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ABSTRACTS

1. PROTECTION OF CITRUS FRUITS AGAINST THE MEDFLY USING ENTOMOPATHOGENIC NEMATODES AND FUNGI. **Abd-Elgawad[1], Mahfouz M. M., A. S. Abdel-Razek[2], and A. E. Abd El-Wahab[3]**. [1]Plant Pathology Dept., [2]Pests and Plant Protection Dept., National Research Centre, Dokki 12622, Giza, Egypt, [3]Dept. of Agricultural Zoology and Nematology, Faculty of Agriculture, Al-Azhar University, Cairo, Egypt.

Two entomopathogenic nematode species *Steinernema riobrave* and *Heterorhabditis bacteriophora* HP88 originally obtained from USA but locally cultured on the Mediterranean fruit fly (the Medfly), *Ceratitis capit*ata, in addition to four Egyptian isolates from each of the fungi *Metarhizium anisopliae* and *Beauveria bassiana* were tested to control the Medfly. Numbers of the Medfly adults emerged from soil treated with the nematode *S. riobrave* or *H. bacteriophora* HP88 in the presence of *C. capitata* prepupae were less ($P \leq 0.05$) than those of the untreated control in natural and sterilized soil. The isolates *B. bassiana* NRC-AB8 and *M. anisopliae* NRC-AM5 could reduce Mediterranean fruit fly emergence from soil by up to 80%. *M. anisopliae* isolate NRC-AM5 at a higher concentration of 1×10^8 was the most effective isolate ($P \leq 0.05$) in reducing *C. capitata* adult emergence when applied to the soil. However, contrary to the applied fungi, the numbers of emerged insect adults from the natural and sterilized soils was not different ($P \leq 0.05$) in soils treated with both nematode species but significantly less than the untreated soils. This is possibly due to the natural enemies found in non-sterilized soils treated with the fungi.

2. MOLECULAR GENOMIC APPROACHES TO NEMATODE-ENVIRONMENT INTERACTION STUDIES. **Adhikari, Bishwo N.[1], D.H. Wall[2], and B.J. Adams[1]**. [1]Department of Biology and Evolutionary Ecology Laboratories, Brigham Young University, Provo, UT 84604. [2]Department of Biology and Natural Resource Ecology Laboratory, Colorado State University, Fort Collins, CO 80523.

The natural environments of organisms present a multitude of biotic and abiotic challenges that require both short-term ecological and long-term evolutionary responses. As one of the most important members of belowground communities and an important component of the terrestrial ecosystem, nematodes are responsive to changing environmental conditions at the ecosystem, community, organismal and genetic levels. Recent advances in genome analysis provide ways to examine the genomic response of nematodes to changes in their environment, and the ways in which the genetic changes affect nematode survival and contribution to ecosystem functioning. Here, we examine the application of genomic approaches to study nematode-environment interactions and describe how these approaches can be used to understand the response of nematodes to environmental perturbations. We will discuss different comparative (i.e. expressed sequenced tag sequencing, subtractive hybridization, quantitative real-time polymerase chain reaction) and functional (i.e. microarray analysis, RNA interference) genomics approaches and show how specific ecological questions can be answered by using such tools. Results from our studies on genomic responses of nematodes to desiccation and freezing stress, transcriptional profiling of nematode trait deterioration and the stoichiometric interaction of soil elemental composition to the nematode genome will be discussed. We also address some of the challenges of using different genomic tools, and how combinations of different research approaches at various functional (i.e. DNA, mRNA) and biological (i.e. individual, population, species) levels can provide a better understanding of the interplay between genes and the environment. Finally, we advocate that an understanding of the genetic mechanisms underlying ecological interactions of nematodes could be used to test predictions of ecological and evolutionary processes across broader taxonomic groups and spatial scales.

3. TEACHING AND LEARNING PLANT-PARASITIC NEMATODE IDENTIFICATION: THE CLEMSON EXPERIENCE. **Agudelo, Paula[1], D.C. Harshman[1] and S.A. Lewis[1]** Department of Entomology, Soils, and Plant Sciences. 114 Long Hall, Clemson University, Clemson, SC 29634.

Every year, since 1981, Clemson University hosts a one-week course on Plant-parasitic Nematode Identification. Course participants typically include research and extension scientists, professional consultants, regulatory personnel, diagnosticians, and graduate students with an interest in plant nematology. The course is structured as short lectures on the biology and ecology of the nematode groups, immediately followed by direct observation of specimens. The emphasis of all the teaching materials is on identification by morphology. The ''recognition'' approach is favored, where all relevant features of a specimen are used simultaneously to identify the genus, as opposed to the sequential approach that taxonomic keys dictate. Participants prepare and observe their own mounts of fresh specimens of the 25 most common plant-parasitic nematode genera in agricultural soils. Each participant is provided with a compound microscope and stereomicroscope, a reference

book, a laboratory workbook, and the necessary tools. The laboratory workbook contains drawings and photographs of diagnostic characters and provides space for students to make their own drawings. The course is designed so that 75% of the time is dedicated to the direct observation of specimens and to answering questions individually at each microscope station. After the course, participants are expected to have the knowledge and skills necessary to identify the most common genera of plant-parasitic nematodes. Two practical identification exams (one in the middle of the course and one at the end of the course) directly assess the completion of this objective. A brief history of the course is presented along with a discussion of the evolution of the teaching techniques.

4. INCIDENCE AND POPULATION DENSITY OF PLANT-PARASITIC NEMATODES INFECTING VEGETABLE CROPS AND ASSOCIATED YIELD LOSSES. **Anwar, Safdar A.[1], M.V. McKenry[2], and N. Javid[1]**. [1]Department of Plant Pathology, University of Agriculture, Faisalabad, Pakistan; [2]Department of Nematology, University of California, Riverside, CA. 92521 USA.

Plant parasitic nematode population densities associated with vegetable crops and the resulting losses were determined during 2007 and 2009. The presence of nematodes in 325 root and soil samples was determined at the harvest of each crop. Nineteen vegetable crops were sampled to collect root and soil from major vegetable production areas over a two-year period. Ten root and soil cores were randomly collected for each crop by walking a zigzag pattern in each field with an Oakfield tube of 2.5-cm diameter to a depth of 18-20 cm. Nematode density in composite root and soil samples was assessed using a Modified Sieving-Baermann Funnel technique. The crops evaluated included: carrot (*Daucus carota*); chilies (*Capsicum annuum*); coriander (*Coriandrum sativum*); crucifers [cabbage (*Brassica oleracea*) and mustard (*Sinapis alba*)]; cucurbits [bitter gourd *(Momordica charantia*), cucumber (*Cucumis sativus*), pumpkin (*Cucurbita argyrosperma*), sponge gourd (*Luffa cylindrica*), melon (*Cucumis melo*)and watermelon (*Citrullus lanatus*)]; eggplant (*Solanum melongena*); lettuce (*Lactuca sativa);,*okra (*Abelmoschus esculentus*); pea (*Pisum sativum*); potato (*Solanum tuberosum*), spinach (*Spinacea oleracea*.), and tomato (*Lycopersicon esculentum*). The most important plant-parasitic nematodes detected, in order of decreasing frequency of infestation (percentage of samples), were *Meloidogyne incognita* (90%), *Pratylenchus penetrans* (30%), T*ylenchorhynchus clarus* (29%*)*, Hoplolaimus columbus (15%), *Paratrichodorus* minor (7.5%), *Xiphinema americanum* (7.1%), M. *javanica* (7%), Belonolaimus longicaudatus (5.6 %), *Longidorus africanus* (5%), and *Helicotylenchus dihystera* (3.2%). Population densities of *Pratylenchus penetrans* and *Meloidogyne incognita* were at potentially damaging levels in most of the vegetable crops surveyed. The criteria used to assess yield losses due to nematodes included grower interviews, visual assessments of foliage growth (necrotic, chlorotic, stunted, and wilted plants), root symptoms and expert opinions. The interview of growers included the condition of crop, quantitative and qualitative yield losses tied to market value and life span of crop. We observed ca 22.4% yield losses from 19 commercially grown vegetable crops. Damage ranged from 2% for cabbage to 45% for squash, which was 35%, 80%, and 46% higher than that from developed countries, USA, and India, respectively. The main reason for more losses in Pakistan appears related to unawareness of growers about the presence of nematodes and damage they cause. Another reason might be non-availability of resistant crop cultivars and nematicides. This study on plant parasitic nematodes and related crop losses will provide important information for extension staff as they create greater awareness about these hidden crop enemies as well as improved management practices.

5. A MAGNETIC BEAD HYBRIDIZATION CAPTURE TECHNIQUE FOR THE DETECTION AND QUANTIFICATION OF *PASTEURIA NISHIZAWAE* IN SOIL. **Atibalentja[1], Ndeme, M. Babadoost[1], and G. R. Noel[2].** [1]Department of Crop Sciences, University of Illinois at Urbana−Champaign, Urbana, IL 61801, [2]USDA ARS, Urbana, IL 61801.

The need for a practical and reliable assay for the detection and quantification of *Pasteuria* spp. in soil has become more pressing with the advent of breakthrough technologies of mass cultivation that would allow for large scale exploitation of *Pasteuria* spp. for the biological control of plant-parasitic nematodes. Although DNA-based detection techniques such as RT-PCR have proven to be very sensitive and specific, their application to *Pasteuria* spp. is hindered by the difficulty of extracting sufficient amounts of *Pasteuria* DNA that is free from PCR inhibitors and contaminating DNA from various sources in the soil. While PCR inhibitors can be effectively removed by using commercial kits for soil DNA isolation, molecules of *Pasteuria* DNA in the extracts often are in such low concentrations that they are overwhelmed in a heterogeneous population of contaminating DNA, leading to failed PCR reactions. The technique reported herein uses a magnetic bead−streptavidin−linked biotinylated specific probe to capture and purify *Pasteuria* DNA from complex soil extracts. The resulting clean DNA is used as a template in RT−PCR reactions. This assay would be especially useful for monitoring the population dynamics of *Pasteuria* spp. in soil and the long-term effects of field applications of *Pasteuria* treatments on the environment.

6. DO AMBUSHER AND CRUISER ENTOMOPATHOGENIC NEMATODES DISPERSE DIFFERENTLY IN SOIL IN THE ABSENCE OF HOSTS? **Bal, Harit K., R. A. J. Taylor, and P. S. Grewal.** Department of Entomology, OARDC, The Ohio State University, Wooster, OH 44691.

A series of elegant laboratory studies show dichotomy in the foraging behavior of entomopathogenic nematodes (EPNs). However, how nematodes with ambusher and cruiser foraging strategies disperse in the field is unknown. Therefore, we

compared the rate of lateral dispersal of a cruise foraging, *Heterorhabditis bacteriophora* and an ambusher, *Steinernema carpocapsae* from infected host cadavers in autoclaved soil (24% w/w) placed in wooden arenas at room temperature (21°C). Soil core samples (2 cm dia and 5 cm deep) were collected in plastic cups at different time intervals (6 to 240 hours) and distances (7 to 61 cm) from a 10-day old cadaver of final instar *Galleria mellonella* infected with the respective nematode species. Nematode movement was estimated using *G. mellonella* bait placed in the collected soil samples. The spatio-temporal data were analyzed by a two-dimensional modified Fick Diffusion Model with least squares method. While, both the species exhibit nearly the same average movement of infective juveniles in soil (6 cm/day), they varied in the pattern of dispersal, whereby *H bacteriophora* vacated the center of the arena and *S. carpocapsae* did not. Number of IJs moving a given distance declined with increasing distance from the cadaver. This study revealed remarkable innate ability of EPNs to move in soil in the absence of a host and also showed how dispersal in the absence of the host by ambusher, *S. carpocapsae* is unexpectedly similar to the cruising nematode, *H. bacteriophora*. While nearly all *H. bacteriophora* IJs moved some distance, most *S. carpocapsae* IJs remained stationary, but with a small proportion moving long distances, even farther than *H. bacteriophora*, which could be considered as emigrants. Little is known about the dispersal pattern of these two species of EPNs in the field. Further investigation of the dispersal pattern of EPNs in the presence of grass will be carried out mimicking the natural field conditions. Knowledge on dispersal capability of EPNs in soil is one of the key aspects to enhancing their use as effective biological control agents of soil borne insect pests.

7. MANURE AND CHEMICAL FERTILIZER EFFECT ON SOYBEAN CYST NEMATODE, NEMATODE COMMUNITY, AND SOYBEAN YIELD IN SCN-SUPPRESSIVE AND CONDUCIVE SOILS. **Bao, Yong, J. Vetsch, S. Chen, and G. Randall.** University of Minnesota Southern Research and Outreach Center, 35838 120th Street, Waseca, MN 56093.

Soybean cyst nematode (SCN), *Heterodera glycines*, is the major yield-limiting pathogen of soybean in North Central USA. Field experiments were conducted in 2009 to determine the effects of liquid swine manure and chemical fertilizer P and K on SCN, nematode community, and soybean yield in the SCN-suppressive and conducive soils. The experiment was a split-plot design with SCN-resistant and susceptible soybean cultivars as main plots and three fertilizer treatments as subplots. The fertilizer treatments were liquid swine manure at 3.74 m^3/ha, P at 112 kg P_2O_5 + K at 112 kg K_2O/ha, and no fertilizer. The manure was injected into the soil 10 cm under each intended row with 30-cm wide sweep injector two weeks before planting soybean. The PK fertilizer was broadcast on the soil surface and then incorporated into soil. Composite soil samples were collected from each 4-row plot at four different times: prior to applying fertilizers, 45 days after treatment, midseason, and harvest to determine population densities of SCN eggs and nematode communities. Soybean yields were measured by harvesting the center two rows of each plot. There was no significant effect of fertilizer treatment on the SCN egg population density, but liquid swine manure treatment resulted in lower SCN second-stage juvenile population density in the SCN-suppressive soil at 45 days after planting, but higher in the SCN-conducive soil at harvest. The application of manure reduced total abundance of plant-parasitic nematodes in SCN-suppressive soil, and increased abundance of bacteria-feeding nematodes in both suppressive and conducive soils. While both manure and PK treatments increased soybean yield, the manure produced highest yield and the fertilizer treatment effect was greater for SCN-susceptible soybean than resistant soybean in the SCN-conducive soil. There was no difference in soybean yield in the SCN-suppressive soil. This study suggests that soil fertility management, especially application of manure, is a useful strategy to alleviate the SCN damage.

8. ABATING ROOT-KNOT NEMATODE DAMAGE IN FRESH CARROT PRODUCTION. **Becker, J. Ole[1], J. Nunez[2], and A. Ploeg[1].** [1]Department of Nematology, University of California, Riverside, CA 92521, [2]University of California Cooperative Extension Kern County, Bakersfield, CA 93307.

Root-knot nematodes (rkn) are by far the most important pathogens in California's fresh carrot production fields because of their wide-spread distribution and the crop's low economic damage threshold. Particularly during the first few weeks after seed germination, rkn injury to the carrot root tip may cause stubbing, forking, and galling. While soil fumigants typically provide excellent efficacy against rkn, their availability in California is likely to further diminish because of regulatory restrictions to mitigate potential air quality and non-target exposure problems. As a potential alternative to soil fumigants, we envisioned a stepwise, multi-strategy approach by reducing the pre-season rkn population with a cover crop and/or biofumigation. Another key component, a biorational, nematicidal seed coating is used to protect young carrot seedlings against early rkn attack. In a *M. javanica*-infested field at the UC Kearney Research and Extension Center, a proof-of concept trial was conducted with a split-plot design. The main treatments were 5 cruciferous cover crops, mustard meal, the soil fumigant dazomet, and a fallow check. The cover crops were grown for about 3 months, cut with a high-speed flail mower and rototilled into the top 15-20 cm soil. Mustard meal (4,483 kg/ha) and dazomet (362.5 kg/ha) were similarly incorporated into the soil. The field was immediately irrigated with overhead sprinklers to field capacity. Two weeks later, carrots (cv PrimeCut 59), coated with a fungicide combination (Dynasty CST) with or without 0.016 mg abamectin/seed were seeded into subplots of the main treatment plots. The initially low rkn populations increased under all cover crops, but

the 2-wk biofumigation then reduced populations by about 50%. In contrast, soil amendment with mustard meal reduced the rkn population density close to detection level. This abated crop damage and resulted in increased yield similar to the dazomet treatment. For all main treatments, carrots derived from abamectin-coated seed were more vigorous, had much less nematode damage and higher yields than those without the nematicidal seed coating. In summary, fresh cruciferous plant material may not provide sufficient biofumigation efficacy to overcome rkn population increase during the crop's growth. In contrast, the very efficacious mustard meal warrants further investigations concerning reduced application rates. Abamectin seed coating improved carrot growth across all pre-season treatments. Soil amendment with mustard meal in combination with abamectin-coated carrot seed appears to be a sound strategy to mitigate rkn damage in carrot production.

9. A SCREENING AND OPTIMIZATION TEST FOR NEMATICIDAL SEED COATINGS. **Becker, J. Ole, and J. Smith Becker.** Department of Nematology, University of California, Riverside, CA 92521.

Potential benefits of nematicidal seed treatments for the establishment of susceptible seedlings in nematode-infested soil were first observed three decades ago, but the technology never gained much attention. This has changed with the successful development and market introduction of Avicta (a.i. abamectin) as a seed coating for seedling protection against early plant-parasitic nematode root attack and consequent damage. An important part in the development of a seed coating product is not only the selection of an efficacious nematicidal compound and its appropriate formulation. Modern commercial seed coatings contain several other ingredients such as insecticides, fungicides and support substances. The combination of those compounds will determine the seed coating's physical and chemical properties such as speed of water uptake, release of active ingredients, seeding flow, and appearance. The performance of such a mixture is difficult to predict and time-consuming to optimize. We developed a so-called QuickScan seedling test that is based on inhibition of root growth by second-stage root-knot nematode juveniles (rkn J2) in sensitive plant species. Tap root and feeder roots of young seedlings are slowed down in growth by invading rkn J2. This is quantifiable before galling becomes apparent. An effective nematicidal seed coating should prevent or mitigate the J2 attack. Thus, its efficacy is reflected in the difference of total root length compared to the non-treated, rkn-infested check as well as to the non-infested control or to a soil treatment with a nematicidal industry standard. Although many plants are suitable for QuickScan, we preferred cucumber as indicator because of their uniform germination, fast growth, sensitive reaction to rkn attack and large seed loading capacity. Seven days after seeding into *M. incognita*-infested sandy soil and incubation at 27 C, roots were washed free of soil. Single root systems were floated in a transparent tray with a shallow water layer, scanned with a flatbed scanner and analyzed with MacRhizo software. The standard QuickScan can be easily modified to evaluate the performance of seed coatings under various conditions such as soil type, pH, and temperature.

10. RECONNAISSANCE OF SOIL-DWELLING NEMATODES OF GREAT SMOKY MOUNTAINS NATIONAL PARK. **Bernard, E.C., M.M. Dee, and P.J. Long**. Dept. of Entomology and Plant Pathology, The University of Tennessee, 2431 Joe Johnson Drive, 205 Plant Sciences, Knoxville, TN 37996-4560.

Nematodes of natural areas have received little attention compared to nematodes of cultivated lands, but they are essential for understanding nematode biogeography. Without a clear recognition of indigenous North American nematodes, issues of nematode distribution and dispersal on a world basis cannot be clarified. However, landscape alteration of much of the continent, coupled with apparent introduction of many exotic nematodes, makes this task difficult. In the eastern U.S., little remains of the great pre-Columbian temperate deciduous and mixed forests. Great Smoky Mountains National Park (GRSM) contains the largest remnants (stands high in virgin attributes) of these forests, primarily at higher elevations where logging was unprofitable. Beginning in the early 1980s, nematodes have been collected intermittently from 24 sites among both these remnant areas and more accessible second-growth (>80 years old) forest. Criconematina is the most diverse taxon, with at least 11 criconematid, 3 paratylenchid, 1 hemicycliophorid, and 1 sphaeronematid species. Females of one Gracilacus sp. become transformed into swollen, persistent, cyst-like sacs, but without internal eggs. Sedentary parasites are represented by 1 Cactodera, 1 Meloidodera, and 2 Meloidogyne spp., all undescribed and all but one root-knot species collected from the same location (Cosby Creek). The root-knot nematode hosts were eastern hemlock, tulip-tree, and hop-hornbeam. Egg masses of the tulip-tree-hop-hornbeam root-knot nematode remain embedded in root tissue. Hoplolaimoid nematodes have been collected only sporadically, but an undescribed genus similar to the western genus Nagelus occurs on Snakeden Ridge. Among Adenophorea, 3 Xiphinema spp. (X. bernardi, X. chambersi, X. americanum-group) have been collected from 1, 1, and 2 sites, respectively, and a Longidorus sp. is known from 2 sites. Trichodorus elefjohnsoni is the most widespread plant-parasitic species, collected from 9 locations throughout the Park. Four genera of Mononchida were identified from the samples: Iotonchus (7 sites), Mononchus (1), Mylonchulus (3), and Prionchulus (2). Elevation does not appear to be a significant factor in distribution of widespread species, but much more systematic sampling is needed to understand the niches of Park nematodes. Representative sampling of a large area such as GRSM is a formidable challenge. Most samples yield only a few specimens, often requiring return to rugged, steep locations for adequate numbers for study. In addition, the logistics of packing out heavy soil samples limits the number that can be collected on a field trip. Targeted sampling of

biodiversity reference plots established by GRSM will aid in recognition of nematode-plant relationships. In addition, application of molecular techniques to specimens collected in the future will enhance our ability to understand the diversity of soil nematodes and reduce the need for repeated collecting.

11. DEDUCING BIOLOGY FROM THE *MELOIDOGYNE HAPLA* GENOME. **Bird[1,2], David McK., E.H. Scholl[1], J.P. Cromer[1], P.M. DiGennaro[1], M. Goshe[3], D. Nielsen[2], V.M. Williamson[4], and C.H. Opperman[1].** [1]Plant Nematode Genomes Group, [2]Bioinformatics Research Center, and [3]Departmental of Structural Biology, NC State University, Raleigh NC. [4]Department of Nematology, University of California, Davis CA.

Through ongoing genome curation we have developed *M. hapla* as a model root-knot nematode (RKN). Initial annotation of the 54Mbp diploid genome identified 14,420 protein-coding genes, from which the proteome (HapPep) was deduced (Opperman et al., 2008. *PNAS*, 105:14802-7). Here we release HapPep4, which incorporates hundreds of mostly minor revisions. Importantly, there is no significant net change in gene number, and many protein predictions have now been experimentally confirmed by mass spectrometry. The annotated genome is available for viewing, analysis and download at www.hapla.org, and the proteome may be queried or downloaded from Superfamily: http://supfam.cs.bris.ac.uk/SUPERFAMILY/cgi-bin/gen_list.cgi?genome=wm. Addition of the *M. incognita* genome sequence to our pipeline has permitted comparative genomics, and we have used this approach to hunt for RKN-encoded mimics of plant peptide hormones. It is well established (Mitchum et al., 2008 *Cur. Opin. Plant Biol.* 11: 75-81) that *Heterodera glycines* encodes a mimic of a Clavata-Like Element (CLE), but CLEs are believed to be absent from RKN. We examined the *M. incognita* and *M. hapla* genomes, revealing five and eight candidate CLE loci respectively. Each is predicted to encode a secreted, 12 amino acid hormone ligand. Mapping these genes to their presumed analogues in Arabidopsis implies that the RKN genomes encode both type A and type B CLEs; these functional classifications are currently being experimentally validated. Interrogation of the *M. incognita* and *M. hapla* genomes also revealed 8 and 9 CEP genes respectively. In Arabidopsis, CEP (C-terminally encoded peptide) genes encode five, 15-amino acid peptide-hormone ligands responsible for root organogenesis. Unlike CLEs, CEP loci are absent from other animal genera, including cyst nematodes. Like their plant analogues, each RKN gene encodes a signal sequence at the amino terminus and single CEP motif at the carboxyl terminus. As is the case for the CLEs, plant CEPs include a domain between the signal sequence and the hormone domain, which most likely indicates a pro-protein that is proteolytically cleaved in the apoplast. Like RKN CLEs, RKN CEPs lack this domain, consistent with injection of functional hormone into the apoplast. We postulate that RKN-encoded CLEs and CEPs work in consort to initiate giant cell and gall formation. To comprehensively dissect transcriptional changes associated with parasitism by RKN we have constructed a 90,000-element microarray. This long-oligo array affords an average of 6-fold coverage for each HapPep gene. In one experiment we compared the transcriptional profile of naïve *M. hapla* J2 to that of J2 exposed to roots. Although it is believed that J2 hatch ''fully armed,'' perception of the host plant initiates expression of endoglucanase and pectinase, as well as genes associated with cell cycle. Using our array, we hope to integrate the cell biology of RKN with activity of its genome.

12. POTENTIAL IMPACT OF THE METAM SODIUM RE-REGISTRATION PROCESS ON POTATO PRODUCTION: WITH SPECIAL REFERENCE TO MICHIGAN. **Bird[1], George W., B. Kudwa[2], D. Sullivan[3], and L. Wernette[1].** [1]Dept. of Entomology 243 Natural Science, Michigan State University, East Lansing, MI 48824, [2]Michigan Potato Industry Commission, Dewitt, MI 48820, [3]Environmental Consulting, Inc., Alexandria, VA 22308.

The potato early-die disease complex, caused by an interaction between *Pratylenchus penetrans* and *Verticillium dahliae,* is a major limiting factor in Michigan potato production. Soil fumigation with metam sodium (sodium methyldithiocarbamate) is commonly used to reduce risk to this problem. While chemigation can be used for metam sodium; today, most applications in MI are made using modern soil injection spray-blade technology. Currently, the U.S. Environmental Protection Agency (EPA) is in the process of re-registering soil fumigants. In 2009, EPA Re-registration Eligibility Decision (RED) recommended significant application buffer zones for metam sodium. Under MI growing conditions, these restrictions have potential to reduce MI potato acreage about 80% for chemigation and 30% for soil injection. During the past year, the potato growers of MI, Wisconsin, Minnesota, North Dakota and Washington have worked with EPA, the product registrants and Sullivan Environmental Consulting, Inc. to obtain a comprehensive atmospheric emissions database for metam sodium for use in the re-registration and labeling processes. The research has been conducted at six commercial potato production sites, two each in MI, WI and WA. At each location, one-acre sites were treated with Sectagon 42 at the maximum labeled rate. Good agricultural practices (GAP) were followed, including one site in each state receiving post-application irrigation. Atmospheric emissions of this chemical were monitored extensively at all locations, before, during and after metam sodium application. In all cases, the emissions were significantly less than the existing data used by EPA for development of the RED recommendations. The results from the MI trials will be discussed in detail. Data from all locations have been submitted to EPA and registrants for use in the final phases of the re-registration process. It is anticipated that the research will result in significant modification of the buffer zone component of the product label. The leadership and resources for this research came directly from the potato growers of MI, WI, WA, MN and ND.

13. CONTRASTING EFFECTS OF ABOVEGROUND PLANT DIVERSITY ON BELOWGROUND NEMATODE COMMUNITY STRUCTURE. **Bliss, TJ[1], T.O. Powers[2], and C.E. Brassil[1]**. [1]School of Biological Sciences 348 Manter Hall, University of Nebraska-Lincoln, Lincoln, NE 68588, [2]Dept. of Plant Pathology 406 Plant Science Hall, University of Nebraska-Lincoln, Lincoln, NE 68583.

How does decreasing aboveground biodiversity influence belowground communities? Studies involving a broad range of aboveground ecosystems and taxonomic groups have shown that changes in diversity in one group within a community can affect the diversity and composition of other groups. Conversely, many soil ecologists have argued that belowground diversity is thought to be more strongly influenced by the presence and identity of the dominant aboveground species than by aboveground diversity itself. To explore the interaction between aboveground diversity and belowground communities, we examined soil nematode diversity and community composition directly under switchgrass (*Panicum virgatum*) in areas of high and low plant diversity. We found that soil nematode diversity under switchgrass in native prairies (high plant diversity) is not significantly different from soil nematode diversity under switchgrass in monoculture (low-to-zero plant diversity). This result indicates that plant diversity itself does not influence nematode diversity in this system. An examination of nematode community composition, however, revealed that plant diversity is linked to compositional shifts in nematode communities under switchgrass. Nematode communities under switchgrass in monoculture were more compositionally similar to each other than they were to nematode communities under switchgrass in native prairies and vice versa. Interestingly, fewer herbivorous nematodes were seen under switchgrass in monoculture than under switchgrass in native prairies, suggesting that nematode herbivory may be reduced in monocultures of switchgrass. Our study shows that both diversity and dominance are important aboveground factors influencing the structure of belowground communities.

14. COMPETITION BETWEEN *GLOBODERA ROSTOCHIENSIS* AND *G. PALLIDA*. **Blok, Vivian, A. Paterson, A. Holt, M. S. Phillips**, Plant Pathology Programme, Scottish Crop Research Institute, Invergowrie, UK DD2 5DA.

The potato cyst nematodes (PCN) *G. rostochiensis* (Woll.) and *G. pallida* (Stone) are the most economically important pests of potato (*Solanum tuberosum* L.) in the UK and are widely distributed in potato growing areas globally. There are few potato cultivars with resistance, particularly to *G. pallida*, and restrictions are increasing on the use of nematicides to control PCN. The withdrawal of the use of the aldicarb for use on potato crops in 2007 by the EU and the potential loss of other nematicides that are under review highlights the need for effective integrated pest management. Increasingly there will be a need to deploy control measures that incorporate an understanding of the role of environmental factors including temperature on PCN population dynamics, inter-specific competition and selection. To this end we have been investigating how the two species compete on potato genotypes that are susceptible or resistant to one or both of the species of PCN and at different temperatures to examine how their life cycles respond to different biotic and abiotic factors. Because the two species frequently occur together in the field, we have been assessing how the two species compete when present as mixtures using quantitative PCR. Biological differences between the two species in hatching and in their responses to different sources of resistance would be predicted to affect how they compete and this will have an impact of inter- and intraspecific selection and population dynamics.

15. INVESTIGATIONS OF THE EXPRESSION OF THE MULTIPARTITE MTDNA OF *GLOBODERA PALLIDA*. **Blok[1], Vivian C., P. Cock[1], M. Hunt[2], P. Hedley[1], J. Morris[1] and J. Jones[1].** [1]Scottish Crop Research Institute, Invergowrie, Dundee, Scotland UK, DD2 5DA, [2]Pathogen Genomics, The Wellcome Trust Sanger Institute, Genome Campus, Hinxton, Cambridge, UK CB10 1SA

The mitochondrial genome of *Globodera pallida* consists of at least 5 small circular molecules in contrast to the typical metazoan mtDNA which comprises a single circular molecule that encodes the 12 or 13 proteins involved in electron transport and oxidative phosphorylation, ribosomal RNAs and tRNAs. The sequencing of 5 scmtDNAs from *G. pallida* has revealed gene duplication, the presence of apparent pseudogenes as well as a substantial noncoding region in each of the scmtDNA. Thus far the *G. pallida* genome sequencing project has confirmed that the existence of several of these scmtDNAs and identified 2 more. The relative representation of each of the scmtDNAs differs in the genome sequence analysis with most hits for scmtDNA IV and VII. To investigate the expression of this unusual mtDNA genome a microarray analysis was conducted of eggs, juveniles and 8 day parasitic stage samples of *G. pallida*. Surprisingly there was limited evidence of overall changes in expression of mtDNA at different life stages compared to the expression of some nuclear encoded genes and the differential expression of different scmtDNAs was also limited. We are now able to compare the microarray analyses with transcriptome sequence analysis of eggs, juveniles, 8 and 15 day parasitic and female stages.

16. VIRULENCE AND MOLECULAR CHARACTERIZATION OF *GLOBODERA PALLIDA* FROM IDAHO. **Blok, Vivian C. and M. S. Phillips**, Plant Pathology Programme, SCRI, Invergowrie, Dundee DD2 5DA Scotland

Biological and molecular experiments were conducted to characterise three isolates of *Globodera pallida* which had been found in Idaho in 2006. A hatching test showed that the cysts recovered had low viability. The use of these original cysts in a

virulence assay gave poor data as reproduction was low. Nonetheless the data suggested that the population was similar to the Pa2/3 virulence group when compared to the European Pathotype Scheme. A second virulence test with a new generation of cysts showed good levels of reproduction. This indicted that there was no inherent lack of fitness in the Idaho population and confirmed that pattern of virulence when tested against resistance from *Solanum vernei* and *S. tuberosum* spp andigena CPC2802 was similar to the Pa2/3 group. Molecular studies comparing the sequences of mitochondrial gene Cytochrome B from South American and European populations indicated that the Idaho sequence had greatest similarity with populations from Southern Peru and Europe. Sequence of the nuclear genes pectate lyase and cathepsin did not distinguish groups of populations as clearly as the mitochondrial sequence but supported those findings.

17. HOST SUITABILITY OF SELECTED MONOCOT PLANTS TO *MELOIDOGYNE MAYAGUENSIS*. **Brito[1], Janete A., M. Hao[1], and D. W. Dickson[2].** [1]Division of Plant Industry, DPI, Gainesville, FL 32614; [2]Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611.

Vegetables, fruit trees, herbs, ornamental and weed plants have all been reported as host of *Meloidogyne mayaguensis* in Florida; however very little is known about this nematode's ability to reproduce on monocot plants. The objective of this study was to determine the host suitability of nine selected monocot genotypes to *M. mayaguensis*. Three root-knot nematode resistant corn germplasm lines (MP 709, MP 711, and MP 712), three susceptible hybrids (Silver Queen, Dixie, Golden Queen) and sorghum 15, sorghum 16 and wheat 'AGS 2000' were evaluated in a duplicated test under greenhouse conditions. Each plant was inoculated with 5,000 eggs. Tomato 'Rutgers' was used as a control for determining inoculum viability. Gall (GI) and egg mass indices (EMI), and number of eggs per gram of fresh root were recorded. No root galling or visible egg masses were observed on any of the monocot genotypes whereas in the control, both GI and EMI were rated 5.0 (scale: 0.0 – 5.0). The corn lines MP 709, MP 711 and MP 712, which have been reported as resistant to both *M. arenaria* and *M. incognita* were nonhost to *M. mayaguensis*. Similarly, two corn hybrids (Silver Queen and Golden Queen) and wheat 'AGS 2000' also were nonhost. Sorghum 15, sorghum 16 and Dixie corn were resistant to this nematode species with 11.67, 1.67 and 1.67 eggs /g of fresh root, respectively. In summary, it appears that some monocot plants are nonhost to *M. mayaguensis*, thus they have the potential to serve as good rotation crops for management of sites infested with this nematode species.

18. ASSESSMENT OF INOCULUM LEVEL, EVALUATION TIME AND VARIABLES FOR SCREENINGS OF *PSIDIUM* SPP. FOR RESISTANCE TO *MELOIDOGYNE MAYAGUENSIS*. **Burla, R.S., R.M. Souza, V.M. Gomes, and F.M. Corrêa.** Dept. of Entomology and Phytopathology, Universidade Estadual do Norte Fluminense Darcy Ribeiro, 28015-620, Campos dos Goytacazes, Brazil.

The difficulty to control *M. mayaguensis* parasitic on guava through chemical or cultural methods suggests that genetic resistance is the best control strategy at a mid-term. Although there has been screenings of *Psidium* spp. in Brazil and elsewhere, no study has been conducted to assess the best inoculum level, evaluation time or variable to be used. In this study, guava seedlings of the *M. mayaguensis*-susceptible cultivar 'Paluma' with four pairs of leaves were inoculated in pots with 2 liters of growth substrate Plantmax® with 500, 2,000, 3,500, 5,000 or 6,500 eggs / seedling, and they were evaluated 45, 90, 135 or 180 days later. The variables assessed were root system fresh mass, final nematode population (Fp), reproduction factor (RF= Fp / inoculum used) and number of eggs + J_2 / gram of root. The experiment was conducted in greenhouse in two periods of the year (spring-summer-fall and winter-spring-summer), in an entirely randomized design with 20 treatments and six replicates (one seedling / pot) per treatment. The transformed (log x) data were analyzed through ANOVA, F test at 5 % and multiple regression. In both periods of the year the variable root system fresh mass showed a positive correlation ($P < 0.05$) with the evaluation time (45 to 180 days after inoculation), but not with the inoculum level. For the variables Fp and number of eggs + J_2 / gram of root, the correlation with the factors evaluation time and inoculum level was either significant or not depending on the period of the year in which the experiment was conducted. In contrast, the variable RF showed a significant positive correlation with the evaluation time and inoculum level in both periods of the year, hence being considered the best variable for screenings of *Psidium* spp. for resistance to *M. mayaguensis*. The results also indicated that high inoculum levels (3,500 eggs / seedling and above) reduce the RF, what could lead to false positive results for resistance to *M. mayaguensis*. The best combination of inoculum level and evaluation time was 500 or 2,000 eggs / seedling and 135 or 180 days after nematode inoculation, respectively.

19. ENTOMOPATHOGENIC NEMATODE ECOLOGY AS A BASIS FOR THEIR USE IN PEST MANAGEMENT. **Campos-Herrera, Raquel[1,2], E. Pathak[1], R.J., Stuart[1], F.E. El-Borai[1,3], C. Gutiérrez[2], J.H. Graham[1], and L.W. Duncan[1].** [1]Citrus Research and Education Center, University of Florida, 700 Experiment Station Rd, Lake Alfred Fl 33850 (USA), [2]Centro de Ciencias Medioambientales, CSIC, Serrano 115 dpdo Madrid, 28006 (Spain), [3]Plant Protection Department, Faculty of Agriculture, Zagazig University (Egypt).

Entomopathogenic nematodes (EPNs) have been applied in Florida citrus orchards since the early 1990s as a component of IPM programs targeting the root weevil, *Diaprepes abbreviatus*. The soil environment modulates biological control of insects

by EPN directly, through physical and chemical properties that affect EPN behavior and longevity, as well as indirectly through the different communities of natural enemies of EPN supported by different soil conditions. However, relatively little is known about EPN ecology or the post-application biology of augmented EPN. Knowledge of interactions between soils, food webs and EPN will provide new opportunities to improve the biological control of insects by endemic or augmented EPN. We designed real-time PCR primer/probe combinations to measure all of the six known EPN species in Florida, six species of nematophagous fungi (NF) known to prey on EPN in Florida citrus orchards, and two *Paenibacillus* species that are phoretic, ectoparasites of EPN. We are using those molecular tools to identify interrelationships between spatial and temporal patterns of these organisms over a range of diverse orchard habitats with various physical soil properties. An ongoing regional survey has shown that the richness and diversity of EPN species are significantly greater in deep, coarse sandy soils of the Central Ridge than in shallower, finer textured soils of the Interior and Coastal Flatwoods. Communities of endoparasitic and trapping NF at these sites, derived from principal components analysis, were closely related to principal components derived from soil texture and chemistry. Both types of NF were inversely related to sand content of soil and endoparasitic NF were also inversely related to soil pH and electrical conductivity when analyzed by stepwise multiple regression. All species of trapping NF were more abundant at depths of 0-15 cm than 15-30 cm at all 3 localities of an ongoing survey of temporal abundance. By contrast, endoparasitic NF and *Steinernema diaprepesi, Heterorhabditis indica* and *H. zealandica* tended to inhabit the deeper soil horizon. In a long-term field trial comparing conventional irrigation/fertilization to daily fertigation of trees, endemic and augmented steinernematids were reduced by daily fertigation, whereas significantly more augmented heterorhabditids were recovered in this treatment. Several NF species were more prevalent in plots that were augmented with *S. riobrave* than with *H. indica* and were more prevalent in fertigated plots when measured 48 and 72 hours after augmenting soil with EPN. Experiments to elucidate the effects of soil moisture and fertilizers on these NF species are ongoing to help determine whether NF play a significant role in how the two cultural practices affect both the endemic EPN communities and the post-application biology of different EPN species.

20. THE USE OF MOLECULAR TOOLS AND TECHNIQUES TO ADDRESS ECOLOGICAL QUESTIONS: TECHNICAL BASIS AND DEVELOPMENT OF EXPERIMENTS. **Campos-Herrera, Raquel[1,2], E. G. Johnson[1], and L. W. Duncan[1].** [1]Citrus Research and Education Center, University of Florida, 700 Experiment Station Rd, Lake Alfred Fl 33850 (USA), [2]Centro de Ciencias Medioambientales, CSIC, Serrano 115 dpdo Madrid, 28006 (Spain).

In recent decades, molecular ecology emerged as a new scientific discipline following the development of new molecular techniques. Their use for addressing different ecological questions has promoted the integration of several sciences including ecology, evolution, conservation biology, biodiversity and behavioural ecology; and provided new perspectives and approaches. Some techniques that were extremely costly and required cutting-edge technology 10 years ago are now considered routine for most laboratories. Other methods are still developing and are cost prohibitive for most researchers. Moreover, the frequency of application of these tools differs among fields, with evolutionary biology being one of the most extensive users. Therefore, the current challenge for zoologists and ecologists is to integrate these tools and ask new questions in ecology or to reinvestigate previous ones from a different perspective. Herein, we present some of the tools and techniques used for the molecular identification of species and individuals in studies of microbial and viral diversity, behavioural ecology (sex ratio and dispersal), population genetics, phylogeography, and conservation genetics in an attempt to enhance their application in Nematology. We will explain the basic concepts of frequently used techniques and provide step-by-step protocols for the design of new and personalized experiments using qPCR, high throughput community analyses, multiple methods of DNA fingerprinting and other useful molecular techniques. Finally, we will address some common problems that arise during method development and alternatives to overcome these difficulties.

21. A NEW DIPLOSCAPTEROIDES (*RHABDITIDA: RHABDITIDAE*) FROM THE SURFACE OF AN ADULT FUNGUS GNAT (*DIPTERA: SCIARIDAE*). **Carta, Lynn K.[1], Z.A. Handoo[1], and W.H. Carlson[2].** [1]United States Department of Agriculture, ARS-BARC-W, Nematology Laboratory, Beltsville, Maryland 20705, USA, [2]United States Department of Agriculture, APHIS-PPQ, Blaine, Washington 98230, USA.

A new association of a new species of Diploscapteroides with an adult fungus gnat is described. Twelve adult nematodes were attached to a single gnat by a secretion of the nematode lip region to all the outer surfaces, including the eyes. The adhesive remained attached to the nematode head even after the nematode was removed with difficulty from the fly. Female morphology is described from alcohol and formalin-fixed specimens. Adults were spiral to J-shaped, with eggs visible near the tail and above the pharyngeal-intestinal junction in some gravid females. Despite some variable, minor longitudinal shrinkage of specimens from alcohol, the new species is discretely shorter in body length and width than the five other known species. It had ‘a’ ratios larger and ‘b’ ratios smaller than most species. Until now Diploscapteroides was found only in rotting plant material or soil, and few specimens were available for most species descriptions. Other nematode associates of Sciaridae flies are members of Tylenchida, Mermithida or juvenile Steinernema and Heterorhabditis that parasitize larval insects.

22. *CATENARIA AUXILIARIS*: A NEW PARASITE OF *ROTYLENCHULUS RENIFORMIS*. **Castillo[1], Juan D., and K. S. Lawrence[1].** [1]Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849.

The reniform nematode (*Rotylenchulus reniformis*) is an economic pathogen of cotton and soybean in tropical and temperate regions of north and South America. Currently, there are no commercially available cotton cultivars resistant to *R. reniformis*; therefore, management practices are limited to crop rotation and chemical nematicides. Although, biological control of *R. reniformis* has never been implemented in cotton or soybean crops in the United States and it is a possible option of nematode management in the future. Previous studies on *R. reniformis* have reported *Arthrobotrys dactyloides* and *Dactylaria brochopaga* are parasites of the nematode vermiform stages, and *Paecilomyces lilacinus* is an egg parasite. Recent microscope observations show the presence of *Catenaria auxiliaris* colonizing various life stages of *R. reniformis*. Vermiform life stages parasitized by this fungus present swellings inside the body that result from the formation of ovoid sporangia. Later sporangia form yellow and circular resting spores with a reticulate appearance with an average diameter of 20-25μm. In advanced stages of infection, uniflagellate oblong zoospores (3x2μm) are released from the nematodes body. *Rotylenchulus reniformis* females with eggs were removed from the roots and observed microscopically. Females confirmed an encystment of the zoospore in the metacarpus region of the nematodes body. Furthermore, cysts are observed inside the eggs. Further studies are being conducting on the biology of the infection at different life stages of *R. reniformis* in cotton roots.

23. VIRULENCE OF THE SOYBEAN CYST NEMATODE HAS INCREASED OVER YEARS IN MINNESOTA. **Chen, S[1], B. Potter[2], and J. Orf[3].** [1]University of Minnesota Southern Research and Outreach Center, Waseca, MN 56093; [2]University of Minnesota Southwest Research and Outreach Center, Lamberton, MN 56152; [3]Department of Agronomy and Plant Genetics, University of Minnesota, St Paul, MN 55108.

The soybean cyst nematode (SCN), *Heterodera glycines*, is a major biotic yield-limiting factor of soybean. Since it was first found in 1978, infestation of SCN in Minnesota gradually spread across the state and now has been found in most (63) soybean-producing counties including a few counties in the Red River Valley in the northwestern region of the state. SCN-resistant soybean cultivars have been used in Minnesota for about two decades. During 2007-2008, a state-wide survey was conducted to determine SCN virulence phenotypes (HG Types) in Minnesota. A total of 252 fields across the state were sampled, and the number of samples taken from each county was based on the soybean acreage in the county. The SCN was detected in approximately 120 soil samples (fields). The reproduction potential measured as Female Index (FI) of the SCN populations was determined on the seven indicator lines PI 548402 (Peking), PI 88788, PI 90763, PI 437654, PI 209332, PI 89772, and PI 548316 with Lee 74 as the susceptible control. Based on the 95 SCN populations that have been completed to date, most of them (71.6%) reproduced well (FI > 10) on PI 88788, the major source of resistance for breeding soybean cultivars in the North Central region. Reproduction of 15.8% of the populations was positive (FI > 10) on PI 548402. PI 90763 and PI 89772 had similar responses to the SCN populations and yielded FI > 10 only in 8.4% of the populations. PI 209332 and PI 548316 yielded FI > 10 for 77.9% and 94.7% of the populations, respectively. Only PI 437654 was resistant to all populations. The average FIs on Peking, PI 88788, PI 90763, PI 437654, PI 209332, PI 89772, and PI 548316 were 5.5, 18.6, 3.1, 0.4, 19.3, 3.2, and 33.2, respectively. Compared with the data in a previous survey conducted in 2002, the virulence of the SCN populations to the resistance source soybean or commercial soybean cultivars has increased dramatically since then. For example, the average FI on PI 88788 increased from 4.5 in 2002 to 18.6 in 2007-2008. Our study suggests that an extensive integrated approach including alternative sources of resistance, appropriate crop rotation, and other cultural and biological control methods are needed for a long-term effective management of the nematode.

24. DIVERSE CLE PEPTIDES FROM CYST NEMATODE SPECIES. **Chen[1], Shiyan, S. Lu[2], H. Yu[1], M.G. Mitchum[3], and X. Wang[1,4].** [1]Dept. of Plant Pathology and Plant-Microbe Biology, Cornell University, Ithaca, NY 14853, [2]USDA/ARS, Cereal Crops Research Unit, Northern Crop Science Laboratory, Fargo, ND 58105, [3]Division of Plant Sciences and Bond Life Sciences Center, University of Missouri, Columbia, MO 65211, [4]Robert W. Holley Center for Agriculture and Health, USDA/ARS, Ithaca, NY 14853.

Plant CLAVATA3/ESR (CLE)-like peptides play diverse roles in plant growth and development including maintenance of the stem cell population in the root meristem. Small secreted peptides sharing similarity to plant CLE signaling peptides have been isolated from several cyst nematode species including soybean cyst nematode (*Heterodera glycines*), sugarbeet cyst nematode (*H. schachtii*), potato cyst nematodes (*Globodera rostochiensis* and *G. pallida*), and tobacco cyst nematode (*G. tabacum*). Interestingly, unlike typical plant and *Heterodera* CLEs that contain a single C-terminal CLE motif, most of the *Globodera CLE* genes encode CLE proteins with multiple CLE motifs. *H. glycines* CLEs have recently been demonstrated to be secreted into nematode-induced feeding cells. Moreover, our in-depth functional characterization demonstrated a functional similarity of nematode CLEs to endogenous plant CLE peptides, suggesting that once nematode CLEs are delivered into plant cells, they can exert their function as endogenous plant CLEs to redirect plant CLE signaling pathways to establish a successful parasitic association with host plants. The wide distribution of CLEs in cyst nematode species indicates that ligand mimicry of plant CLEs is an important mechanism in cyst nematode parasitism of host plants. A better understanding of this extraordinary example of molecular mimicry will advance our knowledge of plant-nematode interactions.

25. MOLECULAR AND MORPHOLOGICAL ANALYSIS OF *PLECTUS* BIODIVERSITY IN THE MCMURDO DRY VALLEYS. **Clayton, Adam L.**[1], **I. Andràssy**[2], **D.H. Wall**[3], **B.J. Adams**[1]. [1]Department of Biology, Brigham Young University, Provo, UT, USA. [2]Department of Systematic Zoology and Ecology of the Eötvös Loránd UniverDepartment of Systematic Zoology and Ecology of the Eötvös Loránd University, Zootaxonomy Research Group of the Hungarian Academy of Sciences, Budapest, Hungary. [3]Department of Biology and Natural Resource Ecology Laboratory, Colorado State University, Fort Collins, CO, USA

Nematodes are known to play important roles in ecosystem functioning, however their specific roles may vary across different taxonomic levels. Correct identification of nematode species is necessary when analyzing their diversity in order to effectively assign their functional role(s). Several different nematode genera are known to inhabit the McMurdo Dry Valleys, Antarctica, but the ecological roles of each of these taxa are still a mystery. One genus of particular interest is *Plectus*. The taxonomic diversity of these Antarctic inhabitants is currently debated. To resolve this we have collected both terrestrial and aquatic samples belonging to the *Plectus* genus, identifying them using morphological and molecular characters. Patterns of lineage divergence and distribution of *Plectus* in the MCM were explored using PCR and sequencing of the 28s, 18s, and ITS ribosomal RNA. Preliminary phylogenetic analyses indicate the presence of at least two genetically distinct species of *Plectus* with additional population-level variation within each species. Potential ecological and geographical factors responsible for the distribution of *Plectus* will also be presented.

26. DGGE FINGERPRINTING TO MONITOR NEMATODE POPULATIONS IN A PEANUT ROTATION SYSTEM. **Conner, Kassie N. and R.N. Huettel**. Dept. of Entomology and Plant Pathology 209 Life Sciences Bldg., Auburn University, AL 36849.

Molecular fingerprinting methods such as DGGE (denatured gradient gel electrophoresis) can generate population specific fingerprints by displaying ribosomal polymorphisms naturally present in a community. In this study genetic profiles of the total nematode community from peanut soils were generated through extraction of nematode DNA, amplification of partial 18S rDNA genes using nematode consensus primers and subsequent separation by DGGE. Samples were collected from four different cropping rotations (continuous peanut, peanut/corn, peanut/cotton, and continuous Bahia) at three sampling periods (pre-plant, mid-season, and harvest) for two consecutive years (2008 and 2009). Unique and common bands within each molecular fingerprint were then excised, re-amplified and sequenced in order to identify populations within the nematode community. Analysis of the genetic profiles revealed seven distinctive groups at 50% or greater similarity for 2008 and 2009. This shows the similarity in nematode populations between repetitions of specific rotation sequences including the continuous peanut rotation for all sampling dates in 2008 and harvest of 2009. There were 103 partial 18S sequences, out of the total 121 samples subjected to sequencing, which were assigned to known sequences found in GenBank. These samples represented 31 separate genera, 64% of which consisted of free-living nematodes. DGGE can be a useful technique to identify nematode populations and monitor shifts within the nematode community under different cultural practices. Understanding the nematode community could lead to management practices aimed at altering or manipulating cropping conditions to increase beneficial nematodes or decrease plant-parasitic nematode populations and ultimately increase crop health.

27. PHYLOGENETIC RELATIONSHIP AMONG NEMATODES OF THE SUBORDER CRICONEMATINA USING 18S-rDNA REGION. **Cordero, M,[1] Robbins, R.[1] and Szalanski, A.[2]** 2010. [1]Department of Plant Pathology. 2601 N. Young Ave. Cralley - Warren Research Lab. University of Arkansas, Fayetteville, AR. 72704. [2]Department of Entomology, AGRI 330B, University of Arkansas, Fayetteville, AR. 72701.mcordero@uark.edu;rrobbin@uark.edu; aszalan@uark.edu

The suborder Criconematina is composed of three Superfamilies: *Criconematoidea*, *Hemicycliophoroidea*, and *Tylenchuloidea*; members of each family share common anatomical features that allow taxonomist to identify and to classify them. However, similarities among them sometimes create ambiguities among genera of different families. Genera from Arkansas, Missouri, North Carolina and Florida were extracted from soil samples using standard methods or received in NaCl 1M or 95% Ethanol. Some of the species collected belong to the genera *Mesocriconema, Nothocriconema, Criconema, Ogma, Xenocriconemella, Paratylenchus, Gracilacus, Tylenchulus* and a population tentatively identified as *Calossia* . Nematodes from each population were crushed in 5 μl of sterile pcr water and storage at -80oC for at least 2 weeks. One individual from each population was PCR amplied using the nuclear rDNA 18S gene primers 18S1.2 and 18Sr2b. Amplicons were sequenced in both directions and a molecular phylogenetic analysis was conducted using maximum parsimony, maximum likelihood and Bayesian analysis.

28. EVALUATION OF *BACILLUS FIRMUS* STRAIN I-1582 AS A BIONEMATICIDE FOR TURF. **Crow, William T.** Entomology and Nematology Dept., University of Florida, Gainesville, FL 32611.

In 2009 two trials were conducted at the University of Florida Plant Science Research Unit in Citra, Florida to evaluate *Bacillus firmus* strain I-1582 as a bionematicide on turf. Both sites were infested with damaging numbers of *Belonolaimus longicaudatus* (sting nematode) at the beginning of the trial. *Bacillus firmus* was formulated as ES TC002 (Bayer

CropSciences, Triangle Park, NC), a wettable granule formulation applied to turf as a spray. This formulation was applied as either: 1) a single application of 78 kg/ha or 2) two applications of 39 kg/ha applied at four week intervals. The *B. firmus* treatments were compared to plots treated with 11.2 kg/ha fenamiphos, and non-treated plots. Effects on population densities of *B. longicaudatus*, turf density, and root length were compared among treatments. In one trial, both *B. firmus* treatments reduced population densities of *B. longicaudatus* relative to the non-treated. In the other trial, population densities of *B. longicaudatus* declined to near zero in all plots over the course of the experiment. In the trial with significant nematode effects, there was a corresponding increase in turf density from *B. firmus* application. In both trials, both *B. firmus* treatments improved root lengths relative to the non-treated as much or greater than did fenamiphos. These trial results indicate that *B. firmus* strain I-1582 could become a useful tactic for management of *B. longicaudatus* on turfgrasses.

29. YIELD DRAG ASSOCIATED WITH RESISTANCE TO *MELOIDOGYNE INCOGNITA* IN HIGH-YIELDING COTTON GERMPLASM. **Davis, R. F., P. W. Chee, and E. L. Lubbers** USDA-ARS, Crop Protection and Management Research Unit, P. O. Box 748, Tifton, GA 31793

In plant breeding, accidental incorporation of deleterious DNA near a desirable gene is called linkage drag; if it reduces yield, it is called yield drag. Yield drag is best documented by comparing near isogenic lines with and without the DNA containing the desired gene to minimize other genetic differences. In a back-cross breeding program to improve the yield and fiber quality of cotton germplasm with resistance to *M. incognita*, near-isogenic lines with and without resistance were selected. In greenhouse tests, the resistant isoline (GA120R1B3) reduced reproduction of *M. incognita* by 95% compared to the susceptible isoline (GA120S1A1). The isolines and the susceptible recurrent parent (PD94042) used in creating the isolines were evaluated for yield drag in a field study with 8 replications. Although they are not isogenic lines, the source of resistance (M-120 RNR) in GA120R1B3 also was tested along with its susceptible recurrent parent (Coker 201). The study was conducted in a field with very low to nil populations of *M. incognita*, and the field was fumigated with 1,3-dichloropropene prior to planting to evaluate plants in the absence of nematode parasitism. No galling was found during this study. Yield of M-120 RNR (1776 kg/ha) did not differ ($P \leq 0.05$) from Coker 201 (1673 kg/ha). This suggests that yield drag may not have occurred with M-120 RNR. However, yield of GA120R1B3 (2087 kg/ha) was less than the yield of either GA120S1A1 (2356 kg/ha) or PD94042 (2405 kg/ha), which did not differ from each other. Although yield and quality of GA120R1B3 are improved compared to its resistant parent (M-120 RNR), yield drag may have been introduced. Yield drag may be more readily observed as yield potential increases. Additional breeding should be able to break this linkage drag thereby further improving yield of this resistant germplasm line.

30. RECENT ADVANCEMENTS IN APPLIED ENTOMOPATHOGENIC NEMATOLOGY IN SOUTH AMERICA. **Dolinski, Claudia.** Universidade Estadual do Norte Fluminense Darcy Ribeiro/CCTA/LEF, Av. Alberto Lamego, 2000, Pq. California, Campos dos Goytacazes, RJ, Brazil, 28015-602

Applied studies using entomopathogenic nematodes (EPNs) in South America started during 90's in different countries focusing diverse soil pests and crops. Important pests from sugar cane, guava, coffee and other crops lead researchers to test different application technology in the field. Different irrigation systems and spray equipment have been tested, also the technique known as insect-cadaver. Operating pressures were also tested since some nematode species may tolerate high pressures without notable damage, whereas other species may require lower pressure limits. Also the ability of the nematode to find a host after receiving high pressure has been observed. Other critical environmental factors including ultraviolet radiation, adequate soil moisture, and appropriate temperature have also being studied. Studies showed that certain fertilizers and chemical pesticides can have positive effects on EPNs efficacy, whereas other agents may have neutral or negative effects. Besides soil application, foliar applications have also been tested showing similar efficacy. A variety of other abiotic and biotic factors have also being tested and in this talk an overview of the main work done in South America will be reported.

31. EFFECTS OF TILLAGE ON THE INTERACTION OF SOYBEAN RHIZOSPHERE ORGANISMS. **Donald, P.A.[1] , and Tyler, D.D.[2].** [1]USDA ARS, Crop Genetics Research Unit, 605 Airways Blvd, Jackson, TN 38301, [2]Biosystems Engineering & Soil Science, University of Tennessee, 605 Airways Blvd, Jackson, TN 38301.

Soybean rhizosphere organisms were monitored in long-term tillage plots established in 1979. The treatments included a glyphosate-resistant soybean monoculture planted both no-till with a winter wheat cover crop and conventionally. In 2002, half of each plot was tilled using a disc to determine the short-term effects of tillage on soybean cyst nematode reproduction. Increased soybean cyst nematode, *Heterodera glycines*, reproduction was observed in the tilled plots initiated in 2002 compared to continuous no-tillage since 1979. Little difference in reproductive rates was observed from 2007 through 2009 for the different tillage treatments. Data were collected on the frequency of active and total bacteria, active and total fungi, protozoans, *H. glycines* egg population density, and grain yield from 2006 through 2009. Although low levels of active fungi were detected, total fungi levels were tenfold higher than active fungi. Bacteria, both active and total, were more abundant than fungi and more abundant in disc treatments regardless of the number of years since the last disc treatment. Protozoans

were predominately flagellates and amoeba. Cilliates were present in much lower numbers. Grain yield was the lowest in 2007 due to severe drought. Low *H. glycines* reproductive rate in 2007 reflected the poor soybean growing conditions and egg population density at planting the following year was also low. *H. glycines* reproductive rate was more variable in the disc tillage treatment than the no-tillage treatment.

32. POTENTIAL ROLES OF GREEN MANURE BIOFUMIGANTS AND SEED EXUDATES IN THE CONTROL OF *GLOBODERA PALLIDA*. **Dossey[1], Zareen, Ekaterini Riga[1,2]. [1]**Plant Pathology Department, Washington State University, Pullman, WA; and **[2]**Washington State University, IAREC, 24106 N. Bunn Rd., Prosser, WA, 99350.

The white potato cyst nematode *Globodera pallida* is a regulated pest in the United States. In 2006, *G. pallida* was discovered in eastern Idaho. The agricultural industry has relied on synthetic nematicides to control nematodes, but their negative impact on the environment has led to their restricted usage. Therefore, the potential use of biofumigation derived from plant residues as an alternative control method was investigated. The biofumigation potential of five plants used as green manures against PCN and the effects of water soluble seed exudates of these plants on the dorsal pharyngeal gland nucleolus (DPGN) of the PCN second stage juvenile (J2) was investigated. The Brassicaceae species used were *Raphanus sativus* 'Terranova', *Brassica napus* 'Greenland', *Sinapis alba* 'Achilles', *Eruca sativa* 'Nemat' and one Poaceae species, *Lolium multiflorum* 'Emmerson' in greenhouse and seed exudate assays. Data from the green manure trials were analyzed using ANOVA. No significant differences ($P>0.05$) were found between treatments and controls. However, the smallest number of cysts was produced in pots amended with *R. sativus* in comparison to other treatments and controls, with a 50% reduction in cyst production. Also, J2s were exposed to seed exudates to determine if any of the plants tested have the potential to mimic PCN egg hatching which is normally induced in the presence of PCN host plants. The responses of DPGN to the seed exudates study were measured using Nomarski microscopy. The data showed significant differences ($P<0.0001$) between treatments and water control. Seed exudates from *B. napus* had the greatest effect, resulting in the largest size nucleolus among the treatments, while exudates from *S. alba* resulted in the smallest size nucleolus. Therefore, *R. sativus, B. napus* and *S. alba* have the potential to reduce PCN and could be used in combination with methyl bromide to aid in PCN eradication.

33. NEW ROOT-KNOT NEMATODE FOUND PARASITIZING CREEPING BENTGRASS ON GOLF COURSES IN VIRGINIA, MARYLAND, AND PENNSYLVANIA. **Eisenback, J. D**. Department of Plant Pathology, Physiology and Weed Science, Virginia Polytechnic Institute and State University, Blacksburg, VA 24061.

Creeping bentgrass plugs collected from the Virginia Tech Turfgrass Research Center failed to thrive when transferred to growth chambers. Examination of the unpotted root system revealed that they were severely infected with root-knot nematodes. Symptoms included chlorosis, stunting, and a reduced root system. Additional research on the morphology and host range revealed several unusual morphological characters and a unique host range indicated that it was a new species. The perineal pattern, shape of the female stylet, and shape of the juvenile tail were unique and different from those of any other described species. Creeping bentgrass was the only host. Populations of root-knot with similar morphology were also found on samples from several golf courses in Virginia, Maryland, and Pennsylvania. Comparison of the rDNA from the 18s ITS region confirmed that these population were identical with each other, but very different from all other Meloidogyne species.

34. PROTECTION STRATEGIES FOR ''SAFE COCOA BEANS HARVEST'' IN NEMATODE-INFESTED PRODUCTION ENVIRONMENTS. **Fademi[1], Olutayo. A., M.O. Ogunlade[1], S.B. Orisajo[1], M.O. Okeniyi[1], B.O. Obatolu[1], O.M. Olatunbosun[2] and A.M. Jolaoso[2]. [1]**Cocoa Research Institute of Nigeria, P.M.B. 5244, Ibadan, Nigeria, [2]Raw Materials Research and Development Council, P.M.B. 232, Abuja, Nigeria.

There is more cocoa consumed in the developed countries than in the developing countries where more than 70% of the cocoa beans are produced. Yet there is intense impasse in demand against production from the developed nations because of the high premium placed on food safety by the developed world. Cocoa bean quality is a major factor influencing the final decision of consumers on what to purchase, yet cocoa producers are at cross roads on level of pesticides use against the many field pests (including nematodes) and diseases of cocoa. Plant-parasitic nematodes including the root-knot nematode (RKN) *Meloidogyne incognita* infests cocoa seedlings, reducing establishment and growth, and yields in later years. A three-year screen house assessment of thirteen cocoa clones which included the two most prevalent on farmers' fields (F_3 Amazon and Amelonado) showed that three (LCTEEN, T12/11, and AMAZ 15.15) and two others (ICS 1 and T65/7) exhibited resistant and tolerant reactions respectively. Further studies on control of the nematodes without recourse to conventional pesticides showed that use of organic amendments nematode development and root galling, also increasing the plant growth parameters of plant height, stem girth, leaf numbers, leaf area, and root/shoot dry weights by 13 – 35% over control. In the greenhouse, cocoa pod husk (CPH) amendment alone at 13,900, 27,800 and 55,600 kg/ha lowered root galling by 88%, 92% and 93% respectively. Mixed with neem leaves, neem seeds or urea, root galling and nematode recovery in roots and soils were reduced by 30 – 35%. Under field conditions, the mixtures suppressed nematode populations by 63.34% - 90.71%. The highest rate of nematode suppression induced by organic amendment ranged from 75 – 100% and 63.34 – 90.71% at 6 and 24

months respectively under field conditions while percent seedling survival in organic-amended soils were higher by 26.09 – 71.74%. This paper reports options for minimum pesticide use for nematode as one step in lowering quantity of pesticides pumped into cocoa on-field.

35. EFFECT OF FALL ANNUAL RYE GRASS SEEDING ON SOYBEAN CYST NEMATODE. **Faghihi, Jamal.[1], R. A. Vierling[2], and V. R. Ferris[1]**. [1]Department of Entomology, Purdue University, West Lafayette, IN 47907, [2]Indiana Crop Improvement Association and Department of Agronomy, Purdue University, West Lafayette, IN 47907.

Soybean cyst nematode (SCN) eggs remain protected inside cysts in the soil over the winter ready to attack the next crop of soybeans. A cover crop that is planted in the fall, which causes the numbers of eggs and infective juveniles to diminish prior to the planting of soybeans in the spring, would be useful in SCN management. Such cover crops can also compete with winter annual weeds, some of which are excellent hosts for SCN. Several researchers have reported that annual rye grass might just be such a crop. In a study in our greenhouse, rye was effective in reducing SCN populations. As a follow-up, we established small field plots in 2008-2010 to determine the efficacy of annual rye in reducing SCN populations. Our attempt to establish annual rye grass in fall of 2008 was not successful and did not reduce SCN populations for the 2009 season. However, following a well established annual rye grass planting in the fall of 2009, the population of SCN was significantly lower as compared with SCN populations in wheat and fallow plots. Similar results were observed in spring of 2010 from the same plots. These plots will be planted with a SCN susceptible soybean cultivar and the SCN populations will be monitored throughout the 2010 growing season.

36. INTRA-GENOMIC VARIATION IN THE RIBOSOMAL REPEATS OF NEMATODES. **Fournier, D.[1], Sung W.[2], Thomas W.K.[2], Bergeron R.D.**[1, 1]Department of Computer Science, University of New Hampshire, Durham, NH 03824, [2]Hubbard Center for Genome Studies, University of New Hampshire, Durham, NH 03824

Ribosomal RNA (rRNA) genes, are common to all biological species and useful for inferring evolutionary relationships and biodiversity. Eukaryotic cells can have hundreds of copies of these genes repeated in tandem arrays. Although, these sequences follow a general pattern of ''concerted evolution'', repeats within an organism can vary substantially in DNA sequence. To investigate these patterns we compared the repeat number and sequences of six nematode species. The number of repeats varies from 50 to over 300 copies and the pattern of variation within a single genome was found to be uncorrelated with patterns of divergence between species. These observations suggest that polymorphisms within a genome are less constrained by selection for function compared to the patterns of divergence between genomes which reflect a strong signature of natural selection for rRNA function. Studying these patterns of variance can help us better understand how mutations affect these genes and how we can use the genes most effectively in evolutionary and ecological studies.

37. COMPARISON OF METHODS FOR ASSESSING RESISTANCE AND TOLERANCE TO *A.FRAGARIAE* IN HOSTA CULTIVARS. **Fu, Zhen[1], P. Agudelo[1] and P. Gerard[2]**. [1]Dept. of Entomology, Soils, and Plant Sciences. 114 Long Hall, Clemson University, Clemson, SC 29634. [2]Dept. of Applied Economics and Statistics. 291 Barre Hall, Clemson University, Clemson, SC 29634.

The use of resistant and tolerant varieties is a desirable approach to manage the foliar nematode (*Aphelenchoides fragariae*). In order to identify tolerance and resistance in commercial hosta cultivars, reliable and efficient screening methods are required. To optimize the screening protocol, a series of greenhouse tests were done using six hosta cultivars and two types of nematode inoculum. The objectives of this study were to: (i) determine the effects of inoculum type, inoculation method, and extraction technique on evaluating *A. fragariae* resistance in hosta, and (ii) optimize the resistance screening protocol used to identify foliar nematode resistant hosta cultivars in the greenhouse.

The pathogenicity and reproduction of *A.fragariae* maintained on fungi vs. maintained on hosta was evaluated with two inoculation methods (with injury and without injury). Marked differences were found for the severity of infection and the levels of reproduction between the two types of inoculum and between the two inoculation methods. Significant differences between two extracting techniques evaluated were also observed. A numerical scale for visual evaluation of severity was developed, and recommendations for a reliable protocol for assessment of resistance and tolerance are discussed.

38. THE EFFECT OF MUSTARD SEED MEAL ON VIABILITY OF *GLOBODERA PALLIDA*. **Gao, X., W. J. Price, C. Bates, J. Worapong, B. King, J. B. Johnson, and R. S. Zemetra**. Department of Plant, Soil, and Entomological Sciences, University of Idaho, Moscow, Idaho 83843.

Pale cyst nematode (PCN), *Globodera pallida*, is a devastating plant-parasitic nematode affecting much of the potato industry worldwide. This nematode was recently detected with limited distribution in the southeastern Idaho. Biofumigation with volatiles from seed meal is widely believed to be a promising alternative to chemical fumigation of soil-borne pathogens. Little information on the efficacy of mustard (*Brassica juncea*) seed meal in suppressing *G. pallida* is known. To examine mustard seed meal as a potential biofumigant for *G. pallida*, we evaluated the impact of mustard seed meal powder on the viability and hatching of *G. pallida*. For each experiment the mustard seed meal was thoroughly mixed at various

concentrations with moist autoclaved sandy soil. Fifteen cysts were enclosed in a nylon bag and placed 2.5 cm below soil surface in a plastic cup containing 100 cm^3 of soil mixed with mustard seed meal at levels of 0, 0.5, 1, and 2 tons/hectare. Thirty milliliter of deionized water was added to each cup to approximately saturate the soil with moisture before it was covered with a lid and sealed to maximize the presence of volatiles released by the seed meal. The cysts of *G. pallida* were exposed to mustard seed meal for 1, 2, or 3 weeks. Viability of *G. pallida* was assessed by staining the eggs with 0.05% Meldona blue. Hatching percentages of *G. pallida* were determined in potato root diffusate for 5 weeks. All 3 treatments containing mustard seed meal reduced the viability and hatching of *G. pallida* eggs. Three weeks from initiation of the biofumigation, egg viability of *G. pallida* were 54.8% without seed meal and 22% - 34.1% in treatments containing seed meal. The egg hatching rate was 22.9% in the untreated check, whereas egg hatching rates in the treatments with seed meal ranged from 0.6%-3.5%. The mustard seed meal is potentially an environmentally beneficial biofumigant for suppressing *G. pallida*.

39. FIRST REPORT OF PINWORMS FROM TERMITE HINDGUTS FROM CENTRAL AMERICA. **Giblin-Davis[1], Robin M., N. Kanzaki[1,2], A. Esquivel[3], K.A. Davies[4], E.A. Herre[5], B. J. Center[1], and R.H. Scheffrahn[1]**. [1]University of Florida-IFAS, Fort Lauderdale Research and Education Center, 3205 College Ave., Davie, FL 33314, [2]Forest Pathology Laboratory, Forestry and Forest Products Research Institute, 1 Matsunosato, Tsukuba, Ibaraki 305-8687 Japan, [3]Escuela de Ciencias Agrarias, Universidad Nacional, Heredia, Costa Rica, [4]Centre for Evolutionary Biology and Biodiversity, School of Agriculture, Food and Wine, The University of Adelaide, Waite Campus, PMB 1, Glen Osmond, South Australia 5064, Australia, and [5]Smithsonian Tropical Research Institute, Apartado 2072, Balboa, Republic of Panama.

During intensive sampling of termites in North and Central America as a focal group for elucidating insect-associated nematode diversity, we found that some termite species in the New World subfamily Syntermitinae are associated at a level of near 100% with thelastomatids in their hindguts. The observed life history is suggestive of the classical symbiotic association known for most pinworms in other invertebrate groups. Unidentified thelastomatids (mostly females and juveniles) were discovered in the hindguts of the humus feeding, mandibulate nasutitermitid, *Embiratermes chagrasi*, from lowland humid rain forests in Costa Rica and Panama. Preliminary morphological work suggests that there may be more than one species associated in different parts of the hindguts of workers and soldiers. The pH of termite hindguts can be highly alkaline which suggests that these nematodes are feeding and reproducing under extreme conditions. Thelastomatids are well known associates of litter inhabiting invertebrates, such as Blattaria (cockroaches), Grylloidea (crickets), Tipulidae (crane flies), Passalidae (bessbugs), Hydrophilidae (water beetles), Lucanidae (stag beetles), Scarabaeidae (scarab beetles) and Diplopoda (millipedes). However, termites have not been previously reported as hosts. Only one male nematode was recovered after dissections of a large number of *E. chagrasi*.

40. GUAVA DECLINE: A COMPLEX DISEASE INVOLVING *MELOIDOGYNE MAYAGUENSIS* AND *FUSARIUM SOLANI*. **Gomes, V.M., R.M. Souza, V. Mussi-Dias, S.F. Silveira, and C. Dolinski.** Dept. of Entomology and Phytopathology, Universidade Estadual do Norte Fluminense Darcy Ribeiro, 28015-620, Campos dos Goytacazes, Brazil.

In Brazil, *Meloidogyne mayaguensis* has become a threat to guava production. About a third of the cultivated area is infested, leading almost inevitably to the orchards‘ decimation. Since parasitized trees present entirely rotten roots as the disease progresses, a study was set up to investigate whether a soilborne pathogen could be involved. From several nematode-free or -infested orchards in different regions in Brazil, nearly 2,000 root fragments were processed for isolation of bacteria and fungi. Positive isolations were obtained from nematode-infested areas only, predominantly of *Fusarium* sp. In a five-month microplot assay, guava seedlings remained uninoculated (as control), or were inoculated with the nematode only, or with the nematode and 21 days later with one of 11 *Fusarium* sp. isolates. A Scott-Knot analysis of several vegetative variables and of the extent of root rot allowed the generation of a dissimilarity dendogram that indicated four *Fusarium* sp. isolates particularly associated with extensive damage to the seedlings. Upon identification of these isolates as *F. solani*, a six-month microplot assay was set up, in which guava seedlings remained uninoculated or were inoculated with one of the following: i) nematode only, ii) four *F. solani* isolates, separately, iii) fungus isolates, combined with physical injury of the roots with a knife, iv) the nematode and 21 days later with the fungus isolates. No root rot and virtually no effect on all variables assessed occurred in the seedlings inoculated with the *F. solani* isolates, with or without physical injury. Major root rot and negative effect on all variables occurred in the seedlings inoculated with *M. mayaguensis* and all four *F. solani* isolates. This characterizes guava decline as a complex disease caused by the synergistic effect of these organisms, in which parasitism by the nematode predisposes the plants to root decay and death caused by the fungus.

41. PARTNERSHIP BETWEEN ENTOMOPATHOGENIC NEMATODES AND BACTERIA. **Grewal, Parwinder S., and R. An**. Department of Entomology, The Ohio State University, Wooster, Ohio 44691.

Entomopathogenic nematodes form symbioses with bacteria in the family Enterobacteriacae. *Heterorhabditis* nematodes are associated with *Photorhabdus* and *Steinernema* with *Xenorhabdus* bacteria. Nematode infective juveniles carrying the bacteria in their gut invade insect hemocoel where the bacteria are released. The bacteria multiply in hemolymph, killing the

insect through septicemia. Nematodes feed on the bacteria and insect tissues and complete 2-3 generations before forming infective juveniles colonized by the bacteria which exit the cadaver in search of a new host. This system presents unique opportunities to study the mechanisms of parasitism/virulence of the nematode-bacteria partnership and mechanisms of symbiosis between the nematode and bacteria.

Using a powerful SCOTS (selective capture of transcribed sequences) technique, we identified 40 genes in *Photorhabdus temperata* and 39 in *Xenorhabdus koppenhoeferi* which are differentially regulated during infection of the white grub *Rhizotrogus majalis*. More than 60% of the identified genes were unique to either bacterium suggesting vastly different molecular mechanisms of pathogenicity used by the two bacterial species. In *P. temperata lysR* gene encoding transcriptional activator was induced, while genes *yijC* and *rseA* encoding transcriptional repressors were induced in *X. koppenhoeferi*. Lipopolysaccharide synthesis gene *lpsE* was induced in *X. koppenhoeferi* but not in *P. temperata*. Except *tcaC* and *hemolysin* related genes, other virulence genes were different between the two bacteria. Genes involved in TCA cycle were induced in *P. temperata* whereas those involved in glyoxylate pathway were induced in *X. koppenhoeferi*, suggesting differences in metabolism between the two bacteria in the same insect host. Upregulation of genes encoding different types of nutrient uptake systems further emphasized the differences in nutritional requirements of the two bacteria. *P. temperata* displayed upregulation of genes encoding siderophore-dependent iron uptake system, but *X. koppenhoeferi* upregulated genes encoding siderophore-independent ion uptake system. *P. temperata* induced genes for amino acid acquisition but *X. koppenhoeferi* upregulated *malF* gene, encoding a maltose uptake system. Further analyses identified possible mechanistic associations between the identified gene products in metabolic pathways, providing an interactive model of pathogenesis for each bacterium species.

We have also used SCOTS technique to uncover molecular mechanisms of symbiosis between *Photorhabdus* and *Heterorhabditis*. Our analyses of differentially expressed *P. temperata* genes in *in vivo* reveal key molecular features reshaped by bacteria to persist in the enduring infective juveniles. In addition to starvation responses, the bacteria adopt major physiological shifts in the nematode infective juvenile minimizing their nutritional dependence on the nematode. We found that the bacteria induce cellular acidification via regulation of proton transport systems, switch to pentose phosphate pathway, and shed motility but form biofilm to persist in the nematode intestine. Our future research on the novel bacterial genes found to be differentially expressed in the nematode in this study may lead to the discovery of additional bacterial persistence and colonization factors. Also, the functional characterization of additional differentially regulated genes will enable further elucidation of molecular networks that allow for successful symbiotic communication between microbes and animals.

42. ARE URBAN VACANT LOTS SUITABLE FOR FOOD PRODUCTION? AN ASSESSMENT OF SOIL NEMATODE FOOD WEBS AND NUTRIENT POOLS IN COMMUNITY GARDENS AND VACANT LOTS**. Grewal, Sharanbir S., Z. Cheng, S. Masih, M. Wolboldt, N. Huda, A. Knight, and P. S. Grewal**. Center for Urban Environment and Economic Development, The Ohio State University, Wooster, Ohio 44691

In the midst of the worst economic crisis since the Great Depression, there is renewed interest in transforming vacant lots into food-producing gardens. This study analyzed whether vacant lots are suitable for food production, by comparing the soil nematode food webs and nutrient pools of vacant lots and community gardens in two post-industrial Ohio cities, Akron and Cleveland. Twelve vacant lots and 12 community gardens were examined in the two cities. All six Akron community gardens were established just prior to the initiation of this study, whereas the six in Cleveland were 15-30 years old. Each site (community garden or vacant lot) was divided into three sections and nine soil cores were collected randomly from each section. All nine cores were then combined to make a composite sample per section, resulting in a total of 72 samples. Soil pH, texture, moisture, organic matter, mineral nitrogen content, microbial biomass, and nematode communities were measured in both cities. Soil decomposition rate was also measured in Cleveland. Results show that the soils of the vacant lots were healthier than those of the young Akron gardens and equal to the soils of the old Cleveland gardens in all of the parameters measured except lower in the amounts of organic matter and nitrate-nitrogen. We conclude that baring any heavy metal contamination, the soil in vacant lots may be suitable for the establishment of food gardens, which can provide many desirable ecosystem services and enhance human well-being.

43. MOLECULAR IDENTIFICATION OF *LONGIDORUS CAMELLIAE* OCCURRING IN CHINA. **Guo[1], Kai, Angelika Matafeo[1], and Jingwu Zheng[1].** [1]Institute of Biotechnology, College of Agriculture & Biotechnology, Zhejiang University, Hangzhou 310029, P.R.China

Needle nematodes (*longidorus* spp.) are an important group of plant parasites that cause serious damage to many plant species. Some of the species in the genus are virus vectors. Act as a possible vector role, the genus has drawn much taxonomic attention. For a long period of time, species identification of *longidorus* is mainly based on morphological characters and morphometric data of females. However, the species in this genus are morphometrical similar, making identification difficult, and the molecular methods have become available to complement morphological identification. In this study, three *longidorus* populations, two collected from the rhizosphere of Chinese red pine (*Pinus massoniana Lamb*) and Chinese sweet gum (*Liquidambar formosana*), Changsha, Hunan province, China, and one from the original type locality of the *Longidorus camelliae*, Fuyang, Zhejiang province, eastern China. The morphology and morphometrics of females and

juveniles were observed and measured, respectively. The results showed that the key morphological characters of the two populations are identical to that of *L. camelliae* (Zheng *et al*, 2000). Molecular analyses of three populations were carried out by using rDNA (ITS1, ITS2, the full length of 18S gene and D2/D3 expansion segments of the 28S gene), and sequence length were 828bp, 687bp, 1778bp and 847bp, respectively. The sequence information of the two populations is highly similar to that of the type population of *L. camelliae*, ranging from 99.1% to 99.6%. These sequence data also provided evidence that two populations are identified as *L. camelliae*. Hunan is a new geographical distribution region of *L. camelliae*, and this is the first report of molecular data of ITS1, ITS2 and the full length of 18S gene of *Longidorus camelliae*.

44. A *WOLBACHIA*-LIKE ENDOSYMBIOTIC BACTERIUM IN THE PLANT-PARASITIC NEMATODE *RADOPHOLUS SIMILIS*. **Haegeman[1], Annelies, B. Vanholme[1], J. Jacob[1], T.T.M. Vandekerckhove[1], M. Claeys[2], G. Borgonie[2], and G. Gheysen[1].** [1]Department of Molecular Biotechnology, Faculty of Bioscience Engineering, Ghent University, Coupure links 653, B-9000 Ghent, Belgium, [2]Department of Biology (Nematology Unit), Faculty of Sciences, Ghent University, K.L. Ledeganckstraat 35, B-9000 Ghent, Belgium.

Wolbachia is an endosymbiotic bacterium widely present in arthropods and animal-parasitic nematodes. The bacterium was first discovered in the mosquito *Culex pipiens*, and was subsequently named *Wolbachia pipientis*. Numerous related *Wolbachia* strains have been described in the last decades. Due to the uncertainty about whether these different strains might represent different species, all strains are classified into taxonomic supergroups and are simply referred to as *Wolbachia*. In insects, these bacteria are true parasites causing reproductive manipulations, while in filarial nematodes they are rather mutualists. Despite previous efforts, *Wolbachia* has never been identified in plant-parasitic nematodes.

Random sequencing of genes expressed by the burrowing nematode *Radopholus similis*, an important nematode pest of banana, resulted in several sequences with similarity to *Wolbachia* genes. Consequently, we investigated the presence of a *Wolbachia*-like endosymbiont in this plant-parasitic nematode using both morphological and molecular approaches. Transmission electron microscopy, fluorescent immunolocalization and DAPI staining confirmed the presence of an endosymbiont within the reproductive tract of female adults. For molecular characterization, *16S rDNA*, *ftsZ* and *groEL* genes derived from the bacterium were cloned. A subsequent phylogenetic analysis showed that the endosymbiont of *R. similis* is distantly related to all currently known *Wolbachia* supergroups. Based on our phylogenetic study and the current literature we have designated the endosymbiont of *R. similis* to a new *Wolbachia* supergroup (supergroup I). Finally, based on our initial success to find sequences of this endosymbiont by screening an expressed sequence tag (EST) dataset, all nematode ESTs were mined for *Wolbachia*-like sequences. Although the retained sequences belonged to seven different nematode species, *R. similis* was the only plant-parasitic nematode with traces of *Wolbachia*. Although the role of *Wolbachia* in *R. similis* currently remains unknown, the endosymbiont was found in all individual nematodes tested, pointing towards an essential function of the bacteria.

The discovery of a *Wolbachia*-like endosymbiont in a plant-parasitic nematode sheds new light on the evolutionary history of this bacterium. Horizontal gene transfer from *Wolbachia* to animal-parasitic nematodes and insects has been proven, and it would be very interesting to find out if an ancestor of *Wolbachia* gave rise to certain genes in plant-parasitic nematodes.

45. DISTRIBUTION OF NEMATODES IN IDAHO CROPS**. Hafez, S.L.[1], P. Sundararaj[1] and Z. A. Handoo[2].** [1]University of Idaho, 29603 U of I Lane, Parma, Idaho 83660, USA. [2]USDA ARS Nematology Laboratory, Beltsville, MD 20705, USA

Ninety three species and 74 genera of nematodes have been recorded in soil samples from 31 crops in 21 counties in Idaho, USA. Among them, 40 species and 36 genera are new records in this region. The highest number of species belongs to the genus *Pratylenchus* represented by *P. agilis, P. brachyurus, P. hexincisus, P. coffeae*, *P. crenatus*, *P. neglectus*, *P. penetrans*, *P. scribneri*, *P. thornei* and *P. vulnus*. *P. neglectus* is the predominant species among all species of the identified genera. The next most predominant genus in Idaho was the ectoparasitic nematode *Helicotylenchus* represented by *H. erythrinae, H. microcephalus, H. bradys, H. crenacauda*, *H. digonicus, H. dihystera, H. platyurus* and *H. pseudorobustus*. There were seven species of *Tylenchorhynchus* comprising of *T. obscurisulcatus, T. clarus and T. silvaticus, T. annulatus, T. cylindricus,* and *T. maximus.* The pale cyst nematode *Globodera pallida* on potato was a new report for the USA. Other nematode species found in this study but their economic importance on potato is not established includes *Longidorella saadi, Mesocriconema rusticum, M. xenoplex, Hemicycliophora obtuse, Ditylenchus valveus, Aphelenchoides fragariae, Criconema mutabilis, Ditylenchus valveus, Heterotylenchus autumnalis, Nothocriconemoides lineolatus, Quinisulcius acti, Q. acutoides, Q. acutus, Q. capitatus, Subanguina balsamophila and Sulphuretylenchus elongatus.* Among the endoparasitic nematodes, the highest percent of occurrence was *Pratylenchus* (29.7) followed by *Meloidogyne* (4.4) and *Heterodera* (3.4).

46. EFFICACY OF MOVENTO COMPARED WITH OTHER NEMATICIDES FOR THE CONTROL OF *PRATYLENCHUS THORNEI* AND *MELOIDOGYNE CHITWOODI* ON POTATOES. **Hafez, S.L., P. Sundararaj and R. Portenier**. University of Idaho, Parma Research and Extension Center, 29603 U of I Ln, Parma, Idaho 83660, USA.

Two field experiments were conducted for the management of lesion and root knot nematodes on potato. In the first experiment efficacy of Movento was compared with Vapam alone or with AdSorb or Temik alone for control of *Pratylenchus*

thornei in potato. Yield of tubers from different treatments indicated that there was an increase in saleable and total yield in different combinations of all treatments compared to the control plots. Maximum saleable and total yield compared to the untreated control was with Vapam treatments followed by Admire+Temik+Movento combination. In general, treatment combinations performed better than the individual treatments. In the second experiment, the efficacy of Movento was tested in combination with Admire Pro for the management of *Meloidogyne chitwoodi*. Yield from different treatments indicated that there was an increase in total yield in different combinations of all treatments as compared to the control plots. Percent of nematode infected tubers in treated plots ranged from 6.6 to 45.7. Lowest level of nematode infection was recorded in the Vapam applied plots followed by Temik in combination with Mocap or Movento applied plots.

47. EFFICACY OF NEMATICIDES IN THE MANAGEMENT OF *MELOIDOGYNE CHITWOODI* ON IDAHO POTATOES. **Hafez, S.L., P. Sundararaj and R. Portenier**. University of Idaho, Parma Research and Extension Center, 29603 U of I Ln, Parma, Idaho 83660, USA.

Two field experiments were conducted to study the efficacy of different formulations of fosthiazate (Experiment-I) or Telone II or Vapam HL or Mocap EC or Vydate C-LV alone or Telone II in combination with Vapam HL (Experiment-II) for control of Columbia root-knot nematode in potato. In the first experiment percent of yield infected with nematodes in treated plots ranged from 8.6 to 28.5. The lowest level of nematode infection was observed in Fosthiozate 10 G 4.5 lbs ai/A applied plots. However, there was no significant increase in total yield in treated plots compared to control plots. This study indicated that the granular formulation of Fosthiozate is more effective in controlling the *M.chitwoodi* than the liquid formulation. In the second experiment, yield of tubers indicated that there was an increase in total yield in all treatments compared to the control plots. Percent nematode infected tubers were significantly reduced by the treatments as compared to control plots. Percent of tubers with nematode infection in treated plots ranged from 1.2 to 38.3. The lowest level of nematode infection was recorded in the Telone 15 g/A + Vapam 30 gal/A.

48. EVALUATION OF FUMIGANT AND NON-FUMIGANT NEMATICIDES IN THE MANAGEMENT OF *MELOIDOGYNE CHITWOODI* ON POTATO**. Hafez, S.L., P. Sundararaj and R. Portenier**. University of Idaho, Parma Research and Extension Center, 29603 U of I Ln, Parma, Idaho 83660, USA.

Three field experiments were conducted at the University of Idaho, Parma Research and Extension Center, Parma, Idaho to demonstrate the effects of Telone II alone and in combination with Vapam HL on *Meloidogyne chitwoodi* management and yield of potato. Yield of tubers from different treatments indicated that there was an increase in total yield and marketable yield in different combinations of all treatments compared to untreated control plots. Nematode infected tubers as well as percent of nematode infection were also significantly reduced by the treatments compared to untreated control plots. Percent of tubers with nematode infection in treated plots ranged from 0.9 to 37.0. Lowest level of nematode infection was recorded in the Telone II 15 gal/A + Vapam HL 30 gal/A, with the maximum marketable yield than other treatments.

49. *Withdrawn* LATEST IN APPLIED ENTOMOPATHOGENIC NEMATOLOGY IN CHINA. **Han, Richou** Guangdong Entomological Institute, 105 Xingang Road West, Guangzhou 510260, China. Email: richou-han@163.net

Entomopathogenic nematodes (EPNs) have been commercialized in China, and ideal pest targets for these nematodes were evaluated. Apart from the important insect pests (such as flee beetle *Phyllotreta striolata*, and chive midge *Bradysia odoriphaga*) in vegetables, new invasive pests such as the oriental fruit fly *Bactrocera dorsalis* (Hendel), asiatic palm weevil *Rhabdoscelus lineaticollis* (Heller) and banana moth *Opogona saccchari* (Bojer) were also controlled by EPNs. Oriental fruit fly attacks many tropical fruits. Asiatic palm weevil is an important pest of palm plants and sugarcane. Banana moth damages various ornamentals and economic crops such as banana, sugarcane and maize. Four species or strains of entomopathogenic nematodes, *S. carpocapsae* All, *S. carpocapsae* A24, *S. feltiae* SN and *H. bacteriophora* H06 were used to control the oriental fruit fly in the laboratory and in the field. *S. carpocapsae* All showed the best control of this fly and 86.3% larval mortality was obtained after 9 days with 300 infective juveniles (IJs) per cm^2 in the soil. A mixture of chlorpyrifos EC, imidacloprid, and *S. carpocapsae* All (4000 IJs/ml) was used for the control of *R. lineaticollis* on palm, and after 7days mortality of the weevil larvae in the combined treatment (98.0%) was significant higher than mortality in single treatments of chlorpyrifos (69.0%), imidacloprid (0%) or *S. carpocapsae* All (68.4-78.6%). *S. carpocapsae* A24 at a dose of 2857 IJs/ml caused 90.9% larval mortality of the banana moth on an ornamental *Dracaena fragrans*. *H. indica* LN2 also gave 88.2% mortality of the fourth instar chive midge *Bradysia odoriphaga*, which destroys Chinese chive, with 400 IJs per larva. These results showed high potential for the control of new target insects by entomopathogenic nematodes.

50. FIRST REPORT OF THE CYST NEMATODE *PUNCTODERA MATADORENSIS* IN THE UNITED STATES. **Handoo[1], Zafar A., A.M. Skantar[1], D.J. Chitwood[1], and L.K. Carta[1].** [1]Nematology Laboratory, USDA-ARS, Beltsville Agricultural Research Center, Beltsville, MD 20705.

In January 2010, a cyst nematode was discovered from soil samples collected in a field near Cavalier (Pembina County), North Dakota. The nematode was found during a survey conducted jointly by the North Dakota State Department of

Agriculture and the USDA Animal and Plant Health Inspection Service through the Cooperative Agricultural Pest Survey program. The morphology of cysts and second-stage juveniles and molecular analysis of juveniles established the identity of the species as *Punctodera matadorensis*. Morphological characters used for identification included cyst shape, characteristics of cyst terminal cone including nature of fenestration, presence of prominent bullae, cyst wall pattern, and the anal-vulval fenestral diameter and distance. The second-stage juvenile morphological characters critical for identification were body and stylet length, shape of stylet knobs, shape and length of tail and hyaline tail terminus, and the number of lines in the lateral field. Measurements of second-stage juveniles included length of body, stylet with prominent forwardly directed to strongly concave knobs, tail, and hyaline tail terminus. The lateral field had four distinct lines. Shapes of tail, tail terminus and stylet knobs were also consistent with *P. matadorensis*. The cysts were light brown in color, ovoid or spherical in shape with small protruding necks. Both vulval and anal areas were marked by circular fenestrae of nearly identical sizes. Several heavy bullae were located between vulval and anal fenestrae or occasionally scattered around fenestral areas. The cyst wall was heavily punctated. Molecular analysis of the population included amplification of the ribosomal internal transcribed spacer (ITS-rDNA), partial 18S small subunit rDNA, 28S large subunit D2-D3 expansion segments, and Hsp90. The ITS-rDNA was also digested with a panel of restriction enzymes in order to identify unique patterns for this species. Detection of *P. matadorensis* in North Dakota represents a new record of this species in the United States.

51. EFFECTS OF SOIL TYPE, IRRIGATION AND *ROTYLENCHULUS RENIFORMIS* ON COTTON YIELD. **Herring, Stephanie L., and Koenning, S. R.** Dept. of Plant Pathology Campus Box 7616, North Carolina State University, Raleigh, NC 27695-7616.

The effects of soil type, irrigation, and population density of *Rotylenchulus reniformis* on cotton were evaluated in a microplot experiment in 2008 and 2009. Six soil types, Fuquay sand (91% sand, 6% silt, 3% clay, 0.6% organic matter), Norfolk sandy loam (84% sand, 12% silt, 4% clay, 1.4% organic matter), Portsmouth loamy sand (72% sand, 18% silt, 10% clay, 3.8% organic matter), Muck (58% sand, 33% silt, 9% clay, >30% organic matter), Ceil sandy loam (53% sand, 18% silt, 29% clay, 2.2% organic matter), and Ceil sandy clay (48% sand, 13% silt, 39% clay, 0.9% organic matter), were arranged in randomized complete blocks with five replications and at least two treatment duplicates per replicates for a total of 240 plots. Irrigation effects on cotton earliness were measured by conducting four separate manual harvests of the crop in 2008 and three separate harvests in 2009. These harvests were combined and ginned to obtain a measure of overall lint yield per plot. Final population (Pf) densities of *R. reniformis* were greatest in the Portsmouth loamy sand. However, in spite of high *R. reniformis* population densities, crops planted in this soil did not have a significant difference in lint yield from a Muck soil that averaged the greatest lint yield per plot. Irrigation did not appear to affect average lint yield for a Cecil sandy clay (P = 0.20), Cecil sandy loam (P = 0.45) or Fuquay sand (P = 0.36). However it did affect yield in Muck (P = 0.0001), Norfolk sandy loam (P = 0.0001) and Portsmouth loamy sandy where it was shown to interact with initial nematode population (Pi) (P = 0.01). The only soil that showed no relationship between yield and Pi was Cecil sandy loam (P = 0.47). Although addition of irrigation did increase yield, the effect on earliness would likely offset any benefits seen from this increase in commercial operation. The Portsmouth sandy loam was one of the greatest yielding soils, had the greatest nematode populations and was the only soil in which an irrigation × Pi interaction occurred. The very great productivity of the Portsmouth loamy sand is likely the reason the Pi × irrigation interaction was only observed in this system. The Fuquay sand was expected to show no effect of irrigation due to its poor moisture retention properties. The opposite may be responsible for the lack of significant differences in irrigated and non-irrigated crops in Cecil sandy loam and Cecil sandy clay; the good soil moisture retention properties of these soils allowed for similar growth in both systems. However, the over all poor productivity of the Cecil sandy loam and Cecil sandy clay soils likely accounts for these results as well.

52. RATE OF GERMINATION AND GROWTH OF *IN VITRO* PRODUCED *PASTEURIA* SPP. PARASITIZING *ROTYLENCHULUS RENIFORMIS*. **Hewlett[1], Thomas E., S. R. Stetina[2], L. M. Schmidt[1],** [1]Pasteuria Bioscience, 12085 Research Dr., Alachua, FL 32615. [2]USDA ARS, 141 Experiment Station Rd., Stoneville MS 38776.

Reniform nematodes (*Rotylenchulus reniformis*) from pot culture were attached with *in vitro* produced *Pasteuria* spp. spores using a centrifuge attachment technique that resulted in 40-50% of the vermiform nematodes with spores adhering to their cuticles. Attached nematodes were placed into small plastic cups filled with 100 cm^3 of an autoclaved mixture of 3 parts sand and 1 part sandy loam soil. Cups, established at two-day intervals for 25 days, were placed in a growth chamber at 28 C with or without a single cotton (*Gossypium hirsutum* cv. Deltapine 444 BG RR) plant. At the end of the experiment, ten nematodes with *Pasteuria* spp. disease symptoms were selected from cups representing each time interval for observation. Nematodes were crushed and observed at 600X for the presence of bacterial cells and sporulating structures. *Pasteuria* spp. germination occurred rapidly after attachment on day 1 with and without cotton plants. Mycelial structures and thalli were present by day 3 in soil with cotton plants and by day 7 in soil alone. Mature *Pasteuria* endospores were formed by day 15 in soil with cotton plants and by day 23 in soil alone. The rate of growth of the *Pasteuria* spp. parasitizing *R. reniformis* (208 degree days) in soil with cotton plants is much more rapid than the growth rate of *P. penetrans* in *Meloidogyne* spp. (408

degree days). *Pasteuria* spp. were observed to infect and complete their life-cycle in juvenile, male and female reniform nematodes.

53. USE OF REMOTE SENSING TO DETECT *HETERODERA SCHACHTII* AND *RHIZOCTONIA SOLANI* INDUCED PLANT STRESS IN SUGAR BEET FIELDS. **Hillnhütter, Christian[1], A.-K. Mahlein[1], T. Mewes[2], R.A. Sikora[1] and E.-C. Oerke[1].** [1]Institute of Crop Science and Resource Conservation (INRES) – Phytomedicine, University of Bonn, Nussallee 9, 53115 Bonn, Germany, [2]Center for Remote Sensing of Land Surfaces (ZFL), University of Bonn, Walter-Flex-Strasse 3, 53113 Bonn, Germany.

The characteristically clustered occurrence and low level of mobility of *Heterodera schachtii* and *Rhizoctonia solani* in the soil and the induction of symptoms in the sugar beet canopy make them perfect targets for precision agriculture tools. Remote sensing in combination with geographic information systems allows instant detection and generation of digital maps that clearly represent the heterogeneous distribution of soil-borne nematodes and pathogens. The objective was to analyze the spatial distribution of *H. schachtii* and *R. solani* and to monitor symptom development of the organisms alone or in combination during the entire growing season. A classification of leaf symptoms induced by the organisms should be obtained by image processing with the computer program ENVI. Therefore, two field experiments were conducted in 2009 with handheld and aerial sensors. Field sites inoculated with *R. solani* and *H. schachtii* and field sites with natural infestations of the organisms were investigated. At mid growing season and at the end of the growing season, two flight campaigns with imaging hyperspectral sensors were conducted. Parallel to the flight campaigns ground truth reflectance was measured with non-imaging spectroradiometers. Furthermore, at several sample points ground truth data, in particular incidence and severity of the diseases were collected and geo-referenced. Canopy reflectance measurements obtained from the flight campaigns and the hand held spectroradiometers clearly discriminated symptoms caused by *H. schachtii* or *R. solani*. By image processing methods (NDVI transformation, supervised classification and change detection) leaf symptoms caused by the organisms were classified. Digital maps containing the spatial distribution, the disease progress, as well as disease severity of the nematode and the pathogen will be presented. The results generated in this study, demonstrated that remote sensing in combination with geographic information system technologies can be used as a tool for the detection of symptoms caused by *H. schachtii* and *R. solani*.

54. LEARNING TO RAISE THE ENTOMOPATHOGENIC NEMATODE *HETERORHABDITIS BACTERIOPHORA* IN SUBMERGED CULTURE. **Holmes[1], Len D. and F.L. Inman III[1].** [1]Department of Chemistry and Physics, The UNCP Biotechnology Research Center, The University of North Carolina at Pembroke, Pembroke, NC 28372.

Heterorhabditis bacteriophora is valuable as a biological agent to control the damage done to crops and turf by the larval stage of insects in the orders *Coleoptera* (beetles) and *Lepidoptera* (moths and butterflies). Submerged nematode cultures in bioreactors will yield sufficient quantities for direct application onto plants threatened by insect pests. *H. bacteriophora* must be grown with its obligate strain-specific symbiont bacteria, *Photorhabdus luminescens*. Culture conditions which promote nematode reproduction, development and survival are not obvious to the new investigator. Although *H. bacteriophora* and other entomopathogenic nematodes have been commercially available for almost two decades, companies keep the details of the process guarded as trade secrets. This presentation will outline the results of our laboratory to discover how nematodes may be grown in suspension culture.

55. EVALUATION OF SOIL INCORPORATION METHODS OF FOSTHIAZATE FOR CONTROL OF *MELOIDOGYNE CHITWOODI* DAMAGE TO POTATO TUBERS. **Ingham[1], Russell, E., B.A. Charlton[2], N.M. Wade[1], and D. Culp[2].** [1]Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR; [2]Klamath Basin Research and Extension Center, Oregon State University, Klamath Falls, OR

Fosthiazate has provided good control of Columbia root-knot nematode (*Meloidogyne chitwoodi*, CRKN) damage to potatoes in experimental trials. This product has typically been applied by broadcasting material on the surface and incorporating it into the soil with a rototiller prior to planting. Concern for worker and environmental exposure between application and incorporation questioned if the product would be efficacious if injected below the soil surface. An experimental trial was conducted in Klamath Falls, OR during 2007 in which 5 kg a.i/ha of fosthiazate 900 EC was applied May 17 and 18 by 1) spraying on the surface and incorporating to a depth of 15 cm with a rototiller, 2) spraying in front of the tines of a rototiller and incorporating to a depth of 15 cm, 3) injecting at depths of 10 and 20 cm from shanks spaced 18 cm apart, 4) treatment 3 followed by rototilling, and 5) injecting at depths of 10, 20, and 30 cm from shanks spaced 18 cm apart. An untreated control was included and treatments were arranged in a randomized block design with five replications of plots 3.7 m wide by 9.1 m long. Potato cv Russet Norkotah was planted May 22 and harvested October 9. A random sample of 25 tubers collected from the center 4.5 m of each plot was evaluated by examining the surface for galling and by peeling tubers and counting CRKN infection sites. External culls were defined as tubers with galls on 10% or more of the surface and internal culls as tubers with six or more infection sites. Infection pressure was high with 59% external culls and 56% internal culls in the untreated control. Nematode control with fosthiazate was excellent when broadcast and rototilled (0% external

and internal culls) or sprayed directly in front of the tines of the rototiller (1% external and internal culls). In contrast, the percentage of external and internal culls in the injection treatments was not significantly less than in the control. Percent external and internal culls were 35% and 44% in treatment 3, 34% and 33% in treatment 4, and 75% and 79% in treatment 5, respectively. These results suggest that fosthiazate does not move from where it is applied and must be thoroughly mixed in the soil to assure contact with target nematodes. Injection treatments presumably produced narrow zones with a high concentration of fosthiazate leaving most of the soil untreated and allowed many nematodes to escape contact with the nematicide. The failure of the rototilling after injection suggests that fosthiazate was mixed vertically and not horizontally which would have left areas of soil untreated. Spraying fosthiazate in front of the tines of a rototiller would be the optimal method for immediate incorporation.

56. PATHOGENICITY OF RING NEMATODES: AN EMERGING PEST IN BLUEBERRIES (*VACCINIUM* SPP.). **Jagdale[1], Ganpati B., P. M. Brannen[1], J. P. Noe[1], B. Cline[2] and A. P. Nyczepir[3]** [1]Department of Plant Pathology, University of Georgia, Athens, GA, 30602, [2]Department of Plant Pathology, North Carolina State University, Horticultural Research Station, Castle Hayne, NC 28429 and [3]USDA-ARS Southeastern Fruit & Tree Nut Res. Lab. 21 Dunbar Road, Byron, GA 31008.

Blueberries, *Vaccinium* spp., are grown in more than 30 states on over 16,400 hectares in the United States. The blueberry industry in Georgia continues to grow rapidly, with substantial acreage increases on a yearly basis. Several plant-parasitic nematode species, including stubby root (*Paratricodorus* sp), spiral (*Helicotylenchus* sp), dagger (*Xiphinema* sp) and ring *Mesocriconema ornata* nematodes, have been reported to be associated with three types of commercially grown blueberries in the United States. However, there is no data available on the effects of plant-parasitic nematodes on the growth and yield of blueberry plants. Therefore, we initiated an experiment in August 2009 to test the pathogenicity of *M. ornata* on Rabbiteye blueberry (*Vaccinium ashei* Reade) variety ''Alapaha'' under greenhouse conditions in Athens, GA and under field microplot conditions in Byron, GA. Single one-year-old blueberry plants were transplanted into plastic pots (surface area 346 cm^2) containing10 kg autoclaved sandy loam soil (pH $\sim$ 5.0). Eight weeks after transplanting, plants were inoculated with five different nematode inoculum levels (treatments) including 0 (water control), 10, 100, 1000 and 10,000 mixed stages of ring nematodes per plant. The nematode population in each pot was assessed 75 and 150 days after inoculation by randomly removing four soil cores (2.5 cm diam. x 10 cm deep) from the root area around each plant. Individual soil cores were combined into a composite sample and nematodes were extracted from 100-g soil sub-samples. Nematode population densities were expressed as numbers of nematodes/pot and their reproduction rates (Pf/Pi) were then calculated by dividing the total number of nematodes per pot (Pf = final population) by the number of nematodes added (Pi = initial population). We found that the nematode populations numerically increased in all the treatments but the rate of their reproduction was greatest in the treatments receiving the lowest initial population level (10 nematodes per plant) 75 and 150 days after inoculation at both locations suggesting that the ring nematode, *M. ornata* is a likely pathogen to blueberry plants. Thus, these preliminary findings demonstrate for the first time that ring nematode, *M. ornata*, is a potential pest of blueberry in Georgia.

57. WHOLE-GENOME SEQUENCING OF BURSAPHELENCHUS XYLOPHILUS AND B. MUCRONATUS AND THEIR COMPARATIVE GENOMICS ANALYSIS. **Jian, Heng,** Department of Plant Pathology, China Agricultural University, Beijing 100193, China, Institute of Vegetables and Flowers, CAAS, Beijing 100081, China, Institute of Plant Protection, CAAS, Beijing 100081, China.

Bursaphelenchus xylophilus, the casual agent of pine wilt disease, lead to a devastating disease of pine forests in Eastern Asia. The nematode has spread over 193 counties in fifteen provinces in China, and about more than 500 million pine trees died already. Recently, this nematode has also been introduced in Europe and southern hemisphere. However, the pathogenicity mechanism of B. xylophilus (B. x) was not clear even now. There were rare effective approaches to control this nematode. The genomic sequencing and assembling of B. xylophilus and its closely related species B. mucronatus (B. m) have been accomplished now. The sequencing depths achieved 101×(B. x) and 156.8×(B. m) by Solexa, and probably represents >95% of the gene region for each nematode. The predicted genomes sizes were about 89.7Mb(B. x) and 77Mb(B. m), the scaffolds N50 values were more than 300Kb and the GC contents were about 40%, respectively. We've also finished the basic annotations. And the protein coding genes were preliminary predicted to be 15,971 and 15,390, respectively. Right now, further annotations, including functional genomics analyses and comparative genomics analysis are underway. We expect the results will help us to understand the molecular pathogenicity mechanisms of B. xylophilus more depth.

58. WHERE'S THE ECOLOGY IN MOLECULAR ECOLOGY? **Johnson, Jerald B., S. M. Peat, and B. J. Adams.** Department of Biology, and Evolutionary Ecology Laboratories, Brigham Young University, Provo, Utah, 84602

Molecular techniques have had a profound impact in biology. Major disciplines, including evolutionary biology, now consistently utilize molecular tools. In contrast, molecular techniques have had a more limited impact in ecology. This discrepancy is surprising. To explore this further we describe the unexpected paucity of ecological research in the field

colloquially referred to as 'molecular ecology.' Publications over the past 15 years from the journals *Ecology*, *Evolution*, and *Molecular Ecology* reveal that much of the research published under the molecular ecology banner is in fact evolutionary in nature, and that comparatively little ecological research incorporates molecular tools. This failure to more broadly utilize molecular techniques in ecology is alarming because several promising lines of ecological inquiry could benefit from molecular approaches. In this presentation I will briefly summarize our analysis of the use of molecular tools in ecology and evolution, and suggest several ways to renew the ecological focus in 'molecular ecology'.

59. REPRODUCTION OF *PRATYLENCUS PENETRANS* IN SOILS WITH AND WITHOUT *PASTEURIA* SPP. **King, Tiara N. and James B. Kotcon**. Division of Plant and Soil Sciences, West Virginia University, P. O. Box 6108, Morgantown, WV 26506.

Pasteuria species are obligate bacterial parasites of plant-parasitic nematodes. Their distribution in the mid-Atlantic region, their occurrence on *Pratylenchus* spp., and their efficacy as a biocontrol agent for *Pratylenchus* spp are unknown. Certified organic agriculture rules prohibit the use of most chemical nematicides, thus biocontrol agents such as *Pasteuria* could assist growers in managing nematode problems. The occurrence of lesion nematode and incidence of *Pasteuria* attachment to *Pratylenchus* were determined in soil samples (~20 kg) collected from 34 fields managed by certified organic growers in the Mid-Atlantic region. No *Pratylenchus* were detected in 11 soils. *Pasteuria* spp. were observed attached to *Pratylenchus* spp. in 6 of the 23 samples with *Pratylenchus*. To assess the effect of *Pasteuria* on reproduction of *Pratylenchus penetrans*, a bench-top laboratory experiment was performed in which soils from six fields with *Pasteuria* (Pasteuria-Infested Soils) were compared to soils from six fields with a similar soil type, but without detectable levels of *Pasteuria*. (Uninfested Soils) All soils were gently steamed to 66±16 C (atmospheric pressure) for 19 hours to reduce the population density of indigenous *Pratylenchus* spp. Soils were placed in 1-L pots and half of the pots from each grower were inoculated with 277 ± 11 *Pratylenchus penetrans* from root explant cultures (Isolate NL-10 kindly provided by Lynn Carta at USDA Beltsville), and a 5-day-old seedling of *Phaseolus vulgaris* cv Jade was transplanted into each pot. Three replicate pots of each soil/treatment combination were harvested after 2 and 8 weeks. Data on number of *Pratylenchus* recovered were analyzed with a three-way ANOVA (*Pasteuria*-Infested vs. Uninfested Soils, With vs Without *Pratylenchus* inoculum, at 2 versus 8 weeks). Few *Pratylenchus* were recovered from pots that were not inoculated, demonstrating that the steam treatment was effective in reducing the population density of indigenous lesion nematodes, thus differences in number of *Pratylenchus* recovered were attributed to effects of treatments on the *Pratylenchus penetrans* inoculum added. Population density of *Pratylenchus* was slightly lower in pots containing *Pasteuria*-Infested Soil (22 *Pratylenchus* per pot), than in Uninfested Soil (29 *Pratylenchus* per pot), however differences were not statistically significant ($P = 0.11$). Population densities of *Pratylenchus* were significantly lower at 8 weeks than 2 weeks after inoculation, suggesting that reproduction was low and survival was poor, regardless of treatment. Up to 10 nematodes per pot were examined for *Pasteuria* spore attachment, however incidence was low, an average of less than one nematode with spores attached was detected per pot, and only from *Pasteuria*-Infested Soils. No spores were observed on nematodes from Uninfested soils. These results suggest that naturally occurring population densities of *Pasteuria* may need to be augmented to provide effective control of lesion nematodes.

60. DELIVERING NEMATOLOGY COURSES TO TRADITIONAL AND NON-TRADITIONAL STUDENTS AT THE UNIVERSITY OF ARKANSAS **Kirkpatrick, T.L.[1], R.T. Robbins[2], R.W. Cartwright[2], R.J. Bateman[1], and J.C. Robinson.[2]** [1]University of Arkansas, SWREC, Hope, AR 71801 and [2]Department of Plant Pathology, University of Arkansas, Fayetteville, AR 71801.

The University of Arkansas Department of Plant Pathology has offered a three credit-hour nematology course (Plant Nematology) to both traditional students on the main campus and non-traditional students statewide since 2002. Lectures in this graduate level course are delivered via compressed video with instructors located at both the UA main campus and at the University of Arkansas Community College at Hope, AR. This method of instruction involves recording lectures and transmitting them through high-speed telecommunications cables to remote sites. The process allows ''real-time'' interaction between instructors and students at the remote sites who are viewing the same course. Students participate in lectures and class discussions in two weekly lecture sessions each lasting 50 minutes. Accompanying laboratory instruction/experience is available for main campus students in the plant pathology departmental teaching laboratory. Off-campus students complete their laboratory activities in all-day intensive laboratory sessions that are offered on two Saturdays during the semester at the Arkansas Cooperative Extension Service Lonoke Agricultural Center in central Arkansas. In addition to our traditional nematology course, we also offer an entirely web-based course, Applied Plant Disease Management, which was taught for the first time in 2008. This course includes considerable emphasis in nematology across several crops via real-time voice lecture sessions using the interactive web conferencing program Elluminate Live©. Course assignments, homework, exams, etc. are accomplished using the Blackboard™ e-Education platform software. Laboratories (required) for all students are completed by participation in four all-day, weekend sessions that include formal laboratory instruction at the Lonoke Agricultural Center and field trips in the central Arkansas area. To date, 55% of the students enrolled in these two courses

have been traditional degree-seeking graduate students in plant pathology, entomology, horticulture, and crop science. The remaining 45% were cooperative extension agents, USDA employees, crop consultants, and agricultural industry employees who were enrolled in the College of Agricultural, Food, and Life Sciences non-thesis Master's degree program.

61. AVICTA® COMPLETE CORN – A COMPREHENSIVE, EFFECTIVE CORN SEED TREATMENT PROGRAM, **Klix, Melanie, Palle Pedersen, David long, Clifford Watrin,** Syngenta.
Nematodes cause severe damage by feeding in or on plant roots, allowing fungi and bacteria to enter and weaken the plant. Damage includes stunting, chlorosis and yield loss.

Most corn nematode species can maintain their populations when soybeans are grown, but repeated cropping of corn can cause a populations increase. Also, use of transgenic, insect-resistant corn hybrids for corn rootworm control may reduce the amount of soil-applied insecticide used in the state. Some have speculated that these insecticides may have provided some suppression of plant-parasitic nematode populations, and reduction in use of soil insecticides also may lead to increases in corn nematode population densities. Finally, the increase in no-tillage and conservation tillage practices may have led to a change where corn nematodes have become an increased threat.

Avicta® Complete Corn nematicide/insecticide/fungicide seed treatment combination offers growers a comprehensive, effective seed treatment program to protect against a wide spectrum of nematodes, insects and diseases to increase plant health and optimize yield and profit potential. As a combination of Avicta seed treatment nematicide, Cruiser® seed treatment insecticide and Apron XL®, Maxim® XL and Dynasty® seed treatment fungicides, Avicta® Complete Corn is innovative and convenient, helping promote plant health and increase yield potential.

In 2007, Syngenta Seedcare initiated an extensive testing of the yield benefit of Avicta® Complete Corn across the Midwest and has found across 3 years of research that growers can expect a 6 bushel per acre average yield increase over Cruiser Extreme 250® insecticide/fungicide on 85 percent of the corn acres planted in the US over time. In a normal season, where crops are subjected to higher temperatures, rainfall and pest pressures that stress the crop Avicta® Complete Corn provides more of a yield increase than in seasons with less stressful growing conditions. Overall applying Avicta® Complete Corn each season provides reliable protection and allows plants to thrive despite adverse growing conditions.

62. ROTATION AND GREEN MANURE CROPS FOR MANAGEMENT OF LESION AND DAGGER NEMATODES. **LaMondia[1], James A. and J. M. Halbrendt[2].** [1]The Connecticut Agricultural Experiment Station, Valley Laboratory, 153 Cook Hill Rd. Windsor CT 06095 and [2]The Pennsylvania State University, Department of Plant Pathology, Fruit Research and Extension Center, Biglerville, PA 17307.

Annual rotation crops of grain pearl millet (*Pennisetum glaucum*) cv. Tifgrain 102, rapeseed (*Brassica napus*) cv. Dwarf Essex, buckwheat (*Fagopyrum* spp.), *Camelina sativa*, *Rudbeckia hirta*, and *Sesame indica* were evaluated as rotation or green manure crops for suppression of dagger (*Xiphinema americanum*) and lesion (*Pratylenchus penetrans*) nematodes in orchard rotation plots in Connecticut and Pennsylvania. Field plots (3 m × 3 m) were established in both states with four replications per treatment. All crops were planted in June 2008. Camelina, rapeseed, buckwheat, grain millet and sesame were planted at 10, 10, 56, 12 and 12 kg seed per hectare, respectively, and *Rudbeckia hirta* plots were established using 5-week-old transplants arranged in five rows of six plants each. Nematodes were counted from soil samples taken pre-plant, mid-season and two weeks after incorporating the crops as green manure in August. Lesion nematodes were also extracted from cover crop roots mid-season in Connecticut. Following the green manure treatment, all plots were planted with cereal rye (*Secale cereale*) as a lesion nematode bait plant in September. Nematodes were extracted from soil samples taken in October in Pennsylvania and from rye roots collected in early December in Connecticut. There were no differences in preplant nematode densities in soil (lesion and dagger nematodes ranged from 0 to 8 and 0 to 61 in Connecticut and 0 to 17 and 0 to 37 in Pennsylvania per 100 cm^3 soil, respectively). The numbers of *Pratylenchus* recovered from rotation crop roots sampled just prior to crop incorporation were also not different. Lesion and dagger nematode population levels in soil sampled prior to crop incorporation did not differ between rotation crops. *Xiphinema* numbers in soil were lowest after incorporation of the Brassica crops *Camelina* and Dwarf Essex. Buckwheat, pearl millet and sesame appeared to be good hosts and *R. hirta* was a moderate host for dagger nematodes. *Pratylenchus* populations recovered from rye roots following sesame were higher than from all other rotation crops. Few lesion nematodes were detected in soil and none in the rye roots following *R. hirta* incorporation. Rotation crops may effectively reduce plant parasitic nematode populations, but can have different impacts against different nematodes.

63. POPULATION VARIABILITY OF *ROTYLENCHULUS RENIFORMIS* IN COTTON AGROECOSYSTEMS. **Leach, Megan[1], P. Agudelo[1], and A. Lawton-Rauh[2].** [1]Dept. of Entomology, Soils and Plant Sciences 114 Long Hall, Clemson University, Clemson, SC 29634, [2]Dept. of Genetics and Biochemistry 100 Jordan Hall, Clemson University, Clemson, SC 29634.

Cotton crop specialists and nematologists report an increase in the incidence and prevalence of reniform nematode (*Rotylenchulus reniformis*) in the United States over the last two decades. It is unknown whether the observed increase in

importance is related to the emergence of novel populations that are more aggressive or have a higher fitness. Understanding the adaptive potential of this species is crucial for the development of durable management strategies. The objectives of this research were to determine the genetic diversity of *R. reniformis* populations representing cotton-growing areas in the United States and to study the effect of temperature and plant host on these populations of the nematode. We developed microsatellite markers to determine the current level of genetic diversity among individuals and populations and how this diversity is distributed in the Southeast. The identified loci exhibited polymorphisms among individuals and populations. We used AFLPs (Amplified Fragment Length Polymorphisms) to determine changes in population structure due to rotations, by subjecting a field population to simulated crop rotations in a greenhouse. The six rotation schemes included susceptible cotton and soybean, resistant soybean, and non-host corn during four planting cycles. Genotype frequencies and population structure were estimated after each cropping cycle. The results suggested that crop rotation schemes impact reniform nematode population structure, with the most distinct population differentiation occurring after corn. The effect of temperature on the fitness of the nematode and how this could influence geographic distribution was studied by measuring the time necessary for completion of embryogenesis in three populations at four temperatures. The time from a one-celled egg to eclosion was measured at 20, 25, 30, and 35°C, for populations isolated from cotton fields in Alabama, Mississippi, and South Carolina. Differences in the temperature requirements for embryonic development were observed. Differences in the calculated theoretical temperature optima and ranges indicate that these populations may respond differently to variations in temperature. Reniform nematode may have the ability to increase its distribution range through variants able to reproduce in a wider temperature range, allowing for increased numbers in new areas.

64. CLONING AND CHARACTERIZATION OF A VENOM ALLERGEN-LIKE PROTEIN GENE CLUSTER FROM THE PINEWOOD NEMATODE BURSAPHELENCHUS XYLOPHILUS. **Lin, Shifeng, Heng Jian*, Haijuan Zhao, Dan Yang, Qian Liu.** Department of Plant Pathology, China Agricultural University, Beijing 100193, China **Corresponding author*.

Pinewood nematode (PWN) is the causal agent of the pine wilt disease. Previous studies have suggested that secretions from the esophageal glands of PWN play an important role in pathogenicity. A cluster of three venom allergen-like protein genes and one pseudogene, Bx-vap-1, Bx-vap-2, Bx-vap-3 and Bx-vap-P, were identified within a 3.7-kb region. Genes vap-1, -2 and -3 are functional and encode three major allelic variants of PWN venom allergen-like proteins. But Bx-vap-P is an untranscribed pseudogene. Genes vap-1, -2 and -3 produce predicted products of 204, 206 and 203 amino acid residues, respectively, including the putative signal peptide sequence at the amino termini. In situ mRNA hybridization analysis showed that the transcripts of genes vap-1, -2 and -3 accumulated exclusively within the oesophageal gland cells of B. xylophilus. Additionally, three modification, transport and regulatory protein genes were also detected in the same flanking region of the Bx-vap gene cluster.

65. IDENTIFICATION OF A *MELOIDOGYNE INCOGNITA*-SUPPRESSIVE SOIL AND ITS POTENTIALLY CAUSAL AGENT. **Loffredo, Angelo[1], J. Yang[2], J. Borneman[2], and J.O. Becker[1].** Departments of [1]Nematology, [2]Microbiology and Plant Pathology, University of California, Riverside, CA 92521.

Dwarf tomato (*Solanum lycopersicum* cv. Tiny Tim) and short-straw wheat (*Triticum aestivum* cv. Yecora Rojo) were grown in soil tubes to screen soil samples from various California fields for suppression of root-knot nematode populations (rkn, *Meloidogyne incognita*). Each test soil was divided into a pasteurized and a non-treated sample. All samples were subsequently infested with 280 J2/100 cm^3, planted with the host crops and arranged in a randomized complete block with 5 replications and grown for 8 weeks at 26°C. Only in one test soil did the two treatments result in significantly different rkn population levels. In a sandy loam the rkn population on tomato and wheat was 5- and 16-fold lower, respectively than in its pasteurized, rkn-infested equivalent. Exposure of the original soil to 50°-60°C for 30 minutes or soil fumigation with methyl iodide eliminated the suppressive effect. Microscopic examinations and fungal rRNA gene analysis of rkn egg masses at the end of those trials identified *Pochonia chlamydosporia* as one of the potential suppressive agents. Isolations from infested rkn eggs yielded four genetically different *P. chlamydosporia* strains that were tested in greenhouse experiments for their efficacy as biological control agents against *M. incognita*. The original suppressive soil was pasteurized, amended with 5,000 chlamydospores/cm^3 soil and infested with 600 eggs of *M. incognita*/100 cm^3 soil. Six weeks after planting to tomato seedlings, root galling and rkn population densities were determined. One of the four *P. chlamydosporia* strains had a significant negative impact on the rkn populations.

66. DETERMINATION OF SOYBEAN CULTIVAR RESISTANCE TO SOYBEAN CYST NEMATODE WITH QUANTITATIVE POLYMERASE CHAIN REACTION. **Lopez-Nicora[1], Horacio D., J.P. Craig[2], T.L. Niblack[1].** [1]Department of Crop Sciences AW-101 Turner Hall, University of Illinois, Urbana, IL 61801. [2]Department of Plant & Microbial Biology 341A Koshland Hall, University of Berkeley, Berkley, CA 94720.

A newly-developed real-time qPCR method for screening *Glycine max* (soybean) for resistance to *Heterodera glycines* was evaluated in two experiments. For the qPCR assay, a primer pair for the single copy gene *Hg*SNO, which codes for a

protein involved in the production of vitamin B6, was selected for *H. glycines g*DNA amplification within soybean roots. In the first experiment, a consistent inoculation method was developed to provide active second-stage juveniles (J2). Two-day-old soybean roots were infested with 0 and 1000 J2/mL. One day after infestation (DAI), the roots were surface sterilized and DNA was extracted with the DNA FastKit (MP Biomedicals, Santa Ana, CA). Consistent detection, amplification, and quantification of *H. glycines g*DNA was achieved in repeated trials. In the second experiment, compatible Lee 74, incompatible Peking and cultivars with different levels of resistance to *H. glycines* were infested with 0 and 1,000 J2/seedlings. One DAI, infected plants were transplanted into pasteurized soil. Subsequently they were harvested at 1, 7, 10, 14, and 21 DAI for DNA extraction. With the qPCR assay, highly resistant cultivars were differentiated from others at 10 and 21 DAI. Quantification of *H. glycines* infection by traditional means (numbers of females produced in 30 days) is time- and labor-intensive; the qPCR method can improve precision in determining infection levels.

67. VERSATILITY OF *IN VITRO*-PRODUCED *PASTEURIA* SPP. ENDOSPORES TO SUPPRESS *BELONOLAIMUS LONGICAUDATUS*. **Luc[1], John E., W. Pang[1], W.T. Crow[1], R. McSorley[1], and R.M. Giblin-Davis[2].** [1]Post-Doctoral Associate, Graduate Student, Associate Professor, and Professors, respectively, Entomology and Nematology Department, University of Florida, Gainesville FL 32611. [2]Professor, University of Florida-IFAS, Fort Lauderdale Research and Education Center, 3205 College Ave., Davie, FL 33314.

In earlier studies, isolates of *P. penetrans* were found to not only exclusively attach to a particular *Meloidogyne* species, but also to have different levels of attachment among isolates within a *Meloidogyne* species. Similarly, the S-1 strain of '*Candidatus* Pasteuria usgae' was shown to have differing levels of attachment to different populations of *Belonolaimus longicaudatus*. Narrow host-isolate specificity could limit the practicality of *Pasteuria* spp. as a commercial biopesticide. The objective of this research was to determine if *in vitro*-produced *Pasteuria* spp. endospores collected from *B. longicaudatus* exhibits host-parasite isolate specificity in its ability to manage *B. longicaudatus*. Five geographically diverse isolates of *Pasteuria* were collected from *B. longicaudatus*, *in vitro*-cultured by Pasteuria Bioscience Inc., and tested for their ability to attach to and suppress two genetically and geographically diverse isolates of *B. longicaudatus* in greenhouse trials. The respective endospore treatments were prepared as a liquid suspension (50 ml) of water, growth media, and endospores at 280,000 endospores/cm^3 of sand. Each endospore treatment was added to a plastic bag containing 400-cm^3 nematode-free United States Golf Association specification sand, gently hand mixed for two minutes, and then potted. 'Penncross' creeping bentgrass was seeded at 98 kg/ha (0.08 g/pot) and allowed to germinate and establish a root system for 13-d before being inoculated with 120 ± 6 mixed-life stages of nematodes. Nematode population densities and root lengths were assessed 84-d after nematode inoculation. All *Pasteuria* isolates attached to and reduced numbers of both *B. longicaudatus* isolates. However, there were no differences among *Pasteuria* isolates. All isolates of *in vitro*-produced *Pasteuria* endospores suppressed *B. longicaudatus* population densities close to 70%, compared to pots with no *Pasteuria* added. These results indicate that a bionematicide utilizing *in vitro*-produced *Pasteuria* endospores is likely to be effective against most populations of *B. longicaudatus*. Similarly, the original geographical source of *in vitro*-produced *Pasteuria* endospores does not appear to affect efficacy.

68. SEQUENTIAL YEARS OF LIQUID HOG MANURE APPLICATIONS TO GRASSED HAYLAND INCREASED THE ENRICHMENT BUT NOT THE STRUCTURE STATUS OF THE SOIL NEMATODE COMMUNITY. **Lumactud, Rhea[1], S. Briar[1], M. Tenuta[1] and K. Ominski[2]**. [1]Department of Soil Science, [2]Department of Animal Science, University of Manitoba, Winnipeg, Manitoba, Canada R3T 2N2.

We investigated the effect of increasing the productivity of a grassed soil used for hay on soil nematode communities. The study was conducted at the La Broquerie Manure and Pasture Productivity Research Site located in south-eastern Manitoba, Canada. Two hayed paddocks were sampled in fall 2007 and spring 2008. Since 2004, one paddock received surface application of liquid hog manure (LHM) at 123 kg plant-available N ha-1 (Manured) in spring and the other no LHM (Non-manured). Sampling in 2007 occurred the fall after LHM application in the spring of that year and in 2008 two weeks after application of manure in spring. Soil from each paddock was sampled on a grid scheme having 15 sample positions. Analysis of nematode trophic groups showed bacterivores were significantly favored ($P< 0.05$) by the Manured treatment while plant feeders were not compared to the Non-manured treatment. The treatments were similar ($P> 0.05$) with respect to other nematode trophic groups including hyphal feeders, plant-associates (Tylenchidae), omnivores and predators. Numbers of dauer juveniles of c-p1 nematodes were much higher in the Manured treatment. The estimated soil food web status based on enrichment and structure index values revealed that the application of LHM enriched the soil food web (increased opportunistic bacterial-feeders) without affecting higher trophic interactions even after four years of manure application or soon after the fifth year of manure application. The results are surprising because hay productivity increased 3-4 times for the Manured than Non-manured treatment. It seems LHM application caused enrichment of the food web through the addition of soluble carbon in the manure increasing bacterial growth. The study is continuing with an examination of the relation between soil profiles of nematode communities to rooting depth changes between manure treatments to test if increased below ground root productivity with manure application increased the abundance of all nematode trophic groups but not the relative abundances of trophic groups.

69. POPULATION DYNAMCIS OF *PRATYLENCHUS PENETRANS* ON CORN AND THE RELATIONSHIP OF NEMATODE POPULATION DENSITIES AND CORN YIELD. **MacGuidwin, A. E.** Dept. of Plant Pathology, 1630 Linden Dr., University of Wisconsin-Madison, Madison, WI 53706.

Nematicide treated corn seed is now available to manage *Pratylenchus spp*. and other nematode pests of corn. Seed treatments, with an efficacy of about 30 days, target early season infection and reproduction by nematodes. We studied the temporal and spatial dynamics of *P. penetrans* over the course of the growing season to determine the importance of early season events to the increase of nematode population densities and corn yield. Four corn fields, one in 2008 and three in 2009, were sampled at 22 randomly-selected sites. About 4 m of row were marked at each site and one corn plant was removed from this area on five sampling dates at 14-16, 20-24, 35-37, 63-65, 79, and 97-100 days after planting. A composite sample of 20 soil cores was collected on the first sampling date. Ten plants from the middle of the designated area at each site were collected for yield determination at the end of the season. Soil samples were assayed using a combination of sieving/centrifugation and incubation techniques. The seminal and adventitious root systems were assayed separately using Baermann funnel incubation for 48 hrs. After the third sampling date, a subsample of adventitious roots was assayed and used to estimate the total root biomass recovered. The initial number of nematodes recovered from soil samples ranged from 53 – 305 per 100 cm^3 soil for the four fields. All sites within the fields were infested with *Pratylenchus spp*., and most were identified as *P. penetrans*. Initial population densities in soil were predictive of nematode densities in roots and, for two fields, were negatively related to corn yield. Most nematodes were in the seminal root system and only on the last sampling date, 97-100 days after planting, did the total number of nematodes in adventitious roots surpass that of the seminal root system. The total number of nematodes and nematodes per gram of seminal roots were positively correlated with nematode densities in adventitious roots for the majority of the fields both within the same date and among different dates. These results confirm that the first five weeks after planting is critical for both nematode population growth and corn yield. Reducing nematodes in soil or seminal roots within the first 5 weeks of the crop is likely to reduce yield loss due to *P. penetrans*. Seminal roots continued to serve as a reservoir for *P. penetrans* and remained densely packed with viable nematodes at the time corn was harvested. Practices that reduce this source of concentrated inoculum are likely to benefit future crops susceptible to *P. penetrans*.

70. AN IMPROVEMENT IN MARIGOLD COVER CROPPING BY TARGETING ACTIVE STAGES OF ROOT-KNOT NEMATODES. **Marahatta, Sharadchandra P., K.-H. Wang, and B.S. Sipes.** Department of Plant and Environmental Protection Sciences, University of Hawaii, 3050 Maile Way, Honolulu, HI 96822.

Marigold (*Tagetes patula*) produces an allelopathic compound, α-terthienyl, known to be toxic to many plant-parasitic nematodes, including root-knot nematodes, *Meloidogyne incognita*. However, a marigold cover crop did not suppress root-knot nematodes consistently in two field trials. We hypothesized that marigold might suppress root-knot nematodes more effectively if the nematodes are in active life stage rather than in dormant life stage. Two greenhouse experiments were conducted where soils infested with *M. incognita* were collected and placed in 15.2-cm-d clay pots. Soils were conditioned either by 1) keeping the soil dry, 2) irrigating with water, or 3) drenching with cucumber (*Cucumis sativus*) leachate for 5-wk. The conditioning was to keep root-knot nematode inactive under the dry condition, and maintain nematode activity with irrigation or cucumber leachate. At the end of conditioning, soils were either planted with cucumber, marigold or remained bare for 10-wk. This 3 × 3 (conditioning × treatment) factorial designed experiment was arranged in randomized complete blocks with 5 replications. At termination of the treatment period, all plants were cut at the soil line and one cucumber seedling was planted in each pot for 3 wk and then nematodes penetration determined by acid fuchsin root staining. Active nematodes were those that moved their bodies when probed with a dental pick. Dry conditioning resulted in the highest number of inactive nematodes, whereas the cucumber leachate drench and irrigated soils had higher number of active nematodes. At termination of the cucumber bioassay, marigold suppressed numbers of root-knot nematode females in cucumber roots if soils were conditioned with irrigation or cucumber leachate, but not in soil under dry conditioning. Thus, marigold suppressed root-knot nematodes most effectively when the nematodes are in active stages.

71. CONTROL OF PLANT-PARASITIC NEMATODES WITH THE BIONEMATICIDE NEMA-Q® AN EXTRACT OF *QUILLAJA SAPONARIA*. **Marais, Lawrence J.,[1] R. Otero[2] and E. Riga[3]. [1]**Monterey AgResources, 3654 S. Willow Ave, Fresno, CA 93745. [2]Desert King International, 7024 Manya Circle, San Diego, CA 92154. [3]WSU, 24106 N..Bunn Rd, Prosser, WA 99350

Nema-Q® a bionematicide containing triterpenoid saponins, polyphenols and tannins, was tested *in vitro*, in the greenhouse and field against important plant parasitic nematodes in California and Washington State : *Xiphinema index, Meloidogyne incognita, M. hapla, M. chitwoodi, Helicotylenchus pseudorobustus, Criconemella xenoplax, Tylenchulus semipenetrans, and Pratylenchus penetrans*. Nema-Q was compared with several synthetic nematicides including Mocap®, Nemacur® and Vydate® as well as the biological nematicide DiTera® in several crops, including potatoes, wine and table grapes and cucumber. Nema-Q was found to be effective in controlling these nematodes at a concentration of 10,000 ppm. Treatment resulted in enhanced quality and yield, an in many cases outperformed all other nematicides tested. In Cabernet

wine grapes, lesion nematodes were reduced from 1200 to 350 per 250-g soil. In Chardonnay wine grapes ring nematode numbers were reduced from 380 to 100 per 250-g soil and yield increased by 15-20 per cent. In potato, *M. chitwoodi* numbers were reduced from 800 to 270 per 250-g soil, with zero culls and only 4 % tuber infection, compared to 59 % culls in untreated controls and 50 % tuber infection. In cucumber greenhouse tests, Nema-Q reduced egg production of *M. incognita* by 60 %.

72. RESPONSE OF *SCOTTNEMA LINDSAYAE* TO ANTARCTIC GLACIAL CYCLES. **Martin[1], Mac T., D.H. Wall[2], S.M. Peat[1], B.N. Adhikari[1], and B.J. Adams[1].** [1]Department of Biology and Evolutionary Ecology Laboratories, Brigham Young University, Provo, UT 84604. [2]Department of Biology and Natural Resource Ecology Laboratory, Colorado State University, Fort Collins, CO 80523.

Glaciologists have suggested that because of the extent of the ice sheets during the last glacial maximum (LGM) in Antarctica no terrestrial species could have survived these harsh conditions. However, substantial evidence suggests that some invertebrate species did indeed survive the LGM. Among these species is the nematode, *Scottnema lindsayae*. The objective of this study is to explore how *S.lindsayae* responded to glacial cycles. To do this, *S.lindsayae* populations were sampled from across northern and southern Victoria Land, Antarctica, including regions that could have served as hypothetical refugia. Areas close to the coast yet high in elevation could have remained ice-free during the LGM, yet not have been inundated during the subsequent sea-level rise concomitant with glacial recession. We hypothesized that extant populations from such areas would yield more genetic variation than low elevation areas that would have been drastically affected by the ice sheets during the LGM, or inundated by sea level rise concomitant with glacial recession. DNA was extracted from collected samples using a DNAzol extraction protocol. Portions of CO1 mtDNA and ITS1 rDNA loci were amplified through PCR and subsequently cloned and sequenced. Model based phylogenetic analyses were performed to infer relationships between populations of *S.lindsayae*. Results show greater genetic variation exists in high altitude coastal *S.lindsayae* populations relative to valley populations, consistent with the elevational refugia hypothesis. Our data suggests that areas close to the coast and high in elevation likely served as source populations for the subsequent colonization of suitable habitat that became available as the glaciers retreated.

73. HATCH DEPRESSION IN TWO SPECIES OF PLANT-PARASITIC NEMATODES: J2 FITNESS AND LEVEL OF SUSCEPTIBILITY TO LOW TEMPERATURE. **Masler[1], Edward P. and S. T. Rogers[1]**. [1]Nematology Laboratory, Agricultural Research Service, U. S. Department of Agriculture, Beltsville, MD 20705.

Exposure of *Heterodera glycines* eggs to low temperature in vitro for 7 days significantly depresses the subsequent total percent hatch, relative to controls, after eggs are returned to normal rearing temperatures (Masler et al., 2008). These studies have been extended to include *Meloidogyne incognita*, and more precisely examine the effects of low temperature egg treatment on hatching behaviors. Test eggs were stored 7 days at 50C and then returned to rearing temperature (210C) for recovery, while control eggs were stored at the rearing temperature throughout 14-day hatch cycle. Initial hatch assays were performed using modified Baermann funnels, and percent hatch calculated daily [(J2 collected/total eggs on funnel) x 100]. With this method, depression of hatch from low temperature-treated *H. glycines* eggs was 35 percent ($P < 0.05$) when measured 14 days after egg recovery at 210C. However, when hatch assays were performed in microtiter plates, which allowed direct visual assessment of all individuals throughout the assay, J2 hatch depression at 14 days was measured at 16 percent ($P < 0.05$). This raises the question of the effects of low temperature treatment of eggs on overall J2 fitness, as well as on hatching per se. In species comparisons, low temperature egg treatment revealed both quantitative and qualitative differences. Microtiter plate assays showed that *H. glycines* hatch depression was rather mild at 7 days after return to 210C (8 percent below controls; $P < 0.05$), increasing to 11 percent ($P < 0.05$) at 10 days and then to16 percent at 14 days. While the extent of hatch depression in H. glycines was time dependent within the recovery period, *M. incognita* hatch depression was constant throughout the same assay period. In addition, *M. incognita* was at least 2-fold more susceptible to low temperature treatment than was *H. glycines* (37 percent depression at 7, 10, and 14 days after return to 210C; $P < 0.01$). The significance of species differential responses to environmental challenges is discussed.

74. SYNCYTIUM GENE EXPRESSION IN GLYCINE MAX (PI 88788) ROOTS UNDERGOING A RESISTANT REACTION TO THE PARASITIC NEMATODE *HETERODERA GLYCINES*. **Matsye, P.D.[1], K. C. Showmaker[2], P. Hosseini[3], N.W. Alkharouf[3], G.W. Lawrence[2], B.F. Matthews[4] and Vincent P. Klink[1]** [1]Department of Biological Sciences, Harned Hall, Mississippi State University, Mississippi State MS, 39762; [2]Department of Entomology and Plant Pathology, Mississippi State University, Mississippi State MS, 39762; [3]Jess and Mildred Fisher College of Science and Mathematics, Department of Computer and Information Sciences, Towson University, 7800 York Road, Towson, Maryland, 21252; [4]United States Department of Agriculture, Soybean Genomics and Improvement Laboratory, Bldg. 006, Beltsville, MD 20705

The plant-parasitic nematode, *Heterodera glycines* is the major pathogen of Glycine max (soybean). *H. glycines* accomplish parasitism by creating a nurse cell known as the syncytium from which it feeds. The syncytium undergoes two

developmental phases. The first is a parasitism phase where syncytia develop. During this earlier phase (4-5 days post infection), syncytia undergoing resistant and susceptible reactions appear the same. The second phase is when the resistance response becomes evident (between 4 and 6 dpi) and is complete by 9 dpi. Analysis of the resistant reaction of G. max genotype PI 88788 to *H. glycines* population NL1-RHg/HG-type 7 is accomplished by laser microdissection of syncytia at 3, 6 and 9 dpi. Comparative analyses are made to pericycle and their neighboring cells isolated from mock-inoculated roots. Direct comparative analyses were also made of syncytia at 6 days post infection to those at 3 dpi (baseline) to identify genes that characterize the resistance phase of the resistant reaction. The most highly induced pathways include components of jasmonic acid biosynthesis, 13-lipoxygenase pathway, S-adenosyl methionine pathway, phenylpropanoid biosynthesis, suberin biosynthesis, adenosylmethionine biosynthesis, ethylene biosynthesis from methionine, flavonoid biosynthesis and the methionine salvage pathway. In comparative analyses of 9 dpi to 6 dpi (baseline), these pathways, along with coumarin biosynthesis, cellulose biosynthesis and homogalacturonan degradation are induced. Custom pathway analyses have revealed other pathways that are induced specifically during the resistance reaction. The experiments presented here strongly implicate the jasmonic acid defense pathway as a factor involved in the resistant reaction of G. max(PI 88788) to H. glycines (NL1-RHg/HG-type 7).

75. UPTAKE AND EXCLUSION OF PLANT-EXPRESSED FLUORESCENT PROTEINS BY THE SOYBEAN CYST NEMATODE *HETERODERA GLYCINES*. **McCarter, James**, **Bingli Gao, John Bradley, Michelle C. Hresko, Amy Caruano-Yzermans, D. Jeremy Williams.** *Divergence Inc., 1005 N. Warson Road, St. Louis MO 63132,* mccarter@divergence.com

Cyst nematodes, major pathogens of soybean, potatoes, sugar beets, and other crops, are sedentary root feeders that limit the size of ingested molecules by use of an organelle-like feeding tube believed to act as a ''molecular sieve'' between the nematode stylet and the surrounding syncytial plant feeding site. The feeding tube constrains the delivery of proteins and nucleic acids from the plant to the nematode. The size cut-off for molecular uptake into the soybean cyst nematode (SCN), *Heterodera glycines*, has never been determined and work on other cyst nematode species provides conflicting values between 20 and 40 kDa. We have therefore studied the uptake of variously sized fluorescent proteins expressed in transgenic roots into *H. glycines* in comparison to another sedentary endoparasite, root knot nematode (*Meloidogyne incognita*) and the migratory lesion nematode (*Pratylenchus scribneri*). We provide the first evidence for uptake of plant-expressed fluorescent proteins into the SCN intestine and describe this uptake by developmental stage. We show how the likelihood of uptake into the intestine decreases with molecular weight establishing a molecular size cut-off for ingestion. The results obtained from this study clarify and expand understanding of SCN host-plant feeding and provide guidance for the development of biotechnology-based strategies for nematode control.

76. INTRODUCTION TO NEMATODES: A NEW MULTIMEDIA PRESENTATION. **McGawley[1], E. C., M.J. Pontif[2], and C. Overstreet[3].** [1 & 3]LSU AgCenter, Dept. of Plant Pathology and Crop Physiology, 302 Life Sciences Bldg., Baton Rouge, LA 70803, [2]LSU AgCenter, Sugarcane Research Station, St. Gabriel, LA 70776.

Introduction to Nematodes is a multimedia presentation that contains 99 multi-layered slides. The presentation contains 481 photographs, 155 illustrations, 17 tables and 14 videos. The presentation is formatted as a Quicktime movie and therefore will play on either a Macintosh or a PC computer. The presentation is accompanied by a 13 page syllabus with notes and credits for each slide, an index of the 18 sections (General, History, Morphology, Body Systems, Symptoms, Loss Estimates, Movement & Dissemination, Sampling, Extraction, Population Dynamics, Thresholds, Management, Taxonomy, Parasitism, Key for Identification, Highlighted Genera, Disease Complexes and Entomogenous Nematodes), a ''read me'' file which contains instructions for obtaining and using the Quicktime player and a set of ''thumbnail views'' of each slide. This presentation can be obtained as a FREE download courtesy of the websites of SON and the Organization of Nematologists of Tropical America.

77. REPRODUCTION AND PATHOGENICITY OF GEOGRAPHIC ISOLATES OF *ROTYLENCHULUS RENIFORMIS*. **McGawley[1], Edward C., M.J. Pontif[2], and C. Overstreet[3].** [1 & 3]LSU AgCenter, Dept. of Plant Pathology and Crop Physiology, 302 Life Sciences Bldg., Baton Rouge, LA 70803, [2]LSU AgCenter, Sugarcane Research Station, St. Gabriel, LA 70776.

The comparative reproduction and pathogenicity of isolates of *Rotylenchulus reniformis* from Alabama, Arkansas, Hawaii, Louisiana, Mississippi and Texas on cotton was evaluated in microplot trials. Prior to initiation of microplot trials, ten clonal populations of each geographic isolate were derived from single egg masses. Reproduction of the clonal populations of each geographic isolate were evaluated in greenhouse studies with LA887 cotton by assessing the numbers of vermiform stages in soil and eggs per gram of root tissue 60 days after inoculation. On the basis of these trials, each repeated once, one clonal population of each of the six isolates was selected for use in microplot trials. Averaged over the two trials, clonal population designations selected for use in microplot trials and their respective reproduction values (R, where R=Pf/Pi) and numbers of

eggs per gram of root were: AL-8 (R=14.9, eggs=202); AR-3 (R=30.4, eggs=525); HI-9 (R=20.2, eggs=183); LA-3 (R=18.2, eggs=517); MS-7 (R=25.7, eggs=602) and TX-10 (R=42.8, eggs=938).

Data From full-season (140-148 days) microplot trials, averaged over 3 years, showed significant differences among isolates of reniform nematode in both reproduction on and pathogenicity to LA887 cotton. Dry plant weight at harvest averaged 370.6g for the non-inoculated control. All isolates except HI-9 produced root weights at harvest that were reduced significantly below that of the control. Harvest weights for plants inoculated with LA-3 and MS-7 were significantly lower than those representing the other four geographic isolates.

78. PERFORMANCE OF SPIROTETRAMAT FOLIAR ON *PRATYLENCHUS VULNUS* INFECTED *JUGLANS* SPP. **McKenry, Michael, T. Buzo and S. Kaku.** Nematology Department, UC Riverside, Riverside CA 92521.

Two hundred and sixteen 25 year-old, 13-m tall Chico walnut trees on *Juglans hindsii* rootstock were set aside for experimentation with 455-ml/ha Movento™ in 1200-L/ha water plus 237-ml/ha Penetrator adjuvant on November 17, 2008. One hundred forty-four trees were sprayed with Movento. Six days later the trunks of seventy-two sprayed trees were severed by chain saw at 45-cm above ground level. An additional 72 trees remained unsprayed within a completely randomized design. At monthly intervals roots from each of 12 replicates were dug and soil adhering to these roots collected, sieved and extracted for three days in mist. Rhizosphere soil collected at 30-days post treatment from severed and non severed trees provided nematode population levels remarkably similar and significantly (P = 0.05) reduced from that of the unsprayed trees. Over the next five-months of rhizosphere sampling it was only the non-severed trees that provided significantly fewer nematodes compared to the unsprayed. These nematode reductions were significant for each of six-months of sampling and the cumulative reduction was 51% compared to nematode counts from unsprayed trees. Roots from trees with severed trunks did not provide significant population reductions after the 30-day soil sampling. During the next five-months the level of nematode control was half that achieved from non-severed trees. The 6-mo mean nematode reduction from severed trees was 24% compared to that achieved from unsprayed trees. Apparently, portions of the enol form of spirotetramat can travel within 6-days from 13-m tall trees to nematodes located in soil 1-m beyond the tree trunk. However, severance of tree trunks six-days after a foliar spray of Movento did not provide adequate time for achieving full lethal impact to *P. vulnus*. It is hypothesized that nematodes that ingested the enol temporarily lost their ability to pass through a double layer of facial tissue when mist extracted at 30-days post treatment but this impact was fleeting or lethal to a lesser portion of the nematode population. Our conclusion is that disruptions to the delivery or maintenance of the active enol metabolite at the site of the nematode can seriously detract from its nematicidal performance. At 120-days after treatment rhizosphere samples were also collected 3-m away from sprayed and unsprayed tree trunks and the 19% population reduction due to spraying was not significant. Previous reports indicate irrigations or rainfall too soon after a Movento spray can reduce nematicidal activity. In conducting these evaluations no irrigations or rains occurred during the two-week period after trees were sprayed. This is the first report of at least six-months of 50% nematode control following a foliar spray of Movento. For growers of perennial crops this level of nematode control is at least equivalent to that achieved at drip sites following drip irrigation delivery of Nemacur at 13-L/ha.

79. POTENTIAL FOR BIOLOGICAL CONTROL OF ROOT-KNOT NEMATODES BY NATURAL PREDATORS IN FLORIDA SOILS. **McSorley[1], Robert, and K.-H. Wang[2].** [1]Dept. of Entomology and Nematology, University of Florida, PO Box 110620, Gainesville, FL 32611-0620, [2]Dept. of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI, 96822-2279.

Root-knot nematodes (*Meloidogyne* spp.) are important pathogens of many vegetables and ornamentals grown commercially or in home gardens. However, naturally-occurring predators that occur in soil may provide some biological control of nematodes. A greenhouse experiment was conducted to evaluate the potential of invertebrate predators in agricultural and natural soils to suppress *M. incognita* on coleus (*Coleus blumei*), an excellent host of this nematode. Soil from adjacent natural and agricultural habitats was collected from three locations in Florida (Quincy, Homestead, Citra). The agricultural sites were previously planted to vegetable crops with prior history of soil fumigation. Each soil was placed into pots, planted with coleus seedlings, inoculated with 2,000 eggs of *M. incognita* race 2, and arranged in a 3 x 2 factorial design (3 locations x 2 habitats). Root-knot nematode numbers were 78-95% lower in natural soils compared to agricultural soils by the end of the 3-month experiment. A variety of invertebrates were monitored in all soils including free-living nematodes, enchytraeid worms, tardigrades, Collembola, mites, japygids, and ants. Omnivorous and predatory nematodes and enchytraeids did not show population patterns consistent with the suppression of root-knot nematodes observed in natural soils. However, Collembola and mites were generally more abundant in natural than in agricultural soils, and increased in numbers over time. Japygids, ants, and tardigrades were also more abundant in all or some natural soils initially, but numbers declined over time. Results support the idea that there are many organisms that occur in Florida soils that may cause nematode suppression. Also, the relative suppression of root-knot nematodes was greater in natural than in agricultural soil, and the occurrence of some invertebrate predators, especially mites and Collembola, was consistent with the observed declines in nematode population density.

80. REPRODUCTION OF ROOT-KNOT NEMATODES ON FOUR SUGARBEET CULTIVARS. **Mendes, Maria de Lourdes, and D. W. Dickson.** Entomology and Nematology Department, University of Florida, Gainesville, FL 32611.

Sugarbeet (*Beta vulgaris*), which is grown commercially for sugar production in the European Union, United States, and Russia, is susceptible to infection by several species of root-knot nematodes. The objective of this research was to evaluate the response of four sugarbeet cultivars (Alota, Bobcat, Mandella, and Trinita) to six species (races) of root-knot nematodes that occur in Florida. Three of the nematodes, *Meloidogyne mayaguensis, M. floridensis,* and *M. javanica* race 3, were recently reported in Florida, whereas the other three, *M. arenaria* race 1, *M. incognita* race 4, and *M. javanica* race 1, are common species (races) of this nematode in Florida. Two experiments were carried out. The first test was conducted in a greenhouse in a completely randomized design with five replicates. 'Rutgers' tomato (*Solanum lycopersicon*) was included as a susceptible host to each nematode species. Plants were inoculated with 5,000 eggs and(or) second-stage juveniles of *M. mayaguensis, M. floridensis,* and *M. javanica* race 3 per plant. Fifty-two days after inoculation the plants were removed and the number of galls, egg masses, and the final population density was assessed. The three nematode isolates produced a large number of galls, and egg masses (>100) on all four sugarbeet cultivars. Reproductive factors on all sugarbeets were similar to tomato (>5.94). The second test was carried out in field microplots previously infested with *M. arenaria* race 1, *M. incognita* race 4, and *M. javanica* race 1. The evaluation was performed over a period of 98 days following the transplant date, and galling and egg mass indices were recorded. All four sugarbeet cultivars were highly susceptible to the three root-knot nematode species averaging more than 100 galls and egg masses/plant. Results indicate that sugarbeet cultivars are highly susceptible to all the root-knot nematode species (races) evaluated.

81. DEVELOPMENT OF SPECIES SPECIFIC PRIMERS FOR MOLECULAR DIAGNOSTICS OF PLANT-PARASITIC NEMATODES ASSOCIATED WITH *MISCANTHUS X GIGANTEUS* AND *PANICUM VIRGATUM* USED FOR BIOFUELS. **MEKETE, Tesfamariam[1], Kimberly REYNOLDS[1], Horacio D. LOPEZ-NICORA[2], Michael E. GRAY[1,2] and Terry L. NIBLACK[1,2]** [1]Energy Biosciences Institute, University of Illinois, 1206 W Gregory Dr, Urbana, IL, 61801, [2]Department of Crop Sciences, University of Illinois, 1102 S Goodwin Ave, Urbana, IL 61801, Corresponding author: Tesfamariam Mekete, tel. +1 2172449480, fax +1 2172443637, email: tmekete@illinois.edu

A survey was conducted in 2008 and 2009 to determine the occurrence and distribution of plant-parasitic nematodes associated with Miscanthus and switchgrass plants used for biofuels. Several plant-parasitic nematode species associated with plants used for biofuels were identified. Of these, *Pratylenchus penetrans, P. scribneri, P. crenatus, Helicotylenchus pseudorobustus, Hoplolaimus galeatus, X. americanum,* and *X. rivesi* are potentially the most damaging pests. These species were identified at several sampling sites. We have developed species-specific primers that discriminate species of *P. penetrans, P. scribneri, P. crenatus, H. pseudorobustus, H. galeatus, X. americanum,* and *X. rivesi*. Our sets of primer pairs could be combined in a single multiplex reaction, and reactions could be run with the primers individually or in various combinations. This will serve as an accurate and fast diagnostic tool for the identification of morphologically close species. Identification and quantification of these nematodes are critical to the development of control strategies.

82. PHYTOTOXICITY OF MUSTARD SEED MEALS ALONE AND IN COMBINATIONS. **Meyer, Susan L.F.[1], I.A. Zasada[2], and S.B. Orisajo[3].** [1]USDA-ARS Nematology Laboratory, Beltsville, MD 20705, USA, [2]USDA-ARS Horticultural Crops Research Laboratory, Corvallis, OR 97330, USA, [3]Crop Protection Division, Cocoa Research Institute of Nigeria, Ibadan, Oyo State, Nigeria.

Mustard seed meal is produced when oil is extracted from brassicaceous seeds. The high glucosinolate content of these seed meals makes them of interest as management agents for weeds and soilborne pathogens. Previous studies indicated that seed meals from *Brassica juncea* and *Sinapis alba* are nematotoxic, with higher application rates of *S. alba* than of *B. juncea* required to reduce nematode population levels. However, there is also the potential for phytotoxicity. It would be advantageous to know the amount of time required between mustard seed meal application and planting to avoid phytotoxicity. Consequently, both meals were tested alone and in combinations to determine toxicity to pepper (*Capsicum annuum*) seedlings. Rates of application (weight meal:weight soil) were: 1) 0.5% *Sinapis alba*, 2) 0.2% *Brassica juncea,* 3) 0.25% *S. alba*:0.25% *B. juncea*, 4) 0.375% *S. alba*:0.125% *B. juncea*, 5) 0.125% *S. alba*:0.375% *B. juncea*, and 6) nontreated. Treated soil was placed into greenhouse pots 5, 4, 3, 2 and 1 weeks prior to transplant, and at transplant (0 weeks). Pots were watered 1-2 times per day, and 6-week old pepper seedlings were then transplanted into each treatment and harvested 12 days later. The experiment was conducted twice. All treatments except controls resulted in 0% plant viability when applied at transplant. The 0.2% *B. juncea* treatment was the only one that resulted in 100% plant viability at all other seed meal application times. This treatment tended to be the least toxic overall to pepper seedlings, including effects on shoot lengths and plant weights. Application of 0.5% *S. alba* resulted in some loss of seedling viability at all application times, and in decreased shoot lengths and shoot and root weights. The 0.125% *S. alba*:0.375% *B. juncea* treatment resulted in the third-highest overall seedling viability after controls and 0.2% *B. juncea*, with 100% live seedlings when treatments were applied 3-5 weeks prior to transplant. Of the combination treatments, 0.375% *S. alba*:0.125% *B. juncea* resulted in the greatest loss of viable seedlings, with no seed meal application date resulting in 100% viability when both trials were combined.

83. THE LATEST DEVELOPMENTS IN APPLIED ENTOMOPATHOGENIC NEMATOLOGY IN EUROPE. **Moens[1], Maurice and R.-U. Ehlers[2].** [1]Institute of Agriculture and Fisheries Research, Merelbeke, Belgium and Laboratory of Agrozoology, Department of Crop Protection, Ghent University, Ghent, Belgium, [2]Institute for Phytopathology, Department of Biotechnology and Biological Control, Christian-Albrechts-University, Raisdorf, Germany

As a result of the reduction of the number of authorized soil insecticides within the European Union and because of increased insect resistance to insecticides, entomopathogenic nematodes (EPN) are increasingly important. During the last few years, EPN have taken the fastest growing market share of the microbial segment of bio-control agents. The species commercialized in Europe are *Heterorhabditis bacteriophora* and *H. megidis*, *Steinernema feltiae*, *S. carpocapsae* and *S. kraussei*. Use of these bio-control agents varies considerably across the EU. In general, traditional markets include control of black vine weevil (*Otiorhynchus sulcatus*) in ornamentals and strawberries, control of grubs (Scarabaeidae) in turf, and control of sciarids (*Bradysia* and *Lycoriella* spp.) in both ornamentals and mushrooms. New markets include control of western flower thrips (*Frankliniella occidentalis*) and white flies in glasshouse grown vegetables, using EPN in combination with other biological antagonists or biological active products like chitosan. Other uses for EPN products include control of the red palm weevil (*Rhynchophorus ferrugineus*) and Mediterranean root borer (*Capnodis tenebrionis*), hazelnut borer (*Balaninus nucum*), codling moth (*Cydia pomonella*), and western corn rootworm (*Diabrotica virgifera*) in maize. Additional markets can only be developed with increasing production capacity and competitive product costs.

84. DOWNWARD MIGRATION OF *ROTYLENCHULUS RENIFORMIS* INFLUENCED BY WATER INFILTRATION AND COTTON ROOT GROWTH. **Moore[1], S. R., K. S. Lawrence[1], E. van Santen[2], and F. J. Arriaga[3].** [1]Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849, [2]Department of Agronomy and Soils, Auburn University, Auburn, AL 36849, [3]National Soil Dynamics Laboratory, USDA-ARS, Auburn, AL 36832.

The presence of *Rotylenchulus reniformis* below plow depth can have negative effects on cotton health. Two trials were established in 7.62-cm diameter by 75-cm deep soil cores to determine 1) the effect of water infiltration on vertical translocation of *R. reniformis*, and 2) the role of root growth in the downward migration of *R. reniformis*. The water infiltration study consisted of three treatments of simulated rainfall amounts, 25.4-mm, 76.2-mm, and 127-mm, and no rainfall. The 25.4-mm rainfall treatment enabled the nematodes to reach a depth of 30-cm. Rainfall of 76.2-mm was required to reach a depth of 45-cm and 127-mm of rainfall was needed to surpass 45-cm. Less than 6% of the initial population was relocated from the top 15-cm by any rainfall amount. To determine the effect of root growth on *R. reniformis,* the nematodes were monitored as a cotton root system developed over time. Cotton roots reached the maximum depth of 75-cm at 60 days after planting (DAP). Vermiform life stages reached 75-cm at 45 DAP, however females were not present in the cotton roots at the 75-cm depth until 90 DAP. Root growth increase at 15 day intervals averaged 390% when *R. reniformis* population increases in the same 15 day interval were the lowest. In contrast, root growth increase averaged only 134% when *R. reniformis* population increases were the highest. Water infiltration minimally affected the downward translocation of *R. reniformis*. Populations were able to increase downward through the profile as a food source became available.

85. PHYLOGENOMIC ANALYSIS IN THE PHYLUM NEMATODA. **Morris[1], Krystalynne, H. Bik[1], P.J. Hatcher[2], W.K. Thomas[1]** [1]Hubbard Center for Genome Studies, University of New Hampshire, 35 Colovos Road, Durham, NH 03824, [2]Department of Computer Science, University of New Hampshire, 33 Academic Way, Durham, NH 03824.

Applying phylogenomic approaches to understanding the relationships within the phylum has been limited by the number of complete genome sequences available for representatives of the Nematoda. However, as advances in DNA sequencing technologies have made it possible to sequences entire genomes rapidly and for little money, the availability of genome sequences from the Nematoda are growing. The first step in phylogenomic analysis is to indentify orthologos genes—genes which duplicated during a speciation event and whose divergence can be used to trace phylogenetic history. Currently, our methodology limits us to predicting putative orthologs from an identified set of single copy, homologs. While these single copy homologs include many orthologs, they also contain genes whose history reflects gene duplications and losses within the genome and not necessarily the phylogenetic history of the species. Because the vast majority of nematodes are unculturable, phylum-wide representation of genome datasets will be greatly advanced by the ability to sequence a genome from single nematodes.

86. SOYBEAN CYST NEMATODE POPULATIONS IN DELAWARE ARE SHIFTING IN RESPONSE TO WIDESPREAD PLANTING OF SOYBEAN CULTIVARS WITH RESISTANCE FROM PI88788. **Mulrooney[1], Robert P., N.F. Gregory[1] and R.D. Heinz[2].** [1]Plant and Soil Sciences Department, University of Delaware, Newark, DE 19716. [2]University of Missouri Extension Nematology Laboratory, Columbia, MO 65211.

The soybean cyst nematode, *Heterodera glycines*, is the most economically important soybean pathogen in Delaware. A survey of SCN populations scattered over a three year period ending in 1995 indicated that 67% of the populations were defined as race 3, 28% as race 1 and 5% races 5, 6, and 9. Thirteen of those populations had an average female index (FI) on PI88788 of 24. More than 90% of the current soybean acreage infested with SCN in Delaware is planted with glyphosate

resistant soybeans with SCN resistance from PI88788. The classic stunting and yellowing symptoms on SCN resistant cultivars are beginning to appear in fields with high initial SCN egg counts and dry growing conditions early in the season. An SCN survey was conducted in 2009 to ascertain the current population levels and HG types and races in Delaware. The survey was conducted to determine if the races in DE soybean fields were changing and at what level the races were found. The results of the survey demonstrated that SCN was found in two of the three counties and egg densities ranged from 72 to 11,448/250cc of soil. Sixty-three samples were taken and SCN was found in 56% of the samples. Fifteen samples which represented 43% of the 35 samples that had SCN present were sent for HG typing and race determination at the University of Missouri Extension Nematology Laboratory. Twelve populations were HG type 2.5.7 and 3 were HG type 1.2.5.7. Race composition was 47% race 1, 33% race 5, and 20% race 2. No race 3 populations were identified. Of those 15 populations 100% had FI greater than 10% on PI88788. The FI on PI88788 ranged from 44-80 with a mean of 67. This limited, but representative sampling of SCN populations indicates a significant shift in the field populations of SCN in Delaware.

87. EVALUATING HATCHING FACTORS OF *GLOBODERA PALLIDA* AND *TABACUM*. **Navarre[1,2], Roy., S. Kumar[1], and R. Zemetra[2].** [1]USDA-ARS, Prosser, WA 99350 [2]Washington State University, Prosser, WA. 99350 [3]University of Idaho, Moscow ID.

Globodera pallida was found in southern Idaho in 2006 and because it is restricted to a small number of fields, an eradication effort is underway. Even in the absence of a host, PCN can persist for decades in fields and largely for this reason is a formidable pest of potatoes. Cysts contain hundreds of eggs that hatch in response to unidentified compounds called hatching factors (HFs) that are present only in root exudates from potatoes and closely related plants. HFs have potential to be used to induce eggs to hatch in the absence of a host, in which case the emerged juveniles will perish, in effect a ''suicide hatch.'' We are exploring the possibility of using hatching factors to induce a suicide hatch and are trying to identify the active components of root exudates using HPLC and LCMS. HPLC fractionation suggests more than one active compound is present in root exudates. The sensitivity and selectivity of LCMS allows additional approaches to identify active compounds, including comparative analysis of exudates from over 30 solanaceous and non-solanaceous plants. Potato root exudates were found to induce hatching in *G. tabacum*, demonstrating that this species can also be used to help identify active compounds.

88. CHARACTERIZING CROP IMPACTS AND FIELD DISTRIBUTION OF STING NEMATODE IN FLORIDA STRAWBERRY. **Noling[1], Joseph W., A.W. Schumann[1], and M. Cody[1].** [1]University of Florida, IFAS, CREC, Lake Alfred, FL 33850.

The Sting nematode, *Belonolaimus longicaudatus*, is a major yield limiting pest of Florida strawberry. Depending on Pi, a patchy field distribution of stunted plants develops after infected plants die, fail to grow or progressively shrink in canopy size. Given the inability to monitor nematode population density and distribution in real time, yield loss maps were developed based on indirect measures, such as plant canopy size and post harvest season counts of fruit stems. Since 2005, over 70 commercial strawberry fields have been studied to characterize field distribution and nematode impact. Experimental objectives were to 1) develop methodologies to map field distributions of sting nematode stunted plants; 2) compare chronological records of total fruit picked and average weight of fruit harvested from plants by specific size category; and 3) to relate the relative impact of canopy size to strawberry yield among the size classes of nematode stunted plants. The numbers of plants in four plant size categories were systematically recorded at 40 to 50 ft intervals throughout most fields. Plant size categories, measured as average canopy diameter, were dead (0), small (<20 cm), medium (>20 and < 30 cm) and large (>30 cm). Using plant sizes, chemical treatment evaluations were determined in over 50 commercial fields with recurring histories of sting nematode problems. Hyperspectral reflectance and other plant, field, and aerial imaging technologies were used to characterize and relate differences in strawberry crop yields to within row, green vegetative cover. A tractor mounted GreenSeeker optical sensor (NTech Industries; Ukiah, Ca) was used to scan strawberry rows to provide estimates of green canopy cover (NDVI) against a backdrop of black plastic mulch covering the raised bed. Cumulative differences in plant numbers and relative yield contribution within each plant size category were then statistically compared between actual measured strawberry plot yields and chemical treatments. Plant stunting and yield losses were well correlated with final harvest soil population density of the nematode. Accurate maps of nematode distribution, crop yields and loss indices were related to the intensity of field sampling and spatial resolution describing nematode, crop, or soil characteristic. Ground truth surveying of plant size distribution repeatedly demonstrated the accuracy of in-field, remotely sensed GreenSeeker information. Strawberry yields from commercially hand harvested large plots were well correlated with relative yield values determined from plants of different sizes within the plots. Differences in plant size distribution and of relative yield also occurred between various alternative to methyl bromide chemical treatments. Overall, field scale changes in strawberry crop productivity due to sting nematode and chemical treatment can be determined, on a farm by farm or industry-wide basis, from post harvest assessments of counts of different plant sizes. The methodology is being used to provide growers guidance and quantitative performance data on alternatives to methyl bromide soil fumigation for nematode management.

89. PREPLANTING TALL FESCUE GRASS FOR CONTROLLING *MELOIDOGYNE INCOGNITA* IN A YOUNG PEACH ORCHARD. **Nyczepir[1], Andrew P., S.L.F. Meyer[2], and J. Cook[3].** [1]USDA-ARS, SE Fruit & Tree Nut Research Laboratory, 21 Dunbar Road, Byron, GA 31008, [2]USDA-ARS, Nematology Laboratory, Beltsville Agricultural Research Center, Beltsville, MD 20705, and [3]University of Georgia Cooperative Extension Service, Butler, GA 31006.

Preplant fumigant nematicides have traditionally been used to control *Meloidogyne* spp. in peach in the southeastern United States. The current preplant nematicides recommended for managing *Meloidogyne* spp. in peach include the soil fumigants, 1,3-dichloropropene and metam sodium. Because the economic hardships afflicting growers in recent years have made it difficult to economically afford these chemicals, finding an alternative nematode control is warranted. Greenhouse trials were initially conducted to evaluate the susceptibility of tall fescue grass (*Festuca arundinacea* = *Schedonorus arundinaceus*) lines with or without endophytes to *M. incognita*. Fescue lines evaluated included, i) Jesup EI (E+, wild-type endophyte present), ii) Jesup EF (E-, no endophyte present), and iii) Max-Q (E+, but non-ergot producing endophyte). Peach (susceptible Lovell rootstock) was included as the control. Nematode reproduction criteria were used in evaluating fescue susceptibility. Peach supported greater ($P \leq 0.05$) reproduction of *M. incognita* than all fescue lines. Differences in reproduction were not detected among the fescue lines and all fescue lines were either poor or nonhosts for *M. incognita*. Furthermore, the presence of the endophyte did not appear to affect nematode reproduction. In a separate greenhouse study, Max-Q did not support *M. hapla* reproduction compared to a tomato control. Additionally, investigation of the effect of 1- and 2-year tall fescue (cv. Max-Q) preplant groundcover rotations for the management of *M. incognita* was initiated in 2005 in a field experiment in central Georgia. In 2008, both fescue rotations suppressed ($P \leq 0.05$) soil population densities of *M. incognita* J2 compared with 2 years of continuous peach (root-knot nematode susceptible Halford rootstock reference control). No differences in suppression of *M. incognita* J2 population density were detected between the 1 and 2- years of fescue rotation. Therefore, a tall fescue rotation has potential as a nonchemical preplant strategy to manage *M. incognita* in peach orchards in the southeastern United States.

90. PLANT-PARASITIC NEMATODE GENOMES: COMPARATIVE ANALYSIS BEGINS TO REVEAL PATHS TO ADAPTATION AND EVOLUTION. **Opperman, Charles H.[1], D.McK. Bird[1], M. Burke[2], V.M. Williamson[3], E.A. Scholl[1], and the Plant Nematode Genome Sequencing Consortium[1,2,3].** [1]Plant Nematode Genetics Group, NC State University, Raleigh, NC 27695, [2]David H. Murdoch Research Institute, Kannapolis, NC, [3]University of California-Davis, Davis, CA.

Just over a decade ago, the first genome sequence of a multicellular animal was reported in the journal Science. Because the animal was a nematode, this milestone had tremendous impact on the Nematology/Parasitology scientific community. The free-living nematode, *Caenorhabditis elegans* posseses a 100 Mb genome with slightly more than 20,000 genes, and has proved to be a robust biological model for many systems, including humans. Indeed, the sequencing consortium assembled for the *C. elegans* genome was largely responsible for the public effort to sequence the human genome. Since the release of the *C. elegans* genome, there has been a growing effort to complete both EST and genome sequences from both plant and animal parasitic nematodes, and as preminisced in 1998, *C. elegans* has proved to be an important tool to guide parasite biologists in the post-genomic era. We have developed a large EST database that has set the stage for numerous genome projects. We completed the genome sequence of *Meloidogyne hapla*, and found that this nematode possesses almost 6,000 fewer genes than the free living nematode, *Caenorhabditis elegans*. Extensive annotation has revealed that these species do share many genes in common, but that there are also genes that appear to be significantly diverged. We will build on that sequence to perform pan-order comparisons. To this end, we have sequenced the genomes of *Radopholus similes, Pratylenchus coffeae*, and are in the process of developing material to sequence *Helicotylenchus multicinctus*. We hypothesize that there may be a core Tylenchid genome, and acquiring these sequences will provide solid data to identify it. This, in turn, may lead to discovery of novel approaches to control plant parasitic nematodes, either by novel chemistry or by transgenic resistance. We are utilizing an evolutionary approach to examine the role of HGT, gene family expansion/contraction, and chromosomal organization in the progression migratory endo-parasitic species to sedentary endo-parasitism. We will also search for genes apparently evolving at a faster rate or under purifying selection. Others species are planned for the future. Individually, these sequences are a digital record of an organism, but collectively they represent a hugely significant resource to study the evolution of parasitism. We are developing these resources in a comprehensive comparative genomics study, which will be made available to the scientific community via our Gbrowse site at www.pngg.org.

91. USING APPARENT ELECTRICAL CONDUCTIVITY AND VERIFICATION STRIPS TO DEFINE NEMATODE MANAGEMENT ZONES IN COTTON. **Overstreet, Charles[1], E.C. McGawley[1], D. Burns[2], and M. Wolcott[3].** [1]Dept. of Plant Pathology and Crop Physiology, LSU Agricultural Center, Baton Rouge, LA 70803, [2]County Agent, LSU Agricultural Center, St. Joseph, LA 71366, [3]Dept. of Biological and Agricultural Engineering, Baton Rouge, LA 70803.

Southern root-knot nematode (*Meloidogyne incognita,* SRKN) and reniform nematode (*Rotylenchulus reniformis,* RN) cause serious losses to cotton in many of the alluvial soils in Louisiana. Variable soil texture found within fields may impact

the areas requiring fumigation. Seventeen trials have been conducted in Louisiana since 2004 to evaluate the use of apparent electrical conductivity (EC_a) as a tool for development of nematode management zones.

All fields had high populations of SRKN, RN, or both nematodes. Verification strips (rows treated with either 1,3-dichloropropene at 28 L/ha or untreated) were laid out so as to go through the various soil zones defined by EC_a. All fields were harvested with cotton pickers having yield monitors. Thirteen of the fields showed a significant yield response to the application the fumigant in one or more zones. The greatest responses to the fumigant generally occurred in the soil zones that had the lowest EC_a values (<25 mS/m). The highest response for any zone was 652 kg/ha lint. In the zones that responded positively to the fumigant, the response averaged 229 kg/ha lint. Four fields showed little or no response to the fumigant in any soil zone. Two of the fields that failed to respond to the fumigant were severely impacted by Hurricane Gustav losing an estimated 30-70% of the crop. The other two fields yielded extremely well (ca. 1400 kg/ha and 1725 kg/ha lint) and never showed nematode injury. The use of EC_a appears to be a good tool to define soil texture profile in the alluvial soils found in Louisiana and in the development of nematode management zones.

92. TOLERANCE AND RESISTANCE OF *CYNODON* SPP. AND *PASPALUM VAGINATUM* TO *BELONOLAIMUS LONGICAUDATUS*. **Pang[1], Wenjing, W.T. Crow[1], and K.E. Kenworthy[2].** [1]Entomology and Nematology Department, Bldg. 970 Natural Area Drive, P.O. Box 110620, University of Florida, Gainesville, FL 32611, [2]Agronomy Department, 304 Newell Hall, P.O. Box 110500, University of Florida, Gainesville, FL 32611.

Bermudagrass (*Cynodon* spp.) and seashore paspalum (*Paspalum vaginatum*) are commonly used warm-season turfgrasses on golf courses in Florida. *Belonolaimus longicaudatus* is the most serious nematode pest on turf grass in Florida. Recent cancellation of fenamiphos has resulted in the need for alternative nematode management tactics. Utilization of resistant or tolerant cultivars is the most efficient and least costly practice for nematode management on turf, but information about the responses of most grass cultivars to *B. longicaudatus* is not available. The objective of this study is to evaluate newer turfgrass cultivars for resistance and tolerance to *B. longicaudatus* and to select *Cynodon* spp. germpalsm accessions with superior nematode responses that can be used in future cultivar breeding and development. Three experiments were conducted to test 17 bermudagrass cultivars, seven seashore paspalum cultivars, and 47 bermudagrass germpalsm accessions, respectively in two sequential trials in a greenhouse in 2009. Aerial stolons were grown in sand-filled plastic conetainers and inoculated with 0 or 50 *B. longicaudatus* per conetainer six weeks after planting. Turf was maintained in a randomized complete block with six replications. Nematode and roots samples were collected 90 days after nematode inoculation. Both trials showed that among the dwarf bermudagrass cultivars, 'Tifdwarf' and 'Emeraldwarf' were tolerant, but susceptible to *B. longicaudatus*; 'Champion', 'Tifeagle', and 'Floradwarf' were resistant, but intolerant to *B. longicaudatus*. The non-dwarf bermudagrass cultivars, 'Tifsport', 'Patriot', and 'Riviera' were both tolerant and resistant to *B. longicaudatus*. The non-dwarf bermudagrass cultivar 'Princess 77' was the most susceptible bermudagrass with a nematode reproductive factor of 5.5. All seashore paspalum cultivars supported the reproduction of *B. longicaudatus*. Highest and lowest population densities were on 'SeaIsle 2000' and 'Aloha', respectively. The root length of 'SeaSpray' was reduced by *B. longicaudatus*. Four African bermudagrass accessions and nine common bermudagrass accessions were selected based on both tolerance and resistances to *B. longicaudatus* for future turf breeding. The development and use of resistant or tolerant cultivars could produce high quality turf grass while reducing the use of nematicides, fertilitizers, and water. These cultivars also can be used as standards for future germplasm accessions screening and testing of newly developed cultivars.

93. CREATING POSSIBILITIES THROUGH NEMATOLOGY NEWSLETTERS. **Parkunan[1], Venkatesan, and B. J. Adams[2].** [1]Hampton Roads Agricultural Research and Extension Center 1444 Diamond springs rd, Virginia Tech, Virginia Beach, VA 23455, [2]Dept. of Biology, and Evolutionary Ecology Laboratories, 775 WIDB, Brigham Young University, Provo, UT 84602.

Nematology resources online are not widespread and represent less than one tenth of the information offered for nematodes on the World Wide Web as compared to other pest or pathogen groups. To address this concern, we analyze the benefits and ways of sharing nematode related information online for effective and quick knowledge transfer through nematology newsletter supplements (NNL portal). Connecting researchers and their research with the public (end users) is critical, but complicated by research articles that are hard for lay audiences to comprehend. For example, some of the most influential scientific findings contain no words, rather figures and illustrations, such as discoveries of x-rays, the structure of DNA, fractal geometry, etc. Communicating nematology findings through figures, photographs, illustrations and other media for education and journalistic purposes would help bridge this gap.

Online techno-friendly gadgets, including social networking sites, discussion forums, blogs, and digital newsletters are gaining importance as they instantaneously deliver research news, findings and interesting information via multimedia resources to multiple audiences. The Nematology Newsletter (NNL) published by the Society of Nematologists every quarter offers interesting tidbits, news, member profiles, announcements, job listings, and excellent columns on nematodes and nematologists from all around the globe. Yet we make little use of social networking sites such as facebook, twitter, and

YouTube, which offer discussion forums, blogs and audio/video clip capabilities. Approximately, half of all Americans currently use these networking sites on a day-to-day basis and their numbers continue to grow.

The NNL has gone a long way since its inception, but little has changed since it began to be distributed electronically. In general, the NNL now tends to use more images, illustrations, photographs, and graphics. But with the availability and popularity of social networking sites among scientific communities and to the public we would like to propose that in addition to the traditional quarterly NNL, as online interactive NNL, or ''NNL portal'', should be implemented. This NNL portal could provide instantaneous, up-to-date, open source information containing video clips, discussion forums, tweets, feeds, podcasts, events and other useful widgets with web portal capability exclusively for quick nematode related information transfer. Several other scientific societies have already implemented these changes. As the Society of Nematologists wishes to improve its standing among scientific societies, and as a discipline, we urge members and non-members to contribute and participate in this NNL portal. Traditional quarterly NNL publication will continue, but with links to the NNL interactive pages. SON Members and general public worldwide should use this service freely employing all possible multimedia resources for more rapid, frequent and effective transfer of nematological information.

94. THE IMPACT OF A NEW TACTIC TO MANAGE A CITRUS DISEASE ON BIOLOGICAL CONTROL OF A CITRUS PEST BY ENTOMOPATHOGENIC NEMATODES. **Pathak[1], Ekta, R. Campos-Herrera[1, 2], R.J. Stuart[1], F. E. El-Borai[1,3], A.W. Schumann[1], J.H. Graham[1], and L.W. Duncan[1].** [1]Citrus Research and Education Center, University of Florida, Lake Alfred Fl 33850. [2]Centro de Ciencias Medioambientales, CSIC, Serrano 115 dpdo Madrid, 28006 (Spain). [3]Plant Protection Dept. Faculty of Agriculture, Zagazig University, Egypt.

Recent introduction of the citrus disease huanglongbing (citrus greening) has dramatically increased rates of tree loss in Florida citrus orchards. Advanced cultural practices that increase young tree growth rates and reduce time to maturity are needed to mitigate production losses. The use of daily, drip-fertigation of young trees (up to several times per day) has been reported to hasten tree maturity and its suitability is currently being studied in Florida. However, this intensively managed citriculture (IMC) could profoundly alter the moisture, chemical and biological nature of the rhizosphere compared to conventional citriculture (CC) in which trees are irrigated up to several times per week using microsprinklers and fertilized periodically. Six species of entomopathogenic nematodes (EPN) are endemic in Florida citrus orchards and are natural control agents of soilborne larvae of a serious citrus pest, the root weevil, *Diaprepes abbreviatus*. We compared the effects of IMC and CC on EPN and some natural enemies of EPN in an ongoing field trial, which began 6 months after the cultural treatments were initiated. Pairs of PVC tubes were inserted 15 cm deep in soil adjacent to IMC irrigation drippers or in the same relative location in CC plots. We introduced 8000 EPN infective juveniles of either *Steinernema riobrave* or *Heterorhabditis indica* into each tube. The soil in the tubes was recovered after 48, 72 or 168 h, and the nematodes were extracted and processed to recover DNA that was subjected to real-time PCR to identify and quantify six species of EPN and five species of nematophagous fungi (NF). Fewer than half as many augmented *S. riobrave* were recovered in IMC than CC ($P<0.05$), whereas augmented *H. indica* were recovered in greater numbers in IMC than CC ($P<0.05$). Moreover, numbers of the endemic EPN *S. diaprepesi* were more than four-fold higher in CC compared to IMC ($P<0.05$). Initially during the time course of the experiment, the trapping NF *Arthrobotrys dactyloides* and *A. musiformis* were significantly more abundant in samples of *S. riobrave* than in *H. indica*; and both of these species and the NF *Monacrosporium gephyropagum* were also more abundant in IMC than CC. However, the NF patterns changed by day seven, perhaps because EPN augmentation induced trophic cascades with different temporal dynamics in the two treatments. *Arthrobotrys oligospora* and *Catenaria* sp. tended to occur in greater numbers in the *S. riobrave* treatment but the differences were not significant. These results demonstrate the need to further evaluate how a potentially important program to manage huanglongbing in Florida citrus orchards impacts biological control of an important citrus pest.

95. COMPARATIVE TRANSCRIPTOMICS: A RESEARCH PROGRAM FOR NEMATODE ECOLOGY AND EVOLUTION. **Peat, Scott M. and B. J. Adams.** Department of Biology, Brigham Young University, Provo, Utah, 84602

Expressed sequence tag (EST) data provide a high upside alternative to whole genome sequencing for investigating gene discovery, function and expression of non-model nematode species. Improvements to sequencing technologies, and cDNA library construction protocols tailored to next generation sequencing methodologies, can now produce more rapid, cost effective, and more robust EST datasets. Comparative analyses of the expressed genes from varying states/stages within or between non-model nematode species facilitates both ecological and evolutionary based studies that inform questions addressing nematode variation, adaptation, and speciation. We detail specific methodologies used to successfully amplify EST data from nematodes using 454 pyrosequencing, address potential problems with and improvements to methodologies as they relate to nematodes, and explore the bioinformatics and molecular evolutionary tools that can be used to address a wide array of ecological and evolutionary based questions. As an example, we utilize the Tylenchomorpha nematode *Deladenus* (=*Beddingia*) *siricidicola* to illustrate how 454 sequencing of EST data and comparative transcriptomics were used to explore the molecular basis for plant and insect parasitism in nematodes and assess the origin and maintenance of plant parasitism genes within the infraorder Tylenchomorpha.

96. ESTIMATE OF THE ECONOMICAL AND SOCIAL IMPACT OF *MELOIDOGYNE MAYAGUENSIS* ONTO THE GUAVA CROP IN BRAZIL. **Pereira, F.O.M., R.M. Souza, P.M. Souza, C. Dolinski, and G.K. Santos.** Dept. of Entomology and Phytopathology, Universidade Estadual do Norte Fluminense Darcy Ribeiro, 28015-620, Campos dos Goytacazes, Brazil.

This study aimed to estimate the impact caused by *M. mayaguensis* Rammah & Hirschmann, 1988 onto Brazilian guava growers. The net present worth (calculated at different discount rates) and the internal rate of return of nematode-free and infested guava orchards were calculated based on the nematode-infested area in five States, the pattern of the disease, the average productivity of infested orchards and the crop's technical coefficients. In the regions examined, the total economic loss caused by *M. mayaguensis* was estimated to be 103.2 million reais (~US$ 61 millions). The decimation of the infested orchards resulted in the loss of 3,703 full-time job positions. This economical and social impact does not take into account the up- and down-stream effects of the widespread decimation of guava orchards, which include decrease in the production and commercialization of fertilizers and other items used for guava production, distribution, processing and reselling of the guava production and tax collection. This study indicates the economic importance of *M. mayaguensis* to Brazil, which may be further increased if other guava-producing regions become infested and/or if other crops become widely parasitized by this nematode.

97. IMPACT OF DAGGER NEMATODE AND CHERRY RASP LEAF VIRUS ON COLORADO'S SWEET CHERRY INDUSTRY. **Pokharel, Ramesh.** Western Colorado Research Center, Colorado State University, 3168 B ½ Road, Grand Junction, CO 81503.

Dagger nematode (*Xiphinema* spp.) is an important nematode in fruit crops because it reduces fruit yield by direct feeding and vectors many plant viruses including Cherry Rasp Leaf Virus (CRLV). CRLV is a main constraint for cherry production in western Colorado and also causes Flat apple disease commonly observed in Colorado but of lesser perceived importance. Different surveys were done during 2008-2009 looking at: incidence of 1) dagger nematode (23 orchards, >100 samples) and 2) CRLV via molecular detection methods (50 samples, approx. 25 orchards), and evaluation of 3) variety (11 varieties) and rootstock (18 rootstocks) for dagger nematode and CRL. Dagger nematodes were observed in 100% of the sweet cherry orchards surveyed (15–70 nematodes / 100 cc soil) in survey #1, 3, & 4. CRLV symptoms (enations / leafy outgrowths on the lower leaf surface) were observed in 52% of the samples with varying symptom severity. In survey #2 (CRLV incidence) 50-yr-old Royal Duke sweet cherry trees had dagger nematodes in the soil beneath it, but no CRLV symptoms nor detectable CRLV infection were found in molecular test despite the fact that adjacent Bing cherry trees had severe CRLV symptoms. In survey #3 (sweet cherry varieties), all varieties had dagger nematode populations (8-50 nematodes / 100 cc soil) and all except Summerset (14-yr-old tree) exhibited CRLV symptoms. In survey #4 (rootstocks), dagger nematodes were present (17–108 nematodes / 100 cc soil) beneath all rootstocks, and CRLV infection was detected in Bing scions on various rootstocks other than those of the Gisela series (up to 11-yr-old trees). This indicated that either these Gisela series rootstocks are resistant to CRLV and/or dagger nematodes or the virus movement is slow in these rootstocks. Results of these studies show that dagger nematodes are more common than CRLV in Colorado. However, the species identification of dagger nematodes found and of those that transmit the virus is unclear. Species identification in this nematode is difficult by the morphometric measurements, and not all dagger species transmit this virus. Thus molecular identification of this nematode from Colorado is underway. The wide-spread dagger nematode distribution (100%), the high incidence of CRLV (52% orchards), and the susceptibility of most varieties and many rootstocks observed in the surveys demonstrate the importance of dagger nematode for the sweet cherry industry in Colorado. Moreover, the direct nematode feeding impact by dagger nematodes in cherry was not included in these surveys which need further investigations. Studies on virus and the nematode complex on the Gisela series rootstocks and other management strategies are in progress in pot studies by inoculating viruliferous nematodes.

98. NEMATODE COMMUNITY ANALYSIS AS A SOIL HEALTH INDICATOR IN ORGANIC VS CONVENTIONALLY MANAGED PEACHES. **Pokharel, Ramesh and Zimmerman, R.** Western Colorado Research Center, Colorado State University, 3168 B ½ Road, Grand Junction, CO 81503.

Free-living and plant-parasitic nematodes (PPNs) may be the most useful biological indicator groups for community indicator analysis of production systems. This is because more information exists on their taxonomy and feeding roles than for other microorganisms. Thus, we studied nematodes as bio-indicators to compare the soil health of conventional and organic production systems in two peach varieties, June Pride and Cresthaven. The trees were planted in 2000-01 in both conventional and organic blocks at Colorado State University's Western Colo. Research Center-Roger Mesa (WCRC-RM) site to compare these two production systems. Organic and conventional blocks were maintained per their respective standard cultural practices. Trees of both varieties in the organic production system exhibited low vigor and yield. Nematode community analysis, soil and plant tissue nutrient analysis, and fruit yield measurements were done to identify potential reasons for the low performance of the organically-grown trees compared to the conventionally-grown trees. Soil samples were taken from five random trees in each variety and production system and bulked to make a single composite sample per variety/production system for subsequent nematode community analysis. The nematode populations (different groups) were

extracted using modified Baermann tray method, identified to genus level, and individuals counted in each bulked sample. Ten bacterial feeder genera (*Alaimus, Cephalobus, Eucephalobus, Plectus, Prismatolaimus, Triphyla, Acrobeles, Geomonhystara, Monhystrella and Panagrolaimus*), 6 fungal feeder genera (*Aporcelaimmellus, Eudorylaimus, Thonus, Tylencholaimus, Microdorylaimus and Tylencholaimellus*), 4 fungal feeder/root feeder genera (*Aphelenchus, Aphelenchoides, Ditylenchus and Filenchus*), 1 predator genus (*Clarkus*), and 5 root feeder genera (*Helicotylenchus, Meloidogyne, Pratylenchus, Paratylenchus and Diphtherophora*) were found in these Colorado peach orchards. PPNs comprised 4-52% of the total nematode populations with the highest populations in conventional Cresthaven peach. Higher diversity of bacterial feeder, fungal feeder, and root feeder nematodes was observed in the organic system irrespective of peach varieties, but this was not the case for fungal feeder/root feeder and predatory nematodes. No predatory nematodes were observed in June Pride peach soils in either the organic or the conventional system. Altogether, five genera of PPNs were observed, with one to two genera associated with each system; densities of total PPNs were higher in organic systems than in conventional system across both varieties. The ratio of free-living nematodes /PPNs (an indicator of better soil health) was higher in soils beneath the organic system trees than beneath the conventional system trees for June Pride, but not for Cresthaven.

99. PRECISION APPLICATION OF 1,3-D IN ARIZONA FIELD CROPS USING ELECTRICAL CONDUCTIVITY MAPPING AND GPS TECHNOLOGY. **Richardson[1], Jesse M., R. Norton[2], M.A. McClure[3], and J.D. Busacca[4].** [1]Dow AgroSciences, Hesperia, CA, [2]Safford Agricultural Center, Safford, AZ, [3]University of Arizona, Tucson, AZ, [4]Dow AgroSciences, Indianapolis, IN.

Electrical conductivity (EC) mapping of soil has been integrated with global positioning system (GPS) technology to define areas within a field where precision application equipment can optimize use of 1,3-dichloropropene to control nematodes. Large-scale field studies were conducted in Eastern Arizona on cotton and field corn to investigate the potential value of this technology under commercial production conditions. In the cotton study at Safford, yield differences attributed to the use of 1,3-D were inconsistent. However, in the field corn study at Bonita, results were dramatic. Prior to planting, a real-time sensor mapped EC in the field that correlated well to a USDA soil survey map. Low EC values also correlated closely to high rootknot nematode densities revealed by soil probe sampling. 1,3-D was shank-injected in 33 of the 60 acres indicated by the EC sensor in the study site. Utilizing these technological advances, corn yield was increased 20-30% in areas impacted by nematodes.

100. BIOFUMIGATION AGAINST ENDOGENOUS AND QUARANTINE PLANT PARASITIC NEMATODES OF VEGETABLE CROPS IN THE PACIFIC NORTHWEST. **Riga[1], Ekaterini, John Wilson[1] and Zareen Dossey[1,2].** [1]IAREC, Washington State University, Prosser, 99350; and [2]Plant Pathology Department, Washington State University, Pullman, WA, USA.

The endophytic fungus, *Muscodor albus*, brassica green manures and mustard seed meals were tested against plant parasitic nematode species including the quarantined potato cyst nematode, *Globodera pallida*, from economically important vegetable crops in the Pacific Northwest. The Columbia root knot nematode, *Meloidogyne chitwoodi,* the northern root knot nematode, *M. hapla,* the stubby root nematode *Paratrichodorus allius,* and the lesion nematode *Pratylenchus penetrans* were exposed in the laboratory, greenhouse and/or field conditions to the above treatments, while *G. pallida* was tested only in the greenhouse. The mean percent mortality of nematodes exposed to *M. albus* in the greenhouse was 82.9% for *P. allius*, 82.1% for *P. penetrans*, and 95% for *M. chitwoodi*; mortality in the controls was 9%, 7%, and 3.9% respectively. Only 21.6% of *M. hapla* juveniles died due to *M. albus* in comparison to 8.9% in controls, however, 69.5% of the treated *M. hapla* juveniles displayed reduced motility in comparison to the controls. Green manures from brassica crops reduced all of the above plant parasitic nematodes between 80 to 100% in the greenhouse and field with exception of *M. chitwoodi*. The Columbia root knot nematode has 5 to 7 life cycles per growing season, therefore, green manure on their own were not able to reduce this species. However, green manures in combination with reduced rates of synthetic nematicides reduced it to similar levels as synthetic fumigants. Mustard seed meals significantly reduced all of the above plant parasitic nematode species in the greenhouse and in the field in comparison to the controls. In addition, potato plants planted in soil containing *G. pallida* and treated with mustard seed meal had no cysts in comparison to the untreated controls which had on average 5.5 cysts per 100 g soil.

101. MORPHOLOGICAL COMPARISON OF *XIPHINEMA BAKERI* POPULATIONS FROM ARKANSAS. **Robbins, Robert[1] and Weimin Ye[2].** [1]Dept. of Plant Pathology, 2601 N. Young Ave, University of Arkansas, Fayetteville, AR 72704, [2]North Carolina Department of Agriculture & Consumer Services, Nematode Assay Section, 4300 Reedy Creek Road, Raleigh, NC 27607-6465.

Xiphinema bakeri is a didelphic species in group 7 of the 1990 polytomous key of Loof and Luc. It was described from British Columbia, Canada specimens in 1961 by Williams. It has been reported in 10 states in the USA as well as Japan. It was found in 30 locations of 828 samples taken in a 1999-2001 Arkansas survey of Arkansas hardwoods of stream banks, mainly in sandy to sandy loam soil on several hardwood and other hosts. Two populations collected prior to the survey are also reported. Morphological data on single males found in two populations and nine males in a second population are given.

The Arkansas populations conform very well with the original description. The common names of the Arkansas associations are: Americam elm, Black cherry, Black locust, Black walnut, Box-Elder, Cottonwood, Cypress, Eastern Red Cedar, Grape, Hackberry, Hickory, Honey locust, Honeysuckle, Oak, Osage orange, Peach, Redbud, Red oak, Red Maple, River birch, River cane, Silver maple, Sweet gum, Sycamore, White oak, Willow, and Winged elm. Selected diagnostic characters from several of the populations are given as well as comparative photos of diagnostic characters.

102. MORPHOLOGICAL COMPARISON OF *XIPHINEMA CHAMBERSI* POPULATIONS FROM ARKANSAS. **Robbins, Robert[1] and Weimin Ye[2]**. [1]Dept. of Plant Pathology, 2601 N. Young Ave, University of Arkansas, Fayetteville, AR 72704, [2]North Carolina Department of Agriculture & Consumer Services, Nematode Assay Section, 4300 Reedy Creek Road, Raleigh, NC 27607-6465.

Xiphinema chambersi is a monodelphic species in group 1 of the 1990 polytomous key of Loof and Luc. It was described from Virginia specimens by Thorne in 1939. It has been reported in 16 states in the USA as well as Japan and Korea. It was found in 18 locations of 828 samples taken in a 1999-2001 Arkansas survey of Arkansas hardwoods of stream banks, mainly in sandy to sandy loam soil on several hardwood hosts. Five populations collected prior to the survey are also reported. Morphological data on a single male from two populations is given. The Arkansas populations conform very well with the original description. The common names of the Arkansas associations are: Americam elm, Blackberry, Box-Elder, Cottonwood, Cypress, Dogwood, Eastern Red Cedar, Grape, Hackberry, Hickory Hackberry, Hickory, Oak, Pecan, Persimmon, Redbud, Red Maple, Red oak, River birch, River cane, Silver maple, Sweet gum, Sycamore, Water oak, White oak, and Willow.

103. MORPHOLOGICAL COMPARISON OF *XIPHINEMA KRUGI* FROM ARKANSAS, HAWAII AND NORTH CAROLINA. **Robbins, Robert[1] and Weimin Ye[2]**. [1]Dept. of Plant Pathology, 2601 N. Young Ave, University of Arkansas, Fayetteville, AR 72704, [2]North Carolina Department of Agriculture & Consumer Services, Nematode Assay Section, 4300 Reedy Creek Road, Raleigh, NC 27607-6465.

Xiphinema krugi is in group 2 of the 1990 polytomous key of Loof and Luc. This species has a partial uterus but lacks an anterior ovary. It was described from specimens Brazil in 1955 by Lordello. It has been reported in 5 states in the USA as well as the following countries worldwide; Argentina, Brazil, Columbia, Malaysia, Martinique, Mauritius, Paraguay, Senegal, South Africa, Sri-Lanka, Surinam, Trinidad, Uruguay, and Venezuela. This species has been found only once by R.T. Robbins in Feburary 1, 1982 from hardwoods by Frog Bayou, south of Alma, Crawford County, Arkansas. Attempts to obtain more specimens from the same location have been unsuccessful. Our specimens agree with Lordello's original 1955 description of this species. A population from North Carolina has a longer tail with a bluntly knobbed tail terminus, whereas another population from Hawaii has a shorter tail with rounded terminus. Our population lies between those two having a conoid tail with a slightly knobbed tail terminus. Fredrick also demonstrated tail variation on specimens from Florida and Alabama.

104. GENETIC AND PHYSICAL MAPPING OF *MELOIDOGYNE INCOGNITA* RESISTANCE ON CHROMOSOME 11 OF ACALA NEMX COTTON. **Roberts[1], Philip A., C. Wang[1], and M. Ulloa[2].** [1]Department of Nematology, University of California, Riverside, CA 92521; [2]USDA-ARS, WICS, Res. Unit, 17053 N. Shafter Ave., Shafter, CA 93263.

Root-knot nematode (RKN, *Meloidogyne incognita*) resistance in *Gossypium hirsutum* 'Acala NemX' cotton is conferred by the recessive gene *rkn1* (locus *Mi$_{2h}$-C11*) on chromosome 11. The concentration of RKN, reniform nematode and other disease resistance determinants on chromosome 11 indicates that much can be gained by molecular genetic and physical mapping analysis of this genomic region. Gene action analysis was conducted by inheritance and quantitative trait loci mapping of the RKN resistance in Acala NemX. Comparative analysis was conducted of the resistance in Acala NemX with RKN resistance in other Upland, Pima, and diploid cotton germplasm sources. The analysis was based on resistance segregation and expression in resistant x resistant and resistant x susceptible crosses, and on DNA sequence information of the multiple alleles of markers linked to RKN resistance and markers framing the chromosome 11 resistance region. The probable ancestral origin and introgression pathways of RKN resistance into the Acala NemX background were revealed, with results supporting artificial (man-made) introgression. Various crosses with RKN resistance sources indicated that allelic interaction, epistasis, and heterosis operate in the expression of resistance depending on resistance source and genetic background. In efforts to develop a physical framework of the resistance region, annotated complete sequence was developed of *G. hirsutum* 'Acala Maxxa' BAC clones anchored to chromosome 11 according to mapped BAC-end sequence derived markers (MUSB). The sequence information is being used to derive additional markers for screening combinatorial pools of an Acala NemX source ('N901') BAC library and for anchoring additional BACs in the resistance region.

105. IDENTIFICATION OF *PASTEURIA* SPP. THAT PARASITIZE HOPLOLAIMUS GALEATUS**.** **Schmidt, Liesbeth M., T. E. Hewlett, J.P. Waters, and A. Green.** Pasteuria Bioscience, Inc. 12085 Research Drive, Suite 185, Alachua, FL 32615.

Second only to *Belonolaimus longicaudatus* in economic importance on turf grass, *Hoplolaimus galeatus* (Lance nematode) can be found along the entire east coast of the United States, the Mississippi River basin, as well as in Colorado,

California and Texas. *H. galeatus* collected from the LaPlaya golf course in Ft. Meyers, Florida were observed to harbor small *Pasteuria*-like endospores attached to their cuticles. Following challenge with anti-*Pasteuria* monoclonal antibody MAb2A41D10 in live immuno-fluorescent assay (IFA), the endospores presenting on the cuticle of these specimens were observed to be positive for the epitope recognized by the *Pasteuria* spp.-specific antibody. Endospores released from spore-filled cadavers also tested positive with MAb2A1D10, thus suggesting attachment and infection of *H. galeatus* by *Pasteuria* spp.. Genomic DNA was extracted from approximately 80 hand-picked spore-filled cadavers. PCR using degenerate primers provided partial coding sequences for *16s rDNA, spoIIAA_AB*, and *atpF*. Phylogenetic analysis of these genes using the Neighbor-Joining method and boot-strap consensus (1000 replications), show the Lance nematode *Pasteuria* spp. shares the greatest degree of homology to *Rotylenchulus reniformis Pasteuria* spp. isolates obtained from Reniform nematodes in Huxford, AL and Quincy, FL, followed by *Pasteuria penetrans*. This finding establishes the first reported immuno-detection, and phylogenetic placement of a *Pasteuria* sp. infecting the Lance nematode. With an estimated 50 million dollar annual market for nematode control on turf grass in the United States; this raises the possibility that these *Pasteuria* may be used for biocontrol of *Hoplolaimus galeatus*.

106. EFFECTS OF SOYBEAN CYST NEMATODE-RESISTANT VARIETIES ON FIELD POPULATIONS OF *HETERODERA GLYCINES* IN MICHIGAN. **Schumacher-Lott[1], Lesley, G. Bird[1], J. Davenport[1], and T. Kendle[2].** [1]Dept. of Entomology 243 Natural Science, Michigan State University, East Lansing, MI 48824, [2]Edwardsburg, MI 49112.

The soybean cyst nematode (*Heterodera glycines*, SCN) is a pest of soybeans throughout most soybean-producing counties in Michigan. SCN management tactics, such as the use of resistant soybean varieties, are ever-increasing on farms with SCN-infested acreage. A resistant soybean variety is one that yields well in a site that has an above threshold initial population density and results in low nematode reproduction. Commercial varieties from only three sources of resistance are available for use in MI: PI 548402, PI 88788, and PI 437654. PI 88788 is planted on about 95% of infested soybean acres in MI. The objective of this research is to demonstrate the effects of SCN resistance source on field populations within single growing seasons. For this research, three farms were selected, based on previous SCN history, for use in 2008 and 2009. The fields were divided into 76.2 m-long sections, with either four or eight row planting widths (38.1 cm or 76.2 cm rows), depending on the location. Six soybean varieties were selected and planted based on their maturity groups and sources of SCN resistance. Two susceptible cultivars as well as two PI 88788, one PI 437654 x PI 88788, and one PI 548402 x PI 88788 were used at each location. Plots were sampled before planting and after harvest using a cone-shaped soil probe (approximate volume of 2000 cm^3), 15.24 cm below the soil surface. Initial and final SCN population densities were determined for both growing seasons. The annual SCN reproduction factor was determined by dividing the final SCN population density by the initial SCN population density for each cultivar from each location. The reproduction factor was used for statistical analysis. In 2008 in Ingham County, the mean reproduction factor for one of the susceptible varieties was 12,196 times greater than the PI 437654 x PI 88788 variety. The mean reproduction factor for the same susceptible variety on the same farm in 2009 was only 6.4 times greater than the PI 437654 x PI 88788 variety. In 2008 in Monroe County, the mean reproduction factor for one of the susceptible varieties was 1,557 times greater than the PI 548402 x PI 88788 variety. In 2009 in Macomb County, the mean reproduction factor for one of the susceptible varieties was 43.6 times greater than the PI 437654 x PI 88788 variety. In 2009 in Cass County, the mean reproduction factor for one of the susceptible varieties was 959 times greater than the PI 437654 x PI 88788 variety. It is probable that these differences result from variations in the aggressiveness of the SCN populations or environmental factors associated with the different growing seasons and locations.

107. NOVEL ENTOMOPATHOGENIC NEMATODE FORMULATIONS AND TARGETS IN NORTH AMERICAN ORCHARDS. **Shapiro-Ilan[1], David I., and Lawrence A. Lacey[2].** [1]USDA-ARS, SE Fruit and Tree Nut Research Laboratory, 21 Dunbar Rd., Byron, GA 31008, [2]USDA-ARS, Yakima Agriculture Research Laboratory, 5230 Konnowac Pass Rd., Wapato, WA, 98908.

Research and commercial application of entomopathogenic nematodes (epns) in North American orchard systems has a long history. In the pursuit of commercial viability, there have been a number of success stories, but also quite a number of dead ends. In this presentation, we focus on new developments and opportunities for epns that are on the horizon. A number of new application approaches and new nematode-pest matchups appear highly promising. For example, in pecan, using a multi-stage/pre-emergence application approach, treatments of *Steinernema carpocapsae* infective juveniles (IJs) resulted in < 1% survival of pecan weevil, *Curculio caryae*. Plum curculio, *Conotrachelus nenuphar*, a major pest of stone and pome fruits, was found to be extremely susceptible to soil applications of *S. riobrave* IJs in peach and other crops. Additionally, research indicated that the peachtree borer, *Synanthedon exitiosa,* can be controlled with *S. carpocapsae* in a curative or preventative manner, e.g., prophylactic applications of *S. carpocapsae* caused similar levels of suppression compared with standard chemical insecticide sprays. Also, novel formulations that that slow desiccation of IJs are being developed will facilitate incorporation and expanded use of nematodes as microbial control agents. For example, a sprayable gel substantially improved epn efficacy for control of the lesser peachtree borer, *Synanthedon pictipes*, on peach limbs. Overwintering

cocooned codling moth, *Cydia pomonella*, larvae make up 100% of the population from late September until the spring when temperatures allow further development. Control of this stage would drastically reduced damage caused by the moth following emergence in the spring. A wood flour foam formulation and formulated epn infected-hosts, caused superior control of overwintering codling moth, *C. pomonella*, on tree trunks and in mulch, respectively. These new formulations (e.g., sprayable gels or foams) may have widespread benefits for epn application, particularly for targeting pests that attack the tree aboveground.

108. CORDON™, A NEW POST-PLANT USE PATTERN FOR 1,3-DICHLOROPROPENE TO MANAGE NEMATODES IN ESTABLISHED GRAPES. **Shatley[1], Deb, Jim Mueller[2], Jesse Richardson[3], Harvey Yoshida[4] and John Busacca[5]** Dow AgroSciences, LLC; [1]Lincoln, CA 95648; [2]Brentwood, CA 94513, [3]Hesperia, CA 92345; [4]Richland, WA 99352, [5]Indianapolis, IN 46268

There are several plant parasitic nematode species that are important pests in grapes that can negatively impact crop vigor and yield.

Limited options are available for post-plant nematode control in perennial crops and the withdrawal of Nemacur™ from the marketplace has eliminated a viable tool. Dow AgroSciences has tested 1,3-dichloropropene (Cordon™ soil fumigant) as a post-plant application to grapes in California and Washington for several years. Research has been conducted on the same trial sites for as many as four consecutive years. Results show that low use rates and concentrations of Cordon™ applied via drip irrigation as pre-bloom and post harvest applications in established grapes can reduce nematode populations significantly and result in yield increases. No injury to the grapevines was observed in these trials.

Cordon is a registered trademark of Dow AgroSciences LLC, Nemacur is a registered trademark of Bayer CropScience.

109. MOLECULAR DIAGNOSTICS OF POTATO CYST NEMATODES (PCN) FROM THE NATIONAL SURVEY. **Skantar, A.M., Handoo, Z. A., Carta, L.K., and Chitwood, D.J.** Nematology Laboratory, USDA-ARS, Plant Sciences Institute, Beltsville, MD 20705

The potato cyst nematodes (PCN) *Globodera rostochiensis* and *G. pallida* are regulated pathogens of potato, a crop worth nearly $3.9 billion in the United States. Since the initial discovery of *G. pallida* in Idaho in 2006, extensive surveys of the major potato growing acreage have been carried out, to determine the extent of PCN distribution and to ensure that appropriate steps are taken to prevent further infestation. Material suspected to contain PCN cysts is typically analyzed by morphological and molecular means at the USDA Nematology Laboratory in Beltsville, MD. Molecular confirmation of species identity has been achieved through amplification of the internal transcribed spacer (ITS-rDNA) using species-specific multiplex PCR, by analysis of restriction site polymorphisms (PCR-RFLPs), and if necessary, through DNA sequencing. Methods that allow discrimination of morphologically similar tobacco cyst nematode (TCN) from PCN have recently been developed. Assay validation, real-time PCR, and issues relevant to the future of PCN diagnostics will be discussed.

110. MORPHOLOGICAL AND MOLECULAR DESCRIPTION OF *HETERODERA ZEAE* FROM A CORN FIELD IN GREECE. **Skantar, A.M.[1], Handoo, Z. A.,[1] Zanakis G.N.[2] and Tzortzakakis, E.A.[3]** [1]Nematology Laboratory, USDA-ARS, Plant Sciences Institute, Beltsville, MD 20705; [2]Pioneer Hi-Bred Hellas S.A., PO BOX 60196, Thermi, Thessaloniki Greece, [3]Nematology Lab, Plant Protection Institute, N.AG.RE.F., PO BOX 2228, 71003, Heraklion, Crete, Greece.

The corn cyst nematode *Heterodera zeae* was first described from India, where it has wide distribution. This nematode has also been reported from Pakistan, Egypt, Thailand, Nepal, and Portugal. Within the U.S., *H. zeae* was first found in Maryland, primarily in heavy silt-clay soils at fairly low densities. It has since been reported in Virginia. There is limited information regarding nematodes attacking cereals in Greece, and thus far, the only cyst nematode reported was *Heterodera avenae* on wheat. In May 2009, a soil sample containing abundant cysts was taken from an organic corn field in northern Greece; the field was under winter fallow at the time of sampling. Soil from the field site was used to fill 1.5L pots, which were planted with corn (*Zea mays*) and grown in a greenhouse. Females appeared after six weeks incubation, and abundant cysts were present after 12 weeks. Morphological and molecular diagnosis confirmed the presence of *H. zeae*. Amplification of ribosomal DNA markers included the 28S large subunit (LSU) D2-D3 expansion segment and internal transcribed spacer (ITS 1&2 rDNA). Analysis of ITS PCR-RFLP patterns revealed nucleotide variation that was further confirmed by analysis of cloned ITS amplicons. These results are in agreement with prior molecular analysis of *H. zeae* populations from the U.S. and India. This study represents the first record of *H. zeae* in Greece and the second report of this nematode in Europe.

111. DISTRIBUTION AND ESTIMATED ECONOMIC IMPACT OF *PRATYLENCHUS NEGLECTUS*, *P. THORNEI*, AND *HETERODERA AVENAE* ON PACIFIC NORTHWEST WHEAT. **Smiley, Richard W., and G.P. Yan.** Oregon State University, Columbia Basin Agricultural Research Center, P.O. Box 370, Pendleton, OR 97801.

Formal surveys and soil samples submitted to nematode diagnostic laboratories from wheat-producing regions of the Pacific Northwest (PNW; Idaho, Oregon and Washington) and Inter-Mountain West (Montana) have revealed that the most prevalent species of plant-parasitic nematodes occurring in wheat fields include *Pratylenchus neglectus*, *P. thornei*, and

Heterodera avenae. Species with more restricted distribution include *Merlinius brevidens*, *Heterodera filipjevi*, *Meloidogyne chitwoodi*, and *M. hapla*. *Pratylenchus neglectus* and/or *P. thornei*, individually or as mixtures, have been detected in 95% of the wheat fields surveyed in low-rainfall regions of Idaho, Montana, Oregon, and Washington. The potential economic impact of these species was estimated from their distribution and population densities, measurements of intolerance (yield increase following aldicarb application) to each species for the most widely produced cultivars in Idaho, Oregon and Washington, mean number of hectares of those same cultivars harvested over a 5-year interval (2002-2006) in each state, and economic data for wheat harvested in each state during that time interval. Calculations included an adjustment for low-rainfall, rainfed wheat acreage estimated to be infested with *Pratylenchus* sp. densities capable of restricting wheat yield; 42% for *P. neglectus* and 15% for *P. thornei*. Eliminating these species as yield constraints was estimated to have the potential for improving wheat production and profitability in the PNW by 361,100 metric tons (= $51.8 million) annually. Losses from *P. neglectus* were estimated at 87,700 metric tons ($13.1 million) for spring wheat and 143,500 tons ($19.6 million) for winter wheat. Losses from *P. thornei* were estimated at 21,900 metric tons ($3.3 million) for spring wheat and 108,000 tons ($14.8 million) for winter wheat. Comparable estimates were made for *Heterodera avenae*, for which very little data is available from rainfed fields that are not typically assayed for plant-parasitic nematodes and especially for cereal cyst nematodes. Based upon our estimates that 0.04% of the total PNW wheat acreage is infested and grain yield is reduced by an average of 10% in infested fields, we calculated that *H. avenae* reduces wheat production in the PNW by at least 21,000 metric tons, for which the farm-gate economic value was $3.4 million in 2007. Further education of farmers and their commercial and public-sector advisors is required to increase the level of awareness of the distributions and potential economic importance of root-lesion nematodes and cereal cyst nematodes in Pacific Northwest wheat fields. Estimates calculated during these investigations also indicate a strong justification for placing additional emphasis on developing resistant and/or tolerant cultivars and on developing modern DNA-based diagnostic procedures capable of reducing the time and expense required to detect, identify and quantify individual *Pratylenchus* and *Heterodera* species using traditional extraction procedures and morphological characteristics.

112. RESISTANCE OF WHEAT, BARLEY AND OAT TO FOUR PACIFIC NORTHWEST POPULATIONS OF *HETERODERA AVENAE*. **Smiley, Richard W.[1], J.N. Pinkerton[2], G.P. Yan[1], A.L. Thompson[1], and J.A. Gourlie[1].** [1]Oregon State University, Columbia Basin Agricultural Research Center, P.O. Box 370, Pendleton, OR 97801; and [2]USDA-Agricultural Research Service, Horticultural Crops Research Laboratory, Corvallis, OR 97331.

The cereal cyst nematode *Heterodera avenae* occurs in at least seven states of the western USA and significantly reduces yields of small grains in localized regions. Six experiments with wheat, barley and oats were conducted to identify effective sources of resistance against populations of *H. avenae* from southeast Idaho, eastern Oregon, western Oregon, and eastern Washington. Screening in the first test was performed by inoculating sterile soil with 100 juveniles at the time of planting and again at four additional times at 3-day intervals. Each subsequent test used naturally infested soils containing more than 1,000 juveniles per kg of soil. The first three tests included the original screening matrix of 23 wheat, barley, and oat differentials from the 'International Test Assortment for Defining Cereal Cyst Nematode Pathotypes', described by Andersen and Andersen in 1982 and acquired from the Nordic Gene Bank in Alnarp, Sweden. The final three tests were performed with subsets of these differentials supplemented with local cultivars and with wheat lines containing resistance genes described during the past 15 years, and acquired from the Global Wheat Program of the International Maize and Wheat Improvement Center (Ankara, Turkey), the National Small Grains Collection of the USDA-Agricultural Research Service (Aberdeen, ID), and other selected wheat breeding programs in Australia and the USA. Production of gravid cysts was consistently greatest on Nidar and Sun II oats and Arminda and Capa wheat. Nidar II was selected as the standard for maximum susceptibility in these tests. One test was inconclusive due to excess variability among replicates. Another test was inconclusive due to low numbers of gravid cysts being produced in the susceptible controls. However, in all experiments there was a near absence of cyst production (averages of fewer than one per root system) on wheat lines containing the *Cre1* resistance gene (such as AUS10894, Ouyen and Loros), on barley lines containing the *Rha2* (Bajo Aragon 1-1, Martin 403-2, KVL191) and *Rha3* (Morocco) resistance genes, and on oats containing the *R1* resistance gene (Pusa Hybrid BS1, = 640318-40-2-1) and an undescribed resistance gene (IGVH 72-646, = MK H. 72-646). Strong partial resistance was exhibited by wheat lines conferring the *Cre5* (VPM1) and *CreR* (6R6D) resistance genes. Crosses were made to introgress *Cre1* from the agronomically desirable Australian wheat cultivar Ouyen into six Pacific Northwest-adapted wheat cultivars. A non-proprietary molecular marker for use by wheat breeders is also being developed to accelerate the pyramiding of *Cre1* with resistances to root-lesion nematodes, Fusarium crown rot, and with other desirable characteristics.

113. TOLERANCE AND RESISTANCE OF PACIFIC NORTHWEST WHEAT AND BARLEY TO *PRATYLENCHUS NEGLECTUS* AND *P. THORNEI*. **Smiley, Richard W., G.P. Yan, J.G. Sheedy, A.L. Thompson, J.A. Gourlie, and S.A. Easley.** Oregon State University, Columbia Basin Agricultural Research Center, P.O. Box 370, Pendleton, OR 97801.

Root-lesion nematodes (*Pratylenchus neglectus* and *P. thornei*) are widely distributed and reduce wheat yields in low-rainfall regions (250 to 400 mm) of the Pacific Northwest (PNW). Experiments were performed to determine if wheat and

barley cultivars differ in tolerance or resistance to these species. Tolerance experiments were conducted by comparing yields in replicated paired plots that were untreated or treated with aldicarb; 4.2 kg a.i./ha banded 2.5 cm below the seed. Field trials were selected at locations infested with either *P. neglectus* (Heppner, OR) or *P. thornei* (Pendleton, OR). *Pratylenchus* densities averaged to a 45-cm depth in the experimental blocks ranged from 1.5 to 8.2 *Pratylenchus*/g of soil. Two-year mean yields in *P. neglectus*-infested soil were improved ($P<0.01$) over a range from 13% to 99% for 39 individual spring wheat entries and from 7% to 20% for six spring barley entries. In *P. thornei*-infested soil aldicarb increased 3-year mean yields from 6% to 39% for 39 spring wheat entries and from 4% to 19% for six spring barley entries. The commercial cultivars, as a group, were more tolerant than the group of breeding lines, suggesting that criteria for registering cultivars includes a previously unrecognized screening of breeding lines for tolerance to *Pratylenchus*. Winter wheat and winter barley entries did not differ significantly ($P>0.70$) in tolerance to either *Pratylenchus* species over a 3-year interval; 45 entries had mean yield improvements ranging from 5% to 23%. Detection of much tolerance differences among spring but not winter cereals was likely due to the protective effect of aldicarb over a greater proportion of the growing season for spring cereals (5 months) compared to winter cereals (10 months). Resistance reactions for 99 wheat and barley were evaluated in six greenhouse or growth chamber experiments using three replicates of pots inoculated with either *P. neglectus* or *P. thornei* produced in carrot cultures or wheat roots. After 16 weeks of incubation at 22°C the multiplication rate (MR) for each entry was compared to unplanted, inoculated soil and to resistant controls. Maximum MRs varied from 23 to 177 in the six tests, indicating that all PNW wheat cultivars tested were susceptible. Resistant wheat controls included the Iranian landraces AUS28451R and Persia 20, each of which had mean MR less than 1.0 for each *Pratylenchus* species, and an Australian hard white spring wheat line GS50A with MR of 7.9 for *P. neglectus* and 3.5 for *P. thornei*. MRs for 81 PNW wheat entries in these tests averaged more than 15 for both species. MRs for 12 barley entries averaged from 6 to 8 for these *Pratylenchus* species, indicating that most PNW barley cultivars are both more tolerant and resistant than wheat cultivars.

114. OBTAINING AND USING NEMATODE GENOME SEQUENCES. **Sternberg, Paul W., Erich Schwarz, Ali Mortazavi, Adler Dillman, Mihoko Kato,** HHMI and Division of Biology, Caltech, Pasadena, CA 91125, pws@caltech.edu

Advances in DNA sequencing technology and bioinformatics now make it possible to obtain and annotate nematode genomes for a relatively reasonable cost. A nematode genome sequence and associated annotation is useful because it provides a protein set, a better understanding of phylogeny, better DNA diagnostics, and reagents for production of antisera, RNAi reagents and transgenic technology, among other things. Examples will be provided based on our own experience. We have sequenced, assembled and annotated three *Steinernema* species (*carpocapsae*, *scapterisci* and *monticolum*) as well as a sixth *Caenorhabditis* genome (strain PS1010; sp. 3, isolated by Robin Giblin-Davis), the other five species having been done by genome sequencing centers. We have characterized the transcriptomes (the collection of transcribed mRNAs) of PS1010 as well as of *Steinernema carpocapsae* eggs and infective juveniles (IJs) using deep sequencing technology. We have also characterized the transcriptome of the *C. elegans* male linker cell, which guides the developing gonad from mid-body to the cloaca; we have identified ~8000 genes expressed in the linker cell and ~800 genes whose expression is enriched in this cell. An argument will be made for much broader efforts to obtain many nematode genomes to better understand the evolution and diversity of this fascinating and important phylum.

115. DIVERSITY AND PHYLOGENETIC RELATIONSHIPS WITHIN THE GENUS *HELICOTYLENCHUS* STEINER, 1945 (TYLENCHIDA: HOPLOLAIMIDAE) AS INFERRED FROM ANALYSIS OF THE D2-D3 OF 28S rRNA GENE SEQUENCES. **Subbotin[1], Sergei A., R.N. Inserra[2], M. Marais[3], P. Mullin[4], T.O. Powers[4], P.A. Roberts[5], E. Van den Berg[3], G.W. Yeates[6], J.G. Baldwin[5].** [1]PPDC, California Department of Food and Agriculture, 3294 Meadowview Road, Sacramento, CA 95832-1448, USA; [2]Florida Department of Agriculture and Consumer Services, DPI, Nematology Section, P.O. Box 147100 Gainesville, FL 32614-7100, USA; [3]National Collection of Nematodes, Biosystematics Programme, ARC-Plant Protection Research Institute, Private Bag X134, Queenswood, 0121 South Africa; [4]Department of Plant Pathology, University of Nebraska, Lincoln, NE 68583, USA; [5]Department of Nematology, University of California, Riverside, CA 92521, USA; [6]P.O. Box 1758, Palmerston North 4440, New Zealand.

The spiral nematodes of the genus *Helicotylenchus* are globally distributed and associated with the root system of diverse groups of plants in cultivated and uncultivated areas. Several species are considered serious parasites of crops. The identification of many *Helicotylenchus* species is not always reliable, in part because many species share very similar diagnostic characters and high intraspecific variation. To verify species identification of geographically distant populations of *Helicotylenchus*, we tested monophyly of some classical morphospecies and studied their phylogenetic relationships; specifically, we conducted sequence and phylogenetic analysis of eighty-nine sequences of the D2-D3 expansion segments of 28S rRNA gene sequences from fifty-four *Helicotylenchus* isolates, including species identified as *H. brevis, H. digonicus, H. dihystera, H. labiodiscinus, H. leiocephalus, H. martini, H. multicinctus, H. platyurus, H. pseudorobustus,* and *H. vulgaris*, and three outgroup taxa. Phylogenetic analysis distinguished nine highly or moderately supported major clades within

Helicotylenchus. Using the molecular approach we were able to confirm congruence with morphological-based identification of samples of *H. dihystera* and *H. multicinctus*. However, sequence and phylogenetic analysis using Bayesian inference and maximum parsimony analysis showed that isolates collected in different countries and morphologically identified as *H. pseudorobustus, H. digonicus* or *H. vulgaris,* were each representative of several different and, sometimes, unrelated lineages. Molecular analysis revealed that fourteen samples were classified as representatives of eleven unidentified species. Molecular characterization of known *Helicotylenchus* species, especially, using populations collected from type localities, is needed to clarify morphological identification of the species.

116. BIODIVERSITY OF NEMATOPHAGOUS FUNGI IN ISMAILIA GOVERNORATE, EGYPT. **Tamer, S. Abd-El-Moneim and Samia I. Massoud,** Suez Canal University, Faculty of Agriculture, Department of Agricultural Botany, Ismailia, Egypt. 41522

A Survey study was conducted during the season of 2008-2009 through 50 locations representing Ismailia governorate, Egypt, to determine the occurrence of the nematophagous fungi. One hundred rhizosphere soil samples were collected from fruit orchards, ornamentals, field crops and vegetables, followed by isolation of nematophagous fungi by sprinkle technique, while, nematode was extracted from soil by using the modified Baerman technique. The soil pH value, soil texture and organic matters percentages of each soil sample were analyzed to investigate the relationship between these environmental factors and the occurrence of nematode-trapping and parasitic fungi. Three species of two nematophagous fungi genera were found and identified in this study. *Dactylaria brochopaga* (nematode trapping fungus) was found parasitized on second stage (J_2) of root-knot nematode (*Meloidogyne incognita)* in 33% of collected soil samples. *D. brochopaga* was found in soil samples containing organic matters more than 2.5%, while, the occurrence of this fungus reduced when amount of organic matter was less than 2% in some collected soil sample. The optimal pH value for growth of *D. brochopaga* in natural habitat was ranged from 6.5 to 8.9 in loamy sand soil texture. In addition, two species of the Chytridiomycetous fungus *Catenaria* (parasitic fungus on nematode and their eggs), were observed in 10% of collected samples. One of them was identified as *C. anguillulae,* which found parasitized on nematode body causing the break down of the nematode cuticle. While, the other identified as *C. auxiliaris,* which destroying nematode eggs in 3% of collected sample. The suitable pH value for fungus *Catenaria* was ranged from 7.8 to 8.3 in sandy soil. On the other hand, the organic matters haven't any effects on the growth of *Catenaria* or the ability of parasitism.

117. BACTERIAL CYANOGENESIS AND ITS ROLE IN BIOCONTROL OF PLANT-PARASITIC NEMATODES. **Taylor, Christopher G.** The Ohio State University, Department of Plant Pathology

Plant-parasitic nematodes are among the most destructive plant pests, causing substantial economic losses to agronomic crops worldwide. Current methods of using bacteria as biocontrol agents for plant-parasitic nematodes have met with limited success in part due to limited knowledge about mechanisms of biocontrol and biotic factors that are important to rhizosphere persistence. Using a *C. elegans* bioassay we have screened over 8,000 bacterial isolates from a variety of natural sources (water, soil, plants) and identified over 60 different isolates of *Pseudomonas* that interfere with nematode growth and development. Nearly a third of these strains exhibit activity in plate and soil assays against root-knot and soybean cyst nematodes. We have characterized the nematode-active *Pseudomonas* isolates for motility, exoprotease activity and production of siderophores, hydrogen cyanide (HCN), polysaccharides, and fluorescence to determine if commonalities exist among plant-parasitic nematode lethal strains. Using a transposon knockout strategy, we identified several *C. elegans* non-lethal isolates for *Pseudomonas* strain 15G2. Testing of the non-lethal transposon tagged isolates for HCN showed significant reduction in HCN production. Loss of HCN production was correlated with reduced capacity to protect plants from plant-parasitic nematodes. These data indicate that HCN is potentially an important compound produced by pseudomonads within the rhizosphere with activity against plant-parasitic nematodes.

118. IDENTIFICATION AND CHARACTERIZATION OF TRANSFER CELL-REGULATED GENES IN ARABIDOPSIS AND MAIZE. **Taylor, Christopher G.** The Ohio State University, Department of Plant Pathology

Plant systems adapt to the mass flow of nutrients across apoplastic spaces through the development of specialized cells called transfer cells. These cells are found in all taxonomic plant groups and are formed during organ development. In maize seed development, transfer cells form in the developing endosperm adjacent to maternal vascular tissues of the cob. In plant pathology, root-knot nematodes (RKN) induce the formation of specialized feeding site consisting of several cells called ''giant cells'' (GC) with transfer cell-like characteristics. Due to their functional similarities, transfer cells from seeds and nematode feeding sites share many of the same morphological characteristics including thickened and highly invaginated cell walls, dense cytoplasm, abundant ER, and numerous small vacuoles and mitochondria. These similarities have motivated us to use a comparative genomic approach towards the identification of genes that are expressed in these cells and determine their function. Using laser microdissection technology we are isolating RNA from a time-course series of transfer cells from developing seeds of maize (basal endosperm transfer layer) as well as GC of Arabidopsis and maize nematode-infested roots. Microarrays of captured RNA are being generated and will be compared to identify transfer cell-specific genes. Molecular

and biochemical analysis of Arabidopsis and maize mutants will provide information on the role of these genes in transfer cells. Reduction in plant growth, seed filling or nematode parasitism is some of the changes we are monitoring.

119. THE EFFECTS OF SOYBEAN CYST NEMATODE INFECTION AND ELEVATED CO_2 ON SOYBEAN GROWTH AND PHYSIOLOGY. **Tefft, Paul M., A. Bertram, S. Martell, H. Pham, M. Chorney, and C.J. Springer.** Biology Department, Saint Joseph's University, Philadelphia, PA 19131

Heterodera glycines, the Soybean Cyst Nematode (SCN), is a common pest of *Glycine max* (soybean) with severe infections resulting in near total crop failure. Despite these large losses in yield, little is known about changes in the physiology and morphology of soybeans as a result of infestation with SCN. Possibly confounding our understanding of parasite-host physiology is increasing atmospheric carbon dioxide concentrations ($[CO_2]$). It is known that increased atmospheric $[CO_2]$ significantly affects the growth, morphology and physiology of higher plants, especially soybean. Surprisingly little is known about the interaction of this important global change phenomenon with this important crop parasite. Our objective was to examine the physiological and morphological effects of elevated carbon dioxide concentration on soybean plants infected with SCN during early and late infection. Soybean plants were grown from seed in sterile sand and inoculated with SCN eggs or water that served as a control and grown at current (400 ppm CO_2) or elevated (1000 ppm CO_2) atmospheric $[CO_2]$. The soybeans were harvested after 4 weeks growth in one set of experiments (short-term study) and after eight weeks of growth in another (long-term study). Just prior to harvesting, we measured photosynthetic CO_2-response curves and chlorophyll. We also measured leaf number, developmental stage, and total leaf area as well as stem, leaf, and root mass. Finally, we recorded the number of juvenile SCN found in plant roots. All data were analyzed using a two-way ANOVA. In the short-term study we found no significant difference in the number of SCN juveniles present in roots between current and elevated CO_2-grown plants. However in the long-term study growth at elevated CO_2 resulted in significantly greater number of juvenile SCN in roots. This differential infection between the short-term and long-term studies resulted in different responses between the two studies as well. For example, in the short-term study the effect of infection with SCN on plant biomass did not depend on the growth CO_2 concentration but in the long-term study we did find a statistically significant interaction between growth CO_2 concentration and SCN infection. Infection was more damaging to plants grown at elevated CO_2 than current CO_2. Total leaf area also exhibited this same pattern of response in both studies. Chlorophyll concentrations were not significantly affected by either SCN infection or growth at elevated CO_2 in either study. The response of photosynthetic capacity to elevated CO_2 was also dependent on SCN infection. This interaction was characterized by no difference in the photosynthetic capacity of non-infected control plants between current and elevated CO_2. However with SCN infection photosynthetic capacity was lower in both current and elevated CO_2-grown plants but this decrease was much more severe under elevated CO_2. Generally, the effects of SCN infection on soybean growth and physiology were more severe under elevated CO_2 than current CO_2.

120. RESISTANT WILD WATERMELON ROOTSTOCKS USEFUL FOR MANAGING *MELOIDOGYNE INCOGNITA* IN GRAFTED WATERMELON. **Thies[1], Judy A., A. Levi[1], J.J. Ariss[1], C.S. Kousik[1], and R.L. Hassell[2].** [1]U.S. Vegetable Laboratory, USDA, ARS, 2700 Savannah Highway, Charleston, SC 29414; [2]Coastal Research and Education Center, 2700 Savannah Highway, Charleston, SC 29414.

The southern root-knot nematode (*Meloidogyne incognita*) significantly reduces watermelon yields in the southern U.S. The primary method for controlling root-knot nematodes in watermelon and other high value vegetable crops has been pre-plant fumigation of soil beds with methyl bromide or 1,3-dichloropropene. However, environmental concerns related to use of fumigant pesticides and increased costs of these products have stimulated interest among growers and watermelon breeders to consider using rootstocks for managing root-knot nematodes in watermelon. 'Tri-X 313' seedless watermelon (*Citrullus lanatus* var. *lanatus*) scions were grafted on ten cucurbit rootstocks and evaluated for their performance in a field infested with *M. incognita* in Charleston, SC in 2009. Five wild watermelon (*Citrullus lanatus* var. *citroides*) germplasm lines developed at the U.S. Vegetable Laboratory (RKVL 301, RKVL 302, RKVL 303, RKVL 316, and RKVL 318), one commercial watermelon rootstock (*C. lanatus* var. *citroides* 'Ojakkyo'), one squash hybrid (*Cucurbita moschata* x *C. maxima* 'Strong Tosa'), one bottle gourd (*Lagenaria siceraria* 'Emphasis'), and three wild tinda (*Praecitrullus fistulosus*) rootstocks were evaluated. Self-grafted 'Tri-X 313' and non-grafted 'Tri-X 313' watermelon were included as controls. *Meloidogyne incognita* infection was severe in 'Strong Tosa' hybrid squash, 'Emphasis' bottle gourd, and the three wild tinda rootstocks with percentages of root system galled ranging from 86 to 100%. The five RKVL wild watermelon lines exhibited significantly lower ($P<0.05$) percentages of galling (range: 9.1 to 16.2%) than non-grafted 'Tri-X 313' (40.9%), 'Emphasis', 'Strong Tosa', and wild tinda rootstocks. Root systems of RKVL 301, RKVL 303, RKVL 316, and RKVL 318 wild watermelon rootstocks had significantly more fibrous roots ($P<0.05$) than those of all other entries. The grafted RKVL 318 produced significantly more ($P<0.05$) fruit (12 per plot) than all other entries (mean = 5.3 per plot), and it produced a heavier ($P<0.05$) fruit yield (29.5 lbs per plot) than all entries except self-grafted 'Tri-X 313' (21.5 lbs per plot). These results suggest that the RKVL wild watermelon rootstocks (*C. lanatus* var. *citroides*) possess durable resistance to root-knot nematodes and may provide a new alternative to pre-plant soil fumigation for managing root-knot nematodes in watermelon.

121. SUITABILITY OF ANNUAL WEEDS AS HOSTS OF *MELOIDOGYNE INCOGNITA* IS UNAFFECTED BY COINFECTION WITH *VERTICILLIUM DAHLIAE*. **Thomas[1], Stephen H., J. M. Trojan[1], J. Schroeder[1], C. Fiore[1], S. Sanogo[1], L. Liess[1], N. Schmidt[2] and L. W. Murray[3]**. [1]Dept. of Entomology, Plant Pathology and Weed Science, and [2]Dept. of Economics and International Business, New Mexico State University, Las Cruces, NM 88003 and [3]Dept. of Statistics, Kansas State University, Manhattan, KS 66505.

In 2007 the three predominant annual weed species encountered in chile pepper (*Capsicum annuum*) fields in Luna County, NM were all found to be infected with *Verticillium dahliae*. These weeds included spurred anoda (*Anoda cristata*, = SA), Wrights groundcherry (*Physalis wrightii*, = WG) and tall morningglory (*Ipomoea purpurea*, = TM), none of which expressed symptoms of Verticillium wilt that were prevalent in surrounding chile plants. All three weeds are also known hosts of *M. incognita*, with symptoms of nematode infection being evident on roots of TM and chile in some of the affected fields. The limited expression of symptoms of *V. dahliae* infection among weeds led us to hypothesize that weeds may serve as refugia that maintain or enhance populations of certain pathogens that can be injurious to chile. Greenhouse experiments were conducted during the summers of 2008 and 2009 to determine the effects of *M. incognita* and *V. dahliae*, alone and in combination, on growth of SA, WG, TM, and chile compared to non-inoculated control plants. *Meloidogyne incognita* reproduction, *V. dahliae* infection confirmed by culturing stem sections and roots, and plant shoot and root growth proportional to that of non-inoculated control plants were measured six weeks post-inoculation. In 2008 preliminary results showed shoot and root biomass of chile were not affected by *M. incognita* alone, but were reduced 26% and 63% respectively by *V. dahliae* and the two pathogens combined. Nematode reproduction on chile was largely unaffected when plants were co-infected with *V. dahliae* either year, despite reductions in chile root biomass in 2008. Overall, *M. incognita* reproduction levels were similar among chile, TM and WG in both studies. Reproduction factors (nematode populations 45 days post-inoculation divided by inoculum level) ranged from 60-83 in 2008, but declined to less than one tenth of that level in 2009 – possibly due to differences in plant size at time of inoculation. Spurred anoda was a poorer host for *M. incognita* than TM or WG in both experiments. Nematode reproduction was slightly greater in all three weed species when plants were co-infected with *V. dahliae* in 2008, but the trend was not evident in 2009. These results demonstrate that all three weeds will support both pathogens, alone or in combination, without suffering pathogenic effects, and that TM and WG support levels of *M. incognita* reproduction similar to those found in highly-susceptible chile plants. Effective management of the weed hosts examined in this study may help reduce populations of both pathogens in future crops.

122. *MELOIDOGYNE HAPLA*, A GENETIC MODEL SYSTEM FOR PLANT PARASITIC NEMATODES **Thomas, Varghese P. and Dr. V.M. Williamson** Department of Nematology, University of California, Davis

Meloidogyne hapla (northern root knot nematode) is an economically important obligate biotrophic pathogen of cool season crops. Availability of a genetic map and completion of the genome sequence makes it an excellent system for investigating inheritance of important nematode traits. In current progress, we were able to identify Single Nucleotide Polymorphisms (SNPs) between parental strains (*M. hapla* strain VW9 and VW 8) used in genetic map construction. 2 sets of 96 SNPs were scored for segregation among 183F2 progenies on Illumina Golden Gate Bead Express System at UC Davis Genome Center Facility. This has helped in merging the genetic map and the genome sequence by positioning physical contigs (contiguous sequences) to the genetic map. Current genetic map contains approximately four hundred AFLP and SNP marker loci and is the densest map produced for any plant parasitic nematode. Important nematode parasitism traits such as attraction, reproduction, galling etc. on a particular host, *Solanum bulbocastanm* (wild potato) were identified to segregate as quantitative traits and have also been mapped to current genetic map. The integration of genetic and physical map along with identification of loci involved in parasitism related traits will facilitate in identifying important nematode genes.

123. ECOMETAGENETICS FOR THE ANALYSIS OF NEMATODE COMMUNITIES IN A TROPICAL RAIN FOREST. **Thomas, W. Kelley[1], D.L. Porazinska[2], R.M. Giblin-Davis[2], W. Sung[1], H. Bik[1], and D.R. Fournier[3].** [1]Hubbard Center for Genome Studies, 35 Colovos Rd., University of New Hampshire, Durham, NH 03824, [2]University of Florida-IFAS, Fort Lauderdale Research and Education Center, 3205 College Ave., Davie, FL 33314, and [3]Department of Computer Science, Kingsbury Hall, University of New Hampshire, Durham, NH 03824.

Because traditional approaches using morphology and single nematode PCR followed by Sanger sequencing are prohibitive for timely nematode community analyses at the species level, we developed an ecometagenetic approach using multiple nematode DNA PCR followed by next generation sequencing. Although ecometagenetics ultimately provides remarkably accurate information about quantitative and qualitative aspects of the nematode community at the species level, issues associated with between-species sequence number variation, within-species sequence variation, OCTU (operational clustered taxonomic unit)-species linkage, and sequence recombinants can obscure and complicate the process of data analysis and understanding. Here, we discuss how these issues can be identified and overcome for a successful analysis of a tropical nematode community.

124. AN 11-YEAR FIELD STUDY WITH *PASTEURIA PENETRANS*: LESSONS LEARNED THE HARD WAY. **Timper[1], Patricia**. [1]USDA ARS, P.O. Box 748, Tifton, GA 31793.

Beginning in 1998, a bioassay using second-stage juveniles (J2) from a greenhouse (GH) population of the root-knot nematode *Meloidogyne arenaria* (Ma) was used to monitor endospore densities of the bacterium *Pasteuria penetrans*, which was parasitizing Ma in a long-term rotation study (begun in 1991). Spore densities of the bacterium were very high in continuous peanuts (21 attached spores/GH J2) and very low when peanut was rotated with non-hosts for Ma (0.5 spores/GH J2). Lesson 1: even the most straightforward studies do not always turn out as planned. In 2000, the rotation sequences were changed to include additional hosts for the nematode. Based on previous studies, spore densities of *P. penetrans* were expected to increase rapidly (<4 years) where hosts for the nematode were frequently planted. However, spore densities did not increase after 8 years in continuous peanut (0.3 spores/GH J2). Lesson 2: do not underestimate the genetic diversity of root-knot nematodes and *P. penetrans*. To determine the susceptibility of Ma to *P. penetrans*, five single egg-mass (SEM) lines from the field population of the nematode were compared to the GH population for acquisition of endospores from the field soil. Four of the five SEM lines acquired 9 to 14 spores/J2; whereas, the GH population and one of the SEM lines acquired 3.5 and 1.8 spores/J2, respectively. These results indicate that the field population of Ma is heterogeneous for attachment of *P. penetrans* spores. Lesson 3: do not rely exclusively on a bioassay to determine abundance of *P. penetrans*. Had native J2 in the soil been examined, an increase in spore attachment over time may have been observed. In 2008, spore densities estimated with the four receptive SEM lines were highest in plots with continuous peanut (14-20 spores/J2), intermediate with two consecutive hosts (6-7 spores/J2), and lowest with only one host in a 3-year rotation (<1 spore/J2). Therefore, spore densities had increased under intensive cropping of hosts for Ma, but the GH population of the nematode was not receptive to spore attachment. However, previously, the GH population was very receptive to spore acquisition from this field site. One explanation for this inconsistency is that the Ma population in the field became resistant to the dominant subpopulation of *P. penetrans* that had been present and this led to the selection of a different subpopulation of the bacterium (now dominant in the field) that is incompatible with the GH population.

125. LOCALIZATION OF MICROBIAL NUCLEIC ACIDS INSIDE SOIL NEMATODES. **Treonis, Amy M., E.H. Michelle, C.A. O'Leary, E.E. Austin, and C.B. Marks.** Department of Biology, University of Richmond, 28 Westhampton Way, Richmond, VA 23173.

Microorganisms (i.e., prokaryotes, fungi) are food sources for soil nematodes, as well as potential mutualists or pathogens. Understanding the linkages between microorganism and invertebrate diversity in soils requires the ability to distinguish between these microbial roles. We tested the potential of a taxa-specific fluorescent *in situ* hybridization (FISH) procedure for identifying and localizing microbial rRNA within the bodies of soil nematodes. Our objective was to determine whether the rate of digestion permitted detection and identification of food source nucleic acids within the nematode digestive system (i.e., pharynges, intestines). First, using laboratory cultures of *Caenorhabditis elegans* maintained on *Escherichia coli*, we were able to localize bacterial rRNA throughout the nematode pharynx with the universal bacterial probe EUB338, although never in the intestines. Second, we applied the fungal rRNA probe FR1 to *Aphelenchus avenae* cultured on the fungus *Rhizoctonia solani*. We were unable to detect fungal rRNA within these nematodes, and it appears that this material may be digested rapidly. Next, we applied our technique to nematodes extracted directly from soils. We were able to localize bacterial rRNA within the pharynges of bacterial-feeding species of nematodes (cephalobs) from Mojave Desert soils. We also localized archaeal rRNA using the probe ARC344. Finally, application of EUB338 to desert soil nematodes revealed the presence of intestinal and ovarian symbionts, albeit inconsistently. This technique has great potential for use in understanding the feeding behavior of bacterial-feeding soil nematodes and also has an important role in studies of nematode:bacterial symbioses.

126. MAXIMIZING EFFECTIVENESS OF EXTENSION EDUCATION EFFORTS. **Tylka, G.L.** Dept. of Plant Pathology, 351 Bessey Hall, Iowa State University, Ames, IA 50011.

Educating growers about plant-parasitic nematodes requires university extension educators to have in-depth knowledge of the biology of nematodes, their host plants and the available management options. Also, extension educators must be able to effectively communicate technical information to nontechnical audiences. But two additional factors can affect the success of extension education efforts. The medium used to deliver information to growers is very important. University and agribusiness personnel routinely access information through computers, smartphones and the Internet. But only 40% of U.S. crop producers surveyed in 2009 reported using a computer ''for farm business.'' Computer usage by growers has steadily increased over the past two decades, however most growers still prefer to receive crop production and protection information in hard copy. Another important factor to consider is the target audience of extension education efforts. Traditionally, extension programs and publications have been aimed at growers. But in some situations, there may be a more important audience to target. In Iowa, more than 90% of surveyed corn and soybean growers indicated they first turn to certified crop advisers, or CCAs, working for agribusinesses to get crop information; 80% of surveyed Iowa CCAs indicated they first turn to Iowa State University for this information. So an efficient way to educate Iowa crop producers is through partnerships with agribusinesses. Grower preferences for receiving information and the structure of the crop industry will vary among

geographic regions and cropping systems. But these basic factors of the educational process should be considered to maximize extension education efforts.

127. BUILDING CAPACITY IN PLANT NEMATOLOGY IN SUB – SAHARAN AFRICA: CONTRIBUTIONS BY NEMATOLOGY INITIATIVE OF EAST AND SOUTHERN AFRICA (NIESA). **Waceke[1], Wanjohi J., J.W. Kimenju[2], Z. Sibanda[3], H. Talwana[4]** [1]Dept of Agricultural Science and Technology, Kenyatta University, P.O. Box 43844 -00100 Nairobi, Kenya, [2]Dept of Crop Science and Protection, University of Nairobi, P.O. Box 29053 - 00625 Nairobi, Kenya, [3]26 Langbourne Avenue. Mt Pleasant, Harare. Zimbabwe, [4]Dept of Crop Science, Makerere University, P.O. Box 7062 Kampala, Uganda

Plant nematology in sub - Saharan Africa continues to lag behind other disciplines mainly because those trained in the discipline spend a significant proportion of their time in other disciplines and lack peer support as most of the trained nematologists work on their own. Further to this, there is inadequate support from policy makers due to the perception that the nematodes are not as important as other biotic and abiotic stresses and lack of awareness by the farmers of even the existence of the nematodes. Due to this, yield losses of up to 60% on cash and food crops are common in the region. A survey conducted in 1996 by CABI Bioscience aimed at identifying sources of taxonomic expertise in nematology revealed that outside of South Africa, there were only two practicing nematode systematists in Africa. Another survey commissioned by Gatsby Charitable Foundation (GCF) in 2003 to determine capacity needs in plant nematology in East and Southern African region revealed similar findings. To address this need, GCF funded a five year regional project (2005- 2010) on capacity building in plant nematology. This project covered Kenya, Malawi, Uganda, Tanzania and Zimbabwe, and received technical support from CABI Bioscience, Rothamsted International and the University of Reading in the UK. One of the project results was the establishment of NIESA in August 2005 with the primary aim of promoting plant nematology in the region. Since its inception, four MSc and three PhD students have completed their studies. Sixty scientists from Tanzania, Kenya, Mozambique, Uganda, Zimbabwe, Malawi, Southern Sudan and Rwanda have also been trained through workshops. The initiative has also developed a plant nematology training manual; established and fully equipped six nematology laboratories in the region; developed an interactive website (http://www.africannematology.info/index.asp); established links with Nematological Society of South Africa (NSSA) and undertaken several joint research projects. The capacity building efforts by NIESA have, however, focused mainly on postgraduate and research scientists, and there is still a need for vocational training targeting extension officers, NGOs and farmer groups. An advocacy program targeting decision and policy makers is needed to convince them about the damaging effects of nematodes, and the need for increased investment on soil health.

Whereas most of the nematology research conducted in the region has centered on the control and management, there is minimal work on taxonomy due to the lack of trained taxonomists. The scope needs to be broadened to cover all aspects of plant nematology and other biotic and abiotic interacting factors within the context of soil health.

128. TELONE II USE FOR NEMATODE CONTROL IN SOUTHEASTERN FIELD CORN. **Weiss, Anthony W.[7], John Busacca[1], Dennis Lane[1], Bob Kemmerait[2], Matt Hoffman[3], Stan Childers[4], Edd Harrision[5], Rome Ethredge[6].** [1]Dow AgroSciences, Indianapolis, IN. [2]University of Georgia, Tifton, GA. [3]Dow AgroSciences, Tallahassee, FL. [4]S & J Tiger Enterprises LLC, Central, SC. [5]County Extension Agent, Camilla, GA. [6]County Extension Coordinator, Donalsonville, GA. [7]Dow AgroSciences, Brandon, FL

Nematodes are an endemic problem in southeastern United States soils for many crops. Seed corn companies have strived for better yields but the new hybrids have not breed in any tolerance to nematodes. As corn acreage and value have grown, the problem of nematode damage has become more important. Therefore, growers are looking for ways to manage this devastating pest with effective and economical control options.

Field trials have been conducted for two years, looking at the effectiveness of Telone® II soil fumigant in managing nematodes in field corn (*Zea mays)*. The three primary nematode species identified are sting (*Belonolaimus gracilis*), stubby root (*Paratrichodorus* minor), and root knot (*Meloidogyne incognita*) nematode. Test results have shown that pre-plant applications of Telone II effectively controls (or reduces nematode populations) nematodes, which results in improved early season crop vigor, earlier crop maturity, increased yields and higher net profits when damaging nematode populations are present. ®*Trademark of Dow AgroSciences LLC*.

129. CORKY RING-SPOT DISEASE OF POTATO CONTROL IN MICHIGAN. **Wernette[1], Loren, G. Bird[1], W. Kirk[2], J. Davenport[1]** [1]Department of Entomology, Michigan State University East Lansing, MI, 48824, [2]Department of Plant Pathology, Michigan State University, East Lansing, MI 49324

Corky ring-spot disease (CRSD) of potato has been identified in two widely separated locations in MI. CRSD causes necrotic ringing and arching of tuber tissue and can lead to crop rejections at processing plants. CRSD is caused by Tobacco Rattle Virus (TRV), which has an RNA genome and a two-part rigid rod shaped particle. TRV is vectored by stubby-root nematodes (SRN). *Trichodorus proximus, T. primitives, T. similis, Paratrichodorus christiei, P. atlanticus, P. prosus,* and *P. pachydermus* are known to exist in MI. The focus of this two-year research project was to evaluate the efficacy of different

nematicide application tactics to control the nematode vector. The research was conducted in White Pigeon, MI, in one of the two fields known to produce CRSD infected tubers. TRV from this field was confirmed and reported in 2007, in Plant Disease Volume 92(11). In 2010, the complete genomic sequence of TRV for this site was published in Archives of Virology 155:621-625. In 2008, we evaluated metam sodium (Sectagon 42), 1,3-D (Telone II) and oxamyl (Vydate C-LV) at different combinations and rates. In 2009, the study was repeated in an adjacent side of the same field using the same nematicides at different timings and rates. 1,3-D was not included due to its lack of efficacy in the 2008 trial. In both years, yields were relatively high across all treatments, with annual means of 458 cwt and 466 cwt, respectively. This is an indication that this field does not suffer from Potato Early Die syndrome, which is common in most potato production areas in MI. After harvest, tubers were stored at 1ÖC and rated multiple times throughout the winter for symptom expression. In 2008, the symptom expression ranged from 0% to 12.5%. In 2009, symptom expression ranged from 0% to 10.5% with the higher symptom expression associated with the non-treated control and lower chemical dosages. Samples of infected tubers were saved after each sampling date and TRV was molecularly confirmed by two separate laboratories. Infected tuber tissue was also observed under transmission electron microscopy for diagnostic confirmation. Throughout the growing seasons, less than 1.0 SRN were recovered per 100 cm^3 of soil in five sets of soil samples. Our research indicates that the most effective treatment to control CRSD is metam sodium. In both years, treatments containing metam sodium had significantly less CRSD symptoms compared to the control and the other treatments. (Oral Graduate Student Competition)

130. SCREENING OF MICROORGANISMS ASSOCIATED WITH CYSTS OF *GLOBODERA PALLLIDA* ISOLATED FROM SOUTHERN IDAHO FIELDS. **Worapong, J., C. Bates, X. Gao, B. King, J. B. Johnson and R. S. Zemetra.** Department of Plant, Soil, and Entomological Sciences, University of Idaho, Moscow, Idaho 83844.

The white Potato cyst nematode (*Globodera pallida*, wPCN,) was recently discovered in potato production fields in Idaho. Chemical fumigations have been used to control the current outbreak of this nematode but a longer term, more environmentally friendly method for management of wPCN such as biological control would be desirable to prevent potential future outbreaks. Therefore, an experiment was initiated to isolate microorganisms from field-collected wPCN cysts that showed a low hatch rate. Each cyst was put into a well of a 96-welled plate containing potato dextrose broth (PDB). Wells with cysts showing mycelium and/or turbidity of PDB were recorded as colonized. Among 65 field cysts, 23 were found with mycelium on the surface of the cysts and 5 wells showed turbidity. In addition, one cyst had mycelium on its surface and flocculate precipitate in its well. Total colonization was 45% that could be divided into 35% with fungal colonizing, 8% bacterial colonizing and 2% with both fungal and bacterial colonizing. Based on ITS1-5.8S-ITS2 sequences, a majority of 9 fungal species and 2 Oomycota species colonizing the surface of PCN cysts were identified as 2 *Cylindrocarpon destructans*, 2 *Cylindrocarpon macrodidymum*, 2 *Fusarium redolens*, 2 *Fusarium tricinctum*, a species of *Microdochium bolleyi*, 3 *Plectosphaerella cucumerina*, 5 *Torula herbarum*, an isolate of *Verticillium dahlia*, a strain of *Volutella ciliate*, an oomycetous species of *Pythium oligandrum* and 3 oomycetous isolates of *Geomyces pannorum*. Based on partial 16S sequences, bacteria colonizing the surface of wPCN cysts were identified as *Flavobacterium* sp., *Paenibacillus amylolyticus*, *Paenibacillus xylanexedens*, *Pseudomonas fluorescens*, *Pseudomonas synxantha* and *Streptomyces tauricus*. Of those *Plectosphaerella cucumerina, P. fluorescens* and *P. synxantha* have been reported as successful wPCN biocontrol agents in Europe and consequently, a potential use for biocontrol agents of wPCN. *In vitro* Koch's postulate tests including pathogenicity tests of these associated microbes on Désirée potato cultivars under greenhouse conditions will be conducted to study the effect of these wPCN associated microorganisms on their ability to colonize the cyst and parasitize the enclosed eggs and juveniles.

131. ISOLATION AND CHARACTERIZATION OF A GHF5 β-1,4-ENDOGLUCANASE FROM THE RENIFORM NEMATODE (*ROTYLENCHULUS RENIFORMIS*). **Wubben[1], Martin J. and F.E. Callahan[1].** [1]USDA-ARS, Crop Science Research Laboratory, Genetics and Precision Agriculture Research Unit, P.O. Box 5367, Mississippi State, MS 39762-5267.

The reniform nematode (*Rotylenchulus reniformis*) is a semi-endoparasitic root pathogen of >300 plant species, including cotton, soybean, and pineapple. Plant-parasitic nematode (PPN) penetration of the root epidermis is facilitated by a collection of cell wall degrading enzymes that are secreted from the esophageal gland cells. Glycosyl hydrolase family 5 (GHF5) β-1,4-endoglucanses, a.k.a. cellulases, comprise a significant portion of this collection. Recently, *R. reniformis* cDNAs were identified as part of an expressed sequence tag project that showed similarity to a Heterodera glycines cellulase cDNA. In this report, we describe the isolation and characterization of a predicted GHF5 β-1,4-endoglucanse gene from *R. reniformis*. The full-length Rr-eng-1 cDNA was 1,341 nucleotides (nt) long and was comprised of a 19 nt 5'-untranslated region (UTR), a 1,242 nt open reading frame (ORF), and an 80 nt 3'-UTR. Forward and reverse PCR primers specific to the 5'- and 3'-UTRs, respectively, amplified an Rr-eng-1 genomic sequence of 2,325 nt. Alignment of the cDNA and genomic sequences revealed 7 introns and 8 exons for Rr-eng-1. BLASTN analysis showed the Rr-eng-1 cDNA was most homologous to the H. glycines cellulase Hg-eng-6 (E=5.0e-121). The *Radopholus similis* cellulase precursor eng1A was the next most homologous sequence at the DNA level with an E value of 2.0e-18. A Southern blot probed with DIG-labeled Rr-eng-1 cDNA suggested a total of three Rr-eng-1-like sequences were present in the *R. reniformis* genome. Translation of the Rr-eng-1 ORF yielded a 414 amino acid peptide having an N-terminal signal sequence for secretion as determined by SignalP3.0. No cellulose

binding domain (CBD) was detected in the RR-ENG-1 protein; however, a putative CBD linker sequence N-terminal to the GHF5 cellulase domain was present. RR-ENG-1 was most homologous to HG-ENG-6 and to cellulases from migratory PPNs. Quantitative reverse-transcription PCR indicated that Rr-eng-1 expression was highest in later juvenile stages and vermiform infective females; however, Rr-eng-1 transcript was detected in total RNAs isolated from R. reniformis eggs, second-stage juveniles, and sedentary parasitic females.

132. DIFFERENTIATION OF *HETERODERA FILIPJEVI* AND *H. AVENAE* USING PCR-RFLP AND CYST MORPHOLOGY. **Yan, Guiping, and R.W. Smiley.** Oregon State University, Columbia Basin Agricultural Research Center, P.O. Box 370, Pendleton, OR, 97801.

The cereal cyst nematodes *Heterodera avenae* and *H. filipjevi* impede wheat production in the Pacific Northwest (PNW). *H. avenae* was first reported in Oregon during 1974 and now occurs in many cereal-producing regions, including the states of Idaho, Oregon, and Washington. High populations of *H. avenae* in commercial fields have reduced winter wheat yields as much as 50% and occasionally destroyed recropped spring wheat in Oregon. *H. filipjevi* was first reported in North America following its discovery during 2008 in a winter wheat field in Oregon. Large patches of stunted plants with up to 90% plant mortality occurred in that field. The use of wheat cultivars that are both resistant and tolerant offers the most effective, economic, and environmentally friendly option to control damage from these nematodes. However, individual wheat cultivars may differ in their ability to resist different species of these nematodes. Accurate identification of cyst nematode species and awareness of high population density in affected fields are therefore essential for selecting cultivars with effective resistance. *H. filipjevi* is closely related to *H. avenae* and only minor morphological differences are available to differentiate them from each other. Distinction between these species, based on morphology, is time-consuming and difficult. Molecular tools have provided the possibility for quick and precise identification of cyst nematode species and subspecies. These species were clearly differentiated using polymerase chain reaction restriction fragment length polymorphism (PCR-RFLP) of internal transcribed spacer (ITS)-ribosomal (r)DNA with up to six restriction endonucleases (*Taq*I, *Hin*fI, *Pst*I, *Hae*III, *Rsa*I, and *Alu*I). The method was validated by inspecting underbridge structures of cyst vulval cones. A distinct underbridge with bifurcated arms was present in vulval cones of the *H. filipjevi* cysts whereas no underbridge was found in *H. avenae*. The species identity of *H. filipjevi* was also confirmed by the sequences of the ITS-rDNA. Grid soil sampling of an Oregon field infested by both species revealed that *H. filipjevi* was present at most of the infested grid sites but mixtures of *H. avenae* and *H. filipjevi* also occurred. These procedures also detected and differentiated *H. filipjevi* and *H. avenae* in soil samples from nearby fields in Oregon and *H. avenae* in samples from Idaho and Washington. Intraspecific polymorphism was not found within *H. filipjevi* or PNW *H. avenae* populations based on the ITS-rDNA. However, intraspecific variation was detected between *H. avenae* populations occurring in the PNW and France. Methods described here will improve detection and identification efficiencies for species of cereal cyst nematodes in fields of wheat, barley, oat and other susceptible small grains

133. GENETIC CHARACTERIZATION OF RESISTANCE TO *PRATYLENCHUS THORNEI* IN WHEAT. **Yan, Guiping, R.W. Smiley, J.A. Gourlie, and A.L. Thompson**. Oregon State University, Columbia Basin Agricultural Research Center, P.O. Box 370, Pendleton, OR, 97801.

The root-lesion nematode *Pratylenchus thornei* is an economically damaging pathogen of wheat in the Pacific Northwest (PNW). This nematode occurs in high numbers in many wheat fields and is hosted by all crops commonly produced in rainfed regions of the PNW. Growing resistant and tolerant cultivars of wheat is the best approach to minimize yield loss. All PNW wheat cultivars tested thus far have been found to be susceptible. To incorporate resistance into PNW-adapted cultivars, crosses were made between three PNW cultivars (Alpowa, Louise, Otis) and GS50a, an Australian wheat line with resistance to *P. thornei*. Resistance evaluations were performed under controlled greenhouse conditions using five check cultivars, parents, and 203 F_2 recombinant inbred lines (RILs) and 194 F_3 RILs of GS50a x Alpowa, 102 F_2 RILs of GS50a x Otis, and 120 F_2 RILs of GS50a x Louise, each inoculated with 1,500 or 2,000 *P. thornei*/kg of soil. RIL populations segregated in a continuous variation indicating that resistance in GS50a was quantitatively inherited. The heritability estimate for the resistance was 0.68. The frequency distribution of F_2 and F_3 progeny suggested that the resistance in GS50a is controlled by more than one gene and is additive in gene action. F_3 RILs of GS50a x Alpowa showing levels of resistance or susceptibility higher than their parents will be used in bulk segregant analysis for molecular marker development. The highly resistant lines obtained in this study may provide a source of resistance for breeding wheat varieties with improved genetic resistance to *P. thornei*.

134. IDENTIFICATION AND QUANTIFICATION OF *PRATYLENCHUS NEGLECTUS* AND *P. THORNEI* FROM SOILS IN THE PACIFIC NORTHWEST USING REAL-TIME POLYMERASE CHAIN REACTION. **Yan, Guiping[1], R.W. Smiley[1], and P.A. Okubara[2].** [1]Oregon State University, Columbia Basin Agricultural Research Center, Pendleton, OR 97801; [2]USDA-Agricultural Research Service, Root Disease and Biological Control Research Unit, Pullman, WA 99164.

Root-lesion nematodes, *Pratylenchus neglectus* and *P. thornei*, are the most important pests restricting productivity of wheat in the Pacific Northwest (PNW). Pre-plant populations of both species are frequently inversely correlated with wheat

yield. It is estimated that these nematodes reduce profitability of farms in Idaho, Oregon and Washington by about $51 million annually. The best approach to control damage from root-lesion nematodes is to grow cultivars that are both resistant and tolerant. Individual wheat cultivars may differ in their ability to resist and tolerate these nematodes. Optimal cultivar selection requires that the lesion nematode species present in each field or region be accurately identified and quantified. It is challenging to discriminate *P. neglectus*, *P. thornei* and other closely related species by morphological characters. It is laborious and difficult to use microscopy to count and identify these nematodes in large numbers of field samples in which other nematode species are also present. A SYBR Green I-based real-time quantitative-polymerase chain reaction assay was developed to facilitate the identification and quantification of individual species in soil. A primer set for *P. neglectus* was designed from the *Pratylenchus* 28S ribosomal RNA gene sequences of the D3 expansion domain. A primer set for *P. thornei* was designed from the internal transcribed spacer ITS1 of the ribosomal DNA. Melting curve analysis revealed that the primer sets were highly specific. The *P. neglectus*-primers did not amplify DNA from 10 isolates of other *Pratylenchus* species and other species and genera of nematodes typically present in the soil communities, and from six fungal species commonly associated with wheat root rot. The analysis of PCR amplification efficiency (E) indicated that DNA extracted from soil samples by a commercial kit (PowerSoil DNA Isolation Kit) can be directly used in real-time PCR without any additional purification step (E = 96% for *P. neglectus*, 97% for *P. thornei*). Standard curves were generated from artificially inoculated soils showing a negative linear regression between threshold cycles (Ct) and Log values of number of nematodes (r^2 = 0.92 for *P. neglectus*, 0.98 for *P. thornei*). Validation tests using 15 field soil samples were conducted to determine the relationship between nematode numbers detected by the real-time PCR assay and the numbers reported by commercial diagnostic laboratories and our research lab using traditional methods. The real-time PCR has the potential to enable laboratories to avoid time-consuming physical separations, microscopic identifications, and counting of these species from field samples with mixed populations of other plant-parasitic and non-plant-parasitic nematodes.

135. IDENTIFICATION AND DISTRIBUTION OF *PRATYLENCHUS* SPP. ON BLUEBERRIES. **Zasada[1], Inga A. and T.A. Forge[2]**. [1]USDA-ARS Horticultural Crops Research Laboratory, 3420 NW Orchard Ave., Corvallis, OR 97330, [2]Agriculture and Agri-Food Canada, Pacific Agri-Food Research Centre, P.O. Box 1000-6947 Hwy 7, Agassiz, BC VOM 1A0.

Pratylenchus spp. are commonly encountered in blueberry (*Vaccinium* spp.) fields in the Pacific Northwest of the United States and coastal British Columbia of Canada. However, in controlled experiments blueberry does not appear to be a host for P. penetrans. We sampled 13 commercial blueberry fields in the spring and fall to determine the species of *Pratylenchus* associated with blueberry plantings, and to determine the geographical distribution of these species. *Pratylenchus* spp. were more commonly found in soil samples (68% and 61% in spring and fall, respectively) than in blueberry root samples (42% and 50% in spring and fall, respectively). Population densities averaged 160 and 201 *Pratylenchus* spp./100 g soil and 82 and 69 *Pratylenchus* spp./g dry root in the spring and fall, respectively. Across locations, *Pratylenchus* spp. were detected in 56% and 54% of the sampled weed roots in spring and fall, respectively, at population densities averaging 1,063 and 1,354 *Pratylenchus* spp./g dry root in spring and fall, respectively. The species of *Pratylenchus* present in blueberry plantings is being determined by amplification of the D2/D3 expansion domains of the nuclear 28S rDNA subunit.

136. EVALUATION OF AMINO ACIDS AGAINST PLANT-PARASTIC NEMATODES. **Zhang, Yun, W. T. Crow**. Entomology and Nematology Department, 970 Natural Area Drive, University of Florida, Gainesville, FL 32611.

Belonolaimus longicaudatus (sting nematode) and *Meloidogyne* sp. (root-knot nematodes) commonly cause economically significant damage to turfgrasses and annual ornamental plants, respectively, in the southeastern United States. However, nematode management on these plants is difficult. DL-methionine and other amino acids could be effective candidates to control nematode problems by interfering with essential intracellular metabolic pathways and enzymes in nematodes. Replicated RCBD bench screen studies were carried out in 2009 to evaluate the effectiveness of DL-methionine several methionine analogues, threonine, and lysine against *B. longicaudatus* and *Meloidogyne* spp. Rates of each treatment (224 and 448 kg amino acid/ha) were comaped to each other and to water-treated controls. Small plastic pots were filled with 200 cm^3 of soil infested with *B. longicaudatus* or J2 of *Meloidogyne* sp. After 72 hours soil was removed from the pots and placed onto modified baermann funnels for 24 hours. Nematodes were collected and counted. The results indicated that all methionine-containing formulations were effective at both rates against *Meloidogyne* sp., whereas only liquid formulations were effective against *B. longicaudatus* in both trials. Threonine and lysine were not as effective as methionine against either nematode. Further studies will focus on testing efficacy and phytotoxicity of methionine formulations against *B. longicaudatus* on *Agrostis palustris* under greenhouse conditions and against *M. incognita* on *Impatiens wallerana* in microplots.

137. INTERACTIONS BETWEEN NEMATODES AND GRAPE ROOTSTOCKS. **Zheng[1], Liang, H. Ferris[1] and A. Walker[2].** [1]Department of Nematology and [2]Department of Viticulture and Enology, University of California, Davis CA 95616.

Resistance and susceptibility to *Meloidogyne* spp. in grape rootstocks are expressed phenotypically in the development and quality of the nematode feeding site. Populations of *M. incognita* race 3 (avirulent on resistant rootstock Harmony), and of

isolates of *M. incognita* and *M. arenaria* that are virulent on Harmony, failed to establish infection sites in five new rootstocks with broad and durable nematode resistance (GRN1-GRN5) unless the roots were incubated at elevated temperature. Low numbers of egg masses (usually 0.1-3% of those on a susceptible control plant) were produced on 12 of 15 rootstock selections, including the GRN series, at temperatures ≥30°C. On pluronic gel medium at 24°C, *M. incognita* race 3 juveniles were equally attracted to roots of both resistant (R) and susceptible (S) cultivars. Juveniles entered, and were visible within, the roots of both R and S cultivars. The juveniles began to establish feeding sites within 2 days in S cultivars but in R cultivars, although they were still visible for several days, there was no evidence of feeding site development or swelling of the nematodes. We assume that juveniles were unable to establish feeding sites in the R cultivars at 24°C. When roots were incubated at higher temperatures, the quality of the giant cell feeding sites in R cultivars seemed inferior to those in S cultivars because, after 13 days, the few developing juveniles in R cultivars had only reached the J3 stage while those in S cultivars were already at the J4 and young adult stage. In studies of life course duration and productivity with plants infected and maintained at 30°C, development was slower in R rootstocks Harmony and GRN1 than in the S cultivar French Colombard. In addition, the adult females were smaller-bodied and produced smaller egg masses in the R than in the S cultivars. In summary, a few individuals of both *M. arenaria* and *M. incognita* can overcome resistance originating from different parentage sources in grape rootstocks. It remains to be seen whether virulent populations of *Meloidogyne* spp. will be selected in field-grown resistant rootstocks and whether that virulence will be maintained and expressed under more usual soil temperatures <30°C.