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ABSTRACTS

DIFFERENTIAL SUSCEPTIBILITY TO OXAMYL OF THREE PLANT PARASITIC NEMATODES. **Ahmed, Saleh[1], P. Agudelo[1], and P. Gerard[2].** [1]School of Agricultural, Forest, and Environmental Sciences, Clemson University. Clemson, SC 29634; [2]Department of Mathematical Sciences, Clemson University, Clemson, SC 29634.

Oxamyl is a carbamate nematicide that controls a broad spectrum of nematodes, insects, and other organisms. It is used on field crops, vegetables, fruits, and ornamental plants and may be applied directly onto plants or the soil. The mode of action of oxamyl is through the inhibition of acetylcholinesterase (AChE), the enzyme that hydrolyzes the excitatory neurotransmitter acetylcholine at the neuromuscular synapse. The objective of this study was to measure the inhibition potency of oxamyl on AChE of *Ditylenchus dipsaci*, *Pratylenchus penetrans*, and *Aphelenchoides fragariae*. A photometric method was used to determine AChE activity of nematode homogenates, using acetyl-thiocholine as a substrate. A range of concentrations of oxamyl was used to determine the concentration that inhibited 50% of nematode AChE activity (IC_{50}) for each species. These observations were supplemented with in vitro toxicity assays in which live nematodes were exposed to eight different doses of oxamyl (ranging from 25 to 6,250 mg/L) for 24 h. Mortality percentage was calculated after 24 h of exposure and after recovery for 24 h in water. Probit analysis was used to determine the LD50 for each species. In the inhibition studies, *Pratylenchus penetrans,* with an IC_{50} of 2.6×10^{-7} M, was the most sensitive to AChE inhibitors of the three species evaluated. *Ditylenchus dipsaci*, with an IC_{50} of 5.7×10^{-6} M, was the least sensitive. In the toxicity assays, there were significant differences between adults and juveniles of the three species. The LD50 of oxamyl for *D. dipsaci* adults and juveniles were 2,170 and 1,376 mg/L, respectively. The LD50 of oxamyl for *A. fragariae* adults and juveniles were 1,168 and 830 mg/L, respectively. The LD50 of oxamyl for *P. penetrans* adults and juveniles were 195 and 140 mg/L, respectively. The lesion nematode was more than 10 times more susceptible than the stem and bulb nematodes. These results suggest the importance of developing different recommended application rates of oxamyl for control of these nematode species.

ROOT ZONE CHEMICAL ECOLOGY; NEW TECHNIQUES FOR BELOW GROUND SAMPLING AND MASS SPECTROMETRIC ANALYSES OF VOLATILE SEMIOCHEMICALS. **Alborn, Hans T[1], F. Kaplan[2], and J.G. Ali[3].** [1]USDA ARS Center for Medical, Agricultural and Veterinary Entomology, Gainesville, FL 32608; [2]Kaplan Schiller Research LLC and Biology Department, University of Florida, Gainesville, FL 32611; [3]Ecology and Evolutionary Biology Department, Cornell University, Ithaca, NY 14853.

The ban of methyl bromide as a soil fumigant has led to an urgent need to develop novel methods of control of soil-dwelling pests. The use of semiochemicals for below-ground insect and nematode control is one such novel avenue of research. New technologies to study semiochemically mediated below-ground plant-insect-nematode-microorganism interactions are critically needed. It is now well documented that roots of many plants release volatile organic compounds (VOCs) in response to herbivore damage and that these volatiles attract entomopathogenic nematodes. Recent research indicates that plant pathogenic nematodes also respond to root (or rhizosphere) volatiles. Above ground, similar VOCs governing multitrophic interactions are well known and are studied using established techniques; however, in contrast to the release of leaf or floral VOCs into a dynamic and constantly changing airspace, root VOCs are released into a virtually static airspace within the soil. Consequently, even VOCs that might be released at a very low rate, or for a short time, will accumulate in the soil surrounding the root and then disperse through the soil airspace solely by diffusion. Root-related VOCs have typically been sampled either by transferring a plant, or just its roots, from a pot to an artificial environment and then drawing most of the air surrounding the roots through an adsorption filter that trap VOCs, or by maceration and solvent extraction of the roots. However, more sensitive and less intrusive sampling is necessary for in vivo studies of below-ground interactions governed by volatile semiochemicals. To accomplish this, probes were designed for direct in-soil sampling that, in combination with improved thermal desorption GC/MS analyses, allow short sampling times and require removal of minimal air volumes. This makes it possible to monitor continuously below-ground release of VOCs without significantly affecting the system. Low impact sampling will also make it possible to follow the dynamics of VOCs induced in response to insect or nematode infestation and to distinguish these from constitutively released VOCs.

EFFECTIVENESS AND DURABILITY OF THE EPN-INDUCED INSECT AND DISEASE RESISTANCE IN TOMATO. **An, Ruisheng[1], D. Orellana[1], L. Phelan[1], and P.S. Grewal[1,2].** [1]Department of Entomology, The Ohio State University, 1680 Madison Ave, Wooster, OH 44691; [2]Entomology and Plant Pathology Department, The University of Tennessee, Knoxville, TN, 37996.

Entomopathogenic nematodes (EPNs) are well known as biological control agents for soil-inhabiting insect pests. Extra benefits from these agents are expected based on previous observations that soil application of EPNs has direct antagonistic effects on root-knot/foliar parasitic nematodes and indirect effects on pests through the activation of defense mechanisms in

hosta and *Arabidopsis thaliana* leaves. Here, we explored the feasibility of using EPNs in induction and maintenance of the systemic resistance in tomato. We applied *Steinernema carpocapsae* infected wax moth (*Galleria mellonella*) cadavers to the soil around the roots of tomato plants and those receiving the freeze-killed cadavers served as the controls. After 3, 7, and 15 d of the treatment (DAT), leaf bioassays were conducted to assess the effects of treatments on the development of insect herbivores and pathogens. The tested pathogens and pests included bacteria *Pseudomonas syringae* pv tomato, silverleaf whitefly *Bemisia tabaci* as a generalist sucking insect, and beet armyworm *Spodoptera exigua* as a generalist chewing pest. We observed that the beet armyworms feeding on the EPN-treated plants 3 and 7 DAT had reduced rate of development relative to those on the control. After 6 d of feeding on the leaves, significantly more beet armyworms remained in the 3rd-instar stage on the treated plants whereas more 4th instars emerged on the control. However, such differences diminished when the beet armyworms were fed on the plants 15 DAT. Similar pattern was observed for the development of the whiteflies and the bacterial pathogen. The whiteflies after 12 d of infestation on the treated plants had significantly more percentage of immature eggs compared with those on the control when the infestation was established on the plants at 3 and 7 but not 15 d postcadaver treatment. For the bacterial disease development, we observed that the number of foliar disease spots was much lower on the treated plants than on the control when the bacteria were inoculated at 3 and 7 d postcadaver treatment. These results confirm our hypothesis that soil application of EPNs carries durable benefit to the tomato plants by enhancing their defensive capability. Thus, this study has the potential to lead to the development of practical approaches for inducing and maintaining systemic resistance for extended periods in economically important plants against diverse pests and pathogens.

ORGANIC AMENDMENTS FOR MANAGEMENT OF ROOT KNOT NEMATODE OF TUNNEL CROPS. **Anwar, Safdar A.[1], M.M. Mahdi[2], and M.V. McKenry[3].** [1]Institute of Agricultural Sciences, University of the Punjab, Quaid-e-Azam Campus, Lahore, 54590, Pakistan; [2]Pest Warning and Quality Control, Agricultural Department, Chinoit, Jhang, Pakistan; [3]Department of Nematology, University of California, Riverside, CA 92521.

Organic amendments have been widely evaluated for management of plant-parasitic nematodes that have become more notable as the use of plastic tunnels has increased. Rapid decline of nematode populations can result for a variety of reasons including release of toxic compounds during decomposition as well as organic benefits that improve soil microbial ecology or crop nutrition. However little information is available on the use of soil amendments against root-knot nematode, *Meloidogyne incognita*, where plants are grown within plastic tunnels. A field experiment was performed in sandy soil (71.2% sand, 12.4% silt, 14.8% clay; 1.6% organic matter) naturally infested with *M. incognita* to assess the effectiveness of compost (@ 4 tons/ha), poultry manure (@ 4 tons/ha), Janter (*Sesbania rostrata* Bremek and Oberm as green manure (28 ton/ha) and cadusafos (Rugby-10G nematicide 8 kg /ha). These treatments were applied alone or in combinations in order to protect Orible cv. of bell pepper, *Capsicum annum* planted within the tunnels. Soil amendments were applied 3 mon before planting, but Rugby nematicide was introduced with via flood irrigation 1 mon after planting. The experiment consisted of a randomized block design with five replications of each treatment. Effectiveness of these treatments was assessed by measurement of the population buildup of pathogenic *M. incognita* as well as the saprophytic nematodes at 6 and 10 mon after incorporation of the amendments. Compared with the untreated, population densities of *M. incognita* declined with the addition of organic matter whether it was applied as a compost, poultry manure or cover crop. Reduction of *M. incognita* populations compared with the untreated persisted for a full 10 mon after application of the organic amendments. By contrast, Rugby alone was not significantly effective in reducing *M. incognita* populations at 6 mon but at 10 mon did show significant population reductions. The population levels of saprophytic nematodes including, *Aphelenchus avenae*, *Acrobeles* spp., *Rhabditis* spp., and *Dorylaimus* spp., significantly increased with passage of time in the rhizosphere. Yield of Orible cv. of bell pepper was significantly increased by soil amendments and nematicide applications. Regardless of how these organic materials and/or known nematicides stimulated improvement in crop nutrition and plant growth, it is apparent that reducing of high plant parasitic nematode populations at the time of planting can be accomplished by several approaches and should become an important consideration when farming in Pakistan.

DISPERSAL AND SPATIAL DISTRIBUTION OF ENTOMOPATHOGENIC NEMATODES IN AN AGROECOSYSTEM WITH A MODIFIED SOIL MANAGEMENT SYSTEM. **Bal, Harit K.[1], N. Acosta[1], Z. Cheng[1,2], P.S. Grewal[1,3], and C.W. Hoy[1,4].** [1]Department of Entomology, OARDC, The Ohio State University, Wooster, OH 44691; [2]Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822; [3]Department of Entomology and Plant Pathology, The University of Tennessee, Knoxville, TN 37996; [4]Agroecosystems Management Program OARDC, The Ohio State University, Wooster, OH 44691.

Previous research has documented patchy distribution of entomopathogenic nematodes (EPNs), and soil management systems that maintain more uniform distributions of EPNs could increase their biocontrol services in agroecosystems. We hypothesized that altered soil fertility and vegetation management in cultivated fields would increase survival and movement of EPNs from the grassy borders to neighboring vegetable fields, leading to more uniform spatial distribution of infective juveniles in the soil, and increase biological control services. The altered soil management system, including reduced tillage, cover crops (clover and barley) and a soil amendment, compost (100 kg of N/ha), was compared with a conventionally

managed system, both planted with carrots at OARDC Muck Crops Research Station, Huron County, OH in 2010 and 2011. Ten-day-old G. mellonella cadavers (45 per plot) infected with Heterorhabditis bacteriophora GPS 11 strain were used as the source of EPNs to establish populations according to field application standards (500,000 IJs per m^2). EPN survival was compared among field plots by releasing H. bacteriophora in late June and recapturing it over a period of 8 wk using in situ bait traps containing five final instar G. mellonella larvae and autoclaved muck soil. EPN dispersal from the grassy border to neighboring cultivated field plots was estimated 2, 9, 16, and 30 d after release in late July and on a grid using the soil baiting technique. To examine the importance of phoretic movement via invertebrates, we compared release areas with and without insecticide applications. Bait traps were removed after 48 h and examined in the laboratory for nematode infection. Repeated measures analysis of variance did not show significant differences between the two soil management regimes in the pattern of H. bacteriophora survival over time in the plots where they were released, or in the numbers of H. bacteriophora moving from grassy borders where they were released into the adjacent field plots. The number of infected G. mellonella larvae in the bait traps and the number of infective juveniles of H. bacteriophora recovered from the dead baits was greater in alternative than conventional field plots in most time points, but this difference was not significant. Spatial structure of *H. bacteriophora* population dispersing from the grassy border into the adjacent cultivated field plots determined by measures of spatial autocorrelation (Moran's I and Geary's c tests of randomness) and spatial analysis by distance indices (SADIE) indicated no significant differences in patterns of distribution of generally low population densities of EPNs. However, a significantly lower proportion of carrots with carrot weevil feeding damage in 2010, lower number of weevil tunnels per damaged carrot and greater carrot root weight in both years was observed in alternative compared with conventional soil management, providing encouraging evidence of improved biocontrol services in alternatively managed systems.

GENETIC SELECTION OF THE AMBUSH FORAGING ENTOMOPATHOGENIC NEMATODE, *STEINERNEMA CARPOCAPSAE* FOR DISPERSAL. **Bal, Harit K.[1], and P.S. Grewal[2].** [1]Department of Entomology, OARDC, The Ohio State University, Wooster, OH; [2]Department of Entomology and Plant Pathology, The University of Tennessee, Knoxville, TN 37996.

The ambush foraging entomopathogenic nematode, *Steinernema carpocapsae* ALL strain was genetically selected for enhanced dispersal in the absence of hosts. Selection was performed by capturing the farthest reaching population of nematode infective juveniles (IJs) emanating from a nematode-infected source *Galleria mellonella* cadaver in microcosms (0.05 m^2) containing autoclaved field soil. The farthest reaching IJs were captured by taking soil core samples at 11.4 cm from the source cadaver between 48 to 84 h at 12 h intervals and baiting with *Galleria mellonella* larvae. The infected baits were placed in White traps to obtain next-generation IJs. The procedure was repeated for a total of 10 rounds of selection. The rate of emergence of unselected and selected lines was estimated to calculate the percentage of IJs dispersing different distances from the source cadavers over time. The effect of selection on mobility was evaluated by comparing the dispersal rates and patterns, sex ratios, and nictation abilities of the selected lines with the foundation population, after five and ten rounds of selection. *S. carpocapsae* showed positive response to selection for dispersal. There was 13- to 23-fold and 8- to 14-fold increase in the mean percentage of IJs dispersed to the farthest distance from the source cadaver, after five and ten rounds of selection, respectively. The average displacement of the selected lines varied from 6.85 to 7.54 cm/d and was significantly greater than the foundation population (5.54 cm/d). The overall mean realized heritability for mobility was 0.60. *S. carpocapsae* reproduction and nictation ability showed a negative response to selection for mobility. Significantly higher proportion ($\sim$ 40 times) of a selected subline with least reproduction potential, 1/20th of the foundation population, dispersed to the farthest distance. The percentage of nictating IJs was significantly lower in the selected lines ($\sim$ 11%) than the foundation population ($\sim$ 90%). The farthest reaching IJs of the selected lines comprised significantly more males (72%) than the foundation population (44%) at most time points. There was no significant change in the mean number of *G. mellonella* baits killed in the microcosms, mean percentage of dispersed IJs, and males between five and ten rounds of selection. Enhancement in dispersal along with high realized heritability (> 0.5) in the selected lines suggested that genetic selection could be an effective approach for enhancing *S. carpocapsae* dispersal despite trade-offs.

MANAGEMENT OF ROOT-KNOT NEMATODES (*MELOIDOGYNE* SPP.) ON *PITTOSPORUM TOBIRA* IN FLORIDA. **Baidoo, Richard[1], R.H. Stamps[2], and W.T. Crow[1].** [1]Entomology and Nematology Department, University of Florida, Gainesville, FL 32611; [2]Mid-Florida REC, University of Florida, Apopka, FL 32703.

Root-knot nematodes (*Meloidogyne* spp.) are among the most devastating pests of cut foliage crops in Florida; hence, control of this nematode is crucial to Florida's green industry. Many *Pittosporum tobira* cut foliage farms have been abandoned owing to severe root-knot nematode infestation, and there is currently no panacea to this menace. Consequently, research is underway by the University of Florida to identify pesticides or biopesticides that could be used to manage these nematodes on perennial cut foliage crops such as *P. tobira*. In this research, a field of 2-yr-old *P. tobira* on a commercial farm naturally infested with *Meloidogyne incognita* was divided into an RCBD with four blocks and four treatments. Treatments were commercial formulations of spirotetramat, furfural, and *Paecilomyces lilacinus* strain 251; and untreated control. Treatment applications were made during the spring and fall seasons according to manufacturer's specifications. Efficacy was

evaluated based on J2/100cm^3 of soil, J2/g of root, and crop yield (kg/plot). Results from the first year of this multiyear trial are presented. Analysis of covariance comparing J2/100cm^3 of soil and J2/g of root from the experimental treatments with the untreated were performed. Yield data was analyzed using ANOVA. Although there were no differences in J2/g of root, J2/100cm^3 of soil were reduced by furfural and *P. lilacinus* treatments following the fall applications. No yield differences were detected after the first year. The experiment is ongoing and is being supplemented by additional small-plot and greenhouse experiments.

DISEASE PROTECTION AGAINST SEDENTARY PLANT PARASITIC NEMATODES THROUGH COMBINATION PRODUCTS WITH AGROCHEMICAL AND BIOLOGICAL CONTROL AGENTS. **Becker, J. Ole.** Department of Nematology, University of California, Riverside, CA 92521.

Certain soilborne microorganisms can significantly reduce the level of plant-parasitic nematode population density and thereby mitigate the pathogens' crop damage potential. In our research, nematode-destroying microorganisms that parasitize late-stage juveniles and young females of cyst or root-knot nematodes were most efficacious in nematode population suppression. In annual crops, however, a potential biological control product based on such organisms is likely to produce unsatisfactory results as the initial attack by mobile infectious juveniles may be insufficiently challenged. Early crop damage during the seedling stages is often more important than nematode-parasitic activity when plants are established. Seed-delivered nematicides, such as Avicta, provide significant seedling protection and mitigation against nematode-caused crop damage but the effective protection time span is limited to a few weeks because of intrinsic product characteristics. A combination of Avicta and nematode-deleterious microorganisms has the potential to improve performance and expand the overall protection period. The active ingredient abamectin, a mixture of *Streptomyces avermectinius* metabolites, is not antimicrobial, making it suitable for a combination product with biological control agents. Avicta-coated seed in combination with strains of *Pochonia chlamydosporia* or *Pasteuria penetrans* provided proof of concept in improving plant health and yield in *Meloidogyne incognita*-infested greenhouse and field mini-plot trials.

EVALUATION OF NOVEL PRODUCTS FOR ROOT-KNOT NEMATODE MANAGEMENT IN PROCESSING TOMATO PRODUCTION. **Becker, J. Ole[1], A. Ploeg[1], and J. Nunez[2].** [1]Department of Nematology, University of California, Riverside, CA 992512; [2]UCCE Kern County, Bakersfield, CA 93307.

Crop damage caused by root-knot nematodes in California's processing tomato production has been estimated to be approximately 10% despite the widespread use of resistant tomato cultivars or nematicides. In addition, the increasing occurrence of Mi-1 gene resistance-breaking root-knot nematode strains in production fields is of serious concern. During the past two seasons we tested a number of compounds for nematode management in both conventional and organic tomato production under Southern California conditions. In both years, trials were conducted during the summer months at the University of California South Coast Research and Extension Center, Irvine, CA (SCREC). The soil was a San Emigdio sandy loam (13% sand, 75% silt, 11.6% clay; 0.4% organic matter; pH 7.3) infested with *M. incognita* (p*i* 2011: 62 J2/100 cm^3; p*i* 2012: 46 J2/100 cm^3). The trials were designed as a randomized complete block with five replications. The soil-applied treatments included Actinovate (*Streptomyces lydicus* WYEC 108), NemaQ (*Quillaja saponaria* extract), MCW-2 480 EC (fluensulfone), RDL-29 SC480 (iprodione), Sesamin EC (sesame oil), Vydate L (oxamyl), and Vydate L followed by foliar spray with Movento 240SC (spirotetramat) plus 0.25% Dyne-amic. Ecozin Plus (azadirachtin), Nortica 5% WP (*Bacillus firmus* I-1582), and Dazitol (mustard/pepper extract) were tested only once. Six weeks after transplanting and at harvest, five root systems per replication were evaluated for root-knot symptoms. Fruits were harvested once when approximately 3/4 were reddish in color. No obvious product tolerance issues were observed. MCW-2 was the only product that in both years reduced tomato root galling at midseason and harvest. It increased tomato yield in the two trials by 36% and 25%, respectively, over the untreated control.

AN UNUSUAL FLAVIVIRUS DISCOVERED IN THE SOYBEAN CYST NEMATODE. **Bekal, Sadia[1], K. Bhalerao[1], J.P. Bond[2], A.M. Fakhoury[2], and K.N. Lambert[1].** [1]University of Illinois at Urbana-Champaign, 1102 South Goodwin Ave. Urbana, IL 61801; [2]Southern Illinois University, Public Policy Institute, Carbondale, IL 62901.

Viruses have not been extensively studied in phytoparasitic nematodes. To date, four negative-polarity RNA viruses that infect *Heterodera glycines*, the soybean cyst nematode (SCN) have been characterized. Recently, a new RNA virus that infects SCN was identified and named soybean cyst nematode virus 5 (ScV5). This new virus was most similar to Pestiviruses in the family Flaviviridae and appears to encode a positive polarity RNA virus with a single ~ 19,000 nucleotide genome. The similarity to Pestiviruses was low, but phylogenetic analysis of the RNA dependent RNA polymerase domain and the conserved C-terminal RNA helicase domain of ScV5 confirmed ScV5 was most similar to Pestiviruses, but was considerably diverged. Other conserved domains in the viral polyprotein included a methlytransferase domain that is found only in Flaviviruses since they use it to produce capped mRNAs. RNA expression analysis was conducted for ScV5 by aligning Illumina SCN cDNA sequencing reads to the viral genome. This analysis showed that a large number of sequence reads matched the viral 3' noncoding region. The massive accumulation of cDNA sequences in the 3'end of the virus is very good

evidence that the virus produces a subgenomic RNA. Interestingly, such subgenomic RNAs are only found in Flaviviruses and not in Pestiviruses. This evidence suggests we have identified a new virus in the family Flaviviridea with properties of both pesti and flavi viruses. RTQPCR studies indicated ScV5 is present in all SCN developmental stages and may be transmitted transovarially (vertically) and sexually (horizontally) by the male. The virus also appears to be very stable once extracted from the nematode, suggesting it may also have an extracellular route for horizontal transmission. Among viruses recently described in SCN, ScV5 is unique in its potential ability to infect SCN via an infectious mRNA thus ScV5 has a superior potential to be used as biological control agent or a useful tool to express or silence SCN genes.

CHEMICAL SIGNALS FROM PLANTS PREVIOUSLY INFECTED WITH ROOT-KNOT. **Benda, Nicole D., H. Alborn, and P.E.A. Teal.** USDA ARS Center for Medical, Agricultural and Veterinary Entomology, Gainesville, FL 32608.

Nematodes are a worldwide problem in agriculture, with losses estimated to $100 billion per year in the United States. Damage caused by root-knot nematodes (*Meloidogyne* spp.) (RKN) disrupts the flow of water and nutrients to the plant and increases the plant's vulnerability to other pathogens. Although studies have shown that chemical signals in the rhizosphere affect nematode behavior, few of these signals have been identified. In addition, little is known about how previous host plant infection alters the behavior of secondary infective juvenile RKN. We found that both susceptible and resistant varieties of cowpeas were equally attractive to RKN. We also found that RKN preferred uninfected susceptible plants relative to previously infected plants. However, when resistant cowpeas were used, RKN were equally attracted to uninfected plants and their previously inoculated counterparts. This suggests that chemical signals associated with successful gall formation results in reduced RKN attraction. Root metabolites were analyzed for differences between infected and uninfected susceptible plants to identify the signals that reduce attraction. Identification of these signals could lead to the development of more sustainable solutions for this major agricultural problem.

NEMATODE SUSCEPTIBILITY RANKINGS FROM SOFT-TISSUE X-RAY IMAGING. **Bernard, Ernest C.[1], D.W. McDonald[2], B.H. Ownley[1], R.B. Michaels[2], and D. Weaver[3].** [1]Entomology and Plant Pathology Department, University of Tennessee, 2505 E. J. Chapman Drive, 370 Plant Biotechnology Building, Knoxville TN 37996-4560; [2]Phenotype Screening Corp., Suite 10, 4028 Papermill Road, Knoxville, TN 37909; [3]Department of Agronomy and Soils, Auburn University, 202 Funchess Hall, Auburn, AL 36849.

In soft-tissue x-ray imaging the entire root volume of a plant is captured in a single high-resolution image. This approach allows for sophisticated comparisons of nematode numbers or egg masses with root volume and length, both of which can be calculated to a high degree of accuracy in this system. A greenhouse experiment was conducted with reniform nematode (*Rotylenchulus reniformis*) and eight cotton selections: six lines from the Auburn University reniform nematode resistance program (two resistant to *R. reniformis*, four susceptible) and two resistant USDA germplasm lines (LONREN-2 and BARBREN-713). Reniform nematode inoculum was obtained from a cotton field in western Tennessee and consisted of mixed adults and juveniles at a density of 1,500 per 500 cm^3 of soil. Seedlings were planted in infested soil in 500-cm^3 Cone-tainers and grown for 6 wk, in a randomized complete block design with eight replicates. After 6 wk, root systems were carefully harvested to minimize egg mass loss, then subjected to X-ray imaging to count egg masses, determine total root length, and calculate root volumes. The overall root system of each plant was characterized to determine its root architecture. Reniform nematode egg masses characteristically appeared on x-rays as hemispheres with a dark basal ring next to the root and a lighter dome, which was distinct from artifacts such as soil particles and root stubs. Each cotton line had a unique distribution of roots as a function of root diameter and as a function of root depth. Each line also had a unique distribution of nematode feeding sites along its root system. Two resistant lines from the Auburn program had low egg mass counts per unit length of root. The USDA line BARBREN-713 had high egg mass counts per unit length of root. The use of x-ray technology and digital image analysis can provide a much more accurate approach to nematode relationships with plant host roots.

PHYSICAL, CHEMICAL AND AGRO-BIOLOGICAL ATTRIBUTES OF COVER CROPS: WITH SPECIFIC REFERENCE TO NEMATODES. **Bird, George.** Department of Entomology, Michigan State University, East Lansing, MI 48824.

There are at least 14 different reasons for using cover crops in agricultural systems: *Managing Cover Crops Profitably*, 2010, Sustainable Agriculture Research and Education Program. These include modification of physical attributes of soil health such as water-stable aggregates, water retention capacity, surface hardness and subsurface hardness (http://soilhealth.cals.cornell.edu/extension/manual.htm). In addition, cover crops are used to alter biologically regulated factors such as soil organic matter, active carbon, and nitrogen mineralization. Cover crops can also be used to optimize soil chemistry; including pH, phosphorus, potassium, and other elements essential for plant growth and development. All of these factors affect microbial community structure and associated processes, including those related to nematodes. Using cover crops to manipulate nematode population densities is one of several system management objectives such as soil erosion prevention, livestock grazing, hay production and general soil health enhancement. There are three fundamental laws for successful use of cover crops: (i) identification of a specific objective for use of a cover crop, (ii) selection of a specific cover crop variety/cultivar, and (iii) management of the cover crop in a manner specifically designed to achieve the desired objective. Successful cover crop use

is variety/cultivar specific and not plant species specific. Use of cover crops for nematode management may involve one or more objectives, including decreasing population densities of plant-parasitic nematodes, increasing population densities of bacterial feeding nematodes for enhanced nutrient mineralization potential or increasing nematode species richness/biological diversity for improving system stability. Using a cover crop to control a population density of a specific species of plant parasitic nematode may require use of a nonhost crop, trap crop, crop-producing toxic chemical exudates, crop that enhances predator/parasite populations, or cover crop in the process of biofumigation. Each of these is a different process, requiring specific cover crop varieties/cultivars and system management practices. Because of the complexities of cover crop use, comprehensive education, facilitation, and persuasion initiatives are essential for long-term adoption of successful cover crop systems for nematode management. Cover crop use in Midwest agriculture is increasing. As a result, the nine-state Midwest Cover Crops Council (www.mccc.msu.edu) has published a Midwest Cover Crops Field Guide to assist in the proper long-term adoption of cover crop technology.

RESISTANT CULTIVARS FOR CONTROL OF *GLOBODERA ROSTOCHIENSIS* IN THE KYRGYZ REPUBLIC. **Bird, G.[1], A. Chakaeva[2], and D. Douches[1].** [1]Michigan State University, East Lansing, MI 48824; [2]Academy of Science, Bishkek, Kyrgyz Republic.

The potato, *Solanum tuberosum* L., is the most important nongrain food crop on a global basis. The Kyrgyz Republic is the largest producer of potatoes in Central Asia. Wide spread pests and diseases, including potato cyst nematodes, prevent Kyrgyz farmers from obtaining high and stable yields of good quality potatoes. In the Kemin District, potato yields have declined significantly during the past five years. In the presence of *Globodera rostochiensis* and possibly other *Globodera* spp., tuber yields are low, ranging from 40 to 110 centners per hectare. Unfortunately, there are currently no potato cyst nematode resistant varieties available to Kyrgyz potato growers. The objective of this project is to identify and evaluate cultivars with resistance to *G. rostochiensis* under Kyrgyz Republic growing conditions. The cultivars Boulder and Missuakee, were selected for comparison with Picasso, the local standard. The trial was planted on 9 May 2012, at the Elbar Farm in the Village of Tegermenti of the Kemin Region. Each cultivar was replicated four times in 0.5-m^2 plots of *G. rostochiensis* infested soil. At-planting and at-harvest nematode population densities, and tuber yields were recorded. The nematode data were collected from 1 liter of soil from each plot. Cysts were extracted using the flotation procedure of Kiryanov and Krall (1969), concentrated into a single drop of water, placed on a microscope slide and crushed by placing a second glass slide on top of the original slide. The resulting eggs and second-stage juveniles (J2s) were washed into a 50-ml beaker of water. The water was flushed with a pipette to obtain a homogeneity. One milliliter of the resulting suspension was taken from a watch glass, and the number of eggs and J2s counted. The data were analyzed in accordance to the methods of Popov (1972). Roots and tubers of each plant were carefully examined at harvest for the presence of *Globodera* females and cysts. Cultivars were considered resistant when no females or cysts or only a single newly formed cyst or female was observed. The at-plant population density of *Globodera* ranged from 395 to 561 eggs and J2s per 100-cm^3 soil. The at-harvest population density was 201 eggs and J2s per 100-cm^3 of soil for Picasso, the susceptible control. It was 25 and 12 for Boulder and Missuakee, respectively. No new cysts or females were observed on Missuakee or Boulder tubers or roots at harvest; whereas, there were 15 to 20 associated with Picasso roots and tubers. Tuber yields associated with Boulder and Missuakee were statistically greater ($P = 0.0289$) than those of Picasso. They were 47% and 38% more for Boulder and Missuakee, respectively, compared with Picasso. The research is being repeated in 2013 with min-tubers grown from tissue culture in Dr. Douches Laboratory at Michigan State University.

DESCRIPTION OF A *TRICHODORUS* SP. ECTOPIC ON THRIPS. **Carta, Lynn, and A.M. Skantar.** USDA ARS, Nematology Laboratory, Beltsville, MD 20705.

A thrips insect (Thysanoptera: Panchaetothripinae) from persimmon fruit (Ebenaceae: *Diospyros* sp.) was intercepted in a passenger bag in November 2012 at the Peace Arch Border Crossing from Canada to Blaine WA by an APHIS-PPQ port inspector. The fruit was from an unknown origin, possibly Asia. Nematodes were attached to the abdomen of the female insect and sent to us in saline. Seven nematodes (five females, two males) were processed for permanent slides and a female and juvenile prepared for PCR. Morphologically these nematodes belonged to the *Trichodorus sparsus* group, and the 28S rDNA D3 sequence showed greatest similarity (94%) to *Trichodorus paragiennensis*. Among other morphological differences, the manubrium of the male spicule in these specimens was clearly larger than in *T. paragiennensis*. Morphological and molecular characteristics of these specimens from an atypical habitat are presented.

EXPANDED HOST RANGE OF *HETERODERA URTICAE* FROM ARKANSAS. **Churamani, Khanal, and R.T. Robbins.** Department of Plant Pathology, Cralley-Warren Research Lab., 2601 N. Young Ave., University of Arkansas, Fayetteville, AR 72704.

Plants of 10 species were inoculated with 500 second-stage juvenile (J2) of the Arkansas (Toad Suck Park) population of *Heterodera urticae* in December 2012 and harvested in March 2013. Of the 10 plant species, at least one cyst containing eggs was found in the soil from seed grown Mouse-Eared Chickweed (*Cerastium fontanum*), Purple Deadnettle (*Lamium purpureum*),

Field Madder (*Sherardia arvensis*), Hairy Bittercress (*Cardamine hirsuta*), and chickweed (*Stellaria media*) indicating they are at least poor hosts. No cysts were found in the soil from *Aridopsis* sp., *Duchesnea indica* (Indian Strawberry), *Plantago lanceolata* (Buckhorn plantain), *Lamium amplexicaule* (Henbit), *Geranium carolinianum* (Carolina geranium), and *Geranium dissectum* (Cutleaf Geranium). The *Heterodera* species identification was made primarily by comparing the molecular analyses of the near-full-length small subunit rDNA gene, D2/D3 expansion segments of the large subunit rDNA gene and internal transcribed spacer revealed this as *H. urticae*. The Arkansas cysts are ambifenestrate like those of *H. urticae*; however, they are more lemon-shaped than round. Most of the J2 morphometrics are slightly smaller but within the description range. The J2 head shape, tail shape, and lateral fields are as for the species description. No males of the Arkansas population have been found, whereas they are common in the European populations. No eggs were found in the small gelatinous matrix that may indicate one generation per year. Further studies of this population are planned.

TAXONOMIC IDENTIFICATION OF SPECIES OF CRICONEMATIDAE FROM THE PERMANENT SLIDE COLLECTION OF R.T. ROBBINS. **Cordero, Marco A., and R.T. Robbins.** Department of Plant Pathology, Crallen/Warren Research Lab., 2601 N. Young Ave., University of Arkansas, Fayetteville, AR 72704.

Several species of the superfamily Criconematoidea were identified from the permanent nematode slide collection of R. T. Robbins. Nematodes from quarantine shipments and field collections from California and Oregon were collected, mounted, and identified to genus by R. T. Robbins while working for the California Department of Food and Agriculture from 1972 to 1979. Samples from Arkansas were also collected, mounted, and identified to genus by R. T. Robbins since 1979. *Ogma* n. sp. 1 is characterized by having a first cephalic annule smooth distinctly wider than a second annule and the last with small scales and 40 rows of digitate scales with smooth ends at the midbody were observed; the tail is elongated sharply conoid with pointed terminus with two to four digitate scales in last annuli in tail. Other species identified in this study include *Ogma fimbriatum, O. hungaricum, O. serratum, Hemicriconemoides californianus, Hemicycliophora californica, H. micoletzkyi, H. nucleata, H. sheri, Gracilacus acicula*, and *Paratylenchus vandenbrandei* are reported. Voucher specimens were deposited in the USDA Nematode Collection at Beltsville, MD.

LEVERAGING *C. ELEGANS* CUE-DEPENDENT BEHAVIOR TO UNDERSTAND THE HOST/PARASITE INTERACTION FOR PLANT PARASITIC NEMATODES. **Crisford, Anna[1], Elizabeth Ludlow[1], Jessica Marvin[2], James Kearn[1], Vincent O'Connor[1], Peter E. Urwin[2], Catherine Lilley[2], and Lindy Holden-Dye[1].** [1]Centre for Biological Sciences, University of Southampton, Southampton SO17 1BJ, UK; [2]Centre for Plant Sciences, Institute of Integrative and Comparative Biology, University of Leeds, Leeds LS2 9JT, UK.

