

ABSTRACTS

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VISUAL ASSESSMENT OF ROOT-KNOT NEMATODE INFESTATIONS OF VEGETABLE SOILS AND DETERMINING THE NEED FOR CONTROL. **Abawi, G. S., J. W. Ludwig, and B. K. Gugino.** Department of Plant Pathology, Cornell University, NYSAES, Geneva, NY 14456.

The northern root-knot nematode (*Meloidogyne hapla*) is a major pathogen of vegetables grown in New York, significantly impacting the quantity and quality of marketable yield of processing and fresh market vegetables. Currently, there are no known resistant commercial varieties and crop rotations are of limited use, as most crops usually included in common vegetable rotations are susceptible to this nematode. Also, it is often not cost-effective to rotate the main vegetable crops with non-host crops which are often less profitable, especially on the high value organic soils. Thus, chemical nematicides are heavily relied upon for the management of plant-parasitic nematodes on vegetables in New York. A simple soil bioassay with lettuce was extensively tested for visual assessment of the soil infestations with *M. hapla* utilizing a root-galling severity (RGS) scale of 1 (no visible galls, healthy roots) to 9 (>80% of roots galled). RGS ratings are then used to determine the need to treat or rotate with a non-host crop based on the susceptibility of the next vegetable crop (e.g. <2 RGS for carrots and >3 RGS for onions). To-date, only 7 out of 17 commercial fields bioassayed with this procedure were found to require treatment, with 4 of these fields requiring only partial treatment due to the uneven infestations of the nematode within the field. Participatory training for growers, consultants, and educators to conduct their own on-farm bioassays and to utilize the results in deciding the need for managing the nematode is currently on-going. Adoption of this cost-effective control strategy will help to reduce nematode damage, reduce nematicide use, and increase yield and profitability.

BIOACTIVE PHYTOCHEMICALS FROM SELECTED ARID LAND FLORA. **Abdel-Rahman, F.,¹ M. H. El-Hamawy,¹ and M. Saleh.²** ¹Departments of Biology and ²Chemistry, Texas Southern University, Houston, Texas 77004.

The biological activity of the phytochemicals isolated from twenty one arid land plants was evaluated. The organic and water extracts of the following plants were studied, *Ruta tuberculata*, *Tamarix nilotica*, *Artemisia judaica*, *Artemisia monosperma*, *Clerodendrum acerbianum*, *Rosmarinus officinalis*, *Cymbopogon citratus*, *Achillea millifolium*, *Lantana camara*, *Kochia indica*, *Geranium cicutarium*, *Cassia acutifolia*, *Matricaria chanomilla*, *Thymus vulgaris*, *Artemisia indica*, *Adhatoda viscum*, *Alpina officinalis*, *Zygophyllum album*, *Eugenia aromatica*, *Colligonum comosum*, and *Peganum harmala*. The effect of organic extracts of the above plant species on the hatching of the eggs of the *Meloidogyne javanica* and on the pathogenic effect of its second-stage larvae were evaluated. *Lantana camara* and *K. indica* were the most effective in reducing egg hatching followed by *R. officinalis*, *G. cicutarium*, and *M. chanomilla*. The *A. judaica* and *A. monosperma* did not reduce the hatching of the eggs but reduced the pathogenicity of the hatched larvae as it was evident from the lower gall counts on tomato roots. The water extracts of *L. camara*, and *P. harmala* reduced the number of hatched larvae and also reduced the pathogenic effect of hatched larvae when exposed to the plant extracts for 24 hr prior to inoculation to tomato seedlings. *Geranium cicutarium* reduced the egg hatching but did not affect the ability of the second-stage larvae of infecting tomato plants and complete the life cycle. The water extract of *M. chanomilla* showed an activation effect on egg hatching because it increased the egg hatching over the control. In another test the three plants with highest phytochemical activities were tested again using 1000 ppm of the organic extract which lowered the hatching of the eggs and reduced the pathogenicity of the second-stage larvae, *L. camara*, had the highest effect followed by *K. indica*, and *A. judaica*.

PHYLOGENETIC RELATIONSHIPS AMONG *MELOIDOGYNE* SPECIES BASED ON RIBOSOMAL DNA SEQUENCES. **Agudelo, P.,¹ S. A. Lewis,¹ A. Szalanski,² and B. Fortnum.¹** ¹Department of Entomology, Soils, and Plant Sciences, Clemson University, Clemson, SC 29634, and ²Department of Entomology, University of Arkansas, Fayetteville, AR 72701.

With the purpose of examining the evolutionary relationships within the genus *Meloidogyne*, a portion of the 18S rDNA gene and the D3 expansion region of the 26S rDNA gene from a collection of isolates representing more than half the species described, were sequenced and analyzed. Although root-knot nematodes exhibit extensive diversity, most studies using molecular methods have focused on the four major species. Several phylogenies have been inferred from comparative analysis of different regions of ribosomal DNA, but a very limited number of species have been included to date, thus presenting a limited analysis of evolutionary relationships. We have included additional representatives across the genus

to construct species-level phylogenies that can provide an indirect record of the speciation events that have led to extant species. Phylogenetic analyses with maximum-parsimony, maximum-likelihood, and neighbor-joining methods were performed. The trees generated indicated several distinct clades. Evolutionary implications and correspondence between morphology-based taxa and inferred evolutionary entities within the genus are discussed.

