeks following the termination of SSgH cover crops. 87.2% energy sorghum variety NX2/NX-5D61 had the highest plant biomass, soil organic carbon, soil moisture, soil microbial respiration (using Solvita CO₂-burst reader), and microbial biomass (using Total Phospholipid Fatty Acids, TPLFA). Canonical correspondence analysis (CCA) showed higher SSgH biomass led to higher microbial biomass, nematode enrichment index, microbial respiration, and gravimetric and volumetric soil moisture. Soil carbon was positively related to nematode structure index, volumetric soil moisture, abundance of omnivorous nematodes, all of which are indicative of a less disturbed soil food web. The plant-parasitic nematodes were positively related to Gram-positive/Gram-negative bacteria which indicated their association with less stress conditions, but negatively related to SSgH biomass, nutrient enrichment, or structured soil food web, as well as total microbial biomass. Both greenhouse and field experiments suggested that NX2 was the most effective SSgH cover crop variety for root-knot nematode suppression, and soil health improvement in Hawaii.

S-30 POPULATION DYNAMICS OF BURSAPHELENCHUS XYLOPHILUS ASSOCIATED WITH PINE FOREST DECLINE. Pires, David1, J. Campão2, J. Branco1, I. Miranda1, T. Calvão2, M. Mota4, and C. Pimentel3. 1Mediterranean Institute for Agriculture, Environment and Development (MED), University of Évora, Portugal; 2Center for Environmental and Sustainability Research (CENSE), Faculty of Science and Technology, New University of Lisbon, Portugal; 3Forest Research Centre (CEF), School of Agriculture, University of Lisbon, Portugal.

The pinewood nematode (PWN), Bursaphelenchus xylophilus, is a quarantine organism in the European Union and the causal agent of pine wilt disease (PWD), a serious threat to pine forests worldwide, leading to rapid decline and death. In Europe, this invasive pest was first reported in Portugal in 1999, on Pinus pinaster. Due to its economic importance and worldwide distribution, an enormous effort has been devoted to research on B. xylophilus and PWD. However, relating the presence and abundance of this pest with actual forest decline and mortality is not straightforward. In the present work, we assessed the interaction between B. xylophilus populations and pine decline at the tree and landscape level. To test this, we determined PWN population densities from different sections of healthy and declining P. pinaster – considered very susceptible to PWN – and P. pinea – considered resistant, in two consecutive years. The studied site was Herdade da Apostiça (Sesimbra, Portugal), a 4 thousand ha forest that exhibits areas of moderate to severe decline. An approximate 500 m long transect was set in four different areas of the experimental site, along which healthy and declining P. pinaster trees were randomly selected. Wood samples from lower (DBH), middle (M) and upper (T) sections of trees were retrieved whenever possible, and kept in individual plastic bags to avoid cross contamination. Twigs from the canopy were also sampled. When present, samples from P. pinea were collected as well. Nematodes were extracted from wood material using a modified Baermann tray method, and counted under a stereoscopic microscope. With only few exceptions, M and T sections consistently displayed the lowest PWN densities, although samples collected at breast height (DBH) also had relatively low numbers of B. xylophilus. On the other hand, twigs had the highest numbers of nematodes in all areas. Surprisingly, nematodes were extracted from the canopy of apparently healthy P. pinaster trees, including the PWN, although in low densities. Expectedly, areas with no visible tree decline had low population densities of B. xylophilus, and declining trees tended to present higher numbers. P. pinea samples were mostly free of the PWN. To further understand the complex dynamics shaping pine forest decline caused by the PWN, more sampling will be carried out in the upcoming months. This will allow us to build predictive models on the spread and damage of PWD.
PRIMARY SUCCESION OF NEMATODE COMMUNITIES FOLLOWING RETREAT OF A HIGH ELEVATION GLACIER.  
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Although many studies have examined the succession of bacterial communities following glacial retreat, how nematode communities assemble within forefields of receding glaciers is still largely unstudied. The deglaciated forefield of Puca Glacier in the Peruvian Andes have been previously characterized as dominated by bacteria with their diversity positively correlated with the age of soils and soil nutrients. As nematode communities feed on microbes, the successional patterns of nematode communities could be tied to the patterns of their primary food sources. Our main objectives were to examine: 1. the diversity and composition of the nematode community along the Puca Glacier chronosequence, and 2. the relationships between nematodes and biotic and abiotic soil factors.

On 23 March 2015, we collected soil samples at three spatial distances from the glacier representing 9-, 24-, and 89-year-old deglaciated soils. We characterized these soils for nematode and microbial communities (i.e., bacteria, fungi, and microbial eukaryotes) using standard 18S rDNA metabarcoding. We also determined soil biogeochemical properties (C, N, moisture, and pH) and rates of microbial enzymes (e.g., phos). Due to known bacterial dominance, high pH, and extremely low C and N content, we hypothesized that: 1. the succession of bacterial-feeding nematodes would precede the succession of other nematode trophic groups, and 2. the diversity of nematode communities would be explained by biotic (especially bacteria) and abiotic (especially C and N) soil factors. Significant differences in alpha diversity, the abundance of trophic groups, and abundance of taxa along the chronosequence were tested using ANOVA followed by Fisher’s LSD. Relationships with microbial diversity were tested using model II regression and with soil geochemical gradients with GDM.

In the 9-year-old soils, only three fungal-feeding and one bacterial-feeding “species” were observed. The nematode communities in the 24-year-old soils were slightly more complex predominantly for fungal-feeding taxa. In contrast, the 89-year-old soils contained all known trophic groups with multiple species representing each trophic group (Figure 1). Only two species were shared among the sites (Figure 2). The alpha diversity (Chao1) of nematode communities correlated strongly with alpha diversity of all microbial communities (Figure 3). Nematode community composition was highly explained by biogeochemistry (83.4% of community dissimilarity) (Figure 4) with microbial C as the strongest predictor.

This study shows the overall expected pattern of primary succession of nematode community along the chronosequence of a retreating glacier, but also indicates the presence of strong relationships with microbes and overall soil biochemistry. The unexpected early predominance of fungal-feeding nematodes suggests a significant fungal biomass must reside in these soils. Further analyses will provide more details on these unique communities.

METABOLIC RESPONSE OF ANTARCTIC NEMATODES TO CHANGING CLIMATE AND LENGTHENING SUMMERS.  
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Plectus murrayi, a hardy antarctic nematode that inhabits semi-aquatic and terrestrial biotopes in the McMurdo Dry Valleys of Antarctica (MCM), can survive extreme freezing conditions and desiccation and then return to full functioning capacity when rehydrated and warmed. This advanced stress response indicates an interesting relationship between temperature and metabolic activity in these antarctic nematodes. Previous studies have shown direct correlation between temperature and metabolic rate in various species. These studies have shown that most organisms undergo peak metabolic activity within a temperature range reflected by their natural habitat. By measuring heat flow per nematode of *P. murrayi* as a function of temperature, we found that *P. murrayi* nematodes consistently increase metabolic activity as a function of temperature until they reach peak metabolic activity at 40°C, a temperature they would never experience in their polar habitat. Due to the effects of lengthening warm seasons in Antarctica, and the increase of average soil temperatures in the MCM, we expect to see a steady increase in duration and magnitude of heightened metabolic activity of *P. murrayi* nematodes in the MCM. This will result in a proportional increase of CO2 production by *P. murrayi* as a result of metabolism. Soil populations of these nematodes, therefore, represent a potential carbon reservoir that will be steadily released as temperatures rise and summers lengthen.

SHIFTS IN SOIL MICROBIAL COMMUNITIES IN DOUBLE-CROPPING SOYBEAN: IMPLICATIONS ON THE MANAGEMENT OF THE SOYBEAN CYST NEMATODE (*Heterodera glycines* L.).  
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In double cropping (DC) systems, fields have two or more crops growing in sequence in a single growing season. For soybean, the crop is commonly planted following winter wheat. The soybean cyst nematode (SCN) (*Heterodera glycines*) is a major soybean pathogen. Several reports in the literature suggested potential suppressive effects of wheat residue on SCN population densities. A field trial was conducted in the 2017-2018 growing season to assess the effect of wheat on SCN populations in DC soybeans. In each field location (N=9), wheat (WT) was planted in strips alternating with strips maintained in fallow (FL) over winter. Soybean followed all strips after wheat harvest. Wheat strips had reduced SCN population densities compared to fallow strips, at soybean growth stage R1 and after soybean harvest. Three field locations with noted SCN suppression were selected for a metagenomics study. Ten subplots were selected (5 WT and 5 FL) from each location. A total of 90 soil samples were selected: 3 fields x 2 treatments x 3 timepoints x 5 replications. For this study, three DNA markers were used to target three distinct microbial groups: bacteria (16S V4-V5), fungi (ITS2) and *Fusarium spp.* (EF-1α). Amplicons were sequenced using an Illumina MiSeq platform (300+300bp paired-end). Sequencing data were processed in R using the DADA2 pipeline.
Statistical analysis was performed using the MicrobiomeAnalyst analyzer. Fungal communities were significantly different between DC and fallow plots at soybean planting and after harvest (P < 0.001). Fungal and bacterial taxa with increased abundance associated with wheat included groups previously reported to parasitize SCN cysts.

A-34 EVALUATING THE SPECIES-SPECIFIC PRIMER FOR PRATYLENCHUS PENETRANS, PPEN, IN WISCONSIN, USA. Saikai, Kananto, and A. E. MacGuidwin1. 1International Institute of Tropical Agriculture, Nairobi, Kenya. 2University of Wisconsin-Madison, WI 53706.

Pratylenchus species is one of the most prevalent genera on agricultural fields in Wisconsin. Among the species found in Wisconsin, P. penetrans is considered more damaging than others on many crops. Species identification for the genus is challenging and usually require parallel identifications by morphological and molecular characters, which can be both time and cost consuming. Species-specific primers have been adopted as a rapid and less expensive diagnostic method for many nematode pathogens although its performance needs to be tested when it is used for the first time in a new region given the possible intraspecific variations. Our objectives were to evaluate the published species-specific primer for P. penetrans, PPEN, for the species we have in Wisconsin and survey the prevalence of P. penetrans and other Pratylenchus species. PPEN was initially tested on 17 axenically cultured single female lines isolated from 12 Wisconsin counties, which have been morphologically and molecularly identified to species level previously. Thirteen of which were P. penetrans and the rest were one P. alleni, two P. fallax and one unknown species isolates. Positive bands of gel electrophoresis with PPEN were only observed for the P. penetrans isolates. PPEN was further evaluated on 120 female and 90 male individuals sampled on 79 fields (1 to 5 specimens/field) from 45 counties in Wisconsin. The majority were corn-soybean rotation field isolates, and the other fields were either planted with wheat, peas, long bean, kidney bean or fallow when they were sampled. These specimens were further identified to species by 28S ribosomal DNA and Cytochrome c oxidase subunit I of mitochondrial DNA. PPEN demonstrated 94.8% success rate in screening for P. penetrans. In total of ten cases of false positives and one case of false negative were observed. 84.4% of the total male specimens were identified to be P. penetrans in this study, suggesting that the presence of males in a soil sample can indicate the high likelihood of the P. penetrans infection in Wisconsin. Pratylenchus penetrans was found on 39 fields from 27 counties in Wisconsin, demonstrating the wide distribution in Wisconsin. Other amphimictic species we found were P. alleni (5 fields), P. fallax (1 field), and the unknown species (3 fields). Among the parthenogenetic species, P. neglectus was the most prevalent (23 fields) followed by P. crenatus (20 fields) and P. scribneri (5 fields). Thirteen fields were found infested by multiple species.

A-35 BRASSICA CARINATA A. BRAUN AS AN ALTERNATIVE WINTER CROP TO MANAGE ROTYLENCHUS RENIFORMIS WITHOUT AFFECTING FREE-LIVING NEMATODE POPULATIONS. Sandoval-Ruiz, Rebecca1, Grabau, Zane J.1, Seepaul, Ramdeo2, Wright, David L.1, and Small, Ian1. 1 Entomology and Nematology Department, University of Florida 2 North Florida Research and Education Center-Quincy, University of Florida.

Given the current outlook on global environmental concerns, restrictions on the use of nematicides have been increased (Chitwood 2003). This has boosted the search for new management alternatives that are safer for the environment and human beings. For this reason, in the present research Brassica carinata was evaluated as a winter rotation crop to an alternative to manage Rotylenchulus reniformis. This nematode is considered one of the most important pathogens in the Southeastern United States. In the same region, B. carinata is an emerging crop, that could contribute to decreasing CO2 emissions as it serves as an alternative source of jet fuel and other products (Cardone et al. 2002). Additionally, carinata has glucosinolates, substances known for affecting soil pathogens (Kruger, Fourie, and Malan 2013). The main objective of this research was to determine the effect of B. carinata, incorporated as a winter rotation crop with oats or fallow and alternating with a summer rotation system (corn, cotton, peanut, soybean) on reniform and free-living nematodes. Winter field systems evaluated were 2-year rotations of carinata-fallow, oats-carinata, and fallow-fallow. These winter rotations were crossed with a 4-year rotation of corn, cotton, peanut, and soybean with each phase present each year. Soil samples were taken from 0-30 cm and 30 -120 cm in four sampling dates after harvest (fall 2017, spring 2018, fall 2018, spring 2019). Nematodes were extracted, counted, and identified by genus. The absolute abundance per sample was measured. By depth, the highest population was seen in the shallow soil profile (first 30 cm of soil). Summer rotation and winter rotation system influence the nematode abundance. For winter rotation, when carinata was present in the system the reniform population was lower. Free-living populations did not decrease with the carinata crop. These results suggest carinata could be considered as a winter cover crop to decrease reniform populations without significantly altering free-living nematodes. More samples will be taken and analyzed over 4 years.

A-36 SINGLE-NEMATODE SEQUENCING: A NEW STEP TOWARDS UNDERSTANDING VIRULENCE IN SOYBEAN CYST NEMATODE. Ste-Croix, Dave T.1,2, A-F. Gendron St-Marseille1,3, E. Lord1, R.R. Bélanger1, J. Brodeur1 and B. Mimee1. 1Saint-Jean-sur-Richelieu Research and Development Centre, Agriculture and Agri-Food Canada, Saint-Jean-sur-Richelieu, Québec, Canada, 2Département de phytologie, Faculté des Sciences de l’Agriculture et de l’Alimentation, Université Laval, Québec, QC, Canada, G1V 0A6, 3Institut de recherche en biologie végétale (IRBV), Université de Montréal, Montréal, Québec, Canada.

The soybean cyst nematode (SCN - Heterodera glycines) remains one of the most damaging pests to the soybean industry in North America causing annually over 1.3B U.S. dollars in yield losses. While resistant cultivars, mainly derived from the parental line PI 88788 and, to a lesser extent, Peking, were quickly adopted as a means of managing this pest, the overuse of these limited genetic sources of resistance has led to a shift towards more virulent SCN populations. We are now seeing higher densities of virulent nematodes to both PI 88788- and Peking-type cultivars. Yet, the underlying genetic mechanisms responsible for this acquired virulence remain unknown. In this study, we applied a modified single-cell transcriptome sequencing approach in order to identify putative virulence genes involved in the breakdown of resistance against both major SCN resistance sources. For the first time, we amplified the full
transcriptome of individual SCN allowing for phenotype-specific contrasts to be carried out between virulent and avirulent individuals. We identified 48 differentially expressed putative effectors (secreted proteins involved in plant parasitism) as well as 40 effectors showing evidence of novel structural variants. Additionally, a stringent transcriptome-wide variant analysis identified 11 effectors containing phenotype-specific sequence polymorphisms. Our differential expression analysis also revealed the presence of co-expressed gene regions containing several putative effectors that could be under the control of a common promoter. Our findings seem to indicate that virulence against Peking requires a multifaceted strategy in which both gene regulation and sequence polymorphisms may play key roles in circumventing resistance. In contrast, the transcriptome of nematodes virulent against PI 88788 was only slightly modified suggesting a distinct mechanism. A follow-up experiment using long-read sequencing of single-nematode derived transcriptomes confirmed that structural variants within genes are much more prevalent in SCN than currently reported. Some of these alternative splice variants were also phenotype-specific suggesting that differential exon usage could be involved in virulence, a hypothesis that is currently being tested.

A-37
MANAGING SOYBEAN CYST NEMATODE THROUGH ROTATION OF THE SOYBEAN CYST NEMATODE RESISTANCE SOURCES (PEKING AND PI88788). *Thapa, Site*, E. Cole*, B. Levene*, and M. Quintanilla*. 1Michigan State University, Department of Entomology, East Lansing, MI. 2Bayer Crop Science, Dewitt, MI.

Soybean cyst nematode (*Heterodera glycines*) is one of the most economically important pathogens of soybean worldwide. In the United States, *H. glycines* is a major pest of soybean causing more yield loss than any other soybean disease causing $469 to $818 million soybean yield loss annually in the United States. The use of nonhost rotation and *H. glycines* resistant soybean varieties are considered primary ways of reducing losses caused by this nematode, but both have some disadvantages. *H. glycines* continues to be a major limiting factor in soybean production despite the availability of dozens of *H. glycines*-resistant soybean cultivars. This project has two main objectives: 1. To evaluate the rotation of two commercially available *H. glycines* resistant varieties (Peking and PI88788) compared to continuous use of a single line of resistance and susceptible soybean, and 2. To determine if the rotations followed shifted the Hg type of existing *H. glycines* populations. A four-year (2017-2020) field trial was recently completed near Saint Charles, Michigan. Each year *H. glycines* population density at planting and harvest as well as soybean yield data was taken. In the first year of the trial, *H. glycines* populations and yields were similar across all treatments. However, in the second year, yield differences were apparent. Rotating resistance from PI88788 to Peking (2017/2018) resulted in the highest yields, while plots with the continuous use of PI 88788 or Peking had yields that were 5% and 8% lower, respectively. Two successive seasons of PI88788 or Peking, however, had slightly greater *H. glycines* levels than when the rotation was rotated. Regardless of the type of *H. glycines* resistant soybean grown in 2017, all of the 2018 plots planted with *H. glycines* susceptible soybeans had the highest levels of *H. glycines* cysts, eggs, and juveniles at harvest with 2-3 times greater levels than the PI88788 to Peking rotation. These trends continued in 2019, with the greatest yield differences observed in plots that rotated sources of resistance. Interestingly, in this study, three continuous seasons utilizing Peking resulted in the lowest yields. Soil samples have been submitted for HG type test to Plant and Pest Diagnostic Laboratory at Michigan State University. We are still completing samples for *H. glycines* field density and soybean yield data is yet to be analyzed for this year.

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Entomopathogenic nematodes (EPN) are excellent biological control agents for soil insect pests. Current biotechnological improvements are allowing their implementation against aerial insect pests. Exploring the intra- and inter-specific variability on EPN virulence can support selecting the best candidates for a particular aerial insect pest. We hypothesized that populations of the same species but with different origins (habitat, geo-region) might differ in their ability to kill the same insect. This study aimed to evaluate the virulence (mortality and time to kill) of various EPN species/populations against the last larval instar of two aerial pests: *Frankliniella occidentalis* (Thysanoptera: Thripidae) and *Tuta absoluta* (Lepidoptera: Gelechiidae). We tested 10 EPN populations belonging to three EPN species: *Heterorhabditis bacteriophora* (Koppert, MG-618b, AM-203, RM-102), *Steinernema feltiae* (Koppert, RS-5, AM-25, RM-107), and *Steinernema carpocapsae* (Koppert, MG-596a). Each EPN population was tested at two concentrations: 160 and 80 IJs/cm² for the *F. occidentalis* trials, and 21 and 41IJs/cm² for the *T. absoluta*. The treatments comprised 5-6 replicates, each with 10 *F. occidentalis* or 8 *T. absoluta*, with food to avoid starvation. Control treatments followed the same experimental procedure but only adding distilled water. The experiments were performed twice, maintained under a controlled environment chamber (60% RH 22°C and 16L:8D). The larval mortality was revised daily up to 6 days post-inoculation. Overall, most EPN populations produced mortalities higher than 50% against *F. occidentalis*, and differences between concentrations were not observed, with few exceptions. *S. carpocapsae* Koppert reached values above 90% mortality after 6 days' exposure, similar to *S. carpocapsae* MG-596a. All the *S. feltiae* populations produced 60-75% mortality. However, on average, *S. carpocapsae* killed in less than 2 days, faster than *S. feltiae* that required 2.2-3.8 days. In the case of *T. absoluta*, all the EPN populations produced >65% mortality. The higher concentration tested resulted in a significantly higher virulence in six out of ten populations. The four *S. feltiae* showed the best results, achieving > 90% mortality even at the concentration of 4 IJs per cm², and killing in less than 2 days. *S. carpocapsae* Koppert also showed similar results. Overall, with a few exceptions, virulence among populations of the same species was not highly variable against the two insects. We consider *S. feltiae* and *S. carpocapsae* very promising for their application against *F. occidentalis* and *T. absoluta* last instar larvae.


A-39

EVALUATION OF ROOT KNOT AND RENIFORM NEMATODE RESISTANT PHYTOGEN COTTON CULTIVARS WITH SUPPLEMENTAL CORTEVA AGRICIENCE NEMATICIDES. Turner, A. Kate 1, Kathy S. Lawrence 1, John Richburg 1, 559 Deval Dr, Auburn, AL 36832 Auburn University, 102 Kimberly Street, Headland AL 36345, Corteva Agriscience.

Root-knot nematode (Meloidogyne incognita) and reniform nematode (Rotylenchulus reniformis) are important agricultural pest in cotton production. Both of these plant-parasitic nematodes significantly reduce cotton yields across the cotton belt. Nematicides and resistant cultivars are the most effective management strategies for reducing these nematodes. The objectives of this study were: 1) to determine the benefit of a root-knot resistance variety PHY360 W3FE and the reniform resistant variety PX3D32 were compared to PHY340 W3FE and reniform populations were 78% lower on PX3D32 compared to PHY340 W3FE. The addition of a nematicide to both susceptible and resistant varieties further reduced nematode egg numbers. Seed cotton yields were 619 lb/A greater for PHY360 W3FE than the PHY340 W3FE susceptible cotton variety in root-knot infested field. Similarly, in the reniform infested field the seed cotton yields were 1692 lb/A greater for PX3D32 than PHY340 W3FE. The best increases due to nematicides were a seed cotton yield of 113 lb/A for the root-knot field and 602 lb/A for the reniform field on the resistant varieties.

A-40

ENHANCING ORGANIC VITICULTURE: INSECTICIDAL EFFECT OF ENTOMOPATHOGENIC NEMATODES AND THE CELL-FREE SUPERNATANT FROM Xenorhabdus AND Photorhabdus BACTERIA AGAINST Philaenus spumarius (HEMIPTERA: APHROPHORIDAE), VECTOR OF Xyella fastidiosa (PROTEOBACTERIA: XANTHOMONADACEAE). Vicente-Diez, Ignacio 1, R. Blanco-Perez 2, M. Chelkha 2, M. Gonzalez-Trujillo 1, A. Pou 1, and R. Campos-Herrera 1. 1 Instituto de Ciencias de la Vid y del Vino (Gobierno de La Rioja, CSIC, Universidad de La Rioja), La Grajera, Ctra. Burgos Km. 6, Salida 13 Lo-20, 26007 Logroño (Spain). 2 Research Team “Lombricidae, Improving Soil Productivity and Environment” (LAPSE), Ecole Normale Superieure (E.N.S.), Centre « Eau, Ressources Naturelles, Environnement et Developpement Durable (CERN2D), Mohammed V University, Avenue Mohammed Bel Hassan El Ouazzani, Takaddoum, Rabat, 5118 (Morocco).

Conventional viticulture is among the most pesticide-consuming agricultural systems worldwide. Providing environmentally sound alternatives to reduce the reliance on chemical control is a primary challenge for winemakers, particularly in organic viticulture, limited if compared with integrated pest management (IPM) programs. Xylella fastidiosa (Proteobacteria: Xanthomonadaceae) is widely known as the cause of Pierce’s disease (PD) of grapevines. Hence, Philaenus spumarius (Hemiptera: Aphrophoridae), the primary vector of X. fastidiosa in Europe, is considered a key pest in vineyards. Using entomopathogenic nematodes (EPNs) for P. spumarius control can be a successful alternative. First, EPNs are well-known biocontrol agents of soil-dwelling insect pests. Thanks to recent advances in application technology, their implementation against aerial insect pests are now possible. Also, the use of the cell-free supernatant derived from EPN-symbiotic bacteria is arising as a powerful bio-tool against aerial insect pests. Thus, this study aimed to evaluate, for the first time, the efficacy of EPN and their symbionts cell-free supernatant applications against the nymphal stages of P. spumarius. In laboratory conditions, we explored the virulence (mortality and time to kill) of EPN species and the cell-free supernatant of their corresponding symbiotic bacteria: Steinernema feltiae RM-107-Xenorhabdus bovienii, S. carpocapsae All-X. nematophila, S. riajaense RM-30-X. kozodoi, and Heterorhabditis bacteriophora RM-102-Photorhabdus luminescens. We filtrated a 3-days Trypstone Soya Broth (TSB) bacterial culture, maintained in agitation and darkness conditions, to obtain the cell-free supernatants. The concentration of 50 IJs/cm² was tested with each EPN species, while for the cell-free supernatant experiments, we tested 10-fold dilution. Each treatment comprised six replicates, each with five nymphs of P. spumarius, supplemented with sucrose suspension to avoid starvation. Control treatments followed the same experimental procedure but only adding distilled water/TSB. Repeated twice, we experimented under a controlled environment chamber (60% RH, 20 °C/14 hL, and 14 °C/10 hD). Subsequently, we recorded the larval mortality daily up to five days post-inoculation. Overall, S. feltiae RM-107 and S. carpocapsae All provided the most promising results, killing 78% and 90% of nymphs after five days of EPN inoculation, respectively. The application of the P. luminescens’ cell-free supernatant killed 64% of the nymphs after five days of exposure. Overall, we demonstrated the great potential of specific EPN and the cell-free supernatant of their symbionts against the nymphal stage of P. spumarius, opening new opportunities to develop them as bio-pesticides components for IMP or organic vineyard productions.

S-41

PRE-PLANT AND IN-SEASON SOIL TREATMENT WITH CHITIN RICH CRUSTACEAN MEAL SUPPRESSED MELOIDOGYNE SPP. AND IMPROVED SOIL HEALTH AND ASPARAGUS GROWTH. Waisen, Philip 1, S. Budhathoki 1, R. Paudel 1, J. Uyeda 1 and K.-H. Wang 1. 1 Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822. 2 Department of Tropical Plant and Soil Sciences, University of Hawaii at Manoa, Honolulu, HI 96822.

Interaction between Meloidogyne spp. and Fusarium oxysporum f. sp. asparagi is considered an important limiting factor for replanting asparagus fields. A field trial was conducted to assess the effects of organic and biological soil amendments on Meloidogyne spp. and soil health. This study examined 5 soil treatments including 1) Actinovate® (AT, Streptomyces lydicus, Noyozyme, Milwaukee, WI); 2) Subtilex® (SB, Bacillus subtilis, BASF, Research Triangle Park, NC); 3) Shrimp Meal (SM, Down To Earth Distributor Inc., Eugene, OR) containing 6-6-0, 10% Ca, 18% chitin and trace minerals derived from ground shrimp shells; 4) Crustacean Meal (CM, PAR 4 Protein Meals, Bridgewell Agribusiness LLC, Clackamas, OR) containing 4-0-0, 12% Ca, 23-30% chitin derived from crab and lobster shells and 5) a soil treatment containing composted horse manure (CMH) and Actinovate®.
Meloidogyne enterolobii

Meloidogyne incognita

Rotylenchulus reniformis

native ecosystems. The objective of this research was to document the host status of Meloidogyne javanica.

A-44

HOST STATUS OF THE HAWAIIAN NATIVE PLANTS VITEX ROTUNDIFOLIA, SIDA FALLAX, IPOMEA PES-CAPRAE BRASILIENSIS, AND PRICHARDIADIATA SP. TO ROTYLENCHULUS RENIFORMIS AND MELOIDOGYNE JAVANICA. Wong, Landon and B. Sipes. University of Hawaii at Manoa, Honolulu, HI 96822, USA.

Diseases caused by fungi, viruses, bacteria, and nematodes affect the health of native plants and can weaken their reintroduction into native ecosystems. The objective of this research was to document the host status of Meloidogyne javanica, a root-knot nematode, and Rotylenchulus reniformis, the reniform nematode, on native plants used in the landscape. In a series of greenhouse experiments, rooted cuttings of Ipomea pes-caprae brasiliensis, Sida fillax and Vitex rotundifolia and seedlings of Prichardidiata sp. were inoculated with eggs of
M. javanica or mixed vermiform and eggs of R. reniformis. M. javanica was not tested on I. pes-caprae brasiliensis. Tomato seedlings were used as a comparative host for M. javanica and cowpea seedlings for R. reniformis. Each test had at least 4 plants and was repeated twice. Three months after inoculation, plants were collected, roots shaken in NaOCl to extract eggs, and nematode reproduction calculated. Nematode reproduction on tomato and cowpea was greater than 1 in all experiments. Prichartidiata, S. fillax, and V. rotundifolia and had reproductive factors less than one for both nematodes. For R. reniformis, I. pes-caprae brasiliensis had a reproductive factor greater than 1. Prichartidiata, S. fillax, and V. rotundifolia are not good hosts to either nematode species. Ipomea pes-caprae brasiliensis is an excellent host for R. reniformis.