The rationale for this study is that elements of the neurobiological underpinnings of *C. elegans* environmentally driven cue-dependent behavior will be conserved in the plant parasitic nematodes (PPNs) and provide a route to new molecular targets for pest control. We have mined the newly available genomic sequence for the potato cyst nematode *Globodera pallida* for orthologues of genes known to contribute to cue-dependent behavior in *C. elegans* and identified candidate genes encoding key components of these signaling pathways. Here we present data for the *G. pallida* orthologue of *tph-1* that encodes tryptophan hydroxylase the synthetic enzyme for the neurotransmitter 5-HT (serotonin). 5-HT is involved in food driven behaviors in *C. elegans* and *tph-1* mutants fail to sustain a high rate of pharyngeal pumping in the presence of food. *G. pallida tph-1* has 66% and 71% identity to *C. elegans tph-1a* and *tph-1b*, respectively. We tested the ability of *G. pallida tph-1* to rescue the aberrant feeding behavior of the *C. elegans tph-1* mutant. *G. pallida tph-1* was amplified from cDNA and cloned into the Gateway expression system using the pan-neuronal promoter *snb-1* to drive expression. The construct was injected into *C. elegans tph-1(mg280)* using *myo-2; gfp* as a transformation marker. Several stable lines expressing *gfp* were generated. Four independent *C. elegans* lines expressing *G. pallida tph-1* in a *tph-1(mg280)* genetic background were completely rescued for the pharyngeal phenotype. This provides evidence that *G. pallida tph-1* encodes the enzyme required for 5-HT synthesis in PPNs. In PPNs, 5-HT stimulates the activity of the stylet that is intimately involved in invasion of the host plant. To assay for 5-HT behavior in PPNs, we modified the electropharyngeogram recording that was established for *C. elegans* pharynx and recorded *G. pallida* electrical waveforms that are coincident with stylet thrusts. 5-HT stimulates stylet activity in a concentration dependent manner. We are currently investigating whether or not we can generate responses from host plant cues in this paradigm and will dovetail this with RNAi experiments for *tph-1* to delineate the role of 5-HT in the PPN host-dependent behavior. Funded by the Biotechnology and Biological Sciences Research Council UK Grant No. BB/K012495/1. We gratefully acknowledge CGC for provision of *tph-1(mg280).*

FLUENSULFONE, A PROMISING NEMATICIDE FOR USE ON TURFGRASSES. **William T. Crow**. Entomology and Nematology Department, University of Florida, Gainesville, FL 32611.

The cancellation of fenamiphos has greatly affected nematode management on turfgrasses. Among the potential replacements for fenamiphos on turf evaluated at the University of Florida is a new active ingredient with a novel mode of action, fluensulfone. A 1.5% granular formulation of fluensulfone is among the most promising nematicides evaluated in our turfgrass trials. Our results indicate that this formulation is effective for management of sting nematode (*Belonolaimus*

longicaudatus) and lance nematode (*Hoplolaimus galeatus*) on lawn and golf course turfgrasses. Our results also reveal consistent improvement in turf health following treatment on established turf, and improved harvestability of sod in turf production. Results from a number of trials on different types of grasses, including turf and nematode response to different rates and application timings, will be presented. The utility of the 1.5% granular formulation in turfgrass nematode management programs will be explored.

THE PATHOGENICITY AND INTERACTIONS OF *HELICOTYLENCHUS PSEUDOROBUSTUS* IN FLORIDA TURFGRASS SYSTEMS. **William T. Crow.** University of Florida Entomology and Nematology Department, Gainesville, FL 32611.

Spiral nematodes of the genus *Helicotylenchus* are among the most ubiquitous plant-parasitic nematodes worldwide. *Helicotylenchus pseudorobustus* is a species common in Florida and the southeastern United States and is frequently found associated with turfgrasses and other grass hosts in the region. On most plants *H. pseudorobustus* is not considered particularly damaging, but recent research at the University of Florida has shown that this species suppresses growth of certain turfgrass hosts. Our research has shown that *H. pseudorobustus* can cause economic damage to some bermudagrass cultivars, and is one of the most important nematode pathogens of seashore paspalum in Florida. The root symptoms of *Helicotylenchus pseudorobustus* are subtle, a general lack of roots compared with noninfested roots. Heavy infection by *H. pseudorobustus* causes a severe reduction in the root system, leading to unthrifty turf. Typical above-ground symptoms include chlorosis and turf decline occurring in patches, often accompanied by proliferation of weeds in the affected areas. Our research also indicates that infection of bermudagrass and seashore paspalum by *H. pseudorobustus* has an inhibitory effect on *Belonolaimus longicaudatus*.

POTENTIAL OF *SOLANUM SISYMBRIIFOLIUM* AND THE BIOLOGICAL CONTROL FUNGI, *TRICHODERMA HARZIANUM* AND *PLECTOSPHAERELLA CUCUMERINA* TO CONTROL *GLOBODERA PALLIDA*, THE PALE CYST NEMATODE. **Dandurand, Louise-Marie[1], G.R. Knudsen[1], C.R. Brown[2], C.J. Filip[1], and P. Gajjar[1].** [1]PSES Department, University of Idaho, Moscow, ID 83844-2339; [2]USDA-ARS, Vegetable and Forge Crops Research Unit, 24106 North Bunn Road, Prosser, WA 99350.

The effect of the trap crop, *Solanum sisymbriifolium,* combined with the biocontrol agents, *Trichoderma harzianum* or *Plectosphaerella cucumerina,* on population decline of *Globodera pallida,* the pale cyst nematode (PCN) were assessed. Effects were determined under three simulated cropping systems (potato, *S. sisymbriifolium* (SNS), or soil only-fallow), amended with either *P. cucumerina*, or *T. harzianum* or nonamended. Soil was infested with PCN at a rate of 5 eggs/g soil and planted with either potato or *S. sisymbriifolium*. The soil-only treatment was amended with the biocontrol agent but not planted. The treatments were as follows: fallow—no agent added, fallow—*P. cucumerina*, fallow—*T. harzianum*, SNS—no agent added, SNS—*P. cucumerina,* SNS—*T. harzianum*, potato—no agent added, potato—*P. cucumerina*, potato—*T. harzianum*. After 21 wk of growth under greenhouse conditions, viability and hatch of eggs retrieved from cysts that were used to infest at initiation of the experiment were determined. The highest number of viable eggs were retrieved from the soil-only treatment. The least number of viable eggs were found in cysts retrieved from the soil-only treatment amended with *T. harzianum*. *P. cucumerina* did not significantly reduce viability of eggs compared with the soil-only treatment. Potato but not SNS reduced viability of eggs compared with soil-only, and there was no difference in viability of eggs after a growth cycle of either potato or SNS. The biological control agents combined with either potato or SNS did not further reduce viability of eggs. No cysts were found in any of the SNS treatments. The biological control agents did not significantly decrease the RF value of cysts in potato.

POTENTIAL OF *SOLANUM SISYMBRIIFOLIUM* AS A TRAP CROP FOR THE CONTROL OF THE PALE CYST NEMATODE, *GLOBODERA PALLIDA*. **Dandurand, Louise-Marie[1], C.R. Brown [2], G.R. Knudsen[1], C.J. Filip[1], and P. Gajjar[1].** [1]PSES Department, University of Idaho, Moscow, ID 83844-2339; [2]USDA-ARS. Vegetable and Forge Crops Research Unit, 24106 North Bunn Road, Prosser, WA 99350.

The efficacy of *Solanum sisymbriifolium* as a trap crop for controlling the pale cyst nematode (PCN), *Globodera pallida* was examined. *S. sisymbriifolium* was shown to induce hatch of PCN eggs and confirmed to be a nonhost for the nematode. A line of *S. sisymbriifolium* was selected on the basis of having fewer spines and confirmed to cause PCN hatch and remain a nonhost. No reproduction occurred on *S. sisymbriifolium* when plants were exposed to PCN at a rate of 5 eggs/g soil for 16 wk under greenhouse conditions, whereas Russet Burbank potato had an Rf value of 20.6. In a separate experiment, the effect of *S. sisymbriifolium* or potato on development of PCN life-stages in roots was determined. Four-week-old plants were inoculated at a rate of 5 eggs/g soil. After 2, 4, 6, 8, 10, and 12 wk of growth, roots of *S. sisymbriifolium* or potato were stained with acid fuschin, and nematode life stages were counted. At 4 wk, both potato and *S.* sisymbriifolium were infected with second-stage juvenile of the nematode but sticky nightshade had significantly fewer juveniles than potato. The nematode continued to develop in the potato to form adult females, males and healthy cysts. However, development of the nematode was arrested by week six in *S. sisymbriifolium*.

UTILIZING MANAGEMENT ZONES FOR *ROTYLENCHULUS RENIFORMIS* IN COTTON: EFFECTS ON NEMATODE LEVELS, CROP DAMAGE, AND *PASTEURIA* SP. **Davis, Richard F.[1], S.K. Aryal[2], C.D. Perry[3], D.G. Sullivan[4], P. Timper[1], B.V. Ortiz[5], K.L. Stevenson[2], G. Vellidis[3], and G. Hawkins[3].** [1]USDA-ARS, P.O. Box 748, Tifton, GA, 31793; [2]Department of Plant Pathology, University of Georgia, 2360 Rainwater Road, Tifton, GA, 31793-5766; [3]Crop and Soil Sciences Department, University of Georgia, 2360 Rainwater Road, Tifton, GA, 31793-5766; [4]TurfScout, Inc., 1015 Carolina St, Greensboro, NC, 27401; [5]Department of Agronomy and Soils, 204 Extension Hall, Auburn University, Auburn, AL 36849.

Nematode management zones (MZs) based on soil electrical conductivity (EC, a proxy for soil texture) have not been published for *R. reniformis*. We tested (i) whether *R. reniformis* levels and the amount of damage caused to cotton differed among MZs; (ii) if the relative effectiveness of nematicides differed among MZs; and (iii) whether the prevalence of *Pasteuria* sp. on *R. reniformis* differed among MZs and nematicide treatments. A field with Dothan loamy sand and Nankin loamy sand soil types was divided into three MZs where MZ3 had sandier soil than MZ1 or MZ2, which were the same, and MZ2 had higher elevation than MZ1 or MZ3, which were the same. Levels of *R. reniformis* near planting in plots not receiving nematicide averaged 1,342 (per 150 cm^3 soil) in 2008, 610 in 2009, and 869 in 2010. Both soil texture and elevation influenced *R. reniformis* population levels with greater reproduction in finer-textured soil and reduced *R. reniformis* levels at higher elevation. Treatment effects on *R. reniformis* levels were the same in all MZs (no MZ × treatment interactions). The effects of texture and elevation on yield were similar to the effects on nematode levels. We observed endospores of *Pasteuria* sp., a bacterial parasite of nematodes, on *R. reniformis* at the field site used for this study. *Pasteuria* sp. generally had greater spore attachment to juvenile *R. reniformis* than to adults with no differences among MZs in percentage of nematodes with endospores, but the number of spores per nematode was lower in MZ3, which had the greatest sand content. The percentage of *R. reniformis* with endospores and the number of attached endospores were reduced by 1,3-dichloropropene + aldicarb. We documented that *R. reniformis* levels are affected by modest differences in soil texture and elevation, but levels of *R. reniformis* were above the action threshold in all MZs, therefore a uniform rate of nematicide would have been recommended and there would have been no cost savings from utilizing MZs in this field.

HIGH THROUGHPUT SEQUENCING OF ROOT-KNOT NEMATODE INFECTED ROOTS YIELDS INSIGHTS INTO PATHOGEN-HOST RESPONSES. **DiGennaro, Peter[1], S. Cha[2], E. Scholl[2], and D. McK. Bird[1,2].** [1]Department of Plant Pathology, NC State University, Raleigh, NC 27695; [2]Bioinformatics Research Center, NC State University, Raleigh, NC 27695.

To initiate and maintain successful feeding sites, root-knot nematode (*Meloidogyne* spp.; RKN) must perceive and respond to changes in host biology. Many plant processes are intimately tied to light, and this manifests as a strong circadian clock. We hypothesize that RKN coordinates its biology with plant circadian rhythms, and this will be mirrored by circadian modulation of the RKN transcriptome. We tested this hypothesis by RNA-Seq analysis of *Medicago truncatula* infected for 2 wk with *M. hapla*, VW9. Plants were arranged in a complete random design in a growth chamber (18-hr light, 25°C). Tissue was collected at six time points over a 24-hr period. This tissue was immediately frozen in liquid nitrogen and mRNA was extracted for high-throughput sequencing. A total of 24 samples (6 time points, 4 replicates) were multiplexed, pooled, and run in parallel on 7 lanes of an illumina GAIIx instrument, generating > 165 million 68 mer high-quality reads. Using TopHat, 97% of these reads were mapped onto the completed genomes of *M. hapla* (6.85%) and *M. truncatula* (93.15%). Initial transcript abundance analysis revealed 147 significantly differentially expressed pathogen loci. Current efforts on this project are aimed at identifying and classifying diurnally regulated genes and placing these loci in the context of plant-pathogen signaling. We are also interested in studying the transcriptional events that accompany the developmental changes throughout the nematode life cycle and the subsequent host responses. To this end, we infected *M. truncatula* with *M. hapla* VW9 and harvested uninfected and infected shoot and roots tissue over a time course (t = 1, 2, 4, 5, 7 d postinoculation) as well as *M. hapla* eggs and prepenetration J2. RNA-seq data was collected for each of the 22 samples yielding > 1 billion high-quality raw reads consisting of 4.37E+08 reads mapped to *M. truncatula* (86.5%) and 6.8E + 07 mapped to *M. hapla* (13.5%). We currently are examining this large and complex data-set to better chart spatio-temporal transcriptional changes in nematode and plant.

EFFICACY OF MCW-2 ON FLORIDA VEGETABLES. **Dickson, D.W., and M.L. Mendes.** University of Florida, Entomology and Nematology Department, P.O. Box 110620, Gainesville, FL 32611-0620.

There is a great need for new chemistry to manage nematode diseases. Over the past 36 years, 12 nematicides have been cancelled or suspended and during that time very little new nematicide chemistry has been registered. Since spring 2008, 15 vegetable trials (carrot, cucumber, egg plant, squash, white and sweet potato, and tomato) have been conducted on MCW-2 (fluensulfone) for root-knot nematode management. These trials incorporated raised bed, polyethylene film, and drip irrigation technology, essentially the same system used in past where methyl bromide was the standard fumigant treatment for soilborne disease management. In most of these trials, MCW-2 was applied as a spray over soil surface and immediately incorporated with a rototiller, followed by a power bedder to build a 9-in. tall bed, and covered with virtually impermeable aluminized metallic reflective polyethylene film. These trials did not include a preplant herbicide; however, a row-middle

herbicide program was included in all trials. Sweet potato and carrot had the best yield increases, whereas cucumber, tomato, eggplant, and squash responded with slightly better or similar yields to the nontreated control. In most trials MCW-2 reduced galling on plant roots or lowered root-knot nematode juveniles as compared with the nontreated control.

ESTABLISHMENT OF BIOCONTROL AGAINST *HETERODERA SCHACHTII* UNDER DIFFERENT SUGAR BEET GENOTYPES. **Eberlein, Caroline, and A. Westphal.** Julius Kühn-Institut, Institute for Plant Protection in Field Crops and Grassland, Messeweg 11/12, 38104 Braunschweig, Germany.

Heterodera schachtii limits the productivity of sugar beet and cruciferous vegetable crops worldwide. In sugar beet systems in Central Europe and North America, *H. schachtii* is managed by crop rotation and cultivating resistant host plants as trap crops. The recent release of a number of nematode-tolerant sugar beet cultivars provides novel management strategies. These cultivars allow less nematode reproduction than susceptible cultivars but more than resistant plants. Tolerant cultivars withstand higher initial population densities of *H. schachtii* than susceptible cultivars before they suffer noticeable nematode damage. Objectives of this study with *H. schachtii* pathotypes Schach0 and Schach1 were to determine: (i) if the obligate bacterial hyperparasite *Pasteuria nishizawae* or the facultative fungus *Dactylella oviparasitica* reduce nematode population densities and protect yield of susceptible, resistant and tolerant cultivars; and (ii) if microbial soil amendments establish consistently under sugar beet monoculture. In a multiyear study, microplots infested with one of the pathotypes of *H. schachtii* were cropped in monoculture with susceptible, resistant, and tolerant sugar beet cultivars. In 2010, plots were either amended with *P. nishizawae* or *D. oviparasitica*; this occurred into natural soil for Schach0, and for Schach1 into previously biocide-treated soil for *D. oviparasitica*. Growth and yield parameters were determined every year, as well as nematode population densities at planting and harvest. In all years, the tolerant genotype yielded the highest. In 2010, seedlings of cultivars amended with *P. nishizawae* in Schach0-infested plots had fewer nematodes in their roots than the control. In these plots, amendment treatments with *D. oviparasitica* or *P. nishizawae* had the largest canopy diameter, but these differences did not result in increased yield. In Schach1 plots, the susceptible and resistant cultivars amended with *D. oviparasitica* had fewer cysts and eggs at harvest compared with the respective control. In 2011, only some Schach0-infested plots received a second amendment with *D. oviparasitica*. In Schach0 plots, the tolerant cultivar planted in soil amended with *P. nishizawae* showed the highest yield; the resistant cultivar, planted in soil containing the second amendment of *D. oviparasitica*, yielded more than the control. In 2012 in Schach0-infested plots, the resistant cultivar treated with either one of the microbial amendments had a lower root penetration than the control. The results of this project suggest that strategies for the sustainable establishment of biocontrol organisms can be developed.

TECHNIQUES FOR PHOTOGRAPHING TECHNIQUES FOR PHOTOGRAPHING NEMATODES IN THE MICROSCOPE. **Eisenback, J.D.** Department of Plant Pathology, Physiology, and Weed Science, Virginia Tech, Blacksburg, VA 24061.

Illustrations dramatically improved during the early part of the 15th century because the optical lens was invented by scientists and used by artists to project images onto the canvas. Tracing these images gave increased understanding of perspective and better representation of detail, especially of highlights and shadows. The lens eventually led to the development of the microscope that allowed mankind to peer into previously unknown worlds. Unfortunately, the images that were seen with the eye had to be drawn with the hand, and often were lacking in both form and detail. The invention of the camera lucida allowed for the projection of the image onto canvas, and drawings of microscopic images greatly improved; however, the development of photography led to the greatest breakthrough in the recording of these images. In the early part of the nineteenth century, light-sensitive paper was developed to capture the shadows of images. Eventually the paper was replaced with transparent film that was placed in a box with a lens that captured negative images of real objects. Film and the photograph were born. At first, the images were in black and white, and later in color. Film reigned supreme for more than a century until it was gradually, and sometimes reluctantly, replaced by the digital camera. Digital cameras contain a charged coupled device that uses photons to make the device emit electrons that are counted and interpreted as one of 256 shades of gray for each pixel. Since each pixel has a numerical value, digital images can easily be manipulated in a variety of ways that are useful for photographing nematodes. Digital images can be used for making high-density range photographs (HDR), they can be added together as a series of photos with differing levels of focus to increase the depth of field of a single image, and many images can be combined to make one large megapixel mosaic photograph that contains high-resolution details of the entire specimen.

GENETICS OF RESISTANCE TO *ROTYLENCHULUS RENIFORMIS* IN *GOSSYPIUM ARBOREUM* ACCESSION PI 529728. **Erpelding, John E., and S. R. Stetina.** USDA ARS, P.O. Box 345, Stoneville, MS 38776.

Reniform nematode (*Rotylenchulus reniformis*) is a serious pathogen of cotton (*Gossypium hirsutum*) in the United States and management of the nematode has been difficult because of the lack of resistant upland cotton varieties. The moderately resistant *G. arboreum* germplasm line PI 529728 was identified as a potential new source of *R. reniformis* resistance. The inheritance of resistance was investigated for PI 529728 through the development of segregating populations for resistance

screening. PI 529728 was crossed with the highly susceptible *G. arboreum* germplasm line PI 529729 to develop BC_1F_1 (PI 529728//PI 529729/PI 529728) and F_2 (PI 529729/PI 529728) populations. The populations were screened for nematode resistance in a plant growth room under controlled environmental conditions. The 10 F_1, 69 BC_1F_1, and 333 F_2 plants were individually inoculated with 1,000 vermiform nematodes and the number of swollen females on the root systems was determined 28 d after inoculation. The F_1 plants were susceptible indicating resistance was a recessive trait. The two populations showed quantitative variation in the number of swollen females per gram of root (FGR). When compared with the resistant parent PI 529728, 30 BC_1F_1 plants showed fewer FGR (mean 30, range 2 to 46). Assuming a 1:1 segregation ratio for the backcross population, classifying these plants as resistant would indicate a single recessive gene is conferring resistance ($\chi^2 = 1.17$, $P = 0.2786$). Assuming a single recessive gene model, 83 F_2 plants would be predicted as resistant, which would indicate plants with 11 or fewer FGR would be classified as resistant. This value would be approximately a 45% reduction in FGR as compared with the mean of the susceptible control PI 529251. Ninety plants had 11 or fewer FGR, which would fit the single recessive gene model ($\chi^2 = 0.73$, $P = 0.3930$). This is the first report of a recessive gene conferring a useful level of *R. reniformis* resistance in *G. arboreum*, suggesting genetic diversity for resistance in the germplasm collection. Additionally, highly resistant plants were observed in the populations suggesting transgressive segregation for resistance. This information will aid in the introgression of *R. reniformis* resistance from PI 529728 into upland cotton for the development of resistant varieties.

UNDERSTANDING THE INTERACTION BETWEEN SOYBEAN CYST NEMATODE AND SUDDEN DEATH SYNDROME IN INDIANA. **Faghihi, Jamal[1], K.A. Wise[2], T.J. Hughes[2], G.J. Bossaer[3], and V.R. Ferris[1].** [1]Department of Entomology, Purdue University, West Lafayette, IN 47907; [2]Department of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907; [3]White County Cooperative Extension Service, 12 N 25 E, Reynolds, IN 47980.

A number of surveys have consistently indicated that sudden death syndrome of soybean (SDS) and soybean cyst nematode (SCN) are two pest issues that can cause significant yield loss on an annual basis in Indiana and most states in the North Central region. In 2010, we observed severe SDS in cultivars that had no SCN resistance, or in which the resistance to SCN was breaking down. Subsequent field studies in 2011 and 2012 confirmed that SDS was more severe in the presence of HG type 2 populations of SCN when the PI88788 source of SCN resistance was no longer effective, even when varieties were reported as resistant to SDS. In these experiments, significantly higher yields were obtained from cultivars with the Peking source of resistance. Despite varied climate differences during the 3-yr period, the SDS-SCN synergistic effect on soybean yield was consistent, and most affected by the type of SCN resistance, whether or not SDS symptom expression was obvious.

LIFE LESSONS LEARNED, AND FROM WHENCE THEY CAME. **Ferris, Howard.** Department of Entomology and Nematology, University of California Davis, CA 95616.

The path of a professional career is shaped by many factors. They include personal interests and expertise, resource availability and associated obligations, and acceptance of positions and assignments by following either the path of least resistance or the only obvious choice. Interactions with four groups of people have been of paramount importance in my career development. One group, of course, was comprised of professors, teachers, and mentors who conveyed the basic understanding and principles of the discipline and related areas of science; another very important group was the long-term research technicians at various institutions who knew how to make things work and how to get things done; a third group was the graduate students and postdoctoral fellows whose evolving ideas shaped my thinking and often dragged me into new directions; and, finally, participation as the nematology component of large-scale, multi-investigator projects, introduced me to the knowledge, skills, and ideas of colleagues from other disciplines and helped mold my own. Sometimes a chance remark in a conversation, or an idea retained from a seminar, has stimulated thinking about the hidden truths concealed within an existing or potential dataset and the questions that are waiting to be asked. The Nematology that I was first introduced to was centered on damage to plants; it still is, but has become much more.

THE IMPORTANCE OF SPECIES DIVERSITY IN SUSTAINING ECOSYSTEM SERVICES. **Ferris, Howard[1], and H. Tuomisto[2].** [1]Department of Entomology and Nematology, University of California Davis, CA 95616; [2]Department of Biology, FI-20014 University of Turku, Finland.

Species diversity is measured as the effective number of species, that is the number of equally abundant species needed to obtain the mean proportional species abundance observed in the dataset of interest. Abundance can be quantified using any relevant measure, such as number of individuals or biomass. Diversity is greatest when all the species in the system are equally abundant according to the chosen measure. In that case, the effective number of species equals the actual number of species observed. Functional guilds are comprised of species with similar enough feeding habits and life course characteristics that they contribute similarly to the same ecosystem service. Nematodes in decomposition food web channels that contribute to nutrient mineralization can be assigned to a set of functional guilds based on the nature of their prey (bacteria or fungi) and life course characteristics (e.g., position along the colonizer-persister gradient). The total species diversity in a dataset can be partitioned into (i) the mean effective number of species per functional guild and (ii) the effective number of

functional guilds of the mean species diversity. The two components can be considered to represent different aspects of functional diversity. The first component, mean within-guild species diversity, may represent both functional redundancy (performance of the same function by multiple species) and functional complementarity associated with physiological and behavioral differences not accounted for in the assignment of species to guilds. Within-guild species diversity may serve as a buffer that ensures that the function of a guild is fulfilled by one species or another as conditions vary across differences in the physical and chemical nature of the habitat. Guild diversity, the second component of total species diversity, quantifies the functional diversity among species that is apparent from major differences in life history or other characteristics. The guilds can be considered complementary in their ecological role, so that high guild diversity ensures the continuity of ecological functions in a system. Both components of functional diversity may be important for sustaining the ecosystem service of the guilds across space and time in, for example, successional contexts.

IDENTIFICATION AND CHARACTERIZATION OF A CLE DOMAIN-CONTAINING PROTEIN FROM *ROTYLENCHULUS RENIFORMIS*. **Gavilano, Lily[1], T.J. Baum[2], W.A. Parrott[3], E.L. Davis[4], and M.J. Wubben[1,5].** [1]Department of Plant and Soil Sciences, Mississippi State University, Starkville, MS 39762; [2]Department of Plant Pathology and Microbiology, Iowa State University, Ames, IA 50010; [3]Department of Crop and Soil Sciences, University of Georgia, Athens, GA 30602; [4]Department of Plant Pathology, North Carolina State University, Raleigh, NC 27607; [5]USDA-ARS, Genetics and Precision Agriculture Unit, Mississippi State, MS 39762.

The reniform nematode, *Rotylenchulus reniformis*, is a sedentary semi-endoparasite that causes significant yield loss on many economically important crops including cotton, soybean, and pineapple. Vermiform infective female nematodes secrete esophageal gland effector proteins, encoded by parasitism genes, into or in proximity to the root cells of their host inducing the formation of a multinucleate nurse cell system known as a syncytium. The identification and characterization of these secreted effector proteins and their respective genes are important for the development of alternative management strategies based on parasitism gene silencing via RNA-interference (RNAi). In this study, we report the isolation and characterization of a full-length candidate parasitism gene isolated from reniform nematode. The isolated gene, designated *Rr-cle-1*, belongs to the CLE (CLAVATA3/ESR-related) domain-containing family of nematode effector proteins. The *Rr-cle-1* cDNA contains an open reading frame of 405 bp that encodes a predicted peptide of 134 amino acids (Rr-CLE-1). Rr-CLE-1contains a single CLE domain at its C-terminal end and a N-terminal signal peptide. The expression of *Rr-cle-1* was confirmed in all reniform nematode stages by RT-PCR and cDNA sequencing; however, transcript levels were greatest in sedentary parasitic females. According to BLASTp analysis, the CLE domain of *Rr-cle-1* showed 83% identity to *Oryza sativa* CLE domain and less than 32% amino acid identity to *Heterodera schachtii* and *H. glycines*. In situ hybridization of *Rr-cle-1* transcripts demonstrated exclusive high level expression in the dorsal esophageal gland cells of *R. reniformis* sedentary females. All vermiform life stages were negative for *Rr-cle-1* transcript using in situ hybridization. Previous studies involving *Heterodera* and *Globodera* spp. have demonstrated that RNAi-mediated silencing of CLE parasitism genes significantly reduced nematode reproduction, thereby, lending themselves as potential targets for nematode control. *Rr-cle-1* gene manipulation toward developing resistant soybean plants via RNAi is currently in progress.

THE EFFECTS OF FERTILIZER, NEMATICIDE, AND TILLAGE ON *HETERODERA GLYCINES*, *PRATYLENCHUS*, *HELICOTYLENCHUS*, AND THE NEMATODE COMMUNITY IN SOYBEAN AND CORN FIELDS IN MINNESOTA. **Grabau, Zane, S. Chen, and J. Vetsch.** University of Minnesota Southern Research and Outreach Center, 35838 120th Street, Waseca, MN 56093.

Soybean Cyst Nematode, *Heterodera glycines*, is the most important pathogen of soybean and there is concern about plant-parasitic nematodes in corn. Additionally, there is increasing interest in soil health and monitoring the nematode community is one way to assess this. A Minnesota field study assessed the impact of tillage, fertilizer, and nematicide on plant-parasitic nematodes, the nematode community, and corn and soybean yields. Field experiments were conducted at adjacent sites in annual soybean to corn and corn to soybean rotations, respectively. Within each site, the experimental design was a randomized complete block with a split-plot arrangement and three replicates. Main plot treatments compared long-term minimum and conventional tillage whereas subplot treatments compared six combinations of swine manure, inorganic fertilizers, and granular nematicide (terbufos or aldicarb). *H. glycines*, *Helicotylenchus* spp, *Xiphinema* spp, and *Pratylenchus* spp were the major plant-parasitic nematodes present at the sites. Tillage had only minor impacts on populations of major plant-parasitic nematode genera. Although aldicarb reduced *H. glycines* and *Helicotylenchus* populations, albeit inconsistently, terbufos did not affect major plant-parasitic nematode populations. Nematicides increased soybean and corn yields under some conditions suggesting plant-parasitic nematodes impacted corn and soybean, although this impact was inconsistent. Tillage, fertilizer, and nematicide impacts on the nematode community were often site- and season-specific. Manure application compellingly shifted the nematode community to one of increased enrichment and decreased community structure. The inorganic fertilizers had minimal impact on the nematode community. Conventional tillage decreased nematode community structure based on some measures, but increased bacterivore and fungivore populations. In contrast, aldicarb nematicide decreased bacterivore and fungivore populations. Effects of terbufos nematicide on nematode populations and community composition were inconsistent.

ANALYZING THE IMPACT OF AGRICULTURAL MANAGEMENT PRACTICES ON SOIL HEALTH USING THE NEMATODE COMMUNITY. **Grabau, Zane, Senyu Chen, and Jeffery Vetsch.** University of Minnesota Southern Research and Outreach Center, 35838 120th Street, Waseca, MN 56093.

In recent years, there has been increased focus on sustainable farming practices in both organic and conventional production. One goal of these practices is to improve and/or maintain soil health. Absent a universal definition of soil health, one challenge is determining what constitutes good soil health and how to measure it. Nevertheless, the main factors generally associated with soil health include biological activity in the soil, composition of the soil community fauna, soil physical/chemical properties, and plant growth. Nematode community analysis can be used as an effective soil biological indicator for measuring these factors. This presentation explores how various agricultural management practices affect soil health as indicated by various ecological parameters derived from the nematode community. Two field experiments are used as case studies. The first study tested inorganic fertilizers, swine manure fertilizer, nematicides, and tillage in Minnesota corn and soybean rotations. In this study, manure application compellingly shifted the nematode community to one of increased enrichment and decreased community structure. The inorganic fertilizers had minimal impact on the nematode community. Conventional tillage decreased nematode community structure based on some measures, but increased bacterivore and fungivore populations. In contrast, aldicarb nematicide decreased bacterivore and fungivore populations. Effects of terbufos nematicide on nematode populations and community composition were inconsistent. The second study tested canola meal, organic soil amendment, and water drainage in Minnesota corn and soybean rotations. Canola meal favored enrichment opportunist nematodes and shifted the nematode community to an earlier stage of ecological succession. Drainage decreased plant-parasitic nematode populations but had few effects on the free-living nematode community.

REDESIGNING URBAN AGRICULTURE FOR LOCAL SELF-RELIANCE: LESSONS FROM NEMATODE COMMUNITY STUDIES. **Grewal, Parwinder.** Department of Entomology and Plant Pathology, University of Tennessee, Knoxville, TN 37966.

Although cities occupy less than 3% of the earth's land surface, they consume more than 75% of total global energy and produce 80% of all greenhouse gas emissions. Daily needs of modern cities for food, water, energy, and other materials are met almost exclusively through importation of goods from distant places, often across continents. The growing urban population and its increasing dependence on transportation for materials and goods are placing earth's life-support systems at risk. Urban agriculture offers a comprehensive framework for local self-reliance and resilience and a means to reducing the ecological footprint of cities. Interest in urban agriculture has escalated recently because of the accumulation of vacant land particularly in postindustrial U.S. cities and motivation to address food insecurity and childhood obesity in disadvantaged neighborhoods. Urban soils are, however, considered highly degraded because of intense human activity. Recent studies on soil nematode communities in urban ecosystems in Ohio reveal interesting spatial patterns and point to their potential implications for monitoring soil health and improving ecosystem services. An overview of the significant findings of these studies are presented.

FIRST COMPLETE GENOME SEQUENCE OF AN ENTOMOPATHOGENIC NEMATODE GOES PUBLIC. **Grewal, Parwinder S.[1], Xiaodong Bai[1], Byron J. Adams[2], Todd A. Ciche[3], Sandra Clifton[4,5], Randy Gaugler[6], Kwi-suk Kim[3], John Spieth[4,5], Paul W. Sternberg[7], and Richard K. Wilson[4,5].** [1]Department of Entomology, The Ohio State University, OARDC, Wooster, OH; [2]Department of Biology and Evolutionary Ecology Laboratories, Brigham Young University, Provo, UT; [3]Department of Microbiology and Molecular Genetics, Michigan State University, East Lansing, MI; [4]Department of Genetics, Washington University School of Medicine, St. Louis, MO; [5]Genome Institute, Washington University School of Medicine, St. Louis, MO; [6]Department of Entomology, Rutgers University, New Brunswick, NJ; and [7]Howard Hughes Medical Institute and Division of Biology, California Institute of Technology, Pasadena, CA.

Entomopathogenic nematodes (Steinernematidae and Heterorhabditidae) have a fascinating biology as they form mutualistic symbioses with bacteria (Enterobacteriacae) and together they serve as important biological control agents of insect pests. Here we reveal the first complete genome sequence of an athogenic nematode species that will serve as a model for future genetic studies in this group and in the phylum Nematoda. The 77 Mbp high-quality draft genome sequence of *Heterorhabditis bacteriophora* reveals significant novelty. Approximately half of the 21,250 putative protein coding genes identified in the genome are novel proteins of unknown function lacking homologs in *Caenorhabditis elegans* or any other sequenced organism. Similarly, 317 of the 603 predicted secreted proteins are novel with unknown function in addition to 19 putative peptidases, 9 peptidase inhibitors, and 7 C-type lectins that may function in interactions with insect hosts or bacterial symbionts. The 134 proteins contained mariner transposase domains, of which there are none in *C. elegans,* suggesting an invasion and expansion of mariner transposons in *H. bacteriophora.* Fewer Kyoto Encyclopedia of Genes and Genomes Orthologies in almost all metabolic categories were detected in the genome compared with nine other sequenced nematode genomes, which may reflect dependence on the symbiont or insect host for these functions. The *H. bacteriophora* genome sequence will greatly facilitate genetics, genomics, and evolutionary studies to gain fundamental knowledge of nematode parasitism and mutualism. It also elevates the utility of *H. bacteriophora* as a bridge species between vertebrate parasitic nematodes and the *C. elegans* model.

CASE STUDIES DEVELOPMENT WORKSHOP. **Guffey, Stan.** University of Tennessee Teaching and Learning Center, 618 Greve Hall, Knoxville, TN 37996.

This 2-hr workshop will review objectives/outcomes driven course or session design and best practices in developing and using case studies to help achieve course or session objectives. Working in groups, participants will formulate a set of learning objectives for a course or learning unit in nematology. Participants will then outline and discuss ideas for developing case studies to achieve specific identified learning objectives.

ASSESSING THE IMPACT OF COMPOST AMENDMENT FOR MANAGING NEMATODES AND SOIL HEALTH IN MINERAL SOIL TO IMPROVE CARROT PRODUCTION. **Habteweld, Alemayehu[1,2], D. Brainard[2], M. Ngouajio[2], S. Kravchenko[3], and H. Melakeberhan[1,2].** [1]Agricultural Nematology Laboratory; [2]Department of Horticulture; and [3]Department of Plant, Soil and Microbial Sciences, Michigan State University, East Lansing, MI 48824.

Without effective and sustainable alternatives to broadly used nematicides, carrot and other vegetable crops have significant challenges in managing plant-parasitic nematode (PPN) and soil health. One way to developing sustainable PPN management practices is to have integrated understanding of the impact of management practices on beneficial nematodes, overall soil health, and carrot yield. This field study investigated the effects of plant- (PC) and animal- (AC) based compost on nematode community structure, PPN suppression and improving soil health under fresh market and processing carrot varieties production. The working hypothesis is that ''*compost amendment will increase carrots yield, enhance mineral soil health and decrease population densities of PPN than non-amended control.*'' PC and AC were applied at recommended (×), 1.5 times (1.5×) and double (2×) rates. Standard rate of urea and no fertilizer treatment served as controls. Although the fields were adjacent, the fresh market variety plots had higher phosphorus (80 ppm), potassium (156 ppm), calcium (1329 ppm), and magnesium (323 ppm) content compared with plots for processing variety. Nematode assemblage analysis was done on soil samples collected at 0, 32, 62, and 78 d after planting for fresh market variety plots and 0, 32, 62, 96, and 132 d after planting for processing variety. Yield data were taken after 80 and 134 d for the fresh market and processing carrot varieties, respectively. The overall results from processing carrot variety plots revealed that compost treatment increased maturity index (MI), MI25, and decreased PPI: MI than noncomposted plots. Colonizer-persister (cp) 2 bacterial feeding, omnivore, and predator nematodes abundance was higher in composted plots than control and urea treated plots. Cp3 plant-parasitic nematodes abundance was also lower in composted plots than control and urea treated plots. Structural index (SI) value increased in compost treated plots more than in urea and nonamended plots, showing progression in food web structure and implying relatively less stressed soil food web. In the fresh market variety plots, 1.5× PC and 1× AC significantly increased yield, suggesting rate-dependent responses. In part, these variable trends between the fresh market and processing cultivars may be explained by the highly nutrient disturbed soil in fresh market variety plots. Overall the first-year results from processing carrot variety plots suggest that compost treatment tended to improve soil health and soil food web, which are keys for soil productivity and increasing carrot yield. This is partially in line with our hypothesis that compost treatment increases beneficial nematodes and suppress PPN that drive to improved soil health and productivity.

EFFICACY OF MCW-2 FOR POTATO AND ONION NEMATODE MANAGEMENT. **Hafez, Saad L.** University of Idaho, Parma, ID 83660.

MCW-2 (fluensulfone), a new nonfumigant systemic nematicide (MANA Inc.), was tested against Columbia root-knot (CRKN), root lesion (RLN), and stubby root (SRN) nematodes on potato and against root lesion nematode on onion in different field and microplot conditions for several years. The field experiments in 2010 and 2011 using MCW-2 alone or in combination with foliar Vydate or Mocap was tested against CRKN on potato. All treatments significantly reduced the percentage of root-knot-infected tubers. The total yield of potato significantly increased in all treatments compared with untreated control. The field and microplot experiments of 2009 and 2010 using MCW-2 alone or in combination with foliar Vydate was tested against RLN on potato. There was a significant reduction in nematode population with an increase of tuber yield in all treated plots compared with untreated control plots. The field experiment of 2011 using MCW-2 alone or in combination with foliar Vydate was tested against SRN and corky ring spot disease on potato. The total yields of potatoes were significantly increased in all treatments as compared with untreated control, and the percentage of tobacco rattle virus (TRV) infected tubers was significantly reduced in all treatments as compared with untreated control. The field experiment in 2011 using MCW-2 was tested against RLN on onion. The total weight of bulb was significantly increased in almost all treatments as compared with untreated control. Jumbo and colossal bulb yields were also increased as compared with the control. In summary, MCW-2 is a highly effective nematicide that can be used for potato and onion nematode management.

THE EFFECT OF DIFFERENT NEMATICIDE COMBINATIONS INCLUDING NEW COMPOUNDS ON COLUMBIA ROOT-KNOT NEMATODE IN POTATO. **Hafez, Saad L.[1], Kelly Luff[2], R. Portenier[1], and M.P. Pudasaini[1].** [1]University of Idaho, Parma Research and Extension Center, 29603 University of Idaho Lane, Parma, ID 83660; [2]Bayer Crop Science, 3554 East 4000 North, Kimberly, ID 83341.

Two separate experiments on chemical management of Columbia root-knot nematode (CRN) in potatoes were conducted in a randomized complete block design on a silt loam soil. The average preplant nematode population density was 0.33 J_2/cc soil

in experiment I and 0.36 J_2/cc in experiment II. Potato cv. Ranger Russet was planted on 11 May 2012. In experiment I, Vapam HL @ 40 gal/ac was applied by a commercial applicator on 25 November 2011 as a standard treatment. Admire Pro @ 8.7 oz/ac was applied in furrow at planting in a 4-in.-wide band over seed pieces. Compound No. 1 @ 10 ml/cwt seed pieces was applied. Compound No. 2 @ 18.5 oz/ac was sprayed at 1- to 2-in. or 6- to 8-in. band in furrow at planting, and compound No. 3 @ 10.95 oz/ac was applied at first irrigation and at monthly intervals for a total of two or four applications. Adsorb @ 1 qt/ac was applied with all chemigation treatments. In experiment II, Mocap 6EC @ 2 gal/ac was preplant incorporated followed by a postplant Vydate C-LV program as the standard treatment. Compound No. 1 @ 0.37 pt/ac or compound No. 4 @ 1.28, 2.14, or 3.00 pt/ac was applied at planting followed by two sequential post-plant applications, one on 6-in. tall plants, and the other a month later. Postplant application of compounds Nos. 1 and 4 were applied through overhead irrigation. In the first experiment, data demonstrates that total yield of potato was significantly increased in most of the treatments as compared with untreated control. Most of the numbered compounds alone or in combination with Admire pro showed similar activity as Vapam in total yield of potatoes. The percentage of infected tuber yield was significantly low in all treatments (0.6% to 3.3%) as compared with untreated control (9.32%). In the second experiment, total yield of potato was increased in compound No. 1 applied sequentially @ 0.37 pt/A at planting, at 6-in. tall plants and 60 d later as compared with untreated control, standard treatment, and other treatments. Other number compounds in experiment II also showed similar activity to the Mocap plus Vydate program in potato yield response. The percentage of infected yield was significantly lower in all treatments (< 1.2%) as compared with the untreated control (9.2%). Under low nematode population densities, these number compounds are showing similar activity to Vapam or the Mocap plus Vydate program in the controlling Columbia root-knot nematode and increasing total yield of potato.

THE ROLE OF ARABIDOPSIS microRNAs IN CYST NEMATODE PARASITISM. **Hewezi, Tarek[1], and Thomas Baum[2].** [1]Department of Plant Sciences, University of Tennessee, Knoxville, TN; [2]Department of Plant Pathology and Microbiology, Iowa State University, Ames, IA.

Plant-parasitic cyst nematodes redifferentiate infected root cells into a complex feeding structure, called syncytium. The formation of this new cell type is induced through sophisticated signaling pathways involving both proteins secreted from the nematode and host factors. Recently, we discovered that cyst nematodes manipulate the transcriptional machinery of infected cells by controlling host microRNA gene expression. Here, we report on a fundamental role of Arabidopsis microRNA genes and their targets in mediating plant susceptibility to the sugar beet cyst nematode *Heterodera schachtii*. We found that overexpression of Arabidopsis miRNA produced plants that are significantly more susceptible to cyst nematode infection than the wild-type control. In contrast, inactivation of miRNA activity through overexpression of mimic sequences of the target sites or through overexpression of noncleavable versions of the target genes produced the opposite phenotype of reduced nematode susceptibility, indicating that miRNA expression is required for host parasitism. Interestingly, knockout mutants of the target genes exhibited a significant increase in nematode susceptibility suggesting a role in suppressing plant defense response against cyst nematode infection.

SCREENING OF ISOLATES OF *PASTEURIA* SPECIES TO PRODUCE VIABLE PRODUCTS FOR A GIVEN NEMATODE/CROP MARKET. **Hewlett, T.E.** Syngenta Pasteuria Lab, Sid Martin Biotechnology Institute, Alachua, FL.

Pasteuria spp. are common nematode pathogens that have been, for more than a century, considered obligate parasites of nematodes. Fermentation methods have been discovered that allow the large-scale production of *Pasteuria* spores. These in vitro spores are capable of attaching and infecting their preferred nematode host. Screening for the most virulent isolates of *Pasteuria* to produce viable products for market for a given nematode/crop market involves four steps: Collection of geographic isolates of target nematodes; collection of new *Pasteuria* isolates of the target nematode genus or species; matrix tests to identify *Pasteuria* isolates that attach and infect a broad geographic range host nematode species; *Pasteuria* 24-hr growth rate in standard fermentation media. Isolates with high growth rate and broad host range are selected for scale-up fermentation and greenhouse and field trials to test efficacy.

HOW TO SELECT A POTENTIAL MICROBIAL CONTROL AGENT FOR *MELOIDOGYNE* SPP.?: *POCHONIA CHLAMYDOSPORIA* VAR. *CATENULATA* A CASE OF STUDIES FROM CUBA. **Hidalgo-Díaz, L.[1], N. Montes de Oca[1], B. Peteira[1], A. Puertas[2], J. Arévalo[1], and W. Ceiro[2].** [1]Centro Nacional de Sanidad Agropecuaria, Apartado 10, San José de las Lajas, Mayabeque, Cuba; [2]Universidad de Granma, Facultad de Ciencias Agrícolas, CP 2140, Granma, Cuba.

Bacterial and fungal parasites, competitors, and antagonists have been evaluated for the control of root-knot nematode (*Meloidogyne* spp.) damage to crops and the regulation of nematode populations in soil. Most microbes show much intra-specific variation and there is a need for careful selection of potential microbial control agents. In 1995-1996, fungal parasites of nematode eggs were isolated from soils of different sites of coffee and tomato plantations infested with a complex of *Meloidogyne* spp. in Cuba. A total of 83 fungal isolates were identified morphologically as *Pochonia chlamydosporia* var. *chlamydosporia*, *P. chlamydosporia* var. *catenulata*, *P. suchlasporia var. catenulata*, *Lecanicillium psalliotae*, and some isolates of *P. chlamydosporia* var. *catenulata* with unusually large chlamydospores. Greater fungal diversity was observed in

Cuba than in similar surveys done in Southern Europe. From these, 24 isolates that represented a range of origins were selected and screened for their ability to parasitize eggs of root-knot nematodes, colonize the rhizosphere on a range of host plants and produce chlamydospores. The strain IMI SD 187 of *P. chlamydosporia* var. *catenulata* was selected, and a low input technology was developed for mass production of inoculum following a Good Manufacturing Practice Guideline. Chlamydospore production is robust and is not affected by repeated subculturing. Single applications of the agent to soil with poor hosts for root-knot nematodes significantly reduced the numbers of healthy root-knot nematodes developing on fully susceptible crops planted subsequently in the crop rotation. Soil factors influence the establishment of the fungus but once established it survives between crops. Real-time PCR has been used to monitor and quantify the fungus after its application in soil. Toxicology and Ecotoxicological tests for the fungus have been completed to EPA standards and a commercial product has been registered in Cuba (KlamiC®). However, there is a need to reduce application dosages, optimize production and formulation methods, as well as to conduct more validation field tests. Part of this work was developed in close collaboration with Professor Brian Kerry and his collaborators from Rothamsted Research, UK. Methods for its exploitation are be presented.

ROOT SCENTS: THE DARK SIDE OF NEMATODE SIGNALING AND PLANT PROTECTION. **Hiltpold, Ivan[1], L. Demarta[1], and B.E. Hibbard[2].** [1]University of Missouri, Columbia, MO; [2]USDA-ARS, University of Missouri, Columbia, MO.

Insect herbivory induces synthesis and release of specific volatile compounds in plants. These volatiles have been shown to be highly attractive to natural enemies of the herbivores, such as predators, parasitic wasps, or entomopathogenic nematodes. In maize, the volatiles emitted upon feeding by leaf- or root-feeding arthropod herbivores have been particularly well studied as well as their effect on beneficial organisms. Key compounds mediating tritrophic interactions between the western corn rootworm (WCR), maize and entomopathogenic nematodes have been identified and several genes and biochemical pathways responsible for the production of these emitted volatiles have been elucidated and described. These advances in understanding volatile emissions in maize roots upon WCR attack and its ecological signaling open novel ways to modify plant volatile blends in order to enhance their attractiveness to natural enemies of the WCR. Furthermore entomopathogenic nematodes have been selected for a better responsiveness to belowground cues or techniques to lure the foraging insect herbivore away from the roots towards nematodes are currently being developed. In the context of Bt-crops (maize), entomopathogenic nematode could be used simultaneously with Bt-products and under certain conditions induce indirect fitness costs to the resistant insect pest. Indeed, because resistance traits result in more insect-damage to the root, it is likely that the induction of defenses in the plant is higher upon attack by resistant insects and so is the emission of nematode attractive volatiles. In the laboratory, such as in the field, certain Bt-maize induced by resistant WCR larvae recruited entomopathogenic nematodes and suffered less damage. This could be particularly interesting in resistance management especially since resistance to genetically modified Bt-plants has been observed in the field. Beside volatile production, roots impact entomopathogenic behavior with their spatial organization. Indeed, nematodes have shown to find their insect host better when the root architecture was simpler. However, the addition of synthetic volatile had a dramatic effect and opposite results were recorded. Therefore root architecture might be another trait to consider while breeding crop plant for the purpose of insect resistance and biological control. These are only few examples on how fundamental knowledge on belowground interactions and ecology offers sustainable tools for insect pest control. As 50% to 90% of the plant photosynthetic products are invested below ground, apprehending soil ecology is pivotal to protect and foster this valuable mankind resource while preserving soil ecosystem functions.

INFLUENCE OF SUNN HEMP AND ORGANIC FERTILIZER ON THE ARTHROPOD AND NEMATODE COMMUNITY IN ZUCCHINI PLANTINGS. **Hooks, Cerruti R.R.[1], J. Hinds[1], and K.-H. Wang[2].** [1]University of Maryland, College Park, MD 20742; [2]University of Hawaii at Manoa, Honolulu, HI 96822.

Plantings of sunn hemp as a cover crop have been shown to improve soil health; reduce insect, weed, and nematode pests; and increase nematode-antagonistic microorganisms. However, these studies have been conducted largely in tropical and subtropical regions. To investigate the impacts of sunn hemp used as a cover crop on arthropod and nematode communities in a temperate region, experiments were conducted in Upper Marlboro, MD, during three field seasons from 2009 to 2011. Field plots were established to investigate the effect of using sunn hemp concurrently as a living and surface mulch (SH) on the arthropod and nematode community and compare it with bare-ground (BG) treatment plots. Additionally, organic (OF) and synthetic fertilizer (SF) treatments were added as subplot treatments to examine their impact on these communities. Treatment impacts on above and below ground organism are discussed.

FUNCTIONAL ANALYSIS OF A GLUTATHIONE S-TRANSFERASE FROM RENIFORM NEMATODE ON SOYBEAN. **Hou, Jing[1], S. Ghabrial[2], A. Kachroo[2], and P. Agudelo[1].** [1]School of Agricultural, Forest, and Environmental Sciences, Clemson University, Clemson, SC, 29634; [2]Department of Plant Pathology, University of Kentucky, Lexington KY 40546.

Nematode glutathione S-transferases (GSTs) have been implicated in plant-nematode interactions as effector proteins with an important role in the establishment of feeding sites. Studies with root-knot nematode in *Arabidopsis thaliana* suggest that

GSTs may protect the nematode against oxidative plant defenses and modulate plant responses to parasitism. Our objective was to study the function of a GST from reniform nematode (*Rotylenchulus reniformis*) in soybean. We used a virus-induced gene silencing (VIGS) system, utilizing a *Bean pod mottle virus* (BPMV)-based vector and a partial sequence of *gsts-1* from *Meloidogyne incognita* to silence the putative reniform nematode homolog. The effect of silencing this gene on reniform nematode infection was evaluated by inoculating treated soybean plants with 3,000 nematodes per plant. The reproduction factor was calculated 35 d after inoculation, and the experiment was conducted twice. Gene silencing was assessed by qRT-PCR at 0, 2, and 4 d after nematode inoculation (dai), using gene specific primers for the reniform nematode *gst* and for soybean tubulin. Hydrogen peroxide concentration in the roots was measured at 0 and 2 dai, using a fluorometrical assay. Roots from the treated and untreated plants were fixed and sectioned for observations on the histopathology of infection. Reproduction on the plants treated with the silencing virus construct was significantly lower than in untreated plants, suggesting this gene of reniform nematode plays an important role in the infection of soybean. Hydrogen peroxide concentration 2 dai in nematode-infected roots with the silenced gene was two times higher than that in roots without the silenced gene. We suggest that one-way plant cells responded to reniform nematode infection was by producing superoxide and its dismutation product, hydrogen peroxide, which are both toxic to plant-parasitic nematodes. The observed behavior of reactive oxygen species (ROS), cell wall thickening, and callose deposition support the possibility of this nematode-secreted protein potentially acting as a microbe-associated molecular pattern.

EFFECT OF TEMPERATURE TREATMENT ON SURVIVAL OF *HETERODERA GLYCINES* AND THE FUNGI ASSOCIATED WITH ITS CYSTS. **Hu**, **Weiming[1,2], S. Chen[1], and X. Liu[2].** [1]University of Minnesota Southern Research and Outreach Center, 35838 120th Street, Waseca, MN 56093; [2]State Key Laboratory of Systematic Mycology and Lichenology, Institute of Microbiology, Chinese Academy of Sciences, Beijing 100101, China.

In studies of nematode-suppressive soil and plant-parasitic nematode management, it is often desired to differentially kill nematodes in the soil but keep the microbial community alive. In this study, the effect of temperature treatment on the survival of the soybean cyst nematode, *Heterodera glycines*, and fungi associated with its cysts was investigated. Separate portions of extracted eggs (eggs), extracted cysts (cysts), and cysts in soil (soil-cysts), were subjected to temperature treatments of -80°C for 24 hr; -20°C for 4 hr followed by -80°C for 24 hr; -20°C for 24 hr; 40°C, 45°C, 50°C, or 55°C in a water bath for 1 hr (eggs and cysts) or 2 hr (soil-cysts); or 4°C storage (control) for 24 hr. Fungal survival was determined by plating treated eggs or cysts on PDA (potato dextrose agar plus 100 ppm streptomycin and 30 ppm chlortetracycline) and monitoring mycelium development up to 2 wk. Following temperature treatment of eggs or cysts, nematode survival was determined by juvenile hatching in vitro and infectivity and development of treated eggs or cysts on soybean in the greenhouse pots. Following any of the treatments of -80°C, -20°C then -80°C, 50°C, or 55°C, no active juveniles hatched and no new nematode females developed on soybean except for one female from a pot with the 50°C soil-cysts treatment. There were no significant differences in juvenile hatch or nematode development on soybean between -20°C, 40°C, 45°C, and 4°C treatments. Fungal survival rate in cysts was reduced following heat treatments at 50°C and 55°C compared with all other treatments, whereas no significant differences were observed between -80°C, -20°C then -80°C, 40°C, and 4°C treatment according to least significant difference (LSD) test at $p > 0.05$. Fungal survival rate was decreased in eggs compared with cysts following temperature-treatments of -20°C and -80°C, and 45°C. A further range temperature test between 44°C and 52 C revealed that no juvenile hatched following 48°C or above, and only one female was found on soybean roots after the treatment at 48°C. Our study demonstrated that freezing treatment of cysts at -80°C can kill *H. glycines* but maintain high fungal community survival.

A MICROFLUIDIC PLATFORM FOR ANALYSIS OF NEMATODE FEEDING BEHAVIOR. **Hu, Chunxiao[1], James Dillon[2], Vincent O'Connor[2], Lindy Holden-Dye[2], and Hywel Morgan[1].** [1]Centre for Hybrid Biodevices; [2]Centre for Biological Science, Institute for Life Sciences, University of Southampton, Southampton SO17 1BJ, UK.

We have developed a microfluidic device that can be used to record the feeding behavior of nematodes. It has been designed for the free-living nematode *Caenorhabditis elegans* but has the capability to be used for other species of nematode, including those of economic and medical relevance. The device works by sequentially trapping individual worms in a fluid-filled channel. The channel consists of two compartments separated by a small aperture that has been fashioned to match the transverse dimensions of the worm. The worm is trapped in this aperture and in the process an electrical seal is generated between the two compartments. Electrodes have been incorporated into the device that measure the potential difference between the two compartments. When the worm is feeding the electrical activity of the neuromuscular system that drives the contraction-relaxation cycle of the pharynx is detected as a potential difference between the two compartments generating a waveform that provides a readout of feeding behavior. The microfluidic platform was first designed for adult *C. elegans* and has now been optimized for smaller larval stages. Size is the key for this microfluidic chip design. Without an optimized trapping channel size for capturing the worm's head, the electrical recordings are of poor quality and cannot deliver key information about neural function or drug effects. Compared with young adult *C. elegans*, the L2 stage worm has a smaller body size ($\sim$ 400 μm in length, $\sim$ 20 μm in diameter) making it more difficult to manipulate it in a conventional way. After

several attempts, the best size for trapping and recording L2 stage nematode was found. By analyzing the video on a frame by frame basis, the correlation between the pharyngeal pumping and signal formation was revealed. The device has capability for relatively high throughput as worms may be fed into the chamber from a reservoir. It also permits the rapid application and removal of compounds and thus a route to chemical screening. It is currently being tested on other species of nematode.

HOST SUITABILITY OF GRAIN SORGHUM TO SOUTHERN ROOT-KNOT, COLUMBIA LANCE AND LESION NEMATODES. **Hu, Mengxuan[1], J.D. Mueller[1], D. Gunter[1], and P. Agudelo[2].** [1]Clemson University, 64 Research Road, Blackville, SC, 29817; [2]Clemson University, 206 Long Hall, Clemson, SC 29634.

The acreage of grain sorghum, *Sorghum bicolor*, in the southeastern United States has increased dramatically in the last several years because of increased demand for grain from the swine and poultry industries. There are contradicting reports on the host status of grain sorghum to *Meloidogyne incognita* and no data is available about its host status to *Hoplolaimus columbus*. Furthermore there is only limited data of the reaction of grain sorghum to lesion nematodes. Host suitability of 10 grain sorghum hybrids, to *M. incognita* (SRK), *H. columbus* (CLN), and *Pratylenchus* spp. (lesion) were evaluated under field conditions at the Edisto REC near Blackville, SC. The hybrids were arranged in three strip-blocks with hybrid as the whole plot and treatment with 28 L ha^{-1} of 1, 3-dichloropropene (1, 3-D) as the subplot. Each subplot consisted of four 10-m-long rows on 96-cm row centers. Two locations were established. The ''Set Irrigation'' field was infested primarily with SRK and CLN and the ''Pole Barn'' field was infested primarily with CLN. Both fields were infested with lesion nematodes. Nematode samples were collected from soil at planting, 6-wk after planting and near harvest. Roots were collected just before harvest for extraction of nematodes in a mist chamber. In the Set Irrigation field, 1, 3-D treatment increased the mean yield over the 10 hybrids by 28% from 2,887 to 3,703 kg ha^{-1} compared with mean yield of the hybrids in the nontreated plots. Yields did not differ among the hybrids. Treatment with 1, 3-D lowered the recovery of CLN at harvest from 54.3 to 10.6 g^{-1} of dry root across hybrids. Hybrids did not differ in the number of CLN in roots at harvest. Recovery of lesion from roots, and all three nematode species from soils at harvest were not affected by either 1, 3-D treatment or hybrid. In the Pole Barn field 1, 3-D treatment did not affect mean yield of the grain sorghum hybrids, but the yield ranged from 3,055 to 6,032 kg ha^{-1} depending on the hybrid. Yield of NK7633 was significantly lower than Pioneer 83P17, Pioneer 83G66, DKS 44-20, DKS 53-67, and NK 7829. Recovery of CLN from roots was higher for untreated than 1, 3-D-treated plants at harvest (110.0 and 35.0 g^{-1} of dry roots, respectively). Recovery of CLN from roots did not differ among hybrids. Recovery of lesion nematodes from roots, and CLN, lesion and SRK from soil at harvest were not affected by either 1, 3-D treatment or hybrid. Based on this study, grain sorghum may prove useful in managing SRK populations but may be susceptible to yield losses because of CLN.

CHARACTERIZATION OF A NOVEL *MELOIDOGYNE* POPULATION ASSOCIATED WITH COFFEE (*COFFEA ARABICA*) IN COSTA RICA. **Humphreys-Pereira, Danny A.[1], L. Flores-Chaves[2], M. Gómez[2], L. Salazar[2], L. Gómez-Alpizar[3], and A.A. Elling[1].** [1]Department of Plant Pathology, Washington State University, Pullman, WA 99164; [2]Laboratory of Nematology-CIPROC, University of Costa Rica, 2060 San Pedro, Costa Rica; [3]Plant Biotechnology Laboratory, Agronomy Research Center, University of Costa Rica, 2060 San Pedro, Costa Rica.

Coffee is one of the three main cash crops in Costa Rica and plant-parasitic nematodes cause significant yield decline of this crop in Costa Rica and other countries in Central and South America. Three root-knot nematode species have been identified in association with coffee trees in Costa Rica, including *M. arabicida*, *M. enterolobii*, and *M. exigua*. In 2012, root samples of coffee seedlings cv. Catuai containing small galls were sampled from southern Costa Rica. Eggs were extracted from the roots and used to inoculate tomato cv. Hayslip under greenhouse conditions for subsequent studies. Morphological characters were measured and compared among a population of 30 adult females, second-stage juveniles (J2s) and eggs each using light and scanning electron microscopy. Overall shape of the perineal patterns was ovoid, with moderately high dorsal arches showing coarse striae. Weak lateral lines were frequently present on both sides. Punctations and striae were absent in the perineum. Females had greater mean body and stylet lengths in comparison with other *Meloidogyne* spp. previously identified in coffee. Greater average values for body and tail length were observed in J2s of this *Meloidogyne* sp. population, but stylet length was similar to that reported for *M. arabicida*. J2s showed a dilated rectum. The region between mitochondrial genes *COII* and *16S rRNA* was amplified, cloned and sequenced. This fragment was 1,370 bp, which differs from previously reported amplicon sizes for other *Meloidogyne* spp. PCR products were digested with *Hinf*I, *Dra*I, and *Alu*I, which revealed unique PCR-RFLP patterns that differed from the most important tropical root-knot nematode species, such as *M. arenaria*, *M. incognita*, and *M. javanica*, and also from other coffee-parasitizing *Meloidogyne* spp. such as *M. arabicida*, *M. izalcoensis*, and *M. paranaensis*. No amplification was obtained when using species-specific primers for *M. arenaria, M. chitwoodi*, *M. enterolobii*, *M. exigua*, *M. hapla*, *M. incognita*, *M. javanica*, and *M. paranaensis*. Phylogenies estimated using Bayesian inference analysis of the region between the *COII* and *16S rRNA* mitochondrial genes indicated that this coffee-associated *Meloidogyne* sp. population was found within a large monophyletic group of other tropical *Meloidogyne* spp. that infect coffee. This tropical clade was distinct from temperate *Meloidogyne* spp. These results provide the first morphological and molecular description of a novel *Meloidogyne* population associated with coffee in Costa Rica and provide preliminary data for future in-depth phylogenetic and biological characterization.

STUDYING THE RESPONSES OF TOMATO GENOTYPES TO AVIRULENT AND *MI*-VIRULENT *MELOIDOGYNE JAVANICA* ISOLATES OCCURRED IN ISRAEL. **Iberkleid, Ionit [1,2], R. Ozalvo[1], L. Feldman[1], M. Elbaz[2], Y. Spiegel[1], and Horowitz S. Brown[1].** [1]Department of Entomology, Nematology and Chemistry Units, Agricultural Research Organization (ARO), the Volcani Center, Bet Dagan, Israel; [2]Research and Development Station the H'absor Research Station.

The behavior of natural selected *Meloidogyne spp.* virulent isolates against the *Mi* resistance gene of tomato occurred in main tomato growing areas in Israel has been studied. At the first stage of this research, the virulence of seven selected isolates over three successive generations on a resistance (*Mi*-carrying) and on a susceptible (non–*Mi*-carrying) tomato cultivar was confirmed. Using diagnostic markers, we verified that *Meloidogyne javanica* is the predominance species among all virulent isolates selected on resistance tomato cultivars and/or rootstocks. For better understanding the determinants involved in nematode selection on *Mi*-carrying plants, reproduction of *Mi*-avirulent and virulent isolates Mjav1 and Mjv2, respectively, measured as eggs per g of root on non–*Mi*-carrying, heterozygous (*Mi*/*mi*) and homozygous (*Mi*/*Mi*) genotypes was evaluated. Although no reproduction of the avirulent Mjav1 isolate was observed on the homozygous genotypes (*Mi*/*Mi*), some reproduction was consistently observed on the heterozygous genotypes (*Mi*/*mi*). Nevertheless, reproduction of the virulent isolate Mjv2 on the homozygous and heterozygous genotypes was similar to that on the susceptible cultivars. These results suggesting that a quantitative effect of the *Mi* gene is reasonable for the avirulence isolate only but not to the selected *Mi*-virulence isolates. Histological examination of giant cells induced by the *Mi*-virulent compared with the avirulent isolate confirmed the high virulence of Mjv2 on all three genotypes tested (*mi*/*mi*; *Mi*/*mi*; *Mi*/*Mi*), allowing the formation of well-developed giant cell systems in spite of the *Mi* gene. Analysis of plant defense response of tomato genotypes carrying different allelic stage of *Mi* (*Mi*/*Mi; Mi*/*mi; mi*/*mi*) to both avirulent and virulent isolates was investigated by quantitative real time PCR. Although the Jasmonate signal pathway is clearly up-regulated by avirulent and virulent isolates on the susceptible (non-carrying *Mi*) and heterozygous plants (*Mi*/*mi*), no change in the jasmonic acid-signaling was observed in the resistance homozygous (*Mi*/*Mi*) line following the incompatible interaction with the avirulent isolate. These results indicating that similar to infection promoted by the avirulent isolate on susceptible genotype, *Mi*-virulent isolate induce JA-dependent pathways that might promote tomato susceptibility during the compatible interaction. Results presented here have important consequences in terms of managing the *Mi* resistance genes in breeding strategies and for ensuring sustainable tomatoes farming.

NEMATICIDE APPLICATION STRATEGIES TO CONTROL NEMATODES IN POTATO. **Ingham[1], Russell, E., P.B. Hamm[1,2], and B.A. Charlton[3].** [1]Department of Botany and Plant Pathology, Oregon State University, 2082 Cordley Hall, Corvallis, OR 97331-2902; [2]Hermiston Agriculture Research and Extension Center, Oregon State University, P.O. Box 105, Hermiston, OR 97838; [3]Department of Crop and Soil Science, Klamath Co. Extension, Oregon State University, 3328 Vandenberg Rd. Klamath Falls, OR 97603-3796.

Successful nematode control often entails finding the right application method for the right nematicide. This concept can be documented using nematodes in potato as examples. Root-knot nematodes (RKN), *Meloidogyne hapla* and *M. chitwoodi* infect potato tubers and cause brown spots to form around the infection sites. These spots are considered quality defects for which there is low tolerance in domestic markets and no tolerance in export markets. Corky ringspot (CRS) produces necrotic arcs and rings in tuber tissues that are caused by Tobacco Rattle Virus vectored to tubers by stubby root-nematodes, *Trichodorus* and *Paratrichodorus* spp. Crops with excessive symptoms from RKN or CRS may be devalued or rejected. Acceptable control of RKN and CRS requires getting maximum performance from nematicides. A number of different approaches to incorporating ethoprop with water were unsuccessful whereas several methods of physical incorporation before planting controlled *M. hapla* damage to tubers. Similarly, application of metam sodium (MS) in irrigation water did not control CRS or RKN, whereas, deeper placement of MS with shank injection suppressed damage from both CRS and RKN under low to moderate disease pressure. Under heavier pressure, deeper placement and a higher rate were required for adequate control. Timing of nematicide application can also be critical. Oxamyl can be used successfully to control CRS and RKN but requires that initial applications are made early in the season. Without applications in-furrow at planting and/or at emergence, control of CRS and RKN with oxamyl is very difficult. Often the best application strategy is using two different nematicides together. 1,3-dichloropropene (1,3-D) is generally very effective for control of CRS and RKN, but occasionally soil conditions are not ideal and some nematodes can escape treatment. Following 1,3-D with MS, ethoprop, or perhaps oxamyl can ensure maximum protection. A tank mix of MS and ethoprop injected at 15 and 30 cm is superior to either product alone or both products applied separately. Control is also improved with combinations of MS and oxamyl, aldicarb and oxamyl, or ethoprop and oxamyl. Design of effective application techniques may be almost as important as development of product chemistry. Nematcides currently being developed may not be as powerful as those that have been lost, so optimum application technology will be critical. New and creative application strategies may be required to achieve optimum performance.

REDESCRIPTION OF *PARASITODIPLOGASTER MAXINEMA* WITH A REPORT OF STOMATAL DIMORPHISM. **Kanzaki, Natsumi[1,2], R.M. Giblin-Davis[1], W. Ye[3], E.A. Herre[4], and Barbara J. Center[1].** [1]Fort Lauderdale Research and Education Center, University of Florida/IFAS, 3205 College Ave, Davie, Florida 33314-7799; [2]Forest Pathology Laboratory, Forestry and Forest Products Research Institute, 1 Matsunosato, Tsukuba, Ibaraki 305-8687 Japan; [3]Nematode Assay

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The nematode species associated with *Ficus maxima* were surveyed at the Smithsonian Tropical Research Institute, Barro Colorado Island, Panama, and several different species were isolated from phase B-D syconia. The isolated nematodes were casually observed under a light microscope for morphotyping and stored in nematode digestion solution for molecular characterization or fixed in TAF for further morphological observation. The morphological and molecular observations revealed that there are three *Parasitodiplogaster* species (*P. maxinema*, *P. yoponema*, and an undescribed species) including four morphotypes, and several different other species, e.g., *Schistonchus* spp. Interestingly, *P. maxinema* has two significantly different stomatal morphotypes, i.e., a stenostomatous and eurystomatous form, and their identities were confirmed with molecular sequencing. The morphology of the stenostomatous form is clearly similar to that of the original description, i.e., the stoma is narrow and tube-like, but in addition, we discovered stick-like dorsal and right subventral teeth protruding from the corresponding metastegostomatal elements that can be seen sticking out from the stomatal opening upon protraction. The stomatal composition of the eurystomatous form is rather similar to that of the *P. laevigata* clade species and *Koerneria* spp., the tentative sister group of *Parasitodiplogaster*, i.e., the stomatal opening is rather wide, occupying more than 60% of the lip diameter (similar to the eurystomatous form of *Koerneria*); baring large claw-like dorsal and right subventral teeth from metastegostomatal elements (known in *P. laevigata* clade and *Koerneria* spp.); and no tooth, denticle, or ridge on the left subventral metastegostomatal sector (as in the *P. laevigata* clade). Stomatal dimorphism is known in several bacterial-feeding diplogastrid genera. The eurystomatous form usually occurs in old culture plates (= unfavorable conditions for the nematodes), and often exhibit predatory behavior. *Parasitodiplogaster* is generally considered an insect-parasitic or necromenic species, and its feeding resource has been hypothesized to be solely limited to their adult fig-wasp hosts (Agaonidae). However, the presence of this stomatal dimorphism suggests that these nematodes are feeding on other resources in the fig syconia, although the detailed nutritional physiology of the nematodes has not been studied thus far.

PHEROMONES REGULATE NEMATODE DISPERSAL. **Kaplan, Fatma[1,2], H.T. Alborn[3], S.H. von Reuss[4], F.C. Schroeder[4], and P.A. Teal[3].** [1]Kaplan Schiller Research LLC. Gainesville, FL; [2]University of Florida, Gainesville, FL; [3]USDA-ARS, Gainesville, FL; [4]BTI/Cornell University, Ithaca, NY.

Dispersal is an important nematode behavior for survival. Upon crowding or food depletion, the free-living bacteriovorus nematode *Caenorhabditis elegans* produces stress resistant dispersal larvae, known as dauer. Other nematodes also have dispersal larvae. In plant parasitic *Meloidogyne* spp., it is called J2 and in insect parasites (entomopathogenic nematodes, EPN), it is known as infective juveniles (IJs). Even though pheromones regulate entry into dispersal larvae in *C. elegans* and insect parasites, it is not known whether pheromones regulate dispersal. We hypothesized that pheromones may regulate dispersal behavior in *C. elegans* and in other nematodes. Liquid chromatography-mass spectrometry analysis of *C. elegans* dauer/dispersal supernatant, which shows strong dispersing activity, revealed four known ascarosides (ascr#2, ascr#3, ascr#8, icas#9). A synthetic pheromone blend at physiologically relevant concentrations dispersed *C. elegans* in the presence of food and also caused dispersion in insect parasite (*S. feltiae*) and plant parasitic (*Meloidogyne* spp). Assay guided fractionation revealed structural analogs as major active components of the *S. feltiae* (ascr#9) and *C. elegans* (ascr#2) dispersal blends. Further analysis revealed that all *Steinernema* spp. and *Heterorhabditis* spp. infected insect host cadavers share a common pheromone, ascr#9, suggesting one species can recognize another's blend. Pheromones are fundamentally important for nematode communication across diverse habitats, and thus may provide sustainable means for control of parasitic nematodes.

IDENTIFICATION OF *PRATYLENCHUS* SPECIES AND DETECTION OF *PRATYLENCHUS SPEIJERI* IN THAILAND. **Khaithong, Tridate[1], and B. Sipes[1,2].** [1]Nematology Section, Plant Pathology Research Group, Plant Protection Research and Development Office, Department of Agriculture, Thailand; [2]Department of Plant and Environmental Protection Sciences, College of Tropical Agriculture and Human Resources, University of Hawaii at Manoa, HI 96822.

Fifteen species of *Pratylenchus* have been reported in Thailand; however, much of the information is dated. A molecular identification of root-lesion nematode populations from across Thailand was therefore undertaken. *Pratylenchus* were obtained from 23 locations mainly from soil surrounding banana. Single-female derived isolates were grown on corn roots in tissue culture. Species-specific primers were used to identify isolates to species level using a PCR protocol. The D2-D3 expansion region of 28S rDNA of 14 isolates was amplified and sequenced. The majority of the isolates were identified as *P. coffeae* with one isolate of *P. brachyurus* and two unidentified isolates from corn. Four isolates appeared related to *P. coffeae* but with differences. Phylogenetic analysis of D2-D3 expansion region revealed close relationship between some of the *Pratylenchus* isolates from Thailand and *P. speijeri*. These samples were further analyzed with species-specific primers for *P. speijeri*. The results of the analysis demonstrated that these isolates were *P. speijeri.* This is the first report of *P. speijeri* in Thailand.

ANAEROBIC SOIL DISINFESTATION (ASD) AND STEAM AS ALTERNATIVES FOR PARASITIC NEMATODE CONTROL IN FLORIDA FLORICULTURE. **Kokalis-Burelle, Nancy[1], E.N. Rosskopf[1], J.C. Hong[1], and D.M. Butler[2].** [1]USDA-ARS, Ft. Pierce, FL; [2]University of Tennessee, Knoxville, TN.

Anaerobic soil disinfestation (ASD) and steam are being investigated for controlling a broad spectrum of pests, including parasitic nematodes and weeds. ASD is a biologically based method that combines organic amendments and solar heat with water saturated soil to create oxygen-depleted soil conditions that favor growth of facultative and obligate anaerobic bacteria. Often, these bacteria produce antimicrobial compounds and acids as secondary metabolites that, in conjunction with anaerobic conditions, can control many pathogens including nematodes. A previous experiment on snapdragon (*Antirrhinum majus*) that compared soil solarization with steam and methyl bromide (MeBr) application established that soil solarization alone does not provide acceptable control of root-knot nematodes (RKN), *Meloidogyne arenaria,* in Florida. However, ASD provided superior control of nematodes and weeds that was not achieved with soil solarization alone. Steam directly affects nematodes, killing them through a combination of moisture and heat. Both ASD and steam can be applied in ornamental and vegetable production. Results from a commercial-grower field trial that included both ASD and steam treatments on naturally occurring populations of RKN are presented. ASD was accomplished using molasses as an easily decomposable soil amendment to stimulate microbial activity. Composted poultry litter (CPL) was used as a nitrogen source and to increase bacterial diversity. Amendments were incorporated into the upper 20 cm of soil. The soil was covered with clear, UV-stabilized polyethylene and saturated with water to create conditions favorable for anaerobic bacterial growth. The polyethylene tarp was left in place for approximately 3 wk, until the soil temperature was elevated and anaerobic conditions were established, based on measurements using oxidation-reduction probes. Steam treatments were applied using standard 7.6 cm perforated tile (steam 1), and custom-drilled drain tile with 1.6 mm holes spaced every 3.8 cm approximately 120° from the center line of the tile (steam 2). Drain tile was buried at 35.5-cm depth and four tiles were installed in each plot. MeBr was included as a control and was applied under metalized film at 225 kg/ha (MeBr: chloropicrin 67:33). All plots were 30.5-m long and 1.8-m wide. The floriculture crops Sweet William (*Dianthus barbatus*), larkspur (*Delphinium x belladonna*), and snapdragon were produced according to standard commercial practices during the winter of 2011–2012. Both ASD and steam provided RKN control in soil at the end of the season comparable with, or exceeding that of MeBr. ASD and steam also reduced RKN populations in snapdragon roots comparable with, or exceeding control with MeBr. MeBr and ASD increased plant growth compared with both steam treatments for larkspur and snapdragon. Although steam provided excellent control of nematodes in this study, plant growth was reduced, indicating a possible deleterious effect of steam on beneficial soil microorganisms. Additional research on different organic amendments for use in ASD, and on supplementing or enhancing natural populations of beneficial soil microorganisms to improve plant growth following steam treatments is currently underway.

NAVIGATING THE PUBLICATION PROCESS FOR THE *JOURNAL OF NEMATOLOGY*. **Kokalis-Burelle, Nancy[1], I. Zasada[2], W.T. Crow[3], J.A. LaMondia[4], S.R. Stetina[5], and A. Westphal[6].** [1]USDA, ARS, Ft. Pierce, FL; [2]USDA, ARS, Corvallis, OR; [3]University of Florida, Gainesville, FL; [4]Connecticut Agricultural Experiment Station, Windsor, CT; [5]USDA, ARS, Stoneville, MS; [6]Julius Kühn-Institut, Münster, Germany.

The *Journal of Nematology* (JON) accepts manuscripts in several areas of nematology including taxomony/systematics, host parasite relations, ecology, biological and chemical control, management, entomopathogenic nematodes, resistance, and physiology. Manuscripts may be formatted as research reports, reviews, or notes. Senior authors should follow the rules of scientific ethics in determining authorship, and manuscripts should be properly formatted for the JON according to the Guide to Authors, available on the Society of Nematologists (SON) website. The manuscript submission and peer review procedures have recently changed to an online process using the Allen Press PeerTrack™ system. Specific details of navigating the PeerTrack™ system will be demonstrated in this workshop. The general PeerTrack™ workflow begins when authors establish a profile and submit a manuscript. Submitting authors are highly encouraged to also register to serve as JON reviewers. Upon submission of a manuscript through PeerTrack™, the Editor-in-Chief (EIC) determines if the subject matter is appropriate for JON, and if the manuscript is of sufficient quality and preparation to be considered for publication. If both criteria are met, the EIC assigns the manuscript to the appropriate subject Editor. The subject Editor then assigns the manuscript to two reviewers and is responsible for making the final determination to accept or reject the manuscript based on the recommendations of the reviewers. The Editors work with authors during the revision process to ensure that all reviewers' comments are considered as the manuscript is revised. The Editor then alerts the EIC when a manuscript is ready for publication. The EIC is responsible for ensuring that accepted manuscripts are ready for publication and for working with the publisher in the production of the journal. This includes submitting manuscripts and related figure files, journal front matter, table of contents, and cover material to the publisher. The EIC is also responsible for ensuring that galley proofs are sent to authors and corrections are made before final proof production. Following production of final proofs the EIC ensures that the journal is available online and to appropriate indexing databases. Members of SON will benefit from utilizing the new online publication system for rapid, broad dissemination of research findings without page charges.

MODE OF ACTION STUDIES ON FLUENSULFONE. **Kearn, James, Elizabeth Ludlow, Vincent O'Connor, and Lindy Holden-Dye.** Centre for Biological Sciences, University of Southampton, Southampton SO17 1BJ, UK.

Fluensulfone is a nematicide of the fluoroalkenyl thioether group that has significantly reduced nontarget toxicity relative to currently used chemical alternatives. The mode of action of fluensulfone is unknown. Here, we have tested whether or not

the model genetic organism *Caenorhabditis elegans* may be used to address this question. Studies in this free-living nematode permit an investigation at all life stages including the adult stage that for sedentary endoparasitic plant parasitic nematodes (PPNs) is resident in the host plant and therefore not readily accessible to study. Furthermore, the genetic tractability of *C. elegans* facilitates a molecular analysis of the impact of fluensulfone on nematode behavior. *C. elegans* is susceptible to the nematicidal effects of fluensulfone, similar to the effects observed on PPNs. More discrete behavioral analyses of the effects of fluensulfone have indicated pleiotropic actions encompassing an inhibition of development, egg laying, and egg hatching. Furthermore, as with the PPNs, the effects of chronic exposure on egg hatching and motility were irreversible. We also observed complex effects on pharyngeal pumping (feeding), locomotion, and responses to food. The effect elicited by fluensulfone was dependent on the dose that was applied to the worms, the duration of exposure and whether the assay was conducted in the presence or absence of food, *E. coli* OP50. For example, *C. elegans* that were placed on agar plates containing fluensulfone and OP50 exhibited an increase in locomotion within 1 hr of exposure, but after 24 hr, locomotion was inhibited. In contrast, when the assay was conducted in the absence of OP50 only an inhibition was observed, exemplifying the context-dependent effects of fluensulfone. This profile of effects of fluensulfone on *C. elegans* behavior is distinct from that of the anticholinesterase aldicarb and the macrocylic lactone ivermectin indicating that it has a different mode of action compared with other nematicidal and nematostatic chemicals. Analysis of *C. elegans* mutants that are resistant to aldicarb and ivermectin has provided further evidence to support this. The most potent effect of fluensulfone on *C. elegans* was on feeding behavior. In *C. elegans* this behavior is amenable to electrophysiological analysis by extracellular recordings from the mouth of the worm that captures the electrical activity of the pharyngeal system; the electropharyngeogram or EPG. The effect of fluensulfone was biphasic, with lower doses stimulating pharyngeal pumping and higher doses causing a profound inhibition. Further studies on *C. elegans* mutants are in progress to delineate the molecular mechanism(s) underpinning these responses to fluensulfone. Acknowledgements: James Kearn is a postgraduate student funded by Makhteshim Agan. We thank Robert Everich and Danny Karmon (Makhteshim Agan Group) for discussion and comments. *C. elegans* mutants were provided by the *C. elegans* Genetics Center (CGC), funded by NIH Office of Research Infrastructure Programs (P40 OD010440).

A GENOMIC STRATEGY FOR DETECTING VIRULENCE GENES IN *HETERODERA GLYCINES*, THE SOYBEAN CYST NEMATODE. **Lambert, Kris N.[1], S. Bekal[2], A.M. Fakhoury[2], and J.P. Bond[2].** [1]University of Illinois at Urbana-Champaign, 1102 South Goodwin Ave. Urbana IL 61801; [2]Southern Illinois University, Public Policy Institute, Carbondale, IL 62901.

The soybean cyst nematode (SCN) is the most damaging pathogen of soybean in the United States. The overuse of SCN resistance derived from PI88788 has allowed the accumulation of virulent SCN that can grow on most commercial soybean varieties. The goal of this project was to identify SNP markers linked to SCN virulence genes. To accomplish this objective, two inbred virulent and avirulent SCN strains were mated and used to generate an F2 SCN population and to form 20 SCN lines derived from the F2. The F2 derived lines were pooled into bulk populations and used to inoculate ten SCN resistant and ten SCN susceptible soybean plants. DNA was extracted from the bulk SCN that grew on the resistant and susceptible soybean. Both F2 DNA samples and those from the bulk SCN were genotyped using the panel of 1536 SCN GoldenGate SNPs. The SNP data was used to generate a draft SCN genetic linkage map and the bulk SCN samples were analyzed for an increase in frequency of SNPs derived from the virulent parent. Most of the SCN SNP calls for the bulk SCN populations were heterozygous as expected, but three SNPs showed a significant shift of frequency favoring the virulent SNP allele for the SCN populations growing on the resistant plants, thus defining these regions as potentially flanking SCN virulence genes. The future identification of virulence genes may allow the development of more durable SCN resistant plants and may help preserve valuable soybean germplasm.

SALT TOLERANCE OF *MELOIDOGYNE SPARTINAE* AND *M. HAPLA*. **LaMondia, James A.[1], and W.H. Elmer[2].** [1]Connecticut Agricultural Experiment Station, Valley Laboratory, 153 Cook Hill Road, Windsor CT 06095; [2]Department of Plant Pathology and Ecology, 123 Huntington St., New Haven, CT 06504.

Meloidogyne spartinae is a parasite of smooth cordgrass, *Spartina alterniflora*, with a distribution from Florida to Maine and has been associated with sudden vegetation dieback (SVD) of *S. alterniflora* in tidal marshes in Connecticut, Massachusetts, and Maine. No single cause has been determined for SVD, but *Fusarium* spp., root-knot nematodes, abiotic factors such as drought and salinity, and herbivory have all been associated with the syndrome. The role of each stressor and possible interactions of stressors in SVD have yet to be determined. The *S. alterniflora* grass and the pathogens in the intertidal marsh areas along marsh creek banks have greater exposure to a range of salinity than in other areas because of freshwater rainfall and drying of salt water. We previously determined that *M. spartinae* were more numerous in cordgrass roots and developed more quickly in the higher elevations of the intertidal zone. We conducted experiments to investigate the ability of *M. spartinae* to survive a wide range of salinity over time. *M. hapla* is closely related genetically to *M. spartinae* and was used in the same experiments for comparison. Juveniles of *M. spartinae* were collected after dissection of naturally infected *S. alterniflora* root galls and *M. hapla* juveniles were recovered from egg masses taken from greenhouse grown *Lobelia* roots

placed on pie pans. Juveniles of both species were placed in separate covered counting dishes in 5 ml of 0.0 (distilled water), 0.1, 0.3, 0.5, 0.7, and 1.0 M (roughly 1.6 × sea water) NaCl concentrations and held at ambient laboratory temperature. Nematode viability was determined after 1, 5, and 12 d by observation of motility. Nonmotile nematodes were probed with a pick to encourage movement when evaluating survival. There were two replicate dishes of each nematode and the first 10 juveniles observed per dish were counted as motile or not. The experiment was conducted three times. *M. hapla* survived best in distilled water and did not survive past 2-wk exposure to 0.3 M NaCl. Maximum survival of *M. hapla* was 100% of juveniles examined. *M. spartinae* survived at all concentrations tested for at least 12 d; maximum survival was 83.5% of juveniles examined, and survival was approximately 60% or greater in salinity ranging from distilled water to 0.5 M NaCl for all times tested, and up to 5 d at 0.7 M or 1 d at 1.0 M. These findings are consistent with the hypothesis that marine organisms in the upper tidal zone that are exposed to extremes in salinity must be able to osmoregulate to withstand a wide range of salinity.

COTTON VARIETIES AND NEMATICIDE COMBINATIONS FOR RENIFORM AND ROOT-KNOT MANAGEMENT IN ALABAMA. **Land, Caroline[1], K.S. Lawrence[1], D. Schrimsher[1], and C.H. Burmester[2].** [1]Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849; [2]Agronomy and Soils Department, Auburn University, Auburn, AL 36849.

Five nematicide combinations were evaluated for *Meloidogyne incognita* (Root-knot nematode) and *Rotylenchulus reniformis* (Reniform nematode) management on three cotton varieties at different locations across Alabama. The field sites were located at the Plant Breeding Unit (PBU) of the E.V. Smith Research Center near Tallassee and the Tennessee Valley Research and Extension Center (TVREC) near Belle Mina. The cotton varieties were treated with nematicide seed treatments. Temik 15G was applied at planting with granular hoppers attached to the planters and Vydate was applied as a foliar spray at the six- to eight-leaf stage. Nematode samples were taken at 45 and 85 d after planting (DAP) at the PBU and at 35 and 65 DAP at the TVREC. Statistically no interaction occurred between the varieties and the nematicides at either location. In both locations FM 1740 B2F supported greater stand counts compared with the Stoneville varieties; also seed cotton yield was significantly greater in the Stoneville varieties compared with the FM 1740 B2F. Root-knot nematode eggs per gram of root were consistently higher on the FM 1740 B2F variety compared with the Stoneville varieties. FM 1740 B2F was supported root-knot densities of 6,443 eggs per gram of root compared with the average of the two Stoneville varieties at 2,265 at 42 DAP, respectively. The two locations differ in the ranking of nematicide effectiveness. At the PBU, Temik 15G produced the greatest average seed cotton yield at 4,488 kg/ha in two of the three varieties followed by Aeris seed treatment that produced an average of 3,735 kg/ha. However, at the TVREC, Vydate CLV produced the greatest seed cotton yield at 3,446 kg/ha in two of the three varieties followed by Temik 15G at 3,067 kg/ha and then the seed treatments. Vydate CLV also decreased the number of reniform eggs per gram of root to 6,424 at 65 DAP compared with Temik 15G that only decreased numbers to 8,823 at 65 DAP. In 2012, the nematicides increased the seed cotton yields on two of the three varieties and did produce enough additional lint yields to pay for the additional nematicide investment.

RENIFORM NEMATODE MANAGEMENT USING SUNN HEMP COVER CROPPING AND POSTPLANT SOIL SURFACTANT APPLICATION. **Leleiwi, Ikaia[1], K.-H. Wang[1], and B.S. Sipes[1].** [1]Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822.

Recently multiple local sweet potato producers have made known their desire for reniform (*Rotylenchulus reniformis*) nematode management options. The following research addresses this problem in a sustainable manner via preplant sunn hemp (*Crotalaria juncea*) application and through the addition of a soil surfactant. We hypothesize that postplant application of soil surfactant will enhance the allelopathic effects of sunn hemp residue left from preplant cover cropping and soil incorporation. Anyhydrobiosis is a dormant state that reniform nematodes can adopt in response to desiccation, allowing the nematodes to withstand many control methods as well as a dry environment. Soil surfactant applications address this issue by promoting a wet soil environment and improving water penetration in the soil. We think that there will be fewer dormant nematodes right after sweet potato harvest, so it is hypothesized that this time would be best to incorporate the sunn hemp cover crop. A 2 × 2 × 2 factorial greenhouse experiment (sunn hemp × surfactant × soil history) was conducted to (i) find the optimum incorporation time for a sunn hemp cover crop in a sweet potato cropping system, and (ii) determine if a soil surfactant aids in nematode suppression. All pots were either filled with sweet potato field soil that had just been harvested or from soil that had been fallow for a year. Half of the pots from each soil type were amended with dry sunn hemp foliage (1% w/w) and half were left without sunn hemp. All of the pots were planted with a single sweet potato slip, at which time half of the pots were then drenched with Transformer soil surfactant (a.i. 19.5% alcohol ethoxylate) at 1 gal/acre. Plants will be grown for 4 mon at which time storage tubers will be harvested, weighed, and graded using the U.S. grading system. Soil samples (250 cm^3) were taken from each soil type at the beginning of the experiment and will be taken from an aggregate sample of each treatment at the end. Nematodes will be soil extracted by elutriation and centrifugal flotation and plant-parasitic nematode populations will be counted using an inverted microscope. From the results gathered, active and dormant reniform nematode population densities are discussed.

PHOTOGRAPHIC BASICS FOR SHOOTING AND EDITING AGRICULTURE PHOTOGRAPHS. **Lawrence, Katherine.** Office of Agricultural Communications, Box 9625, Mississippi State, MS 39762.

Photography basics for shooting and editing photos in an agriculture setting include camera selection, settings, and lens selection, picture composition, lighting, and editing of photographs for use in publications, power point presentations, and media relations. Photography has become easily and more readily available with the conception of digital photography. It is commonplace now to find a camera on all hand-held devices such as cell phones or tablets. Although taking photos may be simple enough, having high-quality photos for use in publications and/or presentation is another story. Basic photo shooting techniques and camera settings are pertinent in capturing high-quality photographs. One of the most important settings when shooting photos for later use is the photo image size and quality. This directly affects how a photo may be used. For any print media, large high-resolution images are required. This means the dots per inch (dpi) of the image must be 300 or higher. These settings can be made directly in camera. Another factor to shooting better images is the camera and lens being used to shoot the photos. DSLR cameras will always give better images than Point-n-Shoot. Lastly, it is important to know how to compose the image. Composing a better image in camera saves time when editing the photograph. Before any image can be published, it must go through an editing process, either by ones self or a photo editor. There are a few programs that can be purchased for this, such as Adobe Photoshop or Lightroom, free programs such as Picassa and Gimp. Basic color correction and cropping should be done in a photo-editing program instead of in camera. These programs allow the user to do things such as cloning out unwanted material, sharpening, dodging and burning, and basic touch-ups. These few basic photographic concepts, when used, provide a more professional quality photograph.

MCW-2 (FLUENSULFONE) A NEW, NOVEL NEMATICIDE: TECHNOLOGY, PERFORMANCE AND REGISTRATION UPDATE. **Long, Dennis, and Herb Young.** Makhteshim-Agan North America, Inc., 3120 Highwoods Boulevard, #100, Raleigh, NC 27604.

MCW-2 (fluensulfone) is a novel broad spectrum, nonfumigant nematicide in global development by Makhteshim-Agan. It belongs to a new chemical class (fluoroalkenyle) and acts by new, yet unknown mode of action. The mode of action is distinct from that of organophosphates, carbamates, and avermectins that represent the vast majority of nematicides used today. MCW-2 is a true nematicide: it kills nematodes through contact causing irreversible nematicidal activity rather than temporary nematostatic activity as seen with organophosphates and carbamates. In the past 5 yr, MCW-2 has been extensively tested all around the world by university researchers, governmental trial stations, private contract researchers, as well as by Makhteshim-Agan team members. Hundreds of trials have been conducted from petri dish in the lab, small plot replicated trials to full-scale grower trials. These trials have been conducted across 23 different countries over all continents in multiple crops and various nematode species. MCW-2 has a superior tox and ecotox profile and will have a signal word of CAUTION on the label. It is currently under global joint review. Registration timelines will be discussed. Petitions currently at EPA are for Fruiting Vegetables and Cucurbits Crop Groups.

EVALUATION OF SOYBEAN FIELDS INFESTED WITH *HETERODERA GLYCINES* AND *MACROPHOMINA PHASEOLINA* IN SOUTHERN OHIO. **Lopez-Nicora, Horacio[1], Anne Dorrance[2], and Terry Niblack[1].** [1]Ohio State University, Department of Plant Pathology, Columbus, OH 43210; [2]Ohio State University, Department of Plant Pathology, Wooster, OH 44691.

Macrophomina phaseolina, causal agent of charcoal rot, is affecting soybean production in areas where it was not previously known to occur. *Heterodera glycines*, the soybean cyst nematode, is a known soybean pathogen throughout the soybean production region of North America. Both pathogens reduce soybean yields more under hot, dry conditions than those in which moisture is not limiting. The objective of this study was to evaluate soybean production in fields infested with low initial population densities of these soilborne pathogens to assess their population dynamics at the beginning of an epidemic. Two field sites (I and II), naturally infested with *H. glycines* and *M. phaseolina,* were planted to soybean in Pike County, OH. Thirty days after planting, a 76.2- × 76.2-m grid was measured and flagged in 7.62- × 7.62-m squares in both fields. Fifteen to twenty soil cores were collected at each grid in both fields on 20 June and 29 September 2012 for initial (Pi) and final (Pf) *H. glycines* egg and *M. phaseolina* CFU counts, respectively. Twenty-five plants were collected and hand-threshed from every grid for yield analysis. In average, *M. phaseolina* density, measured in CFU/g soil, was higher at initial sampling and lower at final sampling for both fields. Reproduction factor (Pf/Pi) for *H. glycines* was greater than one in both fields. Yield reduction because of *M. phaseolina* and *H. glycines* Pi was not observed in either field. Yield was significantly reduced in both fields with increasing *H. glycines* Pf. The same was observed in field II for *M. phaseolina* Pf, which on average was higher than in field I. Evaluation of Pf/Pi indicated increases in inoculum for next season. Prediction of inoculum levels with respect to damage thresholds is essential for cultivar selection and long-term disease management in infested fields.

EFFECT OF MOVENTO PROGRAMS ON COLUMBIA ROOT-KNOT NEMATODE IN THE PACIFIC NORTHWEST. **Luff, Kelly[1], S.L. Hafez[2], and R. Portenier[2].** [1]Bayer CropScience, 3554 East 4000 North, Kimberly, ID 83341; [2]University of Idaho, Parma Research and Extension Center, 29603 U of I Lane, Parma, ID 83360.

Several experiments were conducted in Idaho and Washington during 2011 and 2012 to determine the efficacy of various treatments and combinations of Movento, Vydate, and Vapam on Columbia root-knot nematode. Results from the majority of

these studies show that Vapam alone or Movento in combination with Vydate or with Vapam + Vydate produced the greatest total yield. Movento applied twice, according to label instructions, provided significant suppression of tuber damage but was not sufficiently effective as a stand-alone nematicide. Vapam combinations with Movento and Vydate resulted in the lowest percentage of infected tubers and provided better performance as compared with any of these products alone. Replacing foliar applications of Vydate with Movento resulted in similar percentage of tuber infection as compared with a Vydate foliar program. In conclusion, Movento is a valuable tool that can be used in a nematode management program, providing potato grower the added benefit of aphids, psyllids, whiteflies, mites, thrips larvae, and wireworm activity.

YIELD LOSS OF CORN DUE TO *PRATYLENCHUS SPP*. ESTIMATED BY A NESTED ERROR COMPONENT MODEL. **MacGuidwin, Ann, and B. Bender.** Department of Plant Pathology, University of Wisconsin, Madison, WI 53705.

Root lesion nematodes, *Pratylenchus* spp., are common pests of corn in all soil types and cropping systems in the north central United States. Infection by root lesion nematodes does not always cause identifiable symptoms so yield loss estimates are based on nematode population densities rather than injury. All life stages of *Pratylenchus* spp overwinter and can feed as ecto- or endoparasites within corn roots so there are many opportunities to estimate nematode population densities in soil and roots throughout the life of the crop. We used multiple error component models with unbalanced panel data to describe the relationship between grain yield loss and the population density of root lesion nematodes from soil samples collected on two dates and seminal root and adventitious root samples collected on five dates. The data were from irrigated fields bulk planted with corn at the Hancock Research Station in 2008, 2009, or 2010 representing six field-years. The fields were similar for soil type (Plainfield loamy sand) and presence of *P. penetrans*. Year and field nested within year were specified as random effects in the model to account for variability because of weather and site-specific factors such as corn genotype and field history. Based on the significance level of tests for the estimated slope coefficients and the timing and ease of sample collection, the best of the four significant models is based on soil samples collected at plant growth stage V1/V2. This model implies a 0.0137% yield loss for every nematode recovered from 100-cm^3 soil and roots therein. The mean value for the data set used to construct the model was 201 nematodes per 100-cm^3 soil, suggesting an average yield loss of 2.75%. The experimental error component of the variance was very large, suggesting this damage function is more appropriate for broad-scale estimates of root lesion nematode damage to corn grown in irrigated loamy sand soils than it is for making management decisions. We speculate that an estimate of 2.75% yield loss because of nematodes is conservative given that the corn fields we studied were managed using best practices and achieved yields above the county average.

DEVELOPMENTAL RESPONSES OF *HETERODERA GLYCINES* AND *MELOIDOGYNE INCOGNITA* TO FUNDAMENTAL ENVIRONMENTAL CUES. **Masler, Edward P., S.T. Rogers, and D.J. Chitwood.** Nematology Laboratory, U.S. Department of Agriculture, Beltsville, MD 20705.

Response of plant-parasitic nematodes to the environment involves metabolic and developmental mechanisms that can present novel targets for nematode control. One of the most fundamental environmental cues is temperature. Exposure of *Heterodera glycines* and *Meloidogyne incognita* eggs to low temperature decreased hatch by 50% ($P < 0.05$) in each species. This reduction was associated with developmental arrest at the J1 stage. Reports suggest that levels of catechin polyphenols present in plant-parasitic nematode cysts increase during cold seasons when hatch is low. We questioned whether our low temperature results, particularly with *H. glycines*, might be associated with such polyphenols. A series of eight catechin analogs (catechin, epicatechin, gallocatechin, epigallocatechin, plus the gallated forms of each) was screened against *H. glycines* and *M. incognita* eggs in vitro to assess effects on hatching. Responses of the two species were quantitatively and qualitatively different. Where *H. glycines* hatch was inhibited ca. 25% ($P < 0.05$) by each of the analogs, with no quantitative differences among them, *M. incognita* hatch was inhibited no less than 25% ($P < 0.05$) by any of the analogs, and as much as 90% by the gallated forms. This included the ubiquitous plant polyphenol, epigallocatechin gallate (EGCG). Significant reductions ($P < 0.05$) in hatch were observed by 7 d after exposure in *H. glycines* eggs to 1 mM EGCG, but as early as 5 d in *M. incognita*. Unlike low temperature exposure, hatch inhibition by EGCG is associated with the presence of un-hatched, but viable, J2. This effect is reversed by removal of EGCG and replacement with water. Since EGCG can inhibit mammalian protease activities, we examined the effects of 1mM preparations of EGCG and the seven other catechin analogs on *H. glycines* and *M. incognita* general and specific (chymotrypsin-like, CTL) proteases. General and specific activities were each inhibited more, in each species ($P < 0.05$), by the gallated form of the polyphenol analog than by the corresponding nongallated form. However, species differences were revealed with the CTL substrate, where EGCG inhibited *M. incognita* activity by 75%, significantly greater ($P < 0.05$) than *H. glycines* (45%). We also examined a heat stable component (CE) from *H. glycines* cysts and found that it inhibited nematode protease activities at the equivalent of 0.6 cysts/μl in reactions. Again, *M. incognita* activity was more susceptible to inhibition (92%) than *H. glycines* activity (53% inhibition; $P < 0.05$). The nature of this inhibitor is being examined. Possible connections among environmental cues, plant and nematode sources of regulatory molecules, development and behavior are discussed, and the use of these associations in discovering novel control strategies is presented.

MELOIDOGYNE SPP. POPULATIONS FROM CERRADO AND CULTIVATED AREAS: GENETIC VARIABILITY AND AGGRESSIVENESS TO SOYBEAN. **Mattos, Vanessa da Silva[1,2], J.G.P. Silva[1,2], C. Furlanetto[2], F.R. Sousa[1], A. Jorge-Junior[1], M.R.A. Almeida[1], A.W. Moita[1], P. Castagnone-Sereno[3], and R.M.D.G. Carneiro[1].** [1]Embrapa Recursos Genéticos e Biotecnologia, CEP 70849-970, Brasília, DF, Brazil; [2]Universidade de Brasília, Departamento de Fitopatologia, 70910-900, Brasília, DF, Brazil; [3]INRA/UNSA/CNRS, UMR1301, BP167, 06330 Sophia Antipolis, France.

Conversion to intensive agriculture to meet rising demand for soybean, sugarcane, and livestock products is rapidly occurring over vast areas of the 1.8 million km^2 Brazilian Cerrado, a region that is a natural mosaic of vegetation, climates and soils. Despite the vast biodiversity of the Cerrado, there are few studies focusing on nematodes, especially those of the genus *Meloidogyne*. The purpose of this study was to compare genetic variability and aggressiveness of *Meloidogyne* populations (*M. javanica*, *M. incognita* and *M. morocciensis*) from different physiognomic vegetation of Cerrado and from soybean cropping areas. DNA amplification was performed using 7 AFLP and 30 RAPD primers. Amplified fragments were separated on a 1.5% agarose gel and scored as present or absent from the digitized photographs of the gels. DNA fingerprints of each population were converted into a binary matrix for each of the two marker types. Cluster analyses of this matrix separated *Meloidogyne* species, Cerrado populations, and populations from cultivated areas. The intraspecific variability in *M. javanica*, *M. morocciensis*, and *M. incognita* represented 26.7%, 16.6%, and 15.3% of the polymorphic fragments, respectively. The aggressiveness experiment was conducted under greenhouse conditions using a factorial design of five soybean cultivars, six nematode populations, and eight replications; each plant was inoculated with 5,000 eggs of a given population. Sixty days after inoculation, the nematode reproduction factor (RF) was determined. Populations of *Meloidogyne* spp. from Cerrado and cultivated areas showed similar aggressiveness, except for *M. morocciensis*, whose population from soybean fields was more aggressive than the Cerrado population.

ABUNDANCE AND FREQUENCY OF NEMATODES IN FERRALSOL, LITHOSOL AND NITOSOL SOIL GROUPS IN GHANA, KENYA AND MALAWI. **Maung, Zin Thu Zar[1], S. Yildiz[1], T. Teal[2], J. Gronseth[3], C. Kwoseh[5], T. Adjeigyapong[5], V. Saka[6], M. Lowole[6], G.N. Karuku[4], P.M. Wachira[4], J.W. Kimenju[4], J. Qi[3], T. Schmidt[7], and H. Melakeberhan[1].** [1]Agricultural Nematology Lab, Department of Horticulture; [2]Microbiology and Molecular Genetics; [3]Center for Global Change and Earth Observations, Michigan State University, East Lansing, MI 48824; [4]University of Nairobi, Kenya; [5]Kwame Nkrumah University of Science and Technology, Ghana; [6]University of Malawi, Malawi; [7]Departments of Microbiology and Immunology and Ecology and Evolutionary Biology, University of Michigan, Ann Arbor, MI 48109.

Degrading soil health, which leads to low agricultural production, accelerated food insecurity, global climate change, habitat and biodiversity loss, and finally forcing population migration, is a major challenge in sub-Saharan Africa (SSA). In order to reduce soil health degradation in SSA, an integrated understanding of soil ecosystem services, for which nematode assemblage analyses is a key indicator, will be helpful. Thus, in 2012, a study was conducted with the objectives to compare the nematode community compositions of selected soil groups, regions and landscapes, and to determine the relationships between soil groups and landscapes of Ghana, Kenya, and Malawi. Soil groups (Ferralsol, Lithosol, and Nitosol) were classified and located based on FAO 1974 soil classification and Google Earth data sets. We collected 512 soil samples from two to three fields each under natural and disturbed landscapes in north and south regions, approximately 50 to 300 km apart within the country. Nematodes were identified to genus level and assigned into five trophic groups (herbivores, bacterivores, fungivores, predators, and omnivores). Nematode assemblages were analyzed and compared among soil groups, regions, and landscapes of each country. Among the three countries, the number of herbivores, bacterivores, and fungivores in Malawi was the highest although no significant difference was observed for predators and omnivores. The frequency of occurrence of herbivores was found to be the lowest in Malawi, whereas that of bacterivores and fungivores was the highest among the countries. In Nitosol soils of Kenya and Malawi, herbivores were predominant. In Ghana, the abundance of bacterivores in Ferralsol soil were found the highest; however, no significant difference was observed in Kenya and Malawi soils. Bacterivores and omnivores in north Ghana were significantly higher than those in the south. Numbers of herbivores and bacterivores in Kenya and omnivores in Malawi were significantly higher in the south. In general, nematode populations under natural landscape were higher than under disturbed landscape except omnivores in Kenya and bacterivores in Malawi. The variation of abundance and occurrence frequency of trophic groups found in different soil groups, landscapes, regions, and/or countries indicated the need to reflect the biological facts when considering proper soil amending to improve soil health condition of SSA. Significant interactions of independent variables (soil groups and landscapes) affected nematode assemblages that indicated that the same soil group may have different biological structures and/or functions when land use practices are changed.

NEMATODE COMMUNITY ANALYSES TO ASSESS THE FOOD WEB STRUCTURE AND ECOLOGICAL DISTURBANCES IN FERRALSOL, LITHOSOL, AND NITOSOL SOILS UNDER DIFFERENT LANDSCAPES IN GHANA, KENYA, AND MALAWI. **Maung, Zin Thu Zar[1], S. Yildiz[1], T. Teal[2], J. Gronseth[3], C. Kwoseh[5], T. Adjeigyapong[5], V. Saka[6], M. Lowole[6], G.N. Karuku[4], P.M. Wachira[4], J.W. Kimenju[4], J. Qi[3], T. Schmidt[7], and H. Melakeberhan[1].** [1]Agricultural Nematology Lab, Department of Horticulture; [2]Microbiology and Molecular Genetics;

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Low agricultural production, which is mainly driven by soil degradations, results in conflicting agronomic, ecological, biological, and economic outcomes of sub-Saharan Africa (SSA). To promote agricultural production of the region, it is important to define the levels of soil degradation and types of management options. The objective of this study was to determine if ecological indices are able to differentiate the type and magnitude of disturbance in Ferralsol, Lithosol, and Nitosol soils of Ghana, Kenya, and Malawi. Nematodes were extracted from soil samples collected in undisturbed and disturbed landscapes of selectedsoil groups in northern and southern regions of Ghana (n = 152), Kenya (n = 192), and Malawi (n = 168). Nematodes were identified, assigned into different trophic groups, and colonizer-persister (c-p) values of 1 to 5 were given. Maturity (PPI, MI, MI 2-5, ΣMI, ΣMI 2-5, FI), diversity (H', N0), enrichment (EI), and structure (SI) indices of nematodes in each soil group, region, and landscape within each country were derived. Nematode maturity indices across all countries, soil groups, and landscapes showed that Malawi and Kenya soils were more disturbed than the Ghana soils. In Kenya, Nitosol appear to be less disturbance than Ferralsol and Lithosol, natural than disturbed landscapes, and northern than southern region. In Malawi, Ferralsol appear to be more disturbed than Nitosol, disturbed than undisturbed, and northern than in southern region. Fertility index showed that all the soil groups had similar condition and Kenya and Malawi soils were found within the zone of nutrient disturbance (FI = 0.8 to 1.2). The diversity, both species richness (N0) and Shannon index (H'), was significantly lower in disturbed landscape when compare with natural condition in all countries. In Ghana, Ferralsol and Lithosol were depleted and highly structured in both landscapes, whereas, Nitosol was structured and showed some level of enrichment. In Kenya, the soils were depleted and less structured in both landscapes. In Malawi, the undisturbed landscapes appear to be structured, whereas, those of disturbed landscapes were completely unstructured. The food web structure of all soil groups both under natural and disturbed conditions in the three countries were found similar levels of depletions. Overall, the food web structure of the all three soil groups both under natural and disturbed conditions in all countries were found to have similar levels of depletions to meet the agroecosystem demands, suggesting the need for proper management applications.

TYPES OF BIOLOGICAL AND NUTRITIONAL DEGRADATIONS IN FERRALSOL, LITHOSOL AND NITOSOL GROUPS IN GHANA, KENYA AND MALAWI. **Melakeberhan, Haddish[1], Z.T.Z. Maung[1], S. Yildiz[1], T. Teal[2], J. Gronseth[3], C. Kwoseh[5], T. Adjeigyapong[5], V. Saka[6], M. Lowole[6], G.N. Karuku[4], P.M. Wachira[4], J.W. Kimenju[4], J. Qi[3], and T. Schmidt[7].** [1]Agricultural Nematology Lab, Department of Horticulture; [2]Microbiology and Molecular Genetics; [3]Center for Global Change and Earth Observations, Michigan State University, East Lansing, MI 48824; [4]University of Nairobi, Kenya; [5]Kwame Nkrumah University of Science and Technology, Ghana; [6]University of Malawi, Malawi; [7]Departments of Microbiology and Immunology and Ecology and Evolutionary Biology, University of Michigan, Ann Arbor, MI 48109.

This project is part of a collaborative work toward understanding the biological basis of sub-Saharan Africa (SSA) soil degradations for purposes of developing suitable soil health management strategies that lead to alleviating food insecurity and improving ecosystem health. The objective was to determine the types of biological, nutritional, and physiochemical degradations and nutrient cycling properties of Ferralsol, Lithosol, and Nitosol Soil Groups in rural Ghana, Kenya, and Malawi. More than 500 soil samples were collected from each soil group in two regions of each country separated by 50 to 300 km (north-south) and two to three fields of disturbed (agricultural) and undisturbed landscapes. A combination of traditional soil property, nematode assemblage, and microbial community analyses were conducted to determine the status of the soil groups in the respective countries. Regardless of soil group or landscape, the types of nutritional and physiochemical degradations observed at below-normal and/or present at trace-levels included: soil pH in Ghana and Malawi, organic matter (%OM), and organic nitrogen (%N), and available nitrogen (NO_3-N + NH_4-N), in Kenya and Malawi, and P, K, Ca, Mg, and CEC in Ghana. On the other hand, the types of biological degradations included lower nematode and microbial community diversity in disturbed than in natural landscapes, disturbed ecological and soil fertility indices, and diminished soil food web structure. The relationships between soil community profile (as affected by stress or disturbance = Structure Index) and function (as measured by the turnover of opportunistic bacteriovore and fungivore nematodes = Enrichment Index) across regions and landscapes indicate that the soil food web status is too depleted to meet agroecosystem demands. A combination of the nutritional and biological degradations suggests that the structural and buffering capacity deficits are unlikely to be fixed without building up soil organic matter. The study shows that these soils are indeed very fragile, a significant step forward in our understanding of what types of treatments may be needed, where and at what levels to remedy identified soil problems that will eventually lead to formulating integrated and transformative policies across sectors (economic, social. and health).

HOST STATUS OF SELECTED PEPPER GENOTYPES TO *MELOIDOGYNE FLORIDENSIS* AND *M. JAVANICA* RACE 3 POPULATIONS FROM FLORIDA. **Mendes, Maria L.[1], D.W. Dickson[1], and J.A. Thies[2].** [1]University of Florida, P.O. Box 110620, Gainesville, FL 32611; [2]U.S. Vegetable Laboratory, USDA, ARS, 2700 Savannah Highway, Charleston, SC 29414.

Nine pepper (*Capsicum annuum*) genotypes were evaluated for their reactions against a population of *Meloidogyne floridensis* and a population of *M. javanica* race 3 from Florida. The pepper genotypes were 'CM-334,' 'Olympus,' 'Paladin,' 'Keystone Resistant Giant,' 'California Wonder,' 'Yolo Wonder,' 'Jupiter,' 'Charleston Belle,' and 'Carolina Wonder.' Tomato 'Rutgers' was included as a standard susceptible host. The plants were inoculated with 5,000 eggs and/or second-stage juveniles of either species and grown in a glasshouse. Ninety days after inoculation the plants were uprooted, and the gall (GI) and egg mass (EMI) numbers, final population (Pf = number of eggs per root system), and the reproductive factor (Rf = Pf/Pi) per root system were determined. All genotypes were resistant to *M. javanica* race 3 (GI, EMI, Pf, and Rf = 0). *Meloidogyne floridensis* did not infect 'CM-334,' 'Olympus,' and 'Paladin' (GI, EMI, Pf, and Rf = 0). All other genotypes were highly susceptible to *M. floridensis*. However, 'Keystone Resistant Giant,' 'California Wonder,' and 'Yolo Wonder' exhibited no visible galls but were heavily infected (EMI > 100). 'Jupiter,' 'Charleston Belle,' and 'Carolina Wonder' were severely galled (GI > 100) and showed high nematode reproduction (EMI > 100). The numbers of eggs per root system ranged from 45,520 for 'Keystone Resistant Giant' to 220,720 for 'Carolina Wonder' and reproductive factor ranged from 9.10 for 'Keystone Resistant Giant' to 44.14 for 'Carolina Wonder'. Tomato was highly susceptible to both *M. floridensis* (Pf = 611,093; Rf = 122.22) and *M. javanica* race 3 (Pf = 267,786; Rf = 53.56).

EFFECTS OF FUNGICIDES AND INSECTICIDES ON *BELONOLAIMUS LONGICAUDATUS*. **Mengyi, Gu, and W.T. Crow.** Entomology and NematologyDepartment, University of Florida, Gainesville, FL 32611.

Belonolaimus longicaudatus (Sting nematode) is considered the most destructive plant parasitic nematode on golf course turf in Florida. However, effective management options for this nematode on golf courses are few. Our research sought to identify insecticides or fungicides currently on the market that might be useful for management of *B. longicaudatus* on turf. We evaluated commercial formulations of four insecticides (abamectin, dinotefuran, imidacloprid, and spirotetramet) and two fungicides (iprodione and thiophanate methyl) in greenhouse trials on creeping bentgrass inoculated with *B. longicaudatus*. Each pesticide was evaluated at three rates, and was compared with untreated controls. Four-inch-diam. clay pots were filled with 400 cm^3 of sand and seeded with bentgrass. After germination, 60 mixed life stages of *B. longicaudatus* were inoculated into each pot. Pots were arranged in a randomized complete block design on a greenhouse bench with five replications for each treatment. Two weeks after inoculation treatments were either applied as a drench in 50 ml of water or were sprayed onto the pots following addition of 50 ml water (spirotetramet only). Untreated pots received 50 ml of water. Two weeks after treatment applications *B. longicaudatus* were extracted from the entire soil volume of each pot and counted. Orthogonal contrasts and regression analysis using posttreatment *B. longicaudatus* population densities were used to identify effective treatments. Of the pesticides evaluated, abamectin, spirotetramet, thiophanate methyl, and iprodione reduced population density of *B. longicaudatus* compared with the untreated control. These four pesticides will be included in future field efficacy trials to determine if these results are replicable under field conditions.

THE HUNT FOR *HETERORHABDITIS* IN HAWAII. **Myers, Roxana[1], B.S. Sipes[2], and R.G. Hollingsworth[1].** [1]USDA ARS Pacific Basin Agricultural Research Center, 64 Nowelo Street, Hilo, HI 96720; [2]Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822.

A survey was conducted to collect *Heterorhabditis* spp. in natural environments in Hawaiian soils. Quarantine laws currently prevent the importation of commercial isolates of *Heterorhabditis* into the State of Hawaii for biological control. Documenting natural populations would strengthen the case for the introduction of these organisms and could lead to the discovery of more virulent, better-adapted tropical isolates. Previous surveys in Hawaii showed endemic *Heterorhabditis* populations exist primarily in coastal areas so collection sites at sea level were made. Soil was collected between 5 to 30 cm deep from beneath vegetation within 100 m of the shoreline. Soil samples were placed in a 473-cc plastic container and baited with six *Galleria mellonella*. After 1 wk, morbid larvae were rinsed, placed on white traps, and monitored for emerging nematodes. Of 124 sites sampled, morbid larvae were recovered from 68 samples and 24% of these sites contained some type of entomopathogenic nematode based on morphological observation. PCR was conducted on single EPN specimens using primers to amplify the ITS regions. The resulting products were sequenced and the ITS rDNA sequences compared with existing entries in GenBank. Two specimens were confirmed to be *Heterorhabditis indica*. Further morphological and molecular analysis will be conducted on other specimens tentatively identified as new species of *Heterorhabditis* and *Heterorhabditoides*. Penetration and infectivity assays of promising isolates will aid in determining if these natural isolates are better adapted to tropical environments than current commercially available isolates.

STOP THE CULTIVATION AND THE NATURAL WEB OF SOIL COMMUNITIES RETURNS: IMPACTS OF DISEASE. **Neher, Deborah A.** Department of Plant and Soil Science, University of Vermont, Burlington, VT 05405.

Reduced or no-tillage leaves most of the previous crop residue on the soil surface, and this results in changes in the physical and chemical properties of the soil. Structure of physical habitat affects resource distribution. Further, plants exert control over microbial communities in the rhizosphere, and crop residues impact microbial community composition in the no-till litter. A case study is presented to illustrate natural, but easily disturbed, suppressive soils to afford insights into managing

soybean cyst nematode, *Heterodera glycines* (SCN). Composition and function of nematode communities were characterized in two fields naturally suppressive to SCN in Waseca County, Minnesota, where soybean was grown continuously for more than 15 yr under a no-tillage system. Treatments were chosen to identify management practices that disrupt natural suppression of SCN. Cultivation, application of biocides, and rotation to corn all reduced suppression of SCN, and the impact increased progressively within the 4 yr of the experiment. This result demonstrates that whereas the continuous soybean no-till system is stable, even relatively mild disturbances, such as corn rhizospheres and crop residue, destabilize this system. Cultivation decreased abundance of plant-parasitic and fungivorous nematodes and increased abundance of bacterivorous nematodes. Among plant-parasitic nematodes, the proportion that was *Helicotylenchus* correlated negatively with *Heterodera glycines*. There appears to be some association between *Heterodera glycines* and *Helicotylenchus* in the rhizosphere, perhaps competing for space and/or nutrients. When soybean was rotated to corn, the relative abundance of fungivorous nematodes (especially *Aphelenchoides*) increased. Values of MI25 and trophic diversity indices correlated positively with SCN suppression suggesting that complex soil communities with later succession are indicators of natural disease suppression. Suppression of nematodes appeared limited to SCN and likely surrounding the soybean rhizoshere, but there was no evidence that suppression impacted free-living nematodes outside the rhizosphere. Natural suppressive soils contained greater activity of aminopeptidases associated with collagen. Mannase and arabinase appeared to be related to crop rotation, reflecting different proportions of carbohydrate monomers in the cell wall of corn than soybean. Our results suggest that disease suppression appears to be aligned more closely with fungi than bacteria. Natural suppression of SCN appears to be associated with the microbial community fostered by a combination of no-till and soybean monoculture, and that suppression is easily destabilized by any alteration in the community fostered by soybean monoculture.

NEMATODE MANAGEMENT AND SOIL FUMIGANT RESEARCH : PREFUMIGATION SOIL MOISTURE CONDITIONS. **Noling, J.W.[1], Marjie Cody[1], Danny Johns[2], Steven Lands[3], and Mark Warren[4].** [1]University of Florida, IFAS, Citrus Research and Education Center, Lake Alfred, FL 33850; [2]Potato grower and Owner Blue Sky Farms, Hastings, FL 32145; [3]Courtesy Extension Agent I, St. Johns County Cooperative Extension Service, St. Augustine, FL 32086; [4]Courtesy Extension Agent I, Flagler County Cooperative Extension Service, Bunnell, FL 32110.

In two Hastings, FL, commercial potato field experiments, moisture gradients (dry, semi-wet, wet) were established before shank application of four different soil fumigants (Telone II (61 l/ha), Telone C17 (70 l/ha), Telone C35 (80 l/ha), PicClor 60 (94 l/ha) using multiple tanks of water from a tractor drawn watering tank and drenching boom. Beginning the following day, soil air concentrations of 1,3-D at two depths (6 and 12 in.) and two bed locations (bed center, bed shoulder) were monitored until near complete disappearance from soil. Once the soil dissipation curve for a given fumigant treatment, depth, and bed location was mathematically described, the area under the dissipation curve (AUDC) was calculated to determine cumulative fumigant dosage. In addition to the study of the importance of prefumigation soil moisture conditions, other experimental objectives included evaluations of different fumigant formulations and chloropicrin use rates per acre on potato yield and tuber quality. A significant increase in tuber yield was not observed in either study above a chloropicrin use rate of 15 kg/ha. At the AS farm, under very wet conditions, gravimetric soil moisture content above 12% degraded yield performance of all of the different soil fumigants in a high pressure Corky ringspot (CRS) field where a CRS susceptible potato variety (Red Lasoda) was planted. At the AS farm, Telone C17 under the driest soil moisture regime (12%) produced the highest marketable yield. Under the drier soil conditions at the other commercial DJ farm site, it was the wettest treatment (10.3% soil moisture), which produced the highest fumigant dosages (AUDC) and highest potato yields. At the DJ site, overall fumigant dosages were low and all of the different fumigants disappeared from soil very rapidly. With a CRS resistant yellow cultivar (Fabula), Telone II, a fumigant containing no chloropicrin produced the highest yield and was generally as good as or better than any of the other Telone Chloropicrin mixtures for producing high levels of marketable potato yield. These preliminary studies demonstrate the importance of at least 10% to 12% gravimetric soil moisture content at the time of soil fumigant application. For a typical Florida fine sandy soil, this corresponds to a level of 50% total available water. Under wet conditions, fumigant escape from soil was delayed (higher AUDC), cross bed fumigant movement was retarded and overall effectiveness of fumigant treatment reduced. Under dry conditions, fumigant movement was rapidly upward without significant lateral movement toward the shoulder of the compressed bed.

SUMMARY OF METHYL BROMIDE ALTERNATIVES RESEARCH IN FLORIDA STRAWBERRIES. **Noling, J.W., and M. Cody.** University of Florida, IFAS, Citrus Research and Education Center, Lake Alfred, FL 33850.

The primary focus of the 2007–2012 USDA ARS Areawide funded project was to evaluate cultural and chemical alternatives to methyl bromide soil fumigation within Florida strawberry production. Many different fumigants were evaluated individually or in combination with other herbicides to compare with methyl bromide. Relative yield values were averaged across all years and experimental locations to derive an experiment wide comparison of crop yields using specific chemical treatments and methyl bromide. Strawberry yields were often increased to the near equivalence if not higher than that of methyl bromide using combinations of one or more fumigants such as methyl iodide, chloropicrin, metam sodium or potassium, 1,3-D, and dimethyl disulfide. Regardless of application method, pest control efficacy for the alternative

fumigants was generally a little less than that of methyl bromide and more highly dependent on uniform delivery and distribution. Unlike for methyl bromide, prevailing soil and climatic conditions pre- and postfumigation were important determinants of efficacy and crop response with the alternative chemicals. Therefore, some inconsistency is unavoidable. This work has shown that tank mix applications of various preemergent herbicides are generally needed to effectively broaden the spectrum of weed control. With increasing cost and decreasing availability of methyl bromide, the chloropicrin content of the formulation increased during the 5-yr USDA ARS Areawide project period with transitions from formulations of 98/2 to 67/33 and to 50/50, which was generally considered inadequate for yield, nematode, or weed control efficacy. The problem with this changing reference standard is that, compared with methyl bromide chloropicrin 50/50, every other treatment looks good in terms of strawberry yield potential. Is it for this reason that we continue to hear that there are many effective methyl bromide alternatives, particularly when studies are conducted in fields without pest pressure. Relative strawberry yields were often near equivalent regardless of fumigant treatments when the sting nematode (*Belonolaimus longicaudatus*) was not present (endemic) within the field. In the presence of sting nematode, losses in strawberry yield potential were variable among the different fumigants evaluated. In general, fumigant performance declined on average by 3% to 35% from their respective levels observed at sites where the nematode was not present. Performance inconsistency from one production season to another was demonstrated for some of the fumigant treatments. The top ranking alternative treatment under new plastic mulch for fall production were three-way applications involving Telone C35 or Pic-Clor 60 shank applications followed by minicoulter applications of metam sodium or potassium as a separate bed treatment (4 in. deep) before mulch installation. Among the Telone C35/Inline and Pic-Clor 60/Pic-Clor 60EC treatments, significant differences in relative strawberry yield clearly showed the inferiority of drip compared with shank fumigation treatment, even with two drip tapes per bed into fine sandy soils that typify the Florida strawberry production system. These studies also demonstrated that improved control of plant parasitic nematodes and relative strawberry crop yields are dependent on the adoption of early crop destruction as an IPM practice.

PHEROMONAL CONTROL OF THE PARASITIC LIFE CYCLE OF ENTOMOPATHOGENIC NEMATODES. **Noguez, Jaime H.[1], J.K. Nunnery[1], E.S. Conner[2], Y. Zhou[1], T.A. Ciche[3], J.R. Ragains[2], and R.A. Butcher[1].** [1]Department of Chemistry, University of Florida, Gainesville, FL, 32611; [2]Department of Chemistry, Louisiana State University, Baton Rouge, LA, 70803; [3]Department of Microbiology and Molecular Genetics, Michigan State University, East Lansing, MI, 48824.

Entomopathogenic nematodes survive in the soil as stress-resistant infective juveniles (IJs) that seek out and infect insect hosts. Upon sensing internal host cues, the IJs regurgitate bacterial pathogens from their gut that ultimately kill the host. Inside the host, the nematode develops into a reproductive adult and multiplies until unknown cues trigger the accumulation of IJs. We have shown that the entomopathogenic nematode *Heterorhabditis bacteriophora* uses a pheromone to control IJ development. We developed an IJ recovery assay and purified the pheromone from culture medium using activity-guided fractionation. We then identified the chemical structure of the pheromone using NMR spectroscopy and mass spectrometry. The pheromone, which likely increases in concentration at higher nematode densities, prevents IJ recovery to the J4 stage, allowing IJs to amass late in the infection process. The pheromone is structurally related to the dauer pheromone ascarosides that the free-living nematode *Caenorhabditis elegans* uses to control its development. However, none of the *C. elegans* ascarosides are effective in *H. bacteriophora*, suggesting that there is a high degree of species specificity. More recently, we have developed an IJ recovery assay for another entomopathogenic nematode species, *Steinernema carpocapsae*. Using activity-guided fractionation, we have purified pheromones that suppress IJ recovery in *S. carpocapsae*. Our long-term goal is to understand the role of pheromones in controlling inter- and intraspecies communication in entomopathogenic nematodes. Our work has established that ascarosides are important regulators of development in parasitic nematode species.

IMPACTS OF GLOBAL WARMING ON SOIL FOOD WEB IN RICE PADDY FIELD. **Okada, Hiroaki[1], H. Sakai[1], T. Tokida[1], Y. Usui[1], H. Nakamura[2], and T. Hasegawa[1].** [1]National Institute for Agro-Environmental Sciences, 3-1-3, Kan'nondai, Tsukuba, Ibaraki, 305-8604, Japan; [2]Taiyokeiki CO., 1-12-3, Nakajujo, Kita-ku, Tokyo, 114-0032, Japan.

Soil food web in rice paddy field includes players such as (i) rice plants as primary producers, which provide nutrient and energy to the web via their roots; (ii) microbes as decomposers or primary consumers, which drive nutrient cycling; and (iii) nematodes as primary or secondary consumers, which are pests of rice or prey of other animals. We have been trying to determine the types and strength of the impact of global warming on such food web players during flooded period using a FACE facility, with ambient and FACE (CO_2 concentration is 200 ppm up) treatments in main plots, and normal and elevated temperature (water temperature is 2°C up) treatments in subplots. We hypothesized that the impact of the CO_2 elevation would be dependent on the trophic status of the food web players, i.e., it would appear earlier and greatly on rice plant roots, whereas later and smaller on nematodes. We also hypothesized that the impact of the temperature elevation, which could affect each player simultaneously, would be unpredictable depending on the thermal optima of the players. To estimate the abundance or biomass of the players, we took six soil cores of 100 ml per plot in May (immediately before transplanting), July, and August (immediately before drainage) in 2011, and in May, June, July, and August in 2012. A statistical analysis using GLM mixed models with a repeated sampling design revealed that (i) the elevated CO_2 and the temperature increased

rice root density in both years, but not always clearly and significantly; (ii) the elevated CO_2 did not affect microbial biomass (soil DNA amount, measured in 2012), but the elevated temperature decreased the biomass significantly; (iii) the elevated CO_2 increased the abundance of *Aphelenchoides* species (fungal and plant feeders) but only in 2012, whereas the elevated temperature decreased the abundance of *Filenchus* (fungal and plant feeders) and *Hirschmanniella* species (plant feeders) in summer in both 2011 and 2012. We infer the little impact of the elevated CO_2 on microbes and nematodes could be because of the relatively insensitive growth response of the Japonica rice variety we examined to the CO_2 elevation, which might not provide more nutrition and energy to the heterotrophic organisms in FACE plot soil. We also infer that the thermal constraint of the microbes and the nematode species were directly caused by the elevated temperature, rather than indirectly caused via rice roots. This is because the rice root growth was not suppressed but enhanced, if any, by the temperature. In near future in paddy fields, nematode species of *Filenchus* and *Hirschmanniella* may be reduced by global warming.

STUDIES ON ATTRACTIVE FRACTION DERIVED FROM THE BODY SURFACE OF THE BURROWER BUG, *PARASTRACHIA JAPONENSIS* TO ITS ENTOMOPHILIC NEMATODE, *CAENORHABDITIS JAPONICA* DAUER LARVAE. **Okumura, Etsuko[1, 2], D. Ueno[1], T. Yoshiga[1], and Y. Takeuchi[2].** [1]Department of Applied Biological Sciences, Saga University, Honjo 1, Saga 840-8502, Japan; [2]Graduate School of Agriculture, Kyoto University, Kitashirakawa Oiwake, Sakyo, Kyoto 606-8502, Japan.

A bacteriophagous nematode, *Caenorhabditis japonica* forms a phoretic and necromenic association with the subsocial burrower bug, *Parastrachia japonensis*. *C. japonica* dauer larvae are found on the body surface of *P. japonensis* females. In the previous study, we found that *C. japonica* dauer larvae are attracted to hexane extracts of the body surface components of *P. japonensis* in a species-specific manner. In this study we conducted fractionation of *P. japonensis* hexane-extracts and bioassays to characterize the attractants in the extracts. Hexane extracts prepared by washing *P. japonensis*'s body surface with hexane, were completely dried, dissolved in dichloloethane and separated into acid, basic, and neutral fractions by using NH_3 and HCl as solvents. Each fraction was neutralized before bioassays. Bioassay was done on a 6-cm NGM plate with 1-cm-diam. test and control spots near the edge the plate. After 6 μl of fraction or dicholoroethane was applied onto test or control spots, *C. japonica* dauer larvae were placed onto the center of the plate. We counted the number of nematodes on test and control spots every 10 min for 1 hr. The attractiveness was evaluated as Chemoattraction Index value [(the number of nematode on test—the number of nematode on control)/total number of nematodes]. In addition, we examined the effects of the fractions on arresting dauer larvae dispersal by inoculating *C. japonica* DL on the spot where the fraction was spotted at the center of a 6-cm NGM plate. Although dauer larvae attraction to acid fraction were weak in chemoattraction assay, dauer larvae remained well on acid fraction in the test for the arrest of dispersal experiment. Neutral fraction showed weak attraction and basic fraction showed no attraction. These data suggest that the acid fraction contains attractants that arrest the dispersal of *C. japonica* dauer larvae.

EXPLORING TALLGRASS PRAIRIE DIVERSITY IN THE NEMATODE FAMILY CRICONEMATIDAE. **Olson, Maggie, T.S. Harris, R.S. Higgins, S.M. Paige, K.S. Powers, L.A. Sutton, and T.O. Powers.** Plant Pathology Department, University of Nebraska, Lincoln, NE 68583.

Species and haplotype diversity were compared with two tallgrass prairies in Nebraska. Spring Creek Prairie in Denton, NE, and 9 Mile Prairie in Lincoln, NE, are two remnant prairies existing on the western edge of the Central Tallgrass Prairie Ecoregion. Both prairies have a rich diversity of plant species, although management practices and disturbance have influenced present-day plant species composition. We hypothesize that these factors similarly influence species composition in the plant-parasitic nematode family Criconematidae. Soil samples were taken in spring and summer 2012 from within 40- × 40-m plots in seven management areas at 9 Mile Prairie and from three sections of Spring Creek Prairie. Focal samples from specific plant species were also analyzed to investigate associations between plant hosts and nematode species. Nematode species determinations were based on morphological examination and DNA sequencing of 740 nucleotides of the mitochondrial cytochrome oxidase I (COI) gene. Morphological characterization recognized eight species on 9 Mile Prairie and four on Spring Creek Prairie, with three shared species. Focal sampling suggests that *Ogma decalineatum* is frequently associated with leadplant, *Amorpha canescens*, and *Discocriconemella inarata* is associated with tall dropseed, *Sporobolus asper*. *Mesocriconema xenoplax* is present when management practices permit the development of woody invasive plants in the prairie. DNA sequence indicates that the most common morphospecies, *Mesocriconema curvatum*, is not *M. curvatum*, but instead is composed of two yet to be described cryptic species. Analysis of one management area in 9 Mile Prairie that was cultivated during the 1940s, but allowed to revert to prairie for more than 60 yr, shows the historical legacy of soil disturbance by its altered nematode species composition.

NUTRIENT ENHANCEMENT OF NEMATICIDES IN COTTON WITH CONCOMINANT INFESTATION OF *MELOIDOGYNE INCOGNITA* AND *ROTYLENCHULUS RENIFORMIS*. **Overstreet, Charles[1], E.C. McGawley[1], D.M. Xavier[1], M. Kularathna[1], C.M. Martin[1], and R.A. Haygood[2].** [1]LSU AgCenter, Department of Plant Pathology and Crop Physiology, 302 Life Science Blvd., Baton Rouge, LA 70803; [2]Dow AgroSciences, Collierville, TN 38017.

Our research with site-specific management of reniform (*Rotylenchulus reniformis*) and southern root-knot nematodes (*Meloidogyne incognita*) has shown a correlation between nutrient deficiency and nematode damage. A field study was conducted in 2012 with two nutrient regimes (low fertility consisting of only N at 100 kg/ha and high fertility consisting of dolomitic lime at 2,240 kg/ha, N at 100 kg/ha, P and K at 67.2 kg/ha, S at 9 kg/ha, and B and Zn at 1.1 kg/ha) and four nematicide treatments (seed treatment with Avicta Complete Cotton, the fumigant 1,3-dichloropropene at 28.1 l/ha, a combination of both nematicides, and a no-nematicide control replicated 18 times. Soil samples were collected at-planting and after harvest. Fertility had a significant main effect on cotton yield and was significantly higher in the high fertility regime compared with the low fertility regime (2,738 and 2,625 kg/ha of seed cotton, respectively). Nematicides had a significant main effect on final populations of both nematode species. Application of 1,3-D alone or combined with Avicta Complete Cotton reduced southern root-knot by 62% or 82%, respectively, compared with the no-nematicide treatment. None of the nematicides significantly reduced final populations of reniform nematode. All nematicide treatments produced significantly greater yields than the control. There was a significant interaction between fertility and nematicides with a higher yield with Avicta Complete Cotton in the high fertility regime compared with the low fertility regime. The high fertility regime significantly enhanced yield of the Avicta Complete Cotton compared with the low regime. The fumigant 1,3-D was not significantly affected by fertility regime. The application of adequate or available nutrients may play a role in increasing the benefits of seed treatment nematicides and the use of management zones.

SURVEYS OF THE THELASTOMATID NEMATODES PARASITE IN INVASIVE ALIEN *PERIPLANETA* SPECIES IN JAPAN. **Ozawa Sota[1], T. Yoshiga[2], N. Kanzaki[3], and K. Hasegawa[1].** [1]Department of Environmental Biology, College of Bioscience and Biotechnology, Chubu University, 1200 Matsumoto, Kasugai, 487-8501 Japan; [2]Laboratory of Nematology, Department of Applied Biological Science, Saga University, Saga, 840-8502 Japan; [3]Forest Pathology Laboratory, Forestry and Forest Products Research Institute, 1 Matsunosato, Tsukuba, Ibaraki 305-8687 Japan.

The smokybrown cockroach, *Periplaneta fuliginosa*, and the American cockroach, *P. americana*, has been spreading all over the world, and now is one of the most unwanted invasive alien species in Japan. Because cockroaches are ubiquitously infected by the thelastomatid nematodes, they are traveling around the world with their parasitic nematodes. Little is known about differences or similarity of the parasite's population structures between different areas of the host *Periplaneta* cockroaches. We investigated these two invasive cockroaches to Japan and found that 100% of *P. fuliginosa* individuals were infected with one nematode species *Leidynema appendiculata*, and 100% of *P. americana* individuals were co-infected with two species *Thelastoma bulhoesi* and *Hammerschmidtiella diesingi*. This is the first report of the thelastomatid nematode isolated from the smokybrown cockroach. *L. appendiculata* is a cosmopolitan nematode species, which has been isolated from many Blattaria hosts including *P. americana*. It is hypothesized that these two cockroaches might have been originally sharing parasitic thelastomatid species, but dropped other parasitic species during dispersal. If the communities of parasitic nematode in these cockroaches in the world are examined, it might be able to predict their origin and dispersal history.

LOOKING FORWARD, LOOKING BACK—A NEMATOLOGIST'S VIEW. **Roland N. Perry.** Department of Agro-ecology, Rothamsted Research, Harpenden, Herts. Al5 2jq, UK.

This presentation will attempt to make sense of past changes and how they will affect the future of nematology. The pace of change has accelerated enormously and this is an exciting time for nematology as a science and for individual researchers. Apps and diagnostics through web connections to mobile phones are improving knowledge of nematodes in resource-poor regions. Genomics is a central component for exploiting gene function and unravelling host-parasite relationships. Industry is rapidly buying up molecular engineering and biocontrol companies. Thus, the drivers for effective management of these pests are in place. There is a real possibility of realizing the oft-repeated ''novel control strategy'' that farmers the world over can use. However, change must not ignore past work and will need expertise in certain areas, including taxonomy and field experimentation, that are now under threat.

THE APPLICATION OF FUNGAL BIOCONTROL AGENTS TO CONTROL PLANT PARASITIC NEMATODES. **Lüth, Peter.** Prophyta Biologischer Pflanzenschutz GmbH, Inselstrasse 12, 23999 Malchow, Germany.

Plant parasitic nematodes are amongst the most harmful pests where agricultural and horticultural crops are concerned, although they themselves serve as victims for a large number of ''beneficial'' organisms, such as other nematodes, insects and micro-organisms. For around 30 yr, these have been increasingly employed for the biocontrol of nematodes. Thus fungi and bacteria are artificially reproduced and employed as biological nematicides. In this connection, PROPHYTA (now part of Bayer CropScience) is working in particular on microbial fungi. In most cases, the spores or conidia of fungi are the active ingredients of this modern new class of products. These are generated by means of large fermenters and, after mass culture of the fungi, are subsequently separated from the culture substrate. Cultivation takes place in liquid or solid-state cultures. A maximum yield of stabile spores is always the objective. The spores, which before formulation are either present as a pure spore powder or as a pure spore suspension, are processed by means of various possible procedures, which are known to ensure good durability (shelf life) as well as good applicability. This implies that the vitality of the spores must be retained in

the end product for a prolonged period. Various drying procedures are available for this purpose, e.g., fluidized bed drying, freeze drying (lyophilization) and spray drying, or the previously dried spore powder is simply mixed in a liquid carrier (e.g., a type of oil) or an inert powder. One important objective of the formulation is to obtain the highest-possible concentration of spores in the final product. They must be made available by means of standard application techniques and it must be possible to disperse them in the application broth in such a way that they are present as individual colony forming units. Biological pesticides should be comparable in all evaluating criteria with standard chemical-synthetic pesticides. Apart from a good effect and good applicability, they must also be available at a competitive price. The competitive capability of a product is initially determined by two factors, firstly the productivity of the fungal strain used with regard to spore production and secondly the number of spores per hectare or acre that are required so as to achieve adequately successful control. For example, it can be presumed that good control is possible according to the type of effective antagonist used for application quantities between 1×10^{11} and 1×10^{14} spores per hectare. Accordingly, the spore yield in the fermentation process is crucial for the feasibility of the whole process. Nowadays, when searching for new microbial active ingredients, the biocontrol industry frequently relies on efficacy results obtained from universities or other scientific institutions. Even so, the fungal strains acquired from those institutions must be suitable for the purpose with regard to other features as well. Thus, a low rate per acre, good productivity, good suitability to be formulated, and a good shelf life are essential for the application of a particular species or strain for the purpose of biological nematode control.

SURVEY AND REVISON OF NEMATODES INHABITING NORTH AMERICAN MILLIPEDS. **Phillips, Gary, and E.C. Bernard.** Entomology and Plant Pathology Department, University of Tennessee, 2505 E. J. Chapman Drive, 370 Plant Biotechnology Building, Knoxville TN 37996-4560.

Nematodes that parasitize invertebrates, such as insects and millipedes, are inquiline kleptoparasites that belong to one of four superfamilies: Thelastomatoidea, Rhigonematoidea, Ransomnematoidea, and Rhabditoidea. Species of Thelastomatidae and Rhigonematidae are commonly found in the intestine of many millipeds. An 1853 monograph by Joseph Leidy is still the best reference to these nematodes in North America; currently, only 16 species have been recognized from temperate North American milliped fauna. The goals of this research are a comprehensive taxonomic analysis of these nematodes and their specific host-parasite relationships with millipeds. Millipeds will be collected around the United States with the assistance of volunteers and other interested persons. Nematodes will be dissected from the intestines of millipeds and studied with several different approaches. Some nematodes will be fixed in formalin and processed to glycerin for permanent mounts. Others will be prepared for SEM or for molecular analysis. In dissections undertaken so far, rhabditid dauer juveniles often have been found in the recto-anal zone. These dauers will be placed on agar for development to adults and subsequent identification. Other organisms in the gut of millipedes will be noted, such as ciliated protozoans, gregarines, and trichomycetes. Dissections of about 85 millipeds in the orders Polydesmida, Callipodida, Julida, and Spirobolida have yielded up to 300 nematodes in a single specimen, composed of up to four separate species. Rhigonematids typically are more numerous but thelastomatids are more diverse, with at least 10 species found so far. At least two nematode taxa have been collected that are not yet classifiable to family. In general, nematodes are more numerous at the junction of the midgut and hindgut in the polydesmid milliped *Apheloria montana*, and are absent from the foregut, where gregarine protists are most abundant. Nematodes in the Leidy collection in Philadelphia will be compared with those in our study for proper application of scientific names and for redescription.

DYNAMICS OF GENETIC CONFLICT BETWEEN *CAENORHABDITIS BRIGGSAE* AND THEIR MITOCHONDRIAL DNA IN VARYING EXPERIMENTAL POPULATION SIZES. **Phillips, Wendy S.[1], A.L. Coleman-Hulbert[3], D.K. Howe[2], S. Ping[2], E.S. Weiss[2], S. Estes[3], and D.R. Denver[2].** [1]USDA-ARS Horticultural Crops Research Laboratory, Corvallis, OR 97330; [2]Department of Zoology, Oregon State University, Corvallis, OR 97331; [3]Department of Biology, Portland State University, Portland, OR 97207.

Genetic conflict across levels of biological organization contributes to a variety of basic biological processes including the architecture of genetic systems, genome evolution, and speciation. An example of such genetic conflict is seen in a previously described naturally occurring ~ 870 bp deletion (*nad5*Δ) in the mitochondrial DNA (mtDNA) of *Caenorhabditis briggsae*. This deletion was shown to be a so-called ''selfish'' genetic element as it has a transmission advantage in mass accumulation lines despite having negative effects on organismal fitness. Evolutionary theory predicts that ''selfish'' genetic elements will proliferate when the ''host'' genetic effective population size (N_e) is small, but direct tests of this prediction remain absent. We analyzed the evolutionary dynamics of selfish, deletion-containing mtDNA elements in populations of *C. briggsae* using an experimental evolution approach where population size (N) varied (N = 1, 10, 100, 1,000). Six different natural isolate-derived strains were analyzed across 50 generations, with mtDNA characterized every five generations. We screened for large deletions in mtDNA via two PCR approaches: (i) four overlapping long PCRs that collectively amplified the whole mtDNA genome, and (ii) a PCR targeted at ascertaining low, medium and high levels of *nad5*Δ. Based on all long PCR amplifications, nineteen new deletion events were detected, arising at varying generations in different lines. New deletion occurrences parsed out to just nine new deletion types, ranging in size from 539 to 1,445 bp. In the majority of cases, once a new deletion appeared it persisted through all remaining generations. Population size was correlated with the rate of new deletions, with a higher frequency of new

deletions at smaller population sizes. Likewise, *nad5*Δ levels varied with population size, with deletion frequencies increasing over the 50 generations to high levels only in lines from population sizes of N = 1 and N = 10. A reduction in deletion levels across generations occurred only in lines from the N = 1,000 population size. There was also variation in *nad5*Δ levels between isolate types. At all population sizes, the deletion was detected at only low to medium frequencies in isolates EG4181 and PB800, both originating in the continental United States and both shown to have a compensatory mutation in the direct repeat sequence flanking the deletion. In contrast, the deletion was detected at only medium to high frequencies in isolates HK104 or HK105, both originating from Japan. For all strains, population size N = 100 had the least change in *nad5*Δ levels across generations. This study provides novel, robust, and direct empirical insights into the evolutionary population-genetic parameters that determine when ''selfish'' genetic elements arise and persist, and when they are eliminated by natural selection.

SPECIES ABUNDANCE AND INFLUENCE OF NEMATODES IN URBAN TURFGRASS ECOSYSTEMS IN EAST BATON ROUGE (LA) PARISH. **Plaisance, A.R., E.C. McGawley, C. Overstreet, and Y. Takeuchi.** Louisiana State University AgCenter, Department of Plant Pathology and Crop Physiology, Baton Rouge, LA 70803.

A survey was conducted to characterize plant parasitic nematode communities associated with centipede and St. Augustine turfgrasses in urban ecosystems. To date, 135 lawns in East Baton Rouge Parish, LA, have been sampled. Soil types ranged from clay to sandy clay loam, with the average type being loam (30% sand, 20% clay, 50% silt). Twelve nematode genera were identified from soil and root samples of St. Augustine lawns: *Criconemella* occurred in 91%, *Gracilicus* in 3%, *Helicotylenchus* in 94%, *Hemicycliophora* in 2%, *Hoplolaimus* in 5%, *Meloidogyne* in 46%, *Pratylenchus* in 35%, *Scutellonema* in 2%, *Trichodorus* in 3%, *Tylenchorhynchus* in 25%, *Tylenchus* in 92%, and *Xiphinema* in 15%. Respectively, average nematode densities per 250 cm^3 of soil were 159, 20, 180, 11, 8, 54, 29, 11, 14, 25, 149, and 11. Ten nematode genera were identified from soil and root samples of centipede lawns; *Criconemella* occurred in 86%, *Helicotylenchus* in 78%, *Hoplolaimus* in 25%, *Meloidogyne* in 25%, *Pratylenchus* in 69%, *Scutellonema* in 3%, *Trichodorus* in 14%, *Tylenchorhynchus* in 19%, *Tylenchus* in 92% and *Xiphinema* in 6%. Respectively, nematode densities per 250 cm^3 of soil were 290, 186, 18, 52, 59, 25, 9, 32, 132, and 14. Nematode genera identified from the survey were used to establish microplot trials to evaluate the impact of grass species and soil type on nematode reproduction and pathogenicity. Treatments in microplot trials for 2012 included three soil types (clay [25% sand, 40% clay, 35% silt], loam [45% sand, 25% clay, 30% silt], and sandy loam [75% sand, 10% clay, 15% silt]), three nematode infestation levels (0, 1×, and 10× nematodes) containing seven nematode genera (the 1× infestation rate contains 15 *Pratylenchus*, 12 *Meloidogyne*, 65 *Helicotylenchus*, 164 *Tylenchorhynchus*, 665 *Criconemella*, and 88 *Tylenchus*) and the two grass species. Nematodes did not significantly impact turfgrass growth at densities used in the 2012 experiment. However, both grass species grown in sandy loam soil had significantly lower foliar weights and total plant weights ($p < 0.05$) than those grown in the other two soil types. At 220 days after establishment, nematodes increased 50- and 8-fold, respectively, from the low and high infestation levels on St. Augustine. Similarly, there were 25- and 7-fold community increases, respectively, on centipede. In general, *Meloidogyne* spp. had the greatest reproductive value (R where R= harvest community density/ infestation level). Across all soil types, reproduction of *Pratylenchus* was greater on centipede than on St. Augustine. Across both turfgrass species, reproduction of *Tylenchorhynchus* was greater in microplots containing the clay and loam soil types than those containing the sandy loam soil type. Microplot studies currently in progress employ significantly higher infestation levels than those in 2012.

BETWEEN BARCODE AND MORPHOSPECIES DESIGNATION. **Powers, Thomas[1], E.C. Bernard[2], T.S. Harris[1], R.S. Higgins[1], P. Mullin[1], L.S. Sutton[1], and K.S. Powers[1].** [1]Plant Pathology Department, University of Nebraska, Lincoln, NE 68583; [2]Department of Entomology and Plant Pathology, University of Tennessee, Knoxville, TN 37996.

DNA barcoding with the mitochondrial cytochrome oxidase subunit I gene (COI) has been proposed as a universal approach for animal identification. Accuracy of a barcode-based identification system is dependent on a taxonomically comprehensive reference database and a well-established understanding of species boundaries. Unfortunately neither criterion is commonly met in species of plant-parasitic nematodes. We have been developing a specimen database that includes nematode measurements, evaluation of qualitative morphological characters, biogeographic and ecological information linked to a COI barcode. Species in the criconematid genus *Mesocriconema* provide a useful test case for this integrated taxonomic approach. More than 300 *Mesocriconema* specimens from across North America have been evaluated. For several species (*M. discus*, *M. xenoplax*, *M. ornatum*, and *Discocriconemella inarata*) specimens obtained from type localities have aided the evaluation process. Some species that are characterized by visibly unique morphological features are also strongly supported as unique genetic lineages by COI. *Mesocriconema xenoplax* is supported as a cryptic species complex, but no support exists for the so-called short-stylet forms of the species. COI haplotype analysis identifies numerous groups within the morphospecies *M. curvatum*, but these groups are polyphyletic in phylogenetic assessments.

NEMATODES OF THE GEORGE WASHINGTON MEMORIAL PARKWAY. **Powers, Thomas[1], E.C. Bernard[2], T.S. Harris[1], R.S. Higgins[1], P. Mullin[1], H.S. Smith[2], L.S. Sutton[1], and K.S. Powers[1].** [1]Plant Pathology Department, University of Nebraska, Lincoln, NE 68583; [2]Department of Entomology and Plant Pathology, University of Tennessee, Knoxville, TN 37996.

The George Washington Memorial Parkway is a 7,600-acre national park that parallels the Potomac River from Mount Vernon, VA, to Great Falls Park, approximately 30 miles north. The park includes numerous historical sites such as Arlington House, the residence of record for Robert E. Lee before the Civil War, the Patowmack Canal built for George Washington to circumvent Great Falls, and Fort Marcy, a Civil War fort built to defend Washington, DC. In summer 2012, soil samples were taken from 53 locations in the park with an emphasis on areas of historical disturbance paired with undisturbed reference sites. Notable disturbed sites sampled were the trenches of Fort Marcy, the berm from the canal construction, and fill from the construction of Ronald Reagan National Airport. Nematode community analysis of each sample was based on 150 nematodes identified to genus, 20 keyed to species, and DNA analysis of members of the Criconematina. An average of 30 taxa per sample and 118 total genera have been recorded to date. In general the undisturbed reference sites had higher numbers of taxa and the disturbed sites had higher nematode abundance. Criconematina species are negatively affected by disturbance and they range from 0 to 40% of the estimated nematode abundance in samples. Twenty-eight morphospecies in Criconematina representing 13 genera were recorded. Preliminary DNA analysis based on 740 nucleotides of the mitochondrial COI gene indicates that two haplotype groups of *Mesocriconema xenoplax* are present in the parkway. Both haplotype groups are also found in Great Smoky Mountains National Park suggesting an evolutionary connection between nematodes from both parks.

EFFICACY OF MCW-2 AS A NEMATICIDE FOR FRUITING VEGETABLES (EGGPLANT + OKRA) AND STRAWBERRY. **Sances, Frank[1], B.A. Aglave[1], B. Booker[1], and Dennis Long[2].** [1]Florida Ag Research, 3001 N. Kingsway Road, Thonotosassa, FL 33592; [2]Makhteshim-Agan North America, Inc., 3120 Highwoods Boulevard, #100 Raleigh, NC 27604.

MCW-2 (fluensulfone) is a new nonfumigant nematicide with a novel mode-of-action in global development by Makhteshim Agan. Three field trials were conducted at Florida Ag Research, Hillsbororough County, FL, on strawberry, okra, and eggplant. MCW-2 480EC was sprayed on the bed tops at the different rates from 2.0 pt/a to 3.5 pt/a for fruiting vegetables and 5.34 pt/a to 7.12 pt/a for strawberry. The beds were then rototilled to a depth of 8 in. with a tractor mounted rototiller, reshaped, and then irrigated with 1 acre in. of overhead water for strawberry and 0.4 acre in. for fruiting vegetables. Efficacy was evaluated by assessing crop vigor, yield, root damage, and population density of nematode species at different growth stages of each crop. The studies were conducted to evaluate the effect of MCW-2 against major nematode species (Sting, Root Knot, Lesion, and Spiral nematodes) of strawberry, okra, and eggplant. In all the three studies, MCW-2 reduced the population density of all four nematode species and resulted in higher yields compared with untreated control in strawberries. These results indicate that MCW-2 could become a useful nematode management tool for strawberry and fruiting vegetable crops.

CELLULAR AND MOLECULAR RESPONSES OF THE BACTERIOVOROUS NEMATODE *CAENORHABDITIS ELEGANS* AGAINST THE EPN-MUTUALISTIC BACTERIUM *PHOTORHABDUS LUMINESCENS*. **Sato[1], K., T. Yoshiga[2], and K. Hasegawa[3].** [1]Graduate School of Agriculture, Kyoto University, Sakyo, Kyoto 606-8502, Japan; [2]Department of Applied Biological Sciences, Saga University, Saga 840-8502, Japan; [3]Department of Environmental Biology, College of Bioscience and Biotechnology, Chubu University, 1200 Matsumoto, Kasugai 487-8501, Japan.

The gram negative bacterium *Photorhabdus luminescens* TT01, a mutualistic partner of the entomopathogenic nematode (EPN) *Heterorhabditis bacteriophora* TT01, behaves as a pathogen against not only insects but also nonmutualistic nematodes. However, little is known about its pathogenicity against nonmutualistic nematodes as well as the innate immune responses. We used *Caenorhabditis elegans* as a model host to characterize disease progress and examine the roles of innate immune responses against this pathogen. First, we observed fundamental symptoms and diagnostic characters. Body size, brood size, and lifespan were dramatically decreased when *C. elegans* N2 was fed with *P. luminescens* TT01 instead of their normal laboratory food *E. coli* OP50. *P. luminescens* TT01 cells were not ground down by the terminal bulb and subsequently invaded into the intestinal lumen. However, bacterial multiplication was not observed. Over time, *C. elegans* intestinal cells were gradually damaged and prominent crystal-like structures were formed in the intestinal lumen. Second, we inhibited several signaling pathways regulating antimicrobial responses by RNA interference (RNAi). Survival time was significantly altered by RNAi of p38 MAPK and Insulin/IGF-1–like signaling pathways. Consistent with the change of survival time, the progress of intestinal symptoms was also altered by RNAi. For example, RNAi of *pmk-1* resulted in shorter survival time and faster progress of symptoms than control animals. Our results suggest that *C. elegans* requires the innate immune systems against the pathogenicity of *P. luminescens* TT01.

NEMATICIDE ENHANCEMENTS OF *ROTYLENCHULUS RENIFORMIS* RESISTANT COTTON GENOTYPES. **Schrimsher, D., Lawrence, K., Sikkens, R., and Weaver, D.** Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849.

Reniform nematode (*Rotylenchulus reniformis*) resistance in the cotton LONREN-1 × FM966 breeding lines developed at Auburn University have demonstrated that reniform resistance is accompanied by severe plant stunting and limited plant growth followed by low yields. The objective of this study was to evaluate effects of applying nematicides to selected

reniform nematode resistant breeding lines on reniform nematode populations, early seedling plant stunting, and seed cotton yield. Four resistant breeding lines from the LONREN-1 × FM966 cross, the germplasm lines LONREN-1 and BARBREN 713, one susceptible line from the LONREN-1 × FM966 cross, and the susceptible cultivar DP393 were treated with nematicides and their performances evaluated. In the greenhouse, nematicides increased plant heights in resistant lines. Nematicides further reduced reniform populations in the resistant lines 45 d after planting (DAP). Reniform populations were 50% lower in resistant lines compared with the susceptible lines by the end of the growing period. In microplot and field trials, the phenotypic stunting response of resistant lines was reduced by nematicides with increased plant heights at 30 and 75 DAP. Nematicides reduced early season *R. reniformis* populations in the microplot trial by 41%. By harvest, *R. reniformis* populations in microplot and field trials were 54% and 52%, respectively, higher in the susceptible lines compared with resistant lines. Egg populations in the field trial at 100 DAP were 84% lower in resistant lines compared with susceptible checks. Seed cotton yields in the field trial were increased by nematicides to levels that were comparable with susceptible checks.

ASCAROSIDES REGULATE BEHAVIOR AND PHENOTYPIC PLASTICITY IN DIVERSE NEMATODE SPECIES. **Schroeder, Frank C.** Boyce Thompson Institute, Department of Chemistry and Chemical Biology, Cornell University, Ithaca, NY 14853.

Nematodes are among the most successful animals on earth and include important human pathogens, yet little is known about nematode pheromone systems. In the laboratory model organism *Caenorhabditis elegans* , a family of small molecules called ascarosides have recently been shown to mediate a diverse range of developmental and behavioral phenotypes including mate finding, aggregation, and developmental diapause. Since ascarosides are biosynthesized via highly conserved peroxisomal beta-oxidation, we hypothesized that ascaroside signaling exists outside of the genus *Caenorhabditis*. Analysis of more than 20 different nematode species from several different clades, including free living, animal-, and plant-parasitic species, revealed that nematodes produce species-specific ascaroside mixtures that serve diverse functions. For example, sex-specifically biosynthesized ascarosides serve as male and female sex pheromones in *Panagrellus redivivus* and a number of chemically very complex ascaroside derivatives control adult phenotypic plasticity in *Pristionchus pacificus*. Chemically related small molecules that are based on a slightly modified sugar called paratose instead of ascarylose (''paratosides'' versus ''ascarosides'') mediate dauer formation in *P. pacificus*. Some of the identified signals are of extremely high potency; for example, an ascaroside called hbas#3 induces aggregation in *C. elegans* even at low femtomolar concentrations. Although ascaroside production is generally species-specific and different nematode species respond to distinct and only occasionally overlapping sets of compounds, ascaroside biosynthesis patterns also correlate with phylogeny as well as lifestyle and ecological niche. These observations demonstrate that nematode communication and developmental regulation relies on a conserved family of signaling molecules, despite having evolved to occupy diverse habitats, from free-living to animal- or plant-parasitic. The ascarosides' structural features and their level of conservation are reminiscent of the role of acyl homoserine lactones (AHLs) in bacterial quorum sensing. Paralleling the role of ascarosides as a conserved language of nematodes, AHLs are produced and sensed by many different species of Gram-negative bacteria and regulate diverse aspects of bacterial communication. The identification of species-specific ascaroside mixtures and their signaling functions may enable design of pheromone-based approaches to interfere with the life cycle of parasitic nematodes, which are of increasing concern for agriculture and human health.

THE NUCLEAR OPTION: INVESTIGATING NEMATODE ANATOMY WITH MICROWAVE-FIXATION AND DAPI STAINING. **Schroeder, Nathan, and M.M. Barr.** Dept of Genetics, Rutgers University, 145 Bevier Rd. Piscataway, NJ 08854.

Despite vast ecological habitat and subsequent behavioral differences, the neuroanatomy among nematode species is thought to be well conserved. However, several studies have found differences in neuroanatomy both among different species as well as among stages within a given species. For example, we recently discovered that the inner-labial quadrant neurons undergo dendrite arborization during the dauer stage of *Caenorhabditis elegans*. To further examine the anatomy of *C. elegans* dauers, we developed a microwave fixation technique to modify an existing DAPI (4',6-diamidino-2-phenylindole) nuclear staining protocol. To our knowledge this is the first whole-mount DAPI staining of *C. elegans* dauers. As expected, the nuclei are much more densely arranged during dauer than nondauer stages. However, no other obvious differences in the number of nuclei or relative position of individual nuclei were seen between dauer and nondauer *C. elegans*. To examine anatomical differences among species, we expanded our DAPI staining technique to species with incompletely described anatomies. Recent electron microscopy (EM) studies by Ragsdale et al. described the anterior anatomy of the fungivore *Aphelenchus avenae*. Using microwave fixation, DAPI staining, and DIC Nomarski optics, we identified *A. avenae* nuclei as described by EM. Furthermore, we found differences in the development of ventral nerve cord (VNC) nuclei between *C. elegans* and *A. avenae*. Although the number of nuclei in adults was similar for both species, the number of nuclei in J2 *A. avenae* is approximately half that found in J2 *C. elegans* suggesting differences in VNC developmental timing. An examination of the entomopathogenic nematode *Steinernema carpocapsae* infective juveniles also unveiled differences in VNC nuclei. Although *C. elegans* contain 57 VNC neurons, approximately 80 putative neurons were identified in the *S. carpocapsae* VNC.

Previous research found similar numbers of VNC neurons in *C. elegans* and the large animal parasitic nematode *Ascaris*, suggesting that the number of VNC neurons is not determined by body size. We are currently examining additional *Steinernema* species to determine if infection style (cruiser vs. ambusher) is correlated with VNC nuclei quantity. This method provides direction for further detailed neuroanatomical studies that may elucidate the basis of behavioral differences among nematodes.

THE EFFECT OF TRANSGENIC ENDOGENOUS DEFENSE ELICITORS IN *ARABIDOPSIS* ON THE INFECTION RATE AND LIFE STAGE DEVELOPMENT ROOT-KNOT NEMATODES (*MELOIDOGYNE INCOGNITA*.) **Sekora, David[1], A. Huffaker[2], W. T. Crow[1], and T. Mekete[1].** [1]Entomology and Nematology Department, University of Florida, P. O. Box 110620, Bldg. 970 Natural Area Drive, Gainesville, FL 32611; [2]U.S. Department of Agriculture, Agricultural Research Service Center for Medical, Agricultural and Veterinary Entomology: Chemistry Research Unit, 1700 SW 23rd Drive, Gainesville, FL 32608.

Through the use of genetic manipulation, we are able to enhance a host plant's ability to protect it self from various pathogens and pests. The goal of this project was to test the effect of recently characterized defense signaling genes for their ability to regulate interactions between plant roots and root-feeding nematodes. *Arabidopsis thaliana* was used as a model plant to study the effect on a root-knot nematode (*Meloidogyne incognita)* ability for infection and life stage development. Along with the wild type, five different transgenic lines with either enhanced or compromised defense responses were tested for their potential effect on the plant-parasitic nematodes. The experiments were split into two different but similar trials to test for infection capability or differences in nematode development. For an initial screening trial the test plants were grown on either plant growth media or an autoclaved 50/50 sand soil media both kept in growth chambers for the entirety of the trials. Twenty-five days after germination the test plants were inoculated with 500 second-stage juveniles (J2s) of *M. incognita* and cultivated for 14 d before harvesting. Upon completion of this trial, three of the four enhanced response lines showed a significant reduction in galling of the root tissue ($P < 0.01$) when compared with that of the wild type. Alternatively, to examine the test plants for any ability to inhibit the nematodes ability to progress through its life stages the nematodes were subjected to a time course experiment. Test plants were grown in the same sand/soil mixture as used in the initial screening trials and inoculated at the same time but with only 250 J2s per plant. At regular intervals of 2-d postinoculation (DPI), 4-DPI, 8-DPI, 16-DPI, and 32-DPI; the test plants were harvested and evaluated. The roots of these test plants were stained with acid fuchsin and examined for the various stages of the nematode's development. The results from this trial are currently being evaluated.

BIOLOGICAL CONTROL OF THE ROOT-KNOT NEMATODE, *MELOIDOGYNE HAPLA* WITH AN ANTAGONISTIC BACTERIUM. **Seo, Yunhee, Jiyeong Park, and Young Ho Kim.** Department of Agricultural Biotechnology and Research Institute of Agriculture and Life Sciences, Seoul National University, Seoul 151-921, Korea.

Root-knot nematodes (*Meloidogyne* spp.) are distributed throughout the world and attack more than 2,000 plant species. In Korea, *Meloidogyne incognita, M. arenaria*, and *M. hapla* are major root-knot nematodes (RKNs), among which *M. hapla* damages severely field crops such as carrot and ginseng. This study aimed to investigate biological control of the RKN *M. hapla* using an antagonistic bacterium. RKNs used in experiments were isolated from root galls of ginseng cultivated in Jinan, Korea, and identified *Meloidogyne hapla* through 28S rDNA sequencing analysis. Out of 523 bacterial isolates from soil samples and 19 *Paenibacillus* strains isolated from ginseng roots, a bacterial isolate C1-7 showed a very strong nematicidal activity in vitro against *M. hapla*. In pot experiment on tomato and carrot inoculated with the root-knot nematodes, treatments of C1-7 bacterial culture with an inoculum density of 1×10^8 colony-forming units (CFU)/ml reduced root-galling with control values of 100% and 90% and egg mass formation with control values of 75% and 100%, respectively, compared with those on plants only with RKN inoculation. The bacterial isolate formed circular and flat colonies with undulate margin and was rod-shaped bacilli and gram-positive, which was identified *Bacillus cereus* by the morphological characters, and Biolog program, fatty-acid composition, and 16S rDNA sequencing analyses. Population density of C1-7 were kept high for a long period of time and its nematicidal activity was strong when it was cultured in brain heart infusion broth, compared with other culture media such as nutrient broth, Luria Bertani broth and tryptic soy broth. Culture filtrate of C1-7 from 48-hr culture with or without heat treatment showed strong nematicidal activity even at a concentration of 1%. Light microscopy of RKN- infected root tissues treated with C1-7 showed the inhibition of intact giant cell formation, sometimes accompanying dead nematodes located in the root tissues, whereas RKN alone induced fully matured giant cell formation with profound cell wall ingrowths. All of these results suggest the C1-7 bacterial isolate have a good potential to be developed as a biocontrol agent for the RKN *M. hapla*.

HEAVY METAL CONTAMINATION AND STRUCTURE OF THE SOIL NEMATODE FOOD WEB IN URBAN VACANT LOTS IN OHIO. **Sharma, Kuhuk[1], N.T. Basta[2], and P.S. Grewal[1,3].** [1]Environmental Science Graduate Program, The Ohio State University, OARDC, Wooster, OH 44691; [2]School of Environment and Natural Resource, The Ohio State University, Columbus, OH 43201; [3]Entomology and Plant Pathology Department, The University of Tennessee, Knoxville, TN, 37996.

Increased demand for local produce and food security has escalated interest in utilizing urban vacant lots for food production. However, soil quality and heavy metal contamination are major concerns for urban agriculture. We determined the extent of soil heavy metal contamination in vacant lots in two postindustrial cities and evaluated their relationship with the structure and function of the soil food web. Total concentrations of Pb, Cd, Zn, As, and Cr were determined in 43 vacant lots in two disadvantaged neighborhoods, Hough (Cleveland) and Weinland Park (Columbus), Ohio; along with soil parameters including texture, moisture, pH, active carbon, and organic matter content. Nematode community was used as a surrogate to assess the condition of the soil food web. Human and ecological health risk was assessed using U.S. EPA's generic soil screening levels for human exposure via soil ingestion and Ecological Soil Screening Levels (Eco-SSLs) for ecological receptors. The results showed that all the sites had As concentration above the EPA SSL for human health risk. Also, 33% of the lots in Columbus and 93% in Cleveland had total soil Pb concentrations higher than the Eco SSL for plants whereas, 6% of the lots in Columbus and 53% in Cleveland had Pb concentration higher than the SSL for human health risk. All of the vacant lots in Columbus and 96% in Cleveland had Zn concentrations above the Eco SSL for plants and 87% of the lots in Columbus and 85% in Cleveland had Zn concentration higher than the Eco-SSL for soil invertebrates. Thirty-four nematode genera were identified, with most being plant-parasitic (14) followed by bacteria-feeding (12), omnivorous (5), fungal-feeding (2), and predatory (1). A decrease or absence of the more sensitive, higher trophic levels and increased abundance of plant-parasitic nematodes indicated the disturbed nature of the urban soil food webs. Correlation analysis revealed significant interactions within heavy metals and soil parameters; and between metals, soil, and nematode trophic levels and community indices. Total bacterial and fungal feeding nematodes were negatively correlated with As and Cd concentrations. Nematode food web structural index showed positive correlation with all heavy metals, except Pb, and enrichment index was positively correlated with As and Cr. The ratio of nonplant parasitic to parasitic nematodes was negatively correlated with all studied heavy metals. Multiple regression analysis revealed a combination of As, organic matter and texture as significant predictors of nematode maturity index, structure index, and channel index. We conclude that the opportunistic bacteriovore and fungivore nematodes are more sensitive to metal contamination. Also the soil food web is significantly affected by the interaction of metals and soil physical and chemical parameters.

AZOXYSTROBIN AND ABAMECTIN IMPROVE ROOT HEALTH OF ZOYSIAGRASS INFECTED WITH *TRICHODORUS OBTUSUS*. **Shaver, J. Bradly, P. Agudelo, and S.B. Martin.** School of Agricultural, Forest, and Environmental Sciences, Clemson University, Clemson, SC 29634.

Consistently effective control from the currently available nonfumigant nematicides has been difficult to obtain in turf. New products and management strategies are being studied in hope of finding better alternatives. Preliminary research at Clemson University in 2011 suggested that the addition of azoxystrobin to a nematode management plan enhanced the benefits of certain nematicides. Greenhouse and in vitro studies were conducted to determine the efficacy of an experimental abamectin nematicide and its potential interactions with azoxystrobin. Core samples of 'Empire' zoysiagrass (*Zoysia japonica)*, 10.16-cm diam. × 15.24-cm depth, were taken from a field infested with stubby root nematode *Trichodorus obtusus* in fall 2011. The top 2.54 cm of turf and thatch from each core was removed and set aside. The remaining soil from all cores was bulked, mixed, and packed into 10.16- × 15.24-cm columns. The original 2.54-cm layer of sod was replanted to the columns. Each column served as an experimental unit and was placed in the greenhouse and allowed to establish for 30 d. Treatments included a water control, the experimental abamectin based nematicide, azoxystrobin (as commercial formulation Heritage 50WDG), and a combination of the fungicide and the experimental nematicide. The fungicide was applied two times on a 6-wk interval at 1.2 kg/ha and the nematicide was applied four times on a 3-wk interval at 3.75 L/ha. All treatments, including the control, were applied with a penetrant-type surfactant. Columns were removed from the greenhouse 7 wk after the last application for evaluation. Measurements for each column included dry root weights and nematode counts per 100 cm^3 of soil. The study was repeated in 2012. Results indicate that dry root weights were a more useful measure of treatment effects than nematode population density, and data showed an interaction between the experimental nematicide and the fungicide. In 2013 in vitro toxicity experiments were conducted to compare the toxicity of the experimental nematicide with commercially available formulations of oxamyl (trade name Vydate L) and abamectin (trade name Avid). Nematodes were exposed to a range of concentrations for each treatment for 24 hr, followed by a 24-hr recovery period in water. The percentage of mortality was calculated at 2 and 24 hr of exposure, and after recovery. Probit analysis was used to determine the LD_{50}. Nematicidal and nematostatic dose ranges were established. Nematostatic activity is relevant to explaining why the effect of treatment is often more pronounced in root weights than in nematode population densities. Further, it appears that the addition of a fungicide improved the plants ability to tolerate nematode feeding and its use should be considered as part of an integrated nematode management strategy.

BIOMASS ESTIMATES OF NEMATODE SOIL ENERGY CHANNELS INDICATE CARBON FLOW FOR DECOMPOSITION STUDIES. **Shaw, E. Ashley[1], M.F. Cotrufo[2,3], and D.H. Wall[1,2].** [1]Department of Biology, Colorado State University, Fort Collins, CO 80523; [2]Natural Resource Ecology Laboratory, Colorado State University, CO 80523; [3]Department of Soil and Crop Sciences, Colorado State University, CO 80523.

Many studies use biomass measures of soil fauna trophic groups to estimate changes in soil food web structure and energy flow. We coupled this approach with a decomposition study using stable isotope enrichment to trace the flow of C into nematode trophic groups and to compare this quantified C flow with nematode energy channel biomass measures during decomposition of ^{13}C-labeled big bluestem (*Andropogon gerardii*) root litter. We hypothesized that biomass measures for nematode bacterial and fungal energy channels would indicate the proportion of root litter derived C incorporated into each nematode energy channel. We compared soil nematode food webs in annually burned tallgrass prairie (frequently burned, FB) with 20-yr burn tallgrass prairie (infrequently burned, IB) to assess C flow in differently managed grasslands. In a randomized, replicated greenhouse study, the ^{13}C-labeled dead roots, placed in litter bags, were buried in soil collected from FB and IB areas at the Konza Prairie Long Term Ecological Research site. Nematode biomasses and δ^{13}C values were assessed initially (time 0) and after 180 d of incubation. Results showed the nematode bacterial energy channel dominated over the nematode fungal energy channel in both FB and IB grasslands. Although FB grassland soil had significantly higher nematode bacterial energy channel biomass than IB at time 0, the bacterial energy channel biomass increased significantly after the addition of root litter in both soils and there were no differences in the nematode bacterial channel biomass between the two soils at the final harvest (180 d). There were also no differences between FB and IB soil's nematode fungal energy channel biomass at either time 0 or 180 d, and the addition of litter did not significantly affect total fungal energy channel biomass. ^{13}C analysis of nematodes confirmed our hypothesis, as more root litter-C was concentrated in the bacterial energy channel because of the dominance of this nematode bacterial energy channel in both FB and IB grassland soils. However, the IB soil's nematode bacterial energy channel had significantly more root litter derived C incorporated into its biomass than the FB soil, despite no differences in nematode bacterial energy channel biomasses between the IB and FB soil at the final harvest. Conversely, the FB soil food web showed an opposite effect—the nematode fungal energy channel incorporated more C even though there were no significant differences between the nematode fungal energy channel biomass of FB and IB soils. These results indicate that although energy channel biomass measurements of nematodes give a broad overview of C flow, ^{13}C decomposition tracer studies are more precise, and provide exact measures of C flow through soil food webs for ecosystem research.

UNVEILING THE MOLECULAR ARSENAL WITHIN *ROTYLENCHULUS RENIFORMIS* VIA DE NOVO TRANSCRIPTOME ASSEMBLY. **Showmaker, Kurt C. [1,2], W.S. Sanders[1], M.A. Arick II[1], Z.V. Magbanua[1,3], D.G. Peterson[1,3], and M.J. Wubben[2,3,4].** [1]Institute for Genomics, Biocomputing, and Biotechnology, Mississippi State University, Mississippi State, MS 39762; [2]Department of Biochemistry, Molecular Biology, Entomology and Plant Pathology, Mississippi State University, Mississippi State, MS 39762; [3]Department of Plant and Soil Sciences, Mississippi State University, Mississippi State, MS 39762; [4]USDA-ARS, Genetics and Precision Agriculture Unit, Mississippi State, MS 39762.

We present a transcriptome for the diploid sedentary semi-endoparasite *Rotylenchulus reniformis* derived from RNA isolated from the following developmental stages: egg, second-stage juvenile (J2), J3, vermiform adult (adult), and sedentary female (SF). Paired-end reads were generated on the Illumina MiSeq, then assembled using the Oases de novo transcriptome assembler module for the Velvet short read assembler. A total of 95,687 putative transcripts were generated with a total combined length of 1.47×10^8 nt and an N50 of 2,285 bp. In addition to transcript assembly, raw reads were mapped to 631 previously described *R. reniformis* unigenes that had been assembled from a SF EST library. As expected, more reads from SF were found to map to the 631 unigenes compared with reads from other life-stages. Of the 54,093 transcripts predicted to have Open Reading Frames (ORFs) annotated with the reviewed protein sequences in the UniprotKb, 36,035 and 18,058 transcripts were predicted to have full length ORFs or partial ORFs, respectively. To estimate the number of distinct ORFs, all predicted ORFs were clustered at a 90% identity threshold, which resulted in 18,535 clusters. Importantly, through comparative analysis with other plant-parasitic nematodes (PPN), we have identified *R. reniformis* transcripts that encode peptides that are homologous to known PPN effector proteins including annexin, β-1,4-endoglucanase, chitinase, chorismate mutase, pectate lyase, and ubiquitin extension protein. Of note, we identified two distinct cellulases not previously described for *R. reniformis* in addition to the Rr-ENG-1 cellulase that has been reported. To identify molecular pathways that are present in the assembled transcriptome, *R. reniformis* homologs were identified in *Caenorhabditis elegans* and subsequently used to search the KEGG, reactome, and wikipathways databases for *C. elegans* pathways. This pathway analysis resulted in the identification of *R. reniformis—C. elegans* homologous proteins that function in *C. elegans* pathways involved in mating, development, metabolism, signaling, and response to stimuli. In summary, this transcriptome assembly will serve as a valuable resource for the nematology community and will greatly facilitate the identification of molecular targets within *R. reniformis* that can be exploited to bring about host plant resistance.

MANAGEMENT OF ANTHURIUM DECLINE CAUSED BY *RADOPHOLUS SIMILIS*. **Sipes, B.[1], R. Myers[2], J. Lichty[3], and K. Sewake[1].** [1]Department of Plant and Environmental Protection Sciences. University of Hawaii at Manoa. Honolulu, HI 96822; [2]USDA Pacific Basin Agricultural Research Center, Hilo, HI 96720; [3]Department of Tropical Plant and Soil Sciences. University of Hawaii at Manoa, Honolulu, 96822.

Anthurium decline is a chronic problem in anthurium production in Hawaii. Anthurium decline has worsened with the removal of fenamiphos from the market. Growers need environmentally sound postplant treatments to augment preplant

management tactics. Avermectin (Avid, monthly), thiophanate-methyl (Cleary 3336 once every 3 mon), spinosad (Conserve once every 2 mon), spirotetramat (Movento, once every 6 mon), and imidacloprid (Provado once every 6 mon) were evaluated in a shade-house experiment. Each treatment was replicated over three plots containing 12 plants. Three months after transplanting, *Anthurium andraeanum* were inoculated with *Radopholus similis*. One month after inoculation, treatments were applied at labeled rates. Just before application of the first treatment, a single plant from each plot was arbitrarily selected for a nematode assay. A single plant was collected every 10 wk for the duration of the experiment. By 14 wk after the initial treatment, all the treatments had lower nematode populations per gram root compared with the untreated plants. Avermectin had the highest population and imidacloprid had the lowest nematode populations per gram root. By 6 mon, imidacloprid, spirotetramat, and thiophanate-methyl had the highest nematode populations among those anthurium receiving postplant treatment but all the treatments maintained nematode populations lower than the untreated plants. These treatments were still 69%, 78%, and 84% lower than the untreated plants. All of these products may provide postplant treatment options that can aid in the management of anthurium decline.

FIRST OCCURRENCE OF *MELOIDOGYNE FALLAX* IN NORTH AMERICA, AND MOLECULAR CHARACTERIZATION OF *M. FALLAX* AND *M. MINOR* FROM U.S. GOLF COURSE GREENS. **Skantar, Andrea M.[1], C. Nischwitz[2], Z.A. Handoo[1], M.N. Hult[1], M.E. Schmitt[3], and M.A. McClure[3].** [1]USDA-ARS Nematology Laboratory, Beltsville MD 20705; [2]Department of Biology, Utah State University, Logan UT 84322; and [3]School of Plant Sciences, University of Arizona, Tucson, AZ 85721.

Several species of root-knot nematodes (*Meloidogyne* spp.) are known to have significant presence on turfgrass in golf course greens, especially in the western United States. Nematodes isolated from a golf course in King County, WA, were identified as *Meloidogyne minor* based on analysis of the large ribosomal subunit (LSU 28S D2-D3 expansion segment), the internal transcribed spacers 1 and 2 (ITS-rDNA), the intergenic spacer region 2 (IGS2) and the nuclear protein-coding gene Hsp90. Sequence-characterized amplified region (SCAR) primers that were previously designed to be specific for *M. fallax* were found to cross-react with *M. minor*. A population from California was determined to be *M. fallax* based on juvenile tail morphology and analysis of the ribosomal markers and Hsp90. Using trees based on Hsp90 genomic alignments, the phylogenetic relationships of these populations and known root-knot nematode species were congruent with previous trees based on ribosomal genes. Resolution of *M. fallax* and *M. chitwoodi* using Hsp90 was equivalent to species separation obtained with 28S or 18S rDNA alignments. The strengths and weaknesses of ribosomal and Hsp90 markers, and the use of SCAR PCR as diagnostic tools also are discussed.

COTTON CULTIVARS RESPONSE TO FUSARIUM WILT AND ROOT-KNOT NEMATODE IN ALABAMA. **Smith, Amber[1], K.S. Lawrence[1], D. Schrimsher[1], and K. Glass[2].** [1]Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849; [2]Agronomy and Soils Department, Auburn University, Auburn, AL 36849.

Fusarium wilt (*Fusarium oxysporum* f. sp. *vasinfectum*, ''FOV'') is a serious disease of cotton, and is intensified by the presence of root-knot nematodes (*Meloidogyne incognita*). Twenty total cultivars were tested for Fusarium wilt resistance in addition to susceptible (Rowden) and resistant (M-315) checks in a randomized complete block design located at E. V. Smith Research Center, Plant Breeding Unit, Tallassee, AL. The Fusarium wilt disease pressure was excellent in 2012. The root-knot nematode pressure was ideal at the beginning of the trial, but dry conditions suppressed the nematode numbers later in the season; however, populations were not suppressed below the damage threshold. Initial stand counts were taken in June, and four different counts of wilted/diseased plants were taken throughout the season (twice in July, twice in August), where the wilted plants were removed from the field and reisolated to confirm the presence of the disease. FOV races 1 and 3 were the races of the fungus isolated most frequently in 2013. After these counts, the percentage of wilt incidence was calculated and used to determine the level of resistance in each variety. The varieties showing the highest resistance with the lowest levels of disease incidence were DP0912B2RF, PHYPX443314WRF, PHY367WRF, DG2570B2RF, FM1346GLB2, and FM1348GLB2. The cultivars with lowest populations of root knot eggs per gram of root were PHY367WRF, PHYPX443314WRF, and DP1252B2RF. Cultivars with yields statistically similar to and higher than the M-315 resistant check were DG2570B2RF, FM1346GLB2, and FM1348GLB2, producing 3,369, 3,640, and 3,343 lbs/a of seed cotton, respectively. Although these cultivars had comparable high yields, each supported a higher density of root-knot eggs than the resistant M-315 check. Thus these varieties may have some tolerance to root–knot nematode but not resistance.

THE SPARK THAT LED ME INTO NEMATOLOGY - DISEASE COMPLEXES WITH ROOT-KNOT NEMATODES. **Starr, J.L.** Department of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843-2132.

Like most of us, my decision to become a plant pathologist and to specialize in nematology was the result of a series of decisions, no one of which appeared all that important at the time. My first course in plant nematology was forced on me during my M.S. degree program to ''round out my background.'' Although not particularly interested in the subject matter, I did become intrigued by disease complexes, hence my Ph.D. program was selected because it was a biological phenomenon that intrigued and excited me. Throughout my career it was those topics that excited me that kept me going. In my teaching

efforts I have tried to get my students excited about the biology of whatever system was the topic of the day. Although most of my research efforts have been directed toward improved management, there has always been an element of excitement and fun. Research can be a demanding task master and many procedures require long hours doing repetitive, boring tasks (e.g., counting eggs from hundreds of samples), but it is also lots of fun looking over the data and developing conclusions that lead to yet another hypothesis. Therefore, get excited and enjoy your careers.

NEMATODE FAUNA ASSOCIATED WITH TREES GROWN IN SOUTH LOUISIANA. **Takeuchi, Yuko[1,2], E.C. McGawley[2], C. Overstreet[2], and A.R. Plaisance[2].** [1]Laboratory of Terrestrial Microbial Ecology, Graduate School of Agriculture, Kyoto University, Kyoto 606-8502, Japan; [2]Department of Plant Pathology and Crop Physiology, AgCenter, Louisiana State University, Baton Rouge, LA 70803.

A survey of the nematode fauna associated with trees was conducted in southern Louisiana, stretching from Louisiana State University, East Baton Rouge Parish, LA. Although nematode communities in underground part of trees are generally studied in relation to root diseases, there have been few reports that documented those in aerial part. Considering the case of pine wood nematode, *Bursaphelenchus xylophilus*, which was first described as a general mycophagous species inhabiting trees in Louisiana and later caused a serious forest disease in introduced countries worldwide, it is important to accumulate knowledge of nematode fauna associated with trees, possibly including potential pest species. We took samples of woody tissues from symptomatic and declining trees mainly of coniferous trees, including *Cedrus*, *Cupressus*, *Juniperus*, *Pinus*, and *Quercus* trees and extracted nematodes from them by Baermann funnel method. To date, we have obtained some *Aphelenchoides* and *Bursaphelenchus* species besides free-living species. In the presentation we will show a profile of nematode fauna associated with trees and discuss its properties.

AMENSALISTIC RELATIONSHIP BETWEEN *BURSAPHELENCHUS* SP. AND A JAPANESE NATIVE WEEVIL, *NIPHADES VARIEGATUS*. **Tanaka, Suguru E.[1], R. Tanaka[2], M. Akiba[2], Y. Takeuchi[1], and N. Kanzaki[2].** [1]Laboratory of Terrestrial Microbial Ecology, Graduate School of Agriculture, Kyoto University, Kita-Shirakawa, Kyoto, 606-8502, Japan; [2]Forest Pathology Laboratory, Forestry and Forest Products Research Institute, 1 Matsunosato, Tsukuba, Ibaraki, 305-8687 Japan.

The genus *Bursaphelenchus* consists of biologically variable species. Although the genus contains two lethal plant pathogens, the most *Bursaphelenchus* species are considered as insect-phoretic mycophagous species. However, insect parasitism have been reported or suspected for several species, e.g., *B. hylobianum* was isolated from a weevil species, *Hylobius albosparsus* from Russia as parasitic juvenile, and was originally described as ''*Parasitaphelenchus*'' *hylobianum*. An undescribed *Bursaphelenchus* species was isolated from five different localities in Japan. The nematode was associated with wood or bark of Pinaceae trees (*Pinus thunbergii*, *P. densiflora,* and *Abies veitchii*) as propagative stages, or the tracheal system of a Japanese native weevil species, *Niphades variegatus* as third-stage dispersal (dauer) juveniles (i.e., they did not parasitize the insects). Interestingly, the dauer juveniles associated with the weevil sometimes caused an abnormal expansion to insect trachea (i.e., the overload of nematode caused deformation of the weevil trachea, and it seemingly inhibits weevil's respiration), and therefore, the relationship was hypothesized to be an amensalism. In a collection site, Tama, Tokyo, the nematode infection was confirmed all 17 individual weevils examined (= 100% relevancy), and the number of nematodes isolated from individual beetles varied between 3 and 4,368. The general morphology (typological characters and morphometrics) and molecular barcodes (near-full-length of 18S, D2/D3 expansion segments of 28S and internal transcribed spacer region of ribosomal RNA genes) of the nematode suggest that the species is close to *B. antoniae*, *B. chengi*, and *B. hylobianum* (i.e., the undescribed species formed a well-supported clade with these three species). Except for *B. chengi*, which was isolated from wood, these species are associated with weevils. Thus, the phylogenetic clade is hypothesized as a weevil-associated clade with variable types of insect association including simple phoresy (*B. antoniae*), parasitism (*B. hylobianum*), and amensalistic phoresy (undescribed species).

EVIDENCE OF PLANT-PARASITIC NEMATODE-INDUCED SUPPRESSION OF HOST GENE SILENCING. **Taylor, Christopher G., and E. Walsh.** Department of Plant Pathology, The Ohio State University, Ohio Agricultural Research and Development Center, Wooster, OH 44691.

Plant-parasitic nematodes cause significant damage to crops worldwide. The root-knot nematode (RKN, *Meloidogyne* spp.), one of the most damaging nematodes because of its broad host range, establishes intimate feeding sites (giant cells) within the roots of a variety of plants. How plant-parasitic nematodes, such as RKN, cause such dramatic physiological changes whilst evading plant defenses is unknown. Recently it has been demonstrated that a variety of plant pathogens interfere with their host's gene silencing pathways. This work aims to provide a more refined look into how root-knot nematodes influence their host's silencing pathways during the course of infection. Interference of gene silencing pathways during nematode invasion was indicated in microarray data sets generated from laser-captured giant cells in *A. thaliana* roots. Subsets of genes regulated by small RNAs, as well as silencing machinery components that interact with small RNAs, were upregulated during the infection process. Furthermore, results examining the effects of compromising these pathways in *A. thaliana* and *N. tabacum*,

suggest that these components influence the host's susceptibility to RKN by allowing more adult females to form and increasing fecundity. We have generated multiple transgenic *N. tabacum* lines expressing a silenced reporter gene to detect the disruption of these pathways. During the course of infection, it is evident that the silenced reporter gene, targeted either by dsRNA or miRNA, is recovered specifically within giant cells. Better insight into this interaction will be invaluable to our growing understanding of the roles of host gene silencing during parasitic interactions.

METHODOLOGY DEVELOPMENT FOR PARTIAL RESISTANCE TESTING OF POTATO CULTIVARS RESISTANT TO *M. CHITWOODI*. **Teklu, Misghina Goitom, T.H. Been, and C.H. Schomaker.** Plant Research International, Wageningen University and Research Centre, Plant Sciences Group, NL- 6700 AP Wageningen, The Netherlands.

Meloidogyne chitwoodi was first described in 1995 in The Netherlands. It is now listed as a quarantine organism in the EPPO region with four EU member states officially infested. Since 1996, research has been initiated to identify resistant genes against *M. chitwoodi* from wild species of tuber bearing potatoes and integrate these genes into cultivated potatoes. Currently, several breeding companies successfully produced resistant genotypes against *M. chitwoodi*. Parallel to this, research was started to develop a standard methodology to screen the partial resistance of these genotypes. Population dynamical models were used to estimate their level of resistance, expressed as percentage of relative susceptibility (rs). This methodology provides farmers with quantitative information on the effect of growing resistant potatoes at any initial population density in their field. The models were first tested in a pilot project in 2010 with three resistant potato genotypes in (5 kg) pots at a range of 13 nematode densities. In 2011, another eight genotypes were tested in (10 kg) pots at a range of 12 densities. The results showed that Seinhorst's population dynamical models for nematodes with multiple generations fitted well, except in one genotype tested in 2011 that lacked resistance to *M. chitwoodi*, and a reduction of the number of densities used seems possible. In 2012, research was initiated to investigate whether the pot size can be downscaled from 10 to 5 or even to 2-kg pots—also at 12 densities—without loss of quality of the estimated relative susceptibility. Also, growth, yield loss, and quality damage as root knot index (RKI) were assessed and compared. Genotypes 2011M1, MDG2 and cv. Désirée (control) were the tested potatoes. The population dynamical model fitted well for the genotypes tested. The maximum multiplications rate ''*a*'' and the maximum population density ''*M*'' at 2-, 5-, and 10-kg pots were estimated and used to calculate the rs_a and rs_M values . Despite a decrease in ''*a*'' and ''*M*'' values with increasing pot size the rs_a values were relatively stable. The rs_M values were a bit higher in 5-kg pots. Seinhorst yield models used to describe the fresh tuber weight also fitted well. The RKI values obtained from the three pots sizes were also stable as a quality measure for industrial processing. Overall results indicate the possibility of downscaling the resistance test for *M. chitwoodi* in potato in terms of pot size and number of densities. Implications of the current research in the development of a cheap and reliable resistance test will be discussed.

AFRICAN HORNED CUCUMBER ROOTSTOCKS FOR MANAGING ROOT-KNOT NEMATODES IN GRAFTED MELON. **Thies, Judy, S. Buckner, and A. Levi.** U.S. Vegetable Laboratory, USDA, ARS, 2700 Savannah Highway, Charleston, SC 29414.

Southern root-knot nematodes (*Meloidogyne incognita*) are an important reemerging pest of melon (*Cucumis melo*) in the United States and worldwide. The reemergence of root-knot nematodes (RKN) in melon and many vegetable crops is in large part because of the phase-out of methyl bromide and to the loss of registrations for many other nematicides. Thus, there is a need to develop new technologies such as grafting for managing RKN in melon. Grafting melon to interspecific hybrid squash and bottleneck gourd rootstocks is commonly used in Asia and the Mediterranean for control of soilborne diseases, and is gaining acceptance as a disease management practice in some Central and South American countries. Currently, several U.S investigators also are studying the potential to manage soilborne diseases and nematodes using nonhost or resistant cucurbit rootstocks for grafted melon and watermelon. In 2012, we evaluated the response of 'Athena' melon scions grafted on 10 selected breeding lines of African horned cucumber (*Cucumis metulifer*) in a *M. incognita*-infested field in Charleston, SC. Nongrafted 'Athena' melon and 'Carnivor,' an interspecific hybrid squash rootstock (*Cucurbita maxima* x *C. moschata*), were included as checks. Nine of ten African horned cucumber lines exhibited moderate to high resistance to *M. incognita* and had significantly less galling ($P < 0.05$) (range 4% to 16% galling) than nongrafted 'Athena' (36.8%) and 'Carnivor' (90%). Five African horned cucumber lines exhibited low RKN reproduction (range: 7 to 37 eggs/g fresh root), compared with nongrafted 'Athena' (244 eggs/g fresh root) and 'Carnivor' (547 eggs/g fresh root). Six African horned cucumber lines produced significantly heavier ($P < 0.05$) fruit yields (30.1 to 40.9 kg per plot) than 'Carnivor' (19.6 kg per plot) and nongrafted 'Athena' (15.1 kg per plot). Our results demonstrate the potential of using African horned cucumber as a rootstock for managing RKN in grafted melon. Several of the African horned cucumber lines evaluated here can be used as sources of resistance for the development of rootstocks for grafted melon.

OBSERVATIONS REGARDING A PRESENTLY-UNDETERMINED *MELOIDOGYNE* SPECIES PARASITIZING YELLOW AND PURPLE NUTSEDGES. **Thomas, Stephen[1], J.M. Beacham[1], L. Holland[1], J. Schroeder[1], E. Morris[1], N. Schmidt[2], L. Murray[3], F. Solano-Campos[1], S. Hanson[1], and J.D. Eisenback[4].** [1]Department of Entomology, Plant Pathology and Weed Science, P.O. Box 30003 MSC 3BE; [2] Economics, Applied Statistic and International Business

Department, P.O. Box 30003, MSC 3CQ, New Mexico State University, Las Cruces, NM 88003; [3]Department of Statistics, Kansas State University, Manhattan, KS 66506; [4]Department of Plant Pathology, Virginia Tech, Blacksburg, VA 24061.
In January 2012 during a routine tomato bioassay of tubers from greenhouse cultures of yellow nutsedge (YNS, *Cyperus esculentus* L.) and purple nutsedge (PNS *Cyperus rotundus* L.), prominent galling was observed on PNS roots but was absent on 'Rutgers' tomato roots. This observation was unexpected, because neither PNS nor YNS exhibit readily-apparent galling when parasitized by *Meloidogyne incognita*, the target nematode of the bioassay. NaOCl extraction of roots recovered eggs from PNS but not tomato. Subsequent dissection of PNS root galls revealed small mature *Meloidogyne* females with egg masses that were primarily contained inside the root tissue. Further morphological observation revealed second stage juveniles (J2) with relatively long, thin tails and females with perineal patterns somewhat resembling those characteristic of *M. naasi*. A greenhouse study was conducted to compare the susceptibility of PNS and 'Rutgers' tomato with *M. incognita* and the undetermined *Meloidogyne* species (hereafter referred to as 'nutsedge root-knot nematode' = NSRKN). Results showed PNS to be a host of both NSRKN and *M. incognita*, whereas tomato was a host for *M. incognita* but not NSRKN. A second experiment was conducted to determine the host status of common New Mexico field crops and nutsedges to NSRKN. Alfalfa, chile pepper, corn, cotton, onion, sorghum, tomato, YNS, and PNS were inoculated with 2,000 NSRKN eggs and harvested 57 d later (700 cumulative heat units above 10 C). Galling was observed only on roots of YNS and PNS. Roots of each plant species were individually macerated in a 1.0% NaOCl solution in a blender. Eggs were recovered from YNS and PNS only. From previous reports, both alfalfa and sorghum are hosts for *M. naasi*. In a separate study, DNA was extracted from 56 NSRKN single J2 from multiple-infested nutsedge sources. After direct sequencing of an approximately 550 bp DNA fragment amplified using a primer set that targeted a locus between the mitochondrial cytochrome oxidase subunit II gene (COII) and the 16S rRNA gene (Powers and Harris, 1993, JON 25:1-6), all J2 showed high similarity to each other, but only 90% similarity to *M. graminicola* (closest BLAST hit) in a search of data bases containing *Meloidogyne* mitochondrial sequences. Neither results from the host range study nor direct sequence analysis of the COII-16S rRNA locus appear consistent with data previously reported for *M. naasi*. Further investigation is needed to identify this *Meloidogyne* species infecting nutsedges.

NEGATIVE FREQUENCY-DEPENDANT SELECTION IN *PASTEURIA PENETRANS* AND ITS HOST *MELOIDOGYNE ARENARIA*. **Timper, Patricia.** USDA ARS, P.O. Box 748, Tifton, GA 31793.

In negative frequency-dependant selection (NFDS), parasite genotypes capable of infecting the numerically dominant host genotype are favored, whereas host genotypes resistant to the dominant parasite genotype are favored, creating a cyclical pattern of resistant genotypes in the host population and, after a brief lag, virulent genotypes in the parasite population. The net effect is a selection against the numerically dominant host and parasite genotypes over time. For NFDS to occur there must be multiple host and parasite genotypes in the population, genotypic specificity for parasitism, and a high fitness cost to the host from parasitism. The bacterial parasite *Pasteuria penetrans* substantially reduces egg production of its nematode host, *Meloidogyne* spp. and endospores of the bacterium have a high degree of specificity for attachment to second-stage juveniles (J2). The objective of this study was to determine the changes in endospore attachment to J2 over time in a field population of *P. penetrans* that was parasitizing *M. arenaria*. Once per year from 1998 to 2012, soil was collected from four replicate plots at the Gibbs Farm near Tifton, GA. Juveniles from a greenhouse (GH) culture of *M. arenaria* were added to soil from each plot to bioassay for endospore attachment and, starting in 2010, J2 from a single egg mass line (SEM 6) were also used in the bioassay. From 1998 to 2006, endospore acquisition progressively declined from 95% to 2% of the GH J2 with spores. From 2007 to 2010, endospore acquisition increased from 12% to 87% of the GH J2 with spores. In recent years, endospore acquisition by GH J2 has again started to decline from 67% in 2011 to 53% in 2012, whereas endospore acquisition by SEM 6 J2 was 97 to 100% from 2010 to 2012. Moreover, average spores per J2 increased from 4 to 22 for the SEM 6 line and decreased from 13 to 8 for the GH culture between 2010 and 2011. The GH culture, like the field population of *M. arenaria*, is heterogeneous for endospore attachment; however, the GH culture is not under selection pressure from *P. penetrans*. Consequently, genotypic frequencies in the GH culture should remain constant. The changes in spore acquisition over time likely reflect changes in the frequency of *P. penetrans* genotype capable of adhering to the dominant host genotype in the GH culture and to the single genotype in SEM 6. It appears that the dominant attachment genotype in the GH culture is different from SEM 6.

THE NOVEL GR29D09 EFFECTOR FAMILY FROM THE POTATO CYST NEMATODE *GLOBODERA ROSTOCHIENSIS* SUPPRESSES PLANT IMMUNITY TO PROMOTE NEMATODE PARASITISM. **Tran**, **Tien[1]**, **S. Chen[1]**, **and X. Wang[1,2].** [1]Department of Plant Pathology and Plant-Microbe Biology, Cornell University, Ithaca, NY 14853; [2]USDA-ARS, Robert W. Holley Center for Agriculture and Health, Ithaca, NY 14853.

Potato cyst nematode *Globodera rostochiensis* is a quarantine pest that threatens the U.S. potato industry. This obligate biotrophic pathogen induces feeding cells within the host root that serve as the sole nutrient source for the feeding nematode. It is now believed that effector proteins originated from nematode esophageal gland cells play an essential role in the formation and maintenance of feeding cells. Interestingly, there is emerging evidence that effectors from plant-parasitic

nematodes also have a role in host defense suppression. We have cloned a family of effector genes (*Gr29D09*) from *G. rostochiensis* that showed a specific gland expression and a dramatic upregulation in nematode parasitic stages, suggesting an involvement of this effector family in nematode parasitism. Generated transgenic potato lines overexpressing individual *Gr29D09* genes were found to be more susceptible not only to *G. rostochiensis* but also to *Streptomyces scabies*, which is a bacterial pathogen causing common scab of potato. The increased susceptibility to adapted pathogens is often a result of suppression of pathogen-associated molecular pattern (PAMP)-triggered immunity (PTI). To investigate a role of this effector family in PTI suppression, we evaluated whether members of this effector family can suppress the bacterial PAMP flg22-induced PTI responses in *Nicotiana benthamiana* and *Arabidopsis thaliana*. Using Agrobacterium-mediated transient expression in *N. benthamiana*, we found that Gr29D09 variants suppressed flg22-triggered PTI responses including the production of reactive oxygen species (ROS) and the induction of two PTI marker genes. Similarly, transgenic *Arabidopsis* overexpressing individual *Gr29D09* genes were compromised in ROS production in response to flg22. These results clearly demonstrated an activity of the Gr29D09 family in PTI suppression. PTI appears to be conserved across the plant kingdom. Although the role of PTI in plant-nematode interactions needs to be elucidated, we believe that understanding the function of Gr29D09 effectors would provide insights into the mechanism of PTI in the plant-nematode pathosystem. Members of the Gr29D09 effector family showed significant sequence variations, which may indicate a diversification of the family to expand their virulent targets in host plants. We are currently using different approaches to identify host targets of Gr29D09 variants and the knowledge developed may help discover the specific pathways that this effector family plays in modulating plant immunity.

COVER CROP EFFECTS ON NUTRIENTS, SOIL FAUNA AND SOIL QUALITY. **Donald Tyler[1], and Patricia Donald[2].** [1]University of Tennessee, Biosystems Engineering and Soil Science, West Tennessee Research and Education Center, 605 Airways Blvd., Jackson, TN 38301; [2]USDA/ARS, West Tennessee Research and Education Center, 605 Airways Blvd., Jackson, TN 38301.

In the Southeast United States, cover crops are usually winter annuals planted in the fall, allowed to grow through the winter and spring, and then terminated close to the time of planting of the following row crop. These can be grass or legume species. The grass species commonly used are wheat or cereal rye. The two legumes recommended in Tennessee are crimson clover and hairy vetch. Both types of cover crops have advantages and disadvantage in row crop production. Ideally, cover crops are planted using no-tillage in previous crop residue. The effects of these practices on levels of soybean cyst nematode and microbial and fungal community structure have been studied for a number of years and these findings will be summarized. Cover crops can offer a number of advantages when properly managed in row crop systems. They can supply additional soil cover, which is especially useful in systems such as nonrotated cotton where limited residue remains on the soil surface even in continuous no-tillage. Although the cover crop is growing in the fall, winter, and spring it also captures additional carbon that would not be the case in most fallow situations. This can result in increases in soil carbon storage in no-tillage systems that may lower soil density, enhance aeration, and increase microbial biomass and result in enhanced biodiversity of soil macro-organisms and micro-organisms. Grass cover crops tend to make more fall growth than the legumes when planted after crop harvest. This is especially true when following cotton that in most cases is harvested later than corn or soybeans. This means that the grass covers will provide more additional soil cover and therefore more winter erosion protection than is usually the case with the winter annual legumes. However, the legumes offer other distinct advantages including fixation of atmospheric nitrogen while growing. After growth termination, a part of this nitrogen is recycled to the following row crop as the legume cover crop residue decomposes. We recommend that if either crimson clover or hairy vetch is allowed to grow to midbloom before termination and subsequent row crop establishment that fertilizer nitrogen application can be reduced by 70 to 90 kgN/ha for a number of crops including corn and cotton. The value chosen in this range depends on above ground biomass accumulation that can be related to initial stand establishment, growing conditions, etc. Cover crops can offer residue management enhancement in a number of row crop production systems especially in no-tillage cropping. However, they do cost money, time, and management. If they can be properly managed in the cropping system they can provide a number of soil quality and environmental benefits and in some cases result in higher yields and greater profitability.

DIRECT COMPARISON OF SOYBEAN CYST NEMATODE REPRODUCTION ON RESISTANT SOYBEAN VARIETIES IN GREENHOUSE AND FIELD EXPERIMENTS. **Tylka, Gregory L.[1], M.T. McCarville[2], C.C. Marett[1], G.D. Gebhart[1], D.H. Soh[1], M.P. Mullaney[1], and M.E. O'Neal[2].** [1]Department of Plant Pathology and Microbiology, Iowa State University, Ames, IA 50011; [2]Department of Entomology, Iowa State University, Ames, IA 50011.

Experiments were conducted in 2011 and 2012 to compare reproduction of the soybean cyst nematode (SCN), *Heterodera glycines*, on resistant soybean varieties in the field with reproduction of the nematode in greenhouse experiments using SCN-infested soil taken from the field. Field experiments consisted of 4-row-wide × 5.2-m-long plots in SCN-infested fields at nine locations arranged across three districts (northern, central, and southern) in Iowa each year. Fifty to sixty soybean varieties marketed as resistant to SCN, plus three or four SCN-susceptible varieties, were grown in four blocks in three experiments in each district both years. Ten soil cores were collected from each plot at planting and again at harvest. Cysts of

SCN were extracted from a 100-cm^3 subsample taken from each soil sample, then eggs were extracted from cysts and counted to determine the initial (Pi) and final (Pf) egg population densities in each plot. A reproductive factor was calculated for each plot by dividing Pf by Pi. For one experiment (referred to as the source field experiment) in each district each year, after Pi values were determined from soil samples collected at planting, the remaining soil was combined, mixed well, and used in a greenhouse experiment. Each soybean variety grown in the source field experiment was planted in four replicate cones (21-cm long, 3.8-cm diam.) filled with the SCN-infested soil from the source field. The cones were placed in plastic buckets filled with sand and incubated in a water bath at constant 27°C in the greenhouse under natural and supplemented lighting. Each variety was grown in four replicate blocks. After 30 d, soil was carefully removed from each soybean root, and roots were sprayed with water to dislodge SCN females, which were counted. An SCN female index was calculated for each variety by dividing the mean number of SCN females on the variety by the mean number of SCN females on roots of three susceptible soybean plants in the experiment. Mean SCN female indices from each greenhouse experiment were correlated with mean RF values, after natural log transformation (Ln), for the same varieties in the source field experiment and separately with mean Ln RF values for the varieties in experiments conducted the same year at other locations in the district (called nonsource fields). Correlations between female indices in the greenhouse and Ln RF values from field experiments were highly significant ($P < 0.001$) for all SCN-resistant varieties and for varieties with the PI 88788 source of resistance, but not significant for varieties with the Peking source of resistance. Coefficients of determination (R^2) for significant correlations were 0.36 or less, indicating that SCN female indices in the greenhouse were not highly associated with SCN RF values in the field. Female indices in greenhouse experiments were more highly correlated with RF values from source fields than from nonsource fields in the same growing season. Results indicate that the effects of resistant varieties on SCN population densities in the field are not predicted well by female indices obtained in 30-d greenhouse experiments.

RELATION BETWEEN VIRULENCE AND OXIDATIVE STRESS RESPONSE OF *BURSAPHELENCHUS XYLOPHILUS* AND *B. MUCRONATUS*. **Vicente, Claudia S.L.[1,2], Y. Ikuyo[1], M. Mota[2], and K. Hasegawa[1].** [1]Department of Environmental Biology, College of Bioscience and Biotechnology, Chubu University, Kasugai, 487-8501 Japan; [2]ICAAM-Instituto de Ciências Agrárias e Ambientais Mediterrânicas, Departamento de Biologia, Universidade de Évora, Évora, 7002-554, Portugal.

In plants, oxidative burst (massive production of reactive oxygen species, ROS) is the earliest and fastest defense response against biotic and abiotic stresses. *Bursaphelenchus xylophilus*, the pine wood nematode (PWN), is the causal agent of pine wilt disease. Its aggressiveness to pinelands and consequent environmental and economic impact, made PWN to be considered a worldwide quarantine pest. Little is known about oxidative stress (OS) response of PWN. Our work concerns the study of OS tolerance of virulent and avirulent *Bursaphelenchus xylophilus* isolates and avirulent *Bursaphelenchus mucronatus* and the relation with their pathogenicity (virulence level) to susceptible pine species. Two *Bursaphelenchus xylophilus* isolates, Ka4 (virulent) and C14-5 (avirulent), and one *Bursaphelenchus mucronatus* (avirulent) were tested for OS tolerance using hydrogen peroxide as oxidative agent, in concentrations ranging from 0 to 40 mM H_2O_2. Expression levels of nematode antioxidant enzymes, catalase (*ctl-1*) and superoxide peroxide (*sod-1*), were also quantified using qRT-PCR. Our results show a clear difference in OS tolerance between *B. xylophilus* Ka4 and *B. xylophilus* C14-5/*B. mucronatus*, whereas the virulent PWN is more tolerant in all H_2O_2 concentrations tested as well as presenting a higher expression levels of antioxidant enzymes. We hypothesized a possible positive correlation between the level of OS tolerance and the level of virulence of *Bursaphelenchus xylophilus*, which can be further investigated as a virulence marker to characterize PWN.

INSIGHT INTO KIP-RELATED PROTEINS REVEALS THEIR INVOLVEMENT IN THE CONTROL OF ROOT-KNOT NEMATODE FEEDING SITE DEVELOPMENT. **Vieira, Paulo[1,2], and J. De Almeida Engler[2].** [1]NemaLab/ICAAM, Instituto de Ciências Agrárias e Ambientais Mediterrânicas, Universidade de Évora, Núcleo da Mitra, Ap. 94, 7002-554 Évora, Portugal, [2]Institut National de la Recherche Agronomique, UMR 1355 ISA/Centre National de la Recherche Scientifique, UMR 7254 ISA/Université de Nice-Sophia Antipolis, UMR ISA, 400 Route des Chappes, Sophia-Antipolis, France.

Root-knot nematodes are considered one of the most specialized and harmful plant-parasitic nematodes. The capacity of manipulating the host cell cycle machinery is a fundamental process for the successful reproduction of these plant pathogens. Understanding the molecular mechanism of how root-knot nematodes maneuvers the host cell cycle can bring new opportunities to exploit diverse and novel forms of plant resistance. In plants, the regulation of the cell cycle is driven by the activation of cyclin-dependent kinases (CDKs) through multiple transcriptional and posttranslational mechanisms that control their activity. In this scenario, plant Kip-Related Proteins (KRP inhibitors) are CDK regulators, and as a consequence, modulating KRP activity can be envisaged to block nematode feeding site development. Here we investigated the involvement of this gene family during plant-nematode interaction, and our data robustly support that members of the *ICK/KRP* gene family control gall development in *Arabidopsis*. Our results show that by intensifying CDK inhibition through overexpression of ICK/KRP family members, dramatically affects nematode feeding site development, thus compromising the nematode's life cycle. Hence, a promising manner to control nematode propagation could be achieved by modulating cell cycle regulators in host crop species.

SEQUENCE VARIABILITY OF THE *MSPI* SATELLITE DNA FAMILY OF THE PINEWOOD NEMATODE *BURSAPHELENCHUS XYLOPHILUS* AT DIFFERENT GEOGRAPHIC SCALES. **Vieira, Paulo[1], C. Castagnone[2], S. Mallez[2], M. Espada[1], A. Navas[3], M. Mota[1], and P. Castagnone-Sereno[2].** [1]NemaLab/ICAAM, Instituto de Ciências Agrárias e Ambientais Mediterrânicas, Universidade de Évora, Núcleo da Mitra, Ap. 94, 7002-554 Évora, Portugal; [2]INRA, UNS, CNRS, UMR1355 Institut Sophia Agrobiotech, 400 Route des Chappes, F-06603, Sophia Antipolis, France; [3]Dpto Biodiversidad y Biología Evolutiva, Museo Nacional de Ciencias Naturales, Consejo Superior de Investigaciones Científicas, Calle José Gutiérrez Abascal, 2 Madrid 28006, Spain.

The *Msp*I satellite DNA (satDNA) family of the pinewood nematode *Bursaphelenchus xylophilus* is herein analyzed in an attempt to understand the intraspecific evolutionary dynamics effects related to geographic distribution, structure and repeat-copy variation in the molecular drive process leading to the concerted-evolution pattern that generally occurs for this type of repetitive sequences. A total of 425 *Msp*I monomer units, either PCR-amplified from isolates of local (Peninsula of Setúbal, Portugal) or worldwide origin, or retrieved from the *B. xylophilus* genome sequence, were characterized and compared. Whatever their origin, sliding window analysis of sequence variability patterns among monomers revealed low, moderate, and highly variant domains, indicating that variable levels of evolutionary constraint may act on the entire monomers. The phylogenetic inference based on the different sets of *Msp*I satDNA family for this species shows a broad polymorphism of the individual monomers, which were distributed into four main clusters. However, such clustering appeared independent from the geographic origin of the nematodes, and could not discriminate isolates or groups of geographically close isolates. Rather, the formation of different phylogenetic groups within this satDNA family suggests an *a priori* embodying of a set of diverging repeats from a common ancestor satDNA library, which have been differently amplified along the evolutionary pathway of this species. The present work improves knowledge on the evolutionary dynamics of satDNA at the intraspecific level, and provides new information on satDNA sequence variability among natural populations sampled at a local geographic scale.

EFFECTS OF SURFACE MULCH ON SOIL HEALTH CONDITIONS IN CONSERVATION-TILLAGE SYSTEMS. **Wang, Koon-Hui[1], and C.R.R. Hooks[2].** [1]Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822; [2]Department of Entomology, University of Maryland, College Park, MD 20740.

The benefits of conservation- or no-tillage system on soil health conditions could be multiplied if accompanied by various soil health enhancement practices. Two approaches of soil health enhancement techniques to be discussed separately here are cover cropping and natural farming. In the cover cropping experiment, we compared the effect of four soil surface treatments on nematode communities. Three months before cash crop planting, four preplant soil surface treatments installed were: (i) plant sunn hemp (*Crotalaria juncea*) cover crop and mow SH as surface mulch; (ii) mix planting of SH and rapeseed (*Brassicus napus*) (SH+RS) in rows and strip-tilled rapeseed rows into soil while mowed down SH as surface mulch; (iii) cover the soil with metallic mulch (MM); and (iv) leave the soil surface uncover followed by herbicide treatments (C). Experiment was arranged in randomized complete block design with three replications. Zucchini (*Cucurbita pepo*) seedlings were transplanted at termination of the cover cropping period, and all plants were top dressing with organic fertilizer. Nematode community analysis conducted at the end of zucchini harvest indicated that cover crop mulch (SH or SH+RS) resulted in higher nematode richness, greater opportunistic bacterivorous and total fungivorous nematodes than MM. MM reduced nematode richness compared with the untreated C. Although both cover crop treatments enriched soil nutrients and enhanced richness of nematode communities, they did not improved nematode community structure at the end of the zucchini crop. Thus, mulching with cover crop residues alone did not fully enhance soil health conditions. In the second experiment, soil health conditions were compared between SH organic mulch vs. synthetic weed mat (WM) in a no-till cropping system. In addition, under each mulching system, soil microbial activities were either stimulated by introducing indigenous microorganisms (IMO) prepared according to Korean Natural Farming (KNF) practice or not stimulated. KNF is a farming practice to cultivate IMO collected from undisturbed tree root zones with various organic substrates and then made into compost. To further supplement nutrient, foliar sprays prepared from fermented plant and kitchen waste materials were applied to the plant at weekly interval according to KNF practice. No additional fertilizer was applied in KNF plots. Thus, this experiment was a 2×2 (farming practices $\times$ mulching) designed experiment. The two farming practices were KNF vs. organic farming (received chicken pellets as fertilizer). Cherry tomato seedlings were transplanted. Soil health conditions were monitored over a 5-mon period. KNF enhanced soil health conditions (increased in abundance of bacterivorous, omnivorous and predatory nematodes) at 3 mon after tomato planting only in SH mulched plots but not in WM plots. In conclusion, although adding organic surface mulch enriched soil nutrients in conservation tillage agroecosystems, adding IMO to organic mulching plots could further speed up and complete the process of soil health enhancement in a conservation tillage system.

REAL-TIME PCR FOR SPECIES IDENTIFICATION OF PINE WOOD NEMATODE *BURSAPHELENCHUS XYLOPHILUS* (NEMATODA: PARASITAPHELENCHIDAE). **Ye Weimin[1], and Robin M. Giblin-Davis[2].** [1]Nematode Assay Section, Agronomic Division, North Carolina Department of Agriculture and Consumer Services, Raleigh, NC 27607; [2] Fort Lauderdale Research and Education Center, University of Florida/IFAS, 3205 College Avenue, Davie, FL 33314.

Bursaphelenchus xylophilus, the pine wood nematode (PWN), is the causal agent of pine wilt, one of the most damaging emerging pest problems to forests around the world. It is native to North America where it causes relatively minor damage to native conifers, but is labeled an EPPO A1 pest and a quarantine nematode for many countries outside of the United States. because of its potential for destruction to naïve nonnative conifers. Wood logs and commodities involving softwood packaging materials that are exported each year from the United States now require a lab test for the presence/absence of this regulated nematode species. We describe the development of a real-time PCR method for rapid and accurate identification of PWN targeting the ribosomal DNA internal transcribed spacer region (ITS-1). A total of 86 nematode populations, including 36 populations of *B. xylophilus*, 36 populations of 21 different species of *Bursaphelenchus*, 13 populations of *Aphelenchoides besseyi*, *A. fragariae*, *Aphelenchoides* species and *Aphelenchus avenae*, and one population of mixed nematode species from a soil sample were used to evaluate the specificity and efficacy of this assay. This assay proved to be specific to *B. xylophilus* only and was sensitive to a single nematode specimen. Species confirmation using this approach is critical to provide rapid species identification service to our stakeholders to help manage this pest using zero tolerance regulations in export.

APPLICATION OF VARIOUS ORIENTAL MUSTARD - ALLYL ISOTHIOCYANATE COMPOUNDS TO SUPPRESS SOYBEAN CYST NEMATODES IN ONTARIO. **Welacky, Tom.** Agriculture and Agri-Food Canada, Science and Technology, 2585 County Rd 20, Harrow, Ontario, N0R 1G0, Canada.

Oriental Mustard (OM) seed is grown in western Canada and is processed in several areas of the country primarily for confectionary spice products. Products of OM seed processing are mustard oil, ground mustard flour and bran. OM by-products of oil, bran and seed coats contain various forms of allyl isothiocyanate (AITC) that may range in concentrations of 0.01% to 2%. Several OM materials such as pelleted formulations and ground bran composed of ground seed coat, crushed seed and residual oils were tested on 1.25 m^{-3} field micro-plots infested with Soybean Cyst Nematodes (SCN) *Heterodera glycines* from 2008–2011. Micro-plots were established in 1994 in a field of Harrow Fox sandy loam, cultured with fallow, corn, and soybean rotations with the occasional nonhost crop to develop moderate-high populations of SCN cysts and eggs. In 2008 various rates of pelleted OM were applied, incorporated, and watered as required to create good dissolving conditions for the material. In 2009–2011, OM ground bran was incorporated and plots were watered either with irrigation or natural rainfall to dissolve the material. The 2008 pelleted material contained a range of 1% to 1.5%, 2009 ground bran had 0.2% to 0.9% AITC and 2010–2011 ground bran contained double the AITC concentration as compared with the 2009 bran. Soybeans resistant to SCN were planted in a uniform spaced pattern each year. Plant measurements were taken as required. Micro-plots were soil sampled for SCN cysts before OM application, midseason and late fall. Cysts and eggs were extracted and processed for counting. Across the 4 yr of micro-plot trials, results indicated considerable variation between types of OM materials, applied rates and concentrations of AITC. In general, SCN egg populations were not consistently suppressed by the Oriental Mustard products. Details of the impact of OM products and concentrations on SCN populations are presented.

MCW-2 FOR MANAGEMENT OF ROOT-KNOT NEMATODE ON ANNUAL CROPS. **Westerdahl, Becky[1], D. Long[2], and C.T. Schiller[2].** [1]Department of Entomology and Nematology, University of California, Davis, CA 95616; [2]Makhteshim-Agan of North America, Raleigh, NC 27604.

Five RCB field trials with five replicates were conducted to evaluate the effectiveness ($p = 0.05$) compared with an untreated control (UC) of MCW-2 for management of root-knot nematode (RKN), *Meloidogyne javanica*, on carrot, tomato, squash, cucumber, and cantaloupe. Treatments in all trials were MCW-2 at 2, 3, 4, 6, and 8 kg ai/ha, oxamyl at 4.7 l/ha, metam sodium (MS) at 589 l/ha, 1,3-dichloropropene (1,3-D) at 84 l/ha, and UC. 1,3-D was injected 14-d preplant. MS, MCW-2, and oxamyl were applied 7-d preplant followed by rototilling and sprinkler irrigation. Evaluations were conducted at harvest. The 3 and 8 kg rates of MCW-2 had a higher percentage of marketable carrots. All MCW-2 rates and 1,3-D had fewer RKN. On tomatoes, 4 kg MCW-2 had a greater weight of fruit plus foliage. MS had a greater weight of fruit plus foliage and a greater weight of fruit; 3, 4, and 8 kg MCW-2 and 1,3-D had a lower root gall rating (RG); 4 and 8 kg MCW-2 and 1,3-D had fewer RKN. On cucumbers, 4 and 8 kg MCW-2 had a greater number, weight, and size of fruit. MCW-2 at 2 and 4 kg had a lower RG. All treatments had fewer RKN. On squash, fruit size was greater for 4 and 8 kg MCW-2. MCW-2 at 2 and 4 kg had a lower RG. All treatments had fewer RKN. On cantaloupe, all treatments except 3-kg MCW-2 and oxamyl had a greater fruit weight. MCW-2 at 4 kg, and MS had a larger number of fruit. MCW-2 at 3 kg had larger fruit. At 2 and 8 kg MCW-2 had a lower RG. All treatments had fewer RKN.

EFFECTS OF DEEP-OCCURRING POPULATIONS OF *HETERODERA SCHACHTII* ON SUGAR BEET GENOTYPES. **Westphal, Andreas[1], A. Meinecke[1], A. Hermann[2], K. Ziegler[3], K. Bürcky[4], and M. Daub[1].** [1]Julius Kühn-Institut, Institute for Plant Protection in Field Crops and Grassland, Messeweg 11/12, 38104 Braunschweig, Germany; [2]Bavarian State Research Center for Agriculture, Institute for Plant Protection, Lange Point 10, 85354 Freising; [3]Working Group for Sugar Beet Production Franconia, Würzburger Straße 44, 97246 Eibelstadt, Germany; [4]Südzucker AG, Marktbreiter Straße 74, 97199 Ochsenfurt, Germany.

Heterodera schachtii reduces yields in sugar beet and cruciferous vegetable production worldwide. In Europe, its damage is mitigated by crop sequences that utilize nonhost plants and resistant cover crops. Recent findings have demonstrated the damage potential of populations of the nematode below the plow layer depth of 0 to 30 cm that is typically examined for nematode presence. In recent studies in commercial fields, the occurrence of large population densities in the depth of 30 to 60 cm has been confirmed. The objective of this project was to model the damage potential of populations at these greater depths. Data from several experiments with differential population densities at the 0- to 30-cm and 30- to 60-cm depths were used in addition to one experiment with 30-cm-diam. tubes perpendicularly inserted 0 to 60 cm into the ground. In this experiment, the tubes were filled with steamed soil that was either noninfested or inoculated at the following depths: 0 to 60 cm noninfested; 0 to 60 cm infested; 0 to 30 cm infested, 30 to 60 cm noninfested; and 0 to 30 cm noninfested, 30 to 60 cm infested. For each of these infestation patterns, there were five replications at *H. schachtii* inoculation densities of 200 to 2,000 eggs per 100 g of soil. Each plot was then planted with a susceptible sugar beet cultivar. Root penetration by second-stage juveniles (J2) of *H. schachtii* and canopy diameter at the beginning of the growing season were measured. Canopy diameter data and penetration numbers were negatively correlated. This information was used to develop a predictive model for the contribution of nematodes at different soil depths to root penetration and plant growth. In an extension of these experiments, the model was tested for validity with 39 field experiments conducted in growers' fields. Populations of *H. schachtii* below the 0- to 30-cm plow layer contributed to growth and performance of sugar beet. Not only will the mathematical model for yield predictions depend on nematode population densities but likely also on agroecological parameters, including weather patterns and site-specific characters.

A NON-PATHOGENIC INQUILINE, KLEPTOPARASITIC FUNGUS INHABITING THE INTESTINE OF RHABDITID NEMATODES. **Whitlock, Kimberly J., E.C. Bernard, B. H. Ownley, and A. Bruce.** Entomology and Plant Pathology Department, The University of Tennessee, 2505 E.J. Chapman Drive, 370 Plant Biotechnology, Knoxville, TN 37996-4560.

Rhabditid nematodes are known to infest earthworm cocoons and destroy developing embryos, presumably by introduction of bacteria that consume the yolk-like food reserve. This relationship may impact soil biodiversity and function by reducing earthworm populations. Numerous *Eisenia fetida* cocoons from a compost operation were observed to be infested with nematodes (*Rhabditis* sp. near *blumi*). These cocoons did not contain developing earthworms. Male, female, and juvenile nematodes were all present and easily cultured. This nematode population is obligately amphimictic and dauer juveniles do not nictate; life span of females is about 2 wk. Many of the adult nematodes contained a fungus inside the nematode intestine. The fungus appeared to be one continuous thallus with a spiraling mycelium against the inside intestinal wall. The fungus did not directly harm infected nematodes, implying a nonpathogenic relationship in which the fungus absorbs digested food from the intestinal lumen. This phenomenon has not been documented in published literature. Our immediate objectives were to: (i) confirm presence of the fungus in a significant number of nematodes; (ii) isolate and cultivate the fungus; (iii) identify the fungus with morphological and molecular approaches; and (iv) perform Koch's postulates to confirm the infection. Nematodes from cocoons were cultured on weak cornmeal agar (CMA) and transferred frequently to maintain viable, fungus-infected nematodes. The fungus was obtained in pure culture by surface-sterilizing infected females and rupturing them on antibiotic-amended PDA, or by crushing infected nematodes in sterile water on glass slides and rinsing them onto amended PDA. Identification of the fungus was confirmed with PCR amplification of the internal transcribed spacer (ITS) region of ribosomal DNA and primer pairs ITS4 and ITS5. The 557-bp PCR amplicon was sequenced and had 100% nucleotide identity to *Sarocladium strictum* (= *Acremonium strictum*) isolated as an endophyte (GenBank Accession No. KC172080) from the Chinese herb *Atractylodes lartoea*. This widespread fungus has been isolated from cysts of *Heterodera glycines* but not reported in microbivorous nematodes. The infection occurs in J4s and adults after ingestion of spores; earlier stages appear to have stomas too narrow to allow spore passage. The fungus occurs only in the intestine but does not invade the intestinal wall. Infected females reproduce successfully. Inside nematodes the fungus produces three types of spores: oval 1- to 2-celled conidia (6-μm length), long, slender, seven-septate conidia (65 μm), and crescentiform ascospores that develop in intercalary asci, with four spores per ascus. Asci and slender conidia are formed more abundantly in older, starved nematodes. Ascus formation and ascospore shape somewhat resemble those in *Ascobotryozyma americana*, a nonparasitic ectocommensal fungus on the cuticle of *Panagrellus dubius*. Further research will include host range studies, timing of infection, possible effects on fecundity and life span, roles of the spore types, and a formal description of the perfect state.

INFLUENCE OF SOIL TEXTURE AND COTTON CULTIVAR ON REPRODUCTION AND PATHOGENICITY OF DIFFERENT ISOLATES OF *ROTYLENCHULUS RENIFORMIS*. **Xavier, Déborah Magalhães, C. Overstreet, E.C. McGawley, M. Kularathna, and C.M. Martin.** LSU AgCenter, Department of Plant Pathology and Crop Physiology, 302 Life Sciences Building, Baton Rouge, LA 70803.

Greenhouse studies were conducted to evaluate the influence of soil texture on reproduction and pathogenicity of different isolates of *Rotylenchulus reniformis* (reniform nematode) on different cotton cultivars. A 45-d duration greenhouse experiment confirmed the pathogenicity of an isolate of *R. reniformis* from Avoyelles Parish on Stoneville LA887 cotton. A series of greenhouse experiments were conducted with three geographic isolates of *R. reniformis* (identified as Avoyelles, Evangeline, and

Rapides to indicate the parish of origin) on Stoneville LA887, Stoneville 5288B2F, and Phytogen 375WF cotton growing in soils with varying textures for 60 days. Soil types with sand, silt, and clay contents ranging from 74.4 to 7.8, 20.7 to 66.3, and 4.9 to 25.9, respectively, were employed in this research. Variations in soil texture significantly affected plant height and dry weights in both Stoneville 5288B2F and Phytogen 375WF, but did not have any significant effect on plant growth of Stoneville LA887, except in the 45-d duration experiment. Stoneville 5288B2F plants were significantly taller throughout the experiment in soil with 31.4% sand and 13.3% clay. Phytogen 375WF cultivar showed the same pattern, but the difference in plant height was not observed at harvest. Stoneville 5288B2F and Phytogen 375WF had significantly reduced dry root and shoot weights in sandier soils. Soil type had a significant effect on nematode reproduction on all three cotton cultivars. The interaction between soil type and reniform isolate significantly affected population densities of all reniform isolates tested among the multiple soil types in the cultivars Stoneville 5288B2F and Phytogen 375WF, but no effect was observed in Stoneville LA887 cultivar.

OPTIMIZATION OF CHEMICAL AGITATION FOR DISTINGUISHING LIVE AND DEAD *HETERODERA GLYCINES* JUVENILES *IN VITRO*. **Xiang, N., D. Schrimsher, and K. Lawrence.** Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849.

Multiple methods, stains, and irritants have been used to determine if plant-parasitic nematodes are alive or dead once exposed to a pesticide or biological compounds. The objective of this research project was to select an optimal technique to rapidly distinguish live and dead second-stage juveniles (J2s) of soybean cyst nematode (SCN, *Heterodera glycines*). In vitro assays were used to select the best irritant of 1N sodium carbonate (Na_2CO_3), 1N sodium bicarbonate ($NaHCO_3$), or 1N sodium hydroxide (NaOH). Further test evaluated the optimum pH, the application contention, and time of movement. Tests were established in 100µl 96-well plates or in 50-ml conical tubes. Each trial was repeated twice and each treatment had four replications. A growth chamber experiment to confirm the results of the in vitro screening was also conducted. Results indicated satistically, 1µl 1N Na_2CO_3, 10 µl 1N $NaHCO_3$, and 20µl 1N NaOH at pH = 10 were equally ($P \leq 0.05$) effective at determining live SCN J2s. The 10 µl 1N $NaHCO_3$ and 20µl 1N NaOH solutions increased movement of the nematodes with the normal lateral undulations. The 1µl 1N Na_2CO_3 at pH = 10 caused SCN J2s to display a rapid twisting movement after a 1 min exposure that was easy to visually see. Death also occurred after 30-min exposure in 20µl 1N Na_2CO_3, although 20µl 1N $NaHCO_3$ did not terminate the nematodes. Furthermore, live J2s were confirmed alive and infective in a growth chamber test. The 1µl 1N Na_2CO_3 in100 µl of nematode solution at pH = 10 determined that 80% of the SCN J2s were alive and of those, 46.3% entered soybean roots when they were placed near the root zone. The results confirmed that the optimum sodium agitation technique for rapidly distinguishing live and dead SCN J2s, accurately indicated live viable J2s.

SCREENING MICROORGANISMS TO DISCOVER AND DEVELOP BIOLOGICAL PRODUCTS FOR NEMATODE CONTROL. **Xing, Lijuan, S. Fudali-Alves, K. Song, A. L. Cordova-Kreylos, R. Asolkar, A. Stewart, P. Himmel, and P. Marrone.** Marrone Bio Innovations, Davis, CA 95618.

Marrone Bio Innovations (MBI) screens microbes for pesticidal activity with potential for commercialization. At MBI, a multidisciplinary team of scientists collaborate to discover and develop biological products for nematode control. The ultimate goal is to develop efficacious and environmentally friendly microbial nematicides. Our nematologists collaborate with microbiologists, chemists, and plant scientists to evaluate and select the microbe candidates that produce nematicidal compounds based on direct nematicidal as well as plant based bioassays. Biochemists and fermentation specialists optimize the media in which these microbes are grown to best produce active nematicidal compounds. Physical and chemical properties for commercial scale production are characterized while at the same time, optimization parameters for economical manufacturing are identified. MBI natural product chemists evaluate crude extracts in fermentations of verified microbial candidates to identify pesticidal active fractions in bioassay driven experiments. Ultimately compounds associated with efficacy are identified. The identified compounds become the analytic tool for efficacy measurements and can track the metabolites produced by these microbes in fermentation yield optimization studies and in manufacturing to maintain efficacy without running a bioassay. Finally EPA guidelines are followed by formulation experts to develop stabilized prototypes that are evaluated for efficacy, shelf life, and other parameters that make a product desirable to a customer (e.g., rainfastness, dispersibility). Two microbes with nematicidal activity are now in precommercial development. The developmental process and the candidates of potential nematicidal products will be discussed.

MASS PROPAGATION OF *PRATYLENCHUS NEGLECTUS* AND *P. THORNEI* IN CARROT DISK CULTURES. **Yan, Guiping, and R.W. Smiley.** Oregon State University, Columbia Basin Agricultural Research Center, P.O. Box 370, Pendleton, OR 97801.

Root-lesion nematodes, *Pratylenchus neglectus* and *P. thornei*, are the most important nematodes that restrict productivity of wheat in low-precipitation regions of the Pacific Northwest (PNW). It was estimated that these nematodes reduce farm profits by about $51 million annually in the three PNW states of Idaho, Oregon, and Washington. Management of root-lesion nematodes in dryland wheat fields is best approached by integrating crop rotation and using resistant and tolerant cultivars. However, many crop species or wheat cultivars differ in resistance or tolerance to these *Pratylenchus* species. Resistance

evaluations are therefore essential for selecting the best-performing crops and cultivars for optimal disease management. Large-scale resistance screenings require production of large amounts of nematode inoculum. Carrot disk cultures have been used in our laboratory to culture these two species, both of which are parthenogenic. The highest reproduction rate of *P. neglectus* (15,036 live juveniles and females) was obtained from disks inoculated with five gravid females (Lind, WA) after 8 mon of incubation at 22°C. The highest reproduction rate of *P. thornei* (77,400 live juveniles and females) was obtained from disks inoculated with a single female (Adams, OR) after 6 mon. The ratio of adult females (females/total females plus juveniles) in single nematode progeny populations was 18% and 34% for *P. thornei* (n = 7) and *P. neglectus* (n = 4), respectively. Inoculation with juveniles increased the number of nematodes on disks, but mature females with visible eggs or vulva were more effective. The reproduction rate of *P. thornei* juveniles (n = 4) and adult females (n = 4) from Heppner, OR, were 721 and 5,665 live juveniles and females, respectively, after 5 mon incubation. To increase reproduction potential within a short time period, a thin slice from multiplied carrot culture was transferred to a fresh, surface-sterilized carrot disk in a Petri dish. High numbers of nematodes were harvested from subcultures in 3 mon (38,520 *P. thornei* per disk, n = 5). Nematodes were extracted from carrot by cutting the disks into thin slices and floating the slices in distilled water for up to 60 hr, in which up to an average of 45,555 *P. thornei* live juveniles and females were recovered from each carrot disk (n = 6). Nearly half of the nematodes emerged within the first two hours of extraction. Cultures of *P. neglectus* commonly remained viable for up to 12 mon and super-coiled anhydrobiotic nematodes with dark intestines were observed over the long term storage. The coiled nematodes in the dehydrated and dormant state survived longer in slowly dried carrot disks. Our results confirmed that the carrot culture method is a practical approach to produce large numbers of *P. neglectus* and *P. thornei*. With this method, we produced millions of nematodes for use as inoculum in resistance screening trials.

HATCHING OF *HETERODERA FILIPJEVI* AND *H. AVENAE* POPULATIONS IN OREGON. **Yan, Guiping, R.W. Smiley, and J.A. Gourlie.** Oregon State University, Columbia Basin Agricultural Research Center, P.O. Box 370, Pendleton, OR 97801.

Cereal cyst nematodes, *Heterodera avenae* and *H. filipjevi*, are economically important cyst nematodes that restrict production of cereal crops in the Pacific Northwest (PNW) and elsewhere in the world. In the United States, *H. avenae* and *H. filipjevi* were each first detected in Oregon, in 1974 and 2008, respectively. High populations of *H. avenae* reduced grain yields of susceptible wheat cultivars as much as 50%. It was estimated that this nematode reduces the profit from wheat production by at least $3.4 million annually in the three PNW states: Idaho, Oregon, and Washington. The greatest risk of *H. avenae* to cereal crops occurs during mid- to late spring as infective second-stage juveniles emerge from cysts and penetrate roots of susceptible plants. The timing of juvenile emergence for *H. filipjevi* populations in Oregon was unknown. An experiment was conducted in an outdoor nursery at Pendleton, OR, from January through June 2009 to determine the timing for emergence of juveniles from *H. filipjevi* cysts. The hatching test using cysts in small nylon mesh bags kept in soil incubated at 25-cm depth showed juvenile emergence starting in the early spring (April 7) when the soil temperature was 11°C, and the hatching rate averaged from five replications was 52% (juveniles emerged/total juveniles emerged plus eggs and juveniles in cysts). Maximum emergence occurred on May 5 (12°C) with the hatching rate of 86%. Thereafter, juvenile emergence decreased and reached a minimum of 10% on 16 June (18°C). Soil temperatures correlated with the corresponding hatching rate ($r = 0.596$, $P = 0.041$). In order to obtain enough second-stage juveniles of *H. avenae* as inoculum for resistance evaluation, an experiment was conducted with two populations from Island City, OR, in our laboratory under controlled conditions (10 °C for the first 7 d and 15 °C for the remaining 33 days, at 2- to 3-d intervals) to determine the hatching ability of *H. avenae* eggs. Maximum emergence for both populations (population one with 80 cysts and population two with 94 cysts collected from different cultivars) occurred after 19 d incubation, with about 21% of total juveniles emerged from the cysts. More than 70% of juveniles were collected during 15 d incubation from day 13 to day 28 (85% for population one and 74% for population two). However, the total hatching rates for populations one and two were only 3.1% and 3.2%, respectively. The low hatch rates indicated that collection of juveniles for use as inoculum would be very difficult because of an inability to induce adequate levels of hatching under controlled conditions. Large-scale resistance screenings for *H. avenae* in our greenhouse are therefore performed using naturally infested soil as inoculum.

KEY FACTORS ASSOCIATED WITH MOLECULAR QUANTIFICATION OF *PRATYLENCHUS NEGLECTUS* FROM SOIL. **Yan, Guiping[1], R.W. Smiley[1], P.A. Okubara[2], and A.M. Skantar[3].** [1]Oregon State University, Columbia Basin Agricultural Research Center, Pendleton, OR 97801; [2]USDA-ARS, Root Disease and Biological Control Research Unit, Pullman, WA 99164; [3]USDA-ARS, Nematology Laboratory, Beltsville, MD 20705.

Pratylenchus neglectus is one of the most important root-lesion nematodes that invades plant roots and restricts productivity of wheat in the Pacific Northwest. A quantitative real-time polymerase chain reaction (qPCR) assay was developed to detect and quantify *P. neglectus* directly from DNA extracts of soil. Discrepancies were found between manual counts of *P. neglectus* and nematode numbers quantified using qPCR, which could be because of PCR inhibitors, soil type, soil moisture content, sampling errors, and life stages of *P. neglectus*. To test for inhibitors in soil extracts, plasmid pUC19 (3×10^6 copies) were added to qPCR reactions for water control and DNA extracts of 15 field soils collected from eight locations in Oregon and Washington, including sandy loam, loam, and silt loam. Cycle threshold (Ct) values were either much higher than that

observed in the water control or no plasmid-specific amplification was produced, indicating that PCR inhibitors existed in these soil samples. To reduce the effect of soil inhibitors, we diluted the extracts or added bovine serum albumin (BSA) to the *P. neglectus* qPCR reactions. Dilution was not effective, but BSA (0.4 μg/μL) improved the correlation between the qPCR and counting methods ($R^2 = 0.68$), compared with $R^2 = 0.53$ for the original qPCR data. The qPCR assay was applied to 22 soil samples at natural field moisture content (volumetric soil moisture of 8% to 16%) from a field experiment at Moro, OR. A significant positive correlation ($R^2 = 0.72$) was observed using these soils. Six samples were oven-dried and tested with qPCR. A strong correlation ($R^2 = 0.95$) was obtained between the nematode numbers determined by qPCR and the counting method. The numbers from the counting method were closer to the numbers determined by qPCR using the dry soils than using the moist soils. We evaluated the reproducibility of qPCR in triplicate extracts from six soils. The standard deviation in Ct values among the triplicate extracts was 0.53 to 2.24. These data expressed as nematode counts gave standard deviations ranging from 61 to 2,144. No significant difference in Ct was found among a single adult female, second-stage juvenile, and egg (containing faint outline of developing juvenile), indicating that different life stages of *P. neglectus* contain similar quantities of DNA and proportions of juveniles, adult females and eggs in individual samples are unlikely to affect qPCR quantification. Our data indicate that reproducibility and reliability of qPCR was improved by using BSA and dried soils. Multiple DNA extractions from each soil sample will also reduce variation caused by the small sample size (0.5 g soil) and the heterogenous distribution of nematodes in soil, to further improve the accuracy of the assay.

QUANTITATIVE FIELD TESTING *HETERODERA GLYCINES* FROM SOIL SAMPLES. **Yan, Li[1], G.W. Lawrence[1], V.P. Klink[2], S. Lu[1], and C.J. Balbalian[1].** [1]Department of Biochemistry, Molecular Biology, Entomology and Plant Pathology, Mississippi State University, MS 39762; [2]Department of Biological Sciences, Mississippi State University, MS 39762.

Heterodera glycines (Ichinohe) (soybean cyst nematode [SCN]) is a major pathogen of soybeans, causing more economic damage than any other soybean pathogen. A major problem in managing SCN is that it requires great skill and time to identify and count SCN life stages under the microscope. An objective of this study was to develop quantitative PCR-based *Heterodera glycines* diagnostic techniques. A series of SCN genes were screened for metagenomic analyses of SCN populations found in soil. Genomic DNA was isolated from known quantities (1, 2, 4, 6, 8, 10, 100, and 1,000) of J2 SCN that were used in the PCR analyses. PCR experiments determined that a SCN homolog of the *Caenorhabditis elegans uncoordinated 78* (*unc-78*), denoted Hg-*unc-78*, showed the most reliable amplification. To determine gene specificity, we then isolated genomic DNA from *Rotylenchulus reniformis* and *Meloidogyne incognita*, as well as metagenomic DNA from soil that was not infested with SCN. PCR using the Hg-*unc-78* primers was analyzed on genomic DNA isolated from these samples, showing no amplification. A pair of quantitative PCR (qPCR) primers targeting the gene Hg-*unc-78* were developed, and successfully used for quantitative estimates of the nematode population under laboratory conditions. Currently the qPCR primers and procedure are being evaluated for field testing assays and race-specific molecular marker studies.

MANAGEMENT OF SUGAR BEET CYST NEMATODE WITH TELONE® II USING STRIP-TILLAGE FUMIGATION. **Yoshida, Harvey[1], S. Hafez[2], B. Young[3], and R. Portenier.** [1]Dow AgroSciences, 432 Aimee Drive, Richland, WA 99352, [2]University of Idaho, Parma Research and Extension Center, 29603 University of Idaho at Lane, Parma, ID 83660, [3]Dow AgroSciences, 903 East 1400 North, Shelley, ID 83274.

The sugar beet cyst nematode, *Heterodera schachtii*, is a major pest of sugar beets in Idaho and eastern Oregon. Low densities of *H. schachtii* can cause significant yield reduction and economic losses. A treatment threshold as low as five viable cysts per 500 cc of soil (plus a period of 3 yr or less between sugar beet crops) is recommended for susceptible varieties. Preplant broadcast fumigation has been an effective management option for *H. schachtii*; however, in-row applications may be a more economically attractive option for sugar beet producers. Field studies were conducted using nematode susceptible variety ('Hillshog' 9036RR) at the University of Idaho Research and Extension Center in Parma, ID. The performance of in-row applications of Telone® II (1,3-dichloropropene) for control of *H. schachtii* was evaluated in 2010. Telone® II was applied in-row using a modified 4-row Orthman 1tRIPr strip-tillage system. In 2010, when compared with the untreated, Telone® II applied at 8, 12, and 16 gal/acre resulted in yield increases of 61%, 93%, and 120%, respectively. In contrast, an at-plant treatment of Temik (aldicarb) at 27 lb/acre resulted in a 14% increase in yield over the untreated. The effectiveness of in-row versus broadcast fumigation was evaluated in a 2011 study using the same sugar beet variety and application equipment as in the previous year. With the exception of Telone II applied in-row at 8 gal per acre, all of the Telone treatments (in-row and broadcast at 12 and 16 gallons/acre) resulted in > 40% increase in yield when compared with the untreated. Percentage yield increase for the 8 gal/acre rate of Telone II and Temik at 27 lb/acre was 3.5 and 14, respectively.

TAXONOMIC STUDIES OF *DELADENUS* (TYLENCHIDA: NEOTYLENCHIDAE). **Yu Qing[1], and J. Guo[2].** [1] Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, K1A0C6 ON, Canada; [2] Technical Center, Ningbo Entry-exit Inspection and Quarantine Bureau, Ningbo 315012, Zhejiang, China.

Deladenus Thorne (1941) is remarkable in that the genus consists of species that can have a free-living mycetophagous life cycle, feeding on specific fungal species, or an infective entomophagous cycle that feeds inside wood-boring insects. These

two life cycles produce very different forms—the mycetophagous life cycle results in the Neotylenchidae or more commonly called mycetaphagous form, whereas the infective life cycle results in the Sphaerulariina or infective form. A selected strain of the entomophilic *D. sircidicola* has been used in several countries with great success as a biological control agent for control of the European wood wasp, *Sirex noctilio*, on pines. Understanding the native biodiversity of species of *Deladenus* before introducing a foreign biocontrol agent is critical. In this presentation, three new *Deladenus* species are discussed. One of the new species was found in a wheat field in Ontario, Canada. Only mycetophagous forms were recovered. Although morphologically similar to *D. durus*, the new species differs in having a much more anterior esophagus-intestine junction, at the median bulb. Two other new species were recovered from wood packaging originating from Korea and intercepted in China. Both mycetophagous and infective forms of these new species were found. Both differ from other species of *Deladenus* by having distinct esophagous-intestine valves in mycetophagous females and a degenerated esophagus in mycetophagous males, but one has a much longer body than the other. Additional morphological characters of taxonomic importance are also discussed.

THE WHO AND WHERE OF PLANT-PARASITIC NEMATODES IN SEMI-ARID WASHINGTON WINE GRAPES. **Zasada, Inga[1], A. Howland[2], A. Peetz[1], and R.P. Schreiner[1].** [1]USDA-ARS Horticultural Crops Research Laboratory, 3420 NW Orchard Ave., Corvallis, OR 97330; [2]Department of Horticulture, Oregon State University, Corvallis, OR 97331.

Washington state is the second-largest producer of wine grapes in the United States. Until recently, little was known about the occurrence, identity, distribution, and biology of plant-parasitic nematodes in this grape production system. Research was conducted to fill this knowledge gap. Samples collected from 157 vineyards in four Washington AVAs revealed that plant-parasitic nematodes commonly encountered in vineyards worldwide were also present in Washington, including: *Meloidogyne hapla*, *Xiphinema americanum*, and *Mesocriconema xenoplax*. Both *M. hapla* and *X. americanum* were detected in > 50% of sampled vineyards, whereas *M. xenoplax* was less common being found in only 14% of vineyards. The spatial distribution of plant-parasitic nematodes within the soil profile was also studied at two vineyards: Chardonnay grown on a sandy loam soil and White Riesling grown on silt loam soil. Nematode population densities were determined both horizontally in a 1.5- × 2.1-m area and vertically down to 1 m. *M. hapla*, *Pratylenchus* spp., *Xiphinema* spp., and *Paratylenchus* spp. were found at both vineyards whereas *M. xenoplax* was only detected in the Chardonnay vineyard. At both vineyards, population densities of *M. hapla* were positively related to soil moisture and fine root biomass ($p < 0.0001$). The same trend was observed for *M. xenoplax* at the Chardonnay vineyard ($p < 0.0003$). The horizontal distribution of nematodes varied among the nematode genera with *M. hapla* and *M. xenoplax* concentrated in the vine row near drip-irrigation emitters whereas *Pratylenchus* were aggregated at the edge of the vine row. At both vineyards, *M. hapla* and *Pratylenchus* population densities were concentrated in the upper 45 cm of the soil profile; *M. xenoplax* followed the same trend in the Chardonnay vineyard. Conversely, *Xiphinema* was distributed throughout the soil profile at both vineyards. A higher density of *M. hapla* juveniles near drip-irrigation emitters was associated with increased galling of grapevine fine roots and colonization of symptomatic roots by arbuscular mycorrhizal fungi was less than that of nonsymptomatic fine roots ($p < 0.001$). Molecular tools were employed to explore the species diversity of *Pratylenchus* spp. in semi-arid Washington vineyards and the phylogenetic relationship of *X. americanum* populations from Washington to populations from other regions of the United States.