DRIP IRRIGATION AS A FUMIGANT DELIVERY SYSTEM. Ajwa, H. Department of Plant Sciences, University of California, Davis, CA 95616.

New technologies were developed recently to apply fumigants (such as 1,3-dichloropropene, chloropicrin, and iodomethane) through drip irrigation systems that are used to irrigate crops. Drip fumigation with chloropicrin, InLine (a mixture of 1,3-dichloropropene and chloropicrin), and Midas (a mixture of iodomethane and chloropicrin) applied at high label rates controlled soil pests and produced strawberry yields equivalent to standard methyl bromide plus chloropicrin shank fumigation. Lower rates (60% of label rates) of these fumigants provided good weed and pathogen control when applied under virtually impermeable film or followed by a sequential application of metam sodium. This presentation will discuss techniques for fumigant application through drip irrigation systems, and ongoing studies to determine optimum drip fumigation rates to control various soilborne pathogens and weeds.

TECHNOLOGIES FOR MONITORING SOIL FUMIGANTS IN SOIL. Ajwa, H. Department of Plant Sciences, University of California, Davis, CA 95616.

Accurate measurement of soil fumigant concentration is essential for determining their efficacy to control soil pests and persistence in the environment. Several methods have been developed to determine fumigant concentration in soil. Solvent extraction of soil and trapping of soil fumigants on specific sorbents (such as activated charcoal) followed by solvent extraction are the most widely used. Fumigant concentration in the solvent is then determined by gas chromatography. These methods can result in low and inconsistent measurements due to fumigant volatilization losses during soil sampling and storage or during soil extraction. Therefore, a high speed, portable gas chromatograph (micro GC) was automated to sample and analyze fumigants in the soil gaseous phase. This micro GC method provided real-time, sensitive, accurate, and fast measurements compared to the traditional solvent extraction methods. Advantages and limitations of various methods used for monitoring soil fumigants will be discussed.

OXYCOM APPLICATIONS TO ROOTS COMPARED TO FOLIAGE AND THEIR IMPACT ON TOMATO GROWTH AND NEMATODE DEVELOPMENT. Anwar, S., and M. McKenry. Department of Nematology, University of California, Riverside, CA 92521.

A greenhouse study was conducted to compare the effects of Oxycom root or foliage applications on growth of resistant and susceptible tomato cultivars exposed to two *Meloidogyne incognita* populations. Fifteen-day-old tomato seedlings of resistant cv. Favorita and susceptible cv. Beltran were inoculated with 5000 eggs of virulent San Quentin or avirulent Beltran populations of *M. incognita*. Nematode infected plants received three root or foliar applications at 20-day intervals at the rate of 2500 ppm (V/V) Oxycom. Nematode infected plants receiving only water served as the untreated control and non-infected tomato plants receiving Oxycom served as the growth check. Treatments were arranged in a completely randomized design in a greenhouse at 30°C. Sixty-three days after inoculation nematode reproduction, leaf area, fresh foliage, root and fruit weight were recorded. Oxycom root applications increased the rate of nematode reproduction and improved shoot and root growth over that from foliar applications or the non-treated check plants. Oxycom root applications were more effective in stimulating the growth of susceptible tomato cv. Beltran compared to resistant Favorita in the presence of nematodes. The higher rate of nematode reproduction coupled with improved plant growth suggests a soil drench with Oxycom will provide the preferred opportunity for delivery of nematode tolerance into plants, including those infected with virulent populations of nematodes.

HOST-DEPENDENT SEGREGATION DISTORTION OF LOCI IN SOYBEAN CYST NEMATODE, *HETERODERA GLYCINES*. Atibalentja, N.,¹ S. Bekal,¹ L. L. Domier,² T. L. Niblack,¹ G. R. Noel,² and K. N. Lambert.¹ ¹Department of Crop Sciences, University of Illinois at Urbana-Champaign, 1102 South Goodwin Ave., Urbana, IL 61801, and ²USDA, ARS, Urbana, IL 61801.

Recently, we constructed a genetic linkage map of the soybean cyst nematode, *Heterodera glycines*, by AFLP-fingerprinting a random sample of virgin females from an F₂ population that originated from controlled matings between two highly inbred lines, TN16 and TN20. The females used were reared on the *H. glycines*-susceptible soybean cultivar Lee 74. Several markers, most of which clustered on linkage group I, showed highly skewed segregation in favor of TN16. To investigate the role of plant host on the segregation of markers, we genotyped a second sample of virgin females from the same F₂ population, but reared on the *H. glycines*-resistant soybean PI88788. Most of the markers analyzed in the second sample mapped to the same linkage groups and in the same order as in the sample reared on Lee 74. However, the

segregation patterns of markers on the PI88788-based map were, in general, the converse of the ones observed on the first map. Specifically, markers that exhibited skewed segregation on linkage group I of the first map segregated according to expected Mendelian ratios on the same group of the PI88788-based map. In contrast, markers on linkage group III, which also contains *Hg-cm-1*, a putative virulence gene, showed significant segregation distortion on the PI88788-based map, compared to the first map where they segregated normally. These results indicate that the soybean host mediates the segregation of the nematode genes, which in turn may explain the ability of some populations to defeat resistance.

MOLECULAR GENETIC ANALYSIS OF THE LANCE NEMATODE *HOPLOLAIMUS* (TYLECHIDA:HOPLOLAIMINAE). Bae, C. H.,¹ A. L. Szalanski,² and R. T. Robbins.¹ ¹Departments of Plant Pathology and ²Entomology, University of Arkansas, Fayetteville, AR 72701.

Lance nematode, *Hoplolaimus* spp., is an important nematode pest of various crops in the Southeastern United States. In this study, nuclear ribosomal DNA ITS1, 18S rDNA and mtDNA COI from *Hoplolaimus* sp. were sequenced and compared. Sequence analysis of ITS1 region showed that several genotypes exist in *H. columbus*. Polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) analysis of the first internal transcribed spacer region (ITS1) revealed intra and inter specific variation in *Hoplolaimus* sp. using 10 restriction enzymes. Molecular phylogenetic analysis using ITS1, 18S rDNA, and mtDNA COI was conducted. Based on RFLP profiles, and the phylogenetic analysis, we discuss usefulness of genetic markers for phylogenetic analysis and taxonomic identity of *Hoplolaimus* sp.

MOLECULAR PHYLOGENETIC ANALYSIS OF HOPLOLAIMINAE (NEMATODA: HOPLOLAIMIDAE) BASED ON 18S AND ITS RIBOSOMAL DNA. Bae, C. H.,¹ A. L. Szalanski,² and R. T. Robbins.¹ ¹Departments of Plant Pathology and ²Entomology, University of Arkansas, Fayetteville, AR 72701.

Hoplolaimids are a large group of economically important Tylenchida and undergo part or all of their cycle in the soil. They are obligate plant parasites, and are closely associated with plant roots. The subfamily Hoplolaiminae has eight genera. Some species of Hoplolaiminae are distributed all over world whereas other species have restricted distribution. In this study, Molecular phylogenetic analysis among 5 genera of the subfamily Hoplolaiminae was conducted using 18S rDNA and ITS Ribosomal DNA sequence data. Phylogenies were evaluated using maximum likelihood and maximum parsimony analysis. In addition, combined analyses of the sequences of 18S rDNA and ITS 1 were compared with analyses of the ITS1 alone and 18S alone. The trees showed several distinct clades and from the phylogenetic analysis, we confirmed the hypotheses derived from current classification systems.

EFFECTS OF THE CELL DEATH PROTEIN CED4 ON FREE-LIVING AND PLANT-PARASITIC NEMATODES. Bahaji, A., C. Tristan, T. Padukkavidana, G. Polack, and A. Calderon-Urrea. Department of Biology, California State University, Fresno, CA.

Studies on the nematode *Caenorhabditis elegans* have defined a genetic pathway for the programmed cell death, and also revealed that the protein products of certain cell-death genes (*ced-3*, *ced-4*, and *ced-9*) interact directly. CED4 is a protease-activating factor that acts on a second core protein CED3, and activated CED3 initiates a series of events leading to cell death. Based on these results and evidence suggesting the conservation of cell death pathways in some animals we hypothesized that cell death genes could be used to protect plants against pests. To test our hypothesis we first constructed expression cassettes to generate CED4 protein in heterologous systems and demonstrated by the use of Western blotting and an antibody against CED4, that indeed a CED4 protein is expressed in both bacteria and plant systems. We then provide evidence that by feeding *C. elegans*, either wild type or mutant *ced-3* and *ced-4* worms, with bacteria expressing CED4 protein, a reduced population size in wild type and *ced-4*, compared to *ced-3* mutant can be observed. In addition, when we incubate *Meloidogyne incognita*. J2 worms in H₂O supplemented with different concentration of purified CED4 protein, the number of worms that survive decrease inversely proportional to the increase of CED4 protein concentration. Therefore we concluded that the viability of *C. elegans* and *M. incognita* is decreased when exposed to CED4, perhaps due to a hyper-activated PCD pathway. We then generated transgenic tobacco plants containing the *ced-4* gene as a single copy insert and tested them for their ability to tolerate *M. incognita* infestation. When compared to their control, the *ced-4* homozygous transgenic tobacco plants showed a 40% reduction in the number of galls formed.

TRAP CROP DEVELOPMENT AND USE FOR *HETERODERA SCHACHTII* AND *H. GLYCINES* CONTROL IN MICHIGAN. Bates, C., G. Bird, F. Warner, and J. Davenport. Michigan State University, East Lansing, MI. 48824.

As a specialty crop state, seven *Heterodera* spp. are known to present management challenges in MI. Sugar beet growers have adopted oilseed radish (OSR) cv Colonel or Adagio as traps crop for control of *H. schachtii*. Field trials indicate that a spring crop of OSR before soybeans is a more reliable practice for lowering *H. schachtii* population densities and increasing subsequent sugar beet yields than a late summer planting of OSR following wheat. Producers of other MI commodities have begun to use OSR for general soil quality enhancement, resulting a need to understand the impact of

OSR cultivars on *H. glycines*, *Pratylenchus penetrans* and *Meloidogyne hapla*. Greenhouse trials indicate that trap crop efficacy is cultivar specific. While OSR cv Colonel and Adagio are appropriate trap crops for *H. schachtii*, other cultivars of OSR are not. In addition, OSR cv Colonel was found to be a good host for reproduction of both *P. penetrans* and *M. hapla*. Because of the significantly large number of plants known as poor host of *H. glycines*, these plant species appear to have potential use as trap crops for this species. Several plant species and cultivars including Berseem and Crimson clovers are being evaluated in respect to their efficacy for use as economically viable trap crops for control of *H. glycines* under MI conditions. This situation is complicated by the fact some MI field populations of *Heterodera* can reproduce and be pathogenic on both soybeans and sugar beets. A molecular approach to diagnostics is being developed for clarification of this situation. The work includes identification of field survey populations using Amplified Fragment Length Polymorphisms (AFLP). This should assist in selection of the most appropriate site-specific trap crops for cyst nematode control.

SEED COATING WITH NEMATOCIDES: A NEWFANGLED TECHNOLOGY FOR PLANT PROTECTION. Becker, J. O. Department of Nematology, University of California, Riverside, CA 92521.

Negative perceptions about the usefulness and marketability of seed treatments with nematicidal activity have inhibited the development of this technology. This assessment was influenced by earlier research with carbamate and organophosphate nematicides that demonstrated short-term efficacy against plant-parasitic nematodes but revealed potential problems with phytotoxicity. At the same time, the availability of relatively inexpensive soil-applied nematicides provided no incentive to follow-up with more suitable nematicides. Today, healthy seedling establishment has gained in importance as the cost of hybrid or GM seed has dramatically increased and many soil pesticides are no longer available due to regulatory pressure. Furthermore, in many crops a major part of yield losses due to plant-parasitic nematodes can be avoided if the attack can be delayed for at least the first few weeks. The mitigation of early nematode attack also reduces the invasion of secondary microorganisms or those fungi that take advantage of nematode-induced physiological changes in the plant. Thus, in combination with potent insecticide and fungicide seed treatments, nematicide coating completes a package that provides a new tool for environmentally safe and effective plant protection.

A COMPARATIVE APPROACH TO THE ANALYSIS OF PLANT-PARASITIC NEMATODE CHORISMATE MUTASES. Bekal, S., T. V. Akraiko, T. L. Niblack, and K. N. Lambert. Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

Plant parasitic nematodes secrete chorismate mutases (CM) into plant tissues as they establish an association with their host. CM are enzymes in the shikimate pathway, a primary plant metabolic route that produces the aromatic amino acids and numerous secondary metabolic compounds that control critical plant processes. The shikimate pathway does not exist in animals, but some plant parasitic nematodes secrete this enzyme into the plant tissue to presumably manipulate the regulation of the plant pathway to benefit the nematode. Nematode CM could alter plant cell development, cell wall structure, or host plant resistance. Nematode CM are members of multigene families where different forms are developmentally regulated and differentially expressed. Sequence comparison of root-knot nematode and soybean cyst nematode CM indicate that particular domains of the CM coding region vary between members of the gene family. The CM protein domain is highly conserved within nematode species, but the C-terminal domain of the root-knot nematode CM is variable. Interestingly, the N-terminal domain is variable between CM family members in the soybean cyst nematode. The soybean cyst CM genes are arranged in as tandem repeats, but the functional significance of this gene organization is unclear at the present time. The variation in the protein domains between nematode CM may also imply that members of the nematode gene family have differing enzymatic properties, thus the differential regulation of nematode CM may allow plant nematodes to subtly alter their parasitic ability.

BIODIVERSITY INSIDE OUT: NEMATODE-DOMINATED COMMUNITIES IN MILLIPEDES. Bernard, E. C. Department of Entomology and Plant Pathology, The University of Tennessee, 205 Plant Sciences, Knoxville, TN 37996-4560.

Millipedes collected during the 2002 and 2003 Millipede Marches in Great Smoky Mountains National Park were dissected to collect and record the nematode fauna inhabiting the digestive tract. Our most extensive knowledge of these nematodes was published by Joseph Leidy more than 150 years ago. This century's collection efforts are part of the current All-Taxa Biodiversity Inventory underway in the Park. At least seven species of nematodes in Rhigonematidae and Thelastomatidae were collected via dissection from large cambalid, spirobolid, and xystodesmid millipedes. One nematode taxon could not be identified to family. Smaller millipedes (Andrognathidae, Paradoxosomatidae, Parajulidae, Polydesmidae) did not contain intestinal nematodes. Dissection of millipedes preserved in alcohol yielded distorted and collapsed, but taxonomically usable nematodes. Rhigonematid nematodes often had commensal trichomycete fungi (*Enterobryus* spp.) attached to them. The thallus was attached by means of a holdfast and did not appear to debilitate the nematode.

Trichomycete hyphae in turn usually had colonies of the *Arthromitus* stage of *Bacillus cereus* attached to them. Both larger and smaller millipedes frequently had numerous gregarine protists in their intestine. Estimates of biodiversity in the Park may need to be revised to account for numerous internal parasites and commensals that as yet have scarcely been examined. The ease of collection and the large size and transparency of these nematodes make them excellent candidates for classroom-based exercises.

POTENTIAL OF PREDATORY NEMATODES IN THE BIOLOGICAL CONTROL OF PLANT-PARASITIC NEMATODES. **Bilgrami, A. L.** Department of Entomology, Rutgers University, New Brunswick, NJ 08901.

Predatory nematodes represent a small amount of soil biomass, but their presence across so many trophic levels e.g., plant, fungal and bacterial feeding nematodes and carrion feeders besides other microorganisms is vitally important in soil ecosystem processes. Their future role in nematode management depends greatly on advances made on other control methods, their effectiveness, and the resources provided to establish research programs. Biological control potential and efficacy of predatory nematodes vary with their type. Mononchs because of their long life cycle, low rate of fecundity, susceptibility to changing environmental conditions, difficulty of mass production and cannibalism do not fulfill requirements of efficient biological control agent. The advantage of dorylaim, nygolaim and other stylet bearing predators lie in their wide spread natural occurrence (200-500 million/acre) and ease of mass culturing due to their polyphagous feeding habits. Also, their populations may be elevated in the field by adding organic nutrients. However, more field studies are needed to test this hypothesis. The real possibility of using predatory nematodes in nematode management programs lies in the diplogasterid predators due to their biphasic feeding, high rates of predation and fecundity, short life cycle, ability to search for prey, presence of resistant juvenile stage(s) and rare cannibalism. Despite remarkable similarities with attributes of entomopathogenic nematodes, diplogasterids should not be considered as unilateral inundative agents (i.e., repeated applications for short-term control). The flexible bi-phasic feeding behavior of diplogasterids should endow them with superior persistence; that is, when prey become scarce they should switch to feeding on soil bacteria to maintain themselves. Nematode predators are likely to offer the most promise as augmentative agents in colonization efforts in combination with cultural control tactics, such as rotation, cover cropping, green manuring, organic amendments, and in plant resistance.

TRAIT DETERIORATION IN LABORATORY-CULTURED *HETERORHABDITIS BACTERIOPHORA* AND *STEINERNEMA CARPOCAPSAE*. **Bilgrami, A. L.,¹ R. Gaugler,¹ Y. Wang,¹ D. Shapiro-Ilan,² and B. Adams.³** ¹Department of Entomology, Rutgers University, New Brunswick, NJ 08901; ²USDA-ARS, SE Fruit and Tree Nut Research Lab, 21 Dunbar Rd., Byron, GA 31008; and ³Department of Microbiology and Molecular Biology, Brigham Young University, 775 WIDB, Provo, UT 84602.

Biocontrol agents reared in the laboratory or produced for commercial purposes may show deterioration in important biocontrol traits over time, reducing their field efficacy. To understand deterioration, we characterized phenotypic changes in biocontrol traits e.g., heat tolerance, host finding/nictation, virulence and fecundity of five isolines each for two entomopathogenic nematode species, *Heterorhabditis bacteriophora* and *Steinernema carpocapsae*. One or more, and in some cases all five, nematode isolines showed declines in virulence, reproduction, heat tolerance, host seeking ability, and nictation. *Heterorhabditis bacteriophora* lines deteriorated faster and more extensively than those of *Steinernema carpocapsae*. Rate of deterioration/passage was also higher in *H. bacteriophora*. Genetic analysis will reveal whether the mechanism for these losses is genetically (inbreeding, genetic drift, or inadvertent selection) or non-genetically based (e.g., nutrition, disease).

NRI AWARDEE RESEARCH REPORT: PLANT PERCEPTION OF ROOT-KNOT NEMATODE SIGNALS. **Bird, D.** The Center for the Biology of Nematode Parasitism, North Carolina State University, Raleigh, NC 27695.

Parasitic root-knot nematodes (RKN) and beneficial rhizobacteria induce new organs in the root vasculature: giant cells (GC) and nitrogen nodules, respectively. We used a hormone-responsive gene promoter driving GUS to spatio-temporally profile the influence of cytokinin on these processes in *Lotus japonicus*. Staining was detectible in root hairs shortly after interaction with rhizobia and was evident at the earliest stages of the nodule primordium. In contrast, a cytokinin response was not detectible during root penetration and migration by RKN, nor in the mature GC. Down-regulation of cytokinin levels in planta via transgenic expression of cytokinin oxidase genes yielded roots with significantly fewer nodules. The number of RKN feeding sites also was reduced, suggesting that cytokinin is transiently required for GC initiation, but not for maintenance. Using confocal microscopy we dynamically profiled the GFP-tagged cytoskeleton of living *Lotus* root hairs following exposure to rhizobial Nod factors (NF). Remarkably, RKN elicit an identical cytoskeletal response via a signal able to function at a distance. Neither azide-killed RKN nor *C. elegans* produce this signal. Aspects of the host responses to RKN were altered or abolished by mutations in the NF receptor genes *nfr1* and *nfr5*, suggesting that RKN produce a molecule with functional equivalence to NF, and which we name Nem factor (NemF). A response to NemF was seen in tomato, but not *Arabidopsis*, which lacks components of the NF-receptor/response machinery. Because the ability

of RKN to establish feeding sites and reproduce was markedly reduced in the mutant lines, we propose that RKN have adapted at least part of the symbiont-response pathway to enhance their parasitic ability. Analysis of 100,000 ESTs from 14 tylenchids suggested that RKN acquired rhizobial genes (including those associated with NF biosynthesis) via horizontal gene transfer, insinuating a mechanism for evolutionary adaptation to exploit the host symbiont-response pathway.

SOYBEAN CYST NEMATODE AND SOYBEAN SUDDEN DEATH SYNDROME. Bond, J. P.,¹ T. A. Jackson,² X. B. Gao,³ T. L. Niblack,³ and M. E. Schmidt.¹ ¹Plant, Soil and Ag. Systems, Southern Illinois Univ. Carbondale, IL 62901; ²Department of Plant Pathology, Univ. of Nebraska, Lincoln, NE 68501; and ³Department of Crop Sciences, Univ. of Illinois, Urbana, IL 61801.

Soybean cyst nematode (SCN), *Heterodera glycines*, and the causal agent of soybean sudden death syndrome (SDS), *Fusarium solani* f. sp. *glycines* are two of the most important soybean pathogens in the Midwest. A majority of the production fields in this region are infested with SCN. Many of these same fields are also infested with *F. solani* f. sp. *glycines*. Sudden death syndrome was first observed in the Midwest in 1979; however within 5 years much of the soybean producing area in the Mississippi and Ohio River valleys had experienced frequent epidemics. Previous research has shown that SCN can exacerbate symptoms of SDS. However, there are many questions regarding this interrelationship. From 1988-2004, extensive trials conducted to evaluate variety reaction to SDS, documented the performance of SCN-resistant vs. SCN-susceptible varieties. Initially, SCN-resistant varieties proved to be more resistant to SDS than the SCN-susceptible varieties. In later years this relationship did not hold true. Subsequent studies revealed a genetic linkage between *rhg1*, a major SCN resistance gene, and several SDS resistance genes identified in the cultivar 'Forrest'. Microplot, and greenhouse studies have used near isogenic soybean lines that contrast for resistance or susceptibility to each pathogen. In these studies, the population densities of both pathogens had a significant influence on the severity of foliar symptoms of SDS. Foliar symptoms increased when SCN was present; however this interaction was not always governed by the population density of SCN. In microplots, interactions between these two pathogens were antagonistic with regards to nematode reproduction.

PREDICTIVE VALUES OF NEMATODE COMMUNITY IN ORGANIC AND LOW-TILLED CONVENTIONAL FARMING SYSTEMS. Briar, S. S.,¹ P. S. Grewal,² Deborah H. Stinner,² and S. A. Miller.¹ Departments of ¹Plant Pathology and ²Entomology, The Ohio State University, OARDC Wooster, OH 44691-4096.

Nematodes have the potential to serve as bioindicators of soil health. As opposed to conventional farming, organic farming relies heavily on tillage, which not only increases the production cost but also disturbs the soil community. We hypothesized that the nematodes could serve as indicators of changes in the soil agro-ecosystem. In 2000 a long-term field trial was established at the Ohio Agricultural Research and Development Center representing two farming systems. Conventional plots had corn and soybean rotation and received synthetic inputs. Organic plots had corn, soybeans, oats and hay rotation and received beef and poultry manure. After 4 years, total population of plant-parasitic nematodes and *Pratylenchus crenatus* was significantly reduced in organic as compared to the conventional plots. Maturity index was significantly high in organic system whereas diversity indices were high in conventional system. In the present study a recently proposed model, which provides a framework for soil food web diagnostics, revealed that despite low tillage in the conventional system, organic inputs resulted in a more structured food web in the organic system and indicated both bacterial and fungal decomposition channels. Increased population of omnivore nematodes in the conventional farming was correlated with the low disturbance due to less tillage. In the organic farming system, population of *Xiphinema* sp was high before the tillage in the spring, but in the conventional plots it was high during the fall. This suggests that ectoparasites such as *Xiphinema* sp. can serve as good indicators of disturbance. Therefore, we propose that these nematodes should be included in the model parameters. Our results indicate that the combination of low tillage with organic inputs in the organic system would increase the sustainability of agro-ecosystems.

A NEW ROOT-KNOT NEMATODE INFECTING VEGETABLES IN FLORIDA. Brito, J. A.,¹ J. D. Stanley,¹ R. Cetintas,² J. Hamill,² and D. W. Dickson.² ¹Division of Plant Industry, Gainesville, FL 32614-7100; ²Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620.

Meloidogyne floridensis was recently described as a new species infecting peach (*Prunus persica*) in Alachua Co., Florida. This nematode is of particular importance because it reproduces on root-knot nematode resistant rootstocks, namely Guardian, Nemaguard, Nemared, and Okinawa. Its host range and distribution in Florida and the USA are not known. In 2003-04, root and soil samples were collected from vegetable fields and greenhouses in Florida. Identification of root-knot nematode species was determined using isozymes (esterase and malate dehydrogenase) supplemented by morphological and morphometric analysis when needed. *M. floridensis* was identified from tomato (*Lycopersicon esculentum*) (one sample from Indian River Co., two from Hendry Co.), cucumber (*Cucumis sativus*), eggplant (*Solanum melongena*), and snap bean (*Phaseolus* sp.) from Hendry, Hillsborough, and Alachua Counties, respectively. This nematode

was also found infecting a wild plant, lilac tasselflower (*Emilia sonchifolia*), in Hendry Co. In two sites *M. floridensis* occurred sympatrically with *M. mayaguensis* and *M. incognita*. This is the first report of *M. floridensis* infecting cucumber, eggplant, snap bean, and lilac tasselflower in Florida.

CONVERGENT EVOLUTION OF NEMATODE EYES. Burr, A. H. Jay,¹ C. Burr,¹ I. Tandingan De Ley,² P. De Ley,² and J. G. Baldwin.² ¹Department of Biological Sciences, Simon Fraser University, Burnaby, BC, Canada V5A 1S6, and ²Department of Nematology, University of California, Riverside, CA 92521.

Eyes are found sporadically in widely divergent nematode taxa. However, within taxa, species with eyes occur along with species lacking eyes, and eyes do not occur at all within Rhabditida and certain other groups. Nematode eyes are paired and have only one light sensor, except for the single eye with two photoreceptors in *Mermis nigrescens*. Incoming light is restricted to particular directions by an auxiliary optical structure that occurs at three levels of complexity: a mass of dense pigment ('pigment spot'), a pigment cup or, more rarely, a pigment cup with either a lens or cornea. Previous TEM studies combined with our own molecular phylogenies reveal that the cell type in which shadowing pigment develops differs in representatives of four clades: (1) hypodermal cell in Mermithida (*Mermis nigrescens*), (2) sheath cell in Monhysterida (*Diplolaimella dievengatensis*), (3) pharyngeal radial muscle cell in Oncholaimina (*Oncholaimus vesicarius*), and (4) pharyngeal marginal cell in Enoplina (*Enoplus anisospiculus* and *Deontostoma californicum*). Other differences among these clades include the chemical nature of the pigment or the presence of a ciliary versus multilamellar photoreceptor. In a fifth clade, the Chromadorida (*Chromadorina bioculata* and *C. germanica*), eyespot pigment appears in extensions of marginal cells as in Enoplina. Thus, eye structure may have arisen independently at least four times within Nematoda. Taxonomists have noted eyes in many other nematodes; these need to be reexamined. We are extending the previous TEM studies to other species using DIC microscopy and serial sectioning together with phylogenetic analysis, hoping to deepen our understanding of the evolution of eyes.

CAENORHABDITIS ELEGANS IN NATURE. Caswell-Chen, E. P.,¹ J. Chen,¹ W. H. Moore,¹ S. A. Nadler,¹ G. W. Douhan,² and J. R. Carey.³ Departments of ¹Nematology, ²Plant Pathology, and Entomology, ³University of California, Davis, CA 95616.

Although the nematode *Caenorhabditis elegans* is the world's most completely understood animal, almost all of the research has been conducted in laboratory experiments, and with wild-type strains obtained from the *Caenorhabditis* Genetics Center (CGC). Research on *C. elegans* in natural environments is important, given that environment provides the context for the evolution and genetic control of development. The first objective of our research was to find *C. elegans* in natural environments. We collected *C. elegans* from several geographic locations in California, and identified the worms using a uniform standard and appropriate protocol for isolating and identifying candidate wild-type *C. elegans*. Our protocol used assessment of morphology and the biological characteristics, including the capacity to cross with the canonical wild-type strain N2, dauer formation, and incidence of facultative vivipary. Sequences of 28S rDNA, ITS rDNA, and polymorphic AFLP markers were used to assess the similarity of our isolates to N2 and some CGC wild-type strains. We conclude that: (1) *C. elegans* is not a commonly observed nematode in soil samples, and (2) The ecology and biodemography of *C. elegans* in the wild is very poorly known.

TREATMENT OF WATER WITH OZONE FOR CONTROL OF NEMATODES. Caswell-Chen, E. P.,¹ A. Pryor,² W. H. Moore,¹ J. Chen.¹ ¹Department of Nematology, University of California, Davis, CA 95616, and ²Ozone Process Consultants Inc., Davis, CA 95616.

Agricultural land and nursery soil are often contaminated with nematodes through the use of infested irrigation water. Ozone is a powerful oxidizing agent and has been shown to be an effective fungicidal and bactericidal agent when injected into water. This work assessed the potential for the use of ozone to treat water to eliminate nematodes. Ozone gas generated with a corona discharge ozone generator (Pacific Ozone Technologies, Brentwood, CA) was injected into water containing nematodes using a negative pressure injector (Mazzei Corp., Bakersfield, Ca) placed in the water recirculation loop. Water was pumped from a 1000 ml separatory funnel storage vessel through the injector using a diaphragm pump (Madden Manufacturing Inc., Elkhart Indiana). Ozonated water was returned to the storage tank from which samples were taken for ozone concentration and nematode viability. Ozone of 10.3 and 19.2 g/m³ concentration injected at the rate of 90 ml/min into water recirculating at 2.54 l/min resulted in dissolved ozone concentrations of 0.08 and 0.18 ppm respectively after 5 minutes. Ozone of 13.2 and 26.5 g/m³ concentration injected at the rate of 182 ml/min into water recirculating at 2.54 l/min resulted in dissolved ozone concentrations of 0.215 and 0.38 ppm respectively after 5 minutes. Exposures of *Caenorhabditis elegans* to aqueous ozone concentrations of 0.12 ppm resulted in death of almost all nematodes by 15 minutes. The efficacy of ozonation against other plant-parasitic nematodes will be discussed.

MELOIDOGYNE SPP. FOUND INFECTING WEEDS IN FLORIDA. Cetintas, R.,¹ J. A. Brito,² J. D. Stanley,² and D. W. Dickson.¹ ¹Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620; and ²Division of Plant Industry, Gainesville, FL 32614-7100.

Meloidogyne spp. are widely distributed in Florida and have been found infecting many ornamental, vegetable, and agronomic crops throughout the State. Weed hosts of these nematodes play an important role in their survival and maintenance that have detrimental effects on Florida's agricultural industry. It is important to correctly identify *Meloidogyne* spp. and their host range in order to successfully implement IPM programs. Soil and root samples from weed plants were collected from the major vegetable and agronomic crop producing regions and ornamental nurseries in Florida. Nematode identifications were based on isozymes as well as morphological characters when needed. *M. arenaria* was identified from *Amaranthus spinosus*, *Ipomoea triloba*, *Jacquemontia tamnifolia*, *Macroptilium lathyroides*, *Portulaca oleracea*, *Physalis angulata*, and *Physalis* sp.; *M. graminicola* from *Cyperus rotundus* and *Cyperus* sp.; *M. incognita* from *Amaranthus spinosus*, *Bidens alba*, *Desmodium* sp., *Urena lobata*, *Indigofera* sp., *Medicago lupulina*, *Melilotus albus*, *Pilea macrophylla*, *Portulaca oleracea*, and *Solanum americanum*; *M. javanica* from *Cyperus* sp., *Indigofera* sp., and *Portulaca oleracea*; *Meloidogyne* spp. from *Amaranthus hybridus*, *Amaranthus spinosus*, *Bidens alba*, *Cyperus rotundus*, *Cyperus esculentus*, *Eclipta prostrata*, *Jacquemontia tamnifolia*, *Medicago lupulina*, *Melilotus albus*, *Phyllanthus tenellus*, *Poinsettia heterophylla*, *Portulaca oleracea*, and *Solanum americanum*. The ability to correctly identify weed hosts of *Meloidogyne* spp. will benefit Florida's Agricultural Industry.

PATHOGENICITY OF MELOIDOGYNE MAYAGUENSIS COMPARED WITH M. INCOGNITA, M. ARENARIA, M. JAVANICA, AND M. FLORIDENSIS ON TOMATO IN MICROPLOTS IN FLORIDA. Cetintas, R.,¹ D. W. Dickson,¹ J. A. Brito,² and A. P. Nyczepir.³ ¹Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620; ²Division of Plant Industry, Gainesville, FL 32618-7100; and ³USDA-ARS SE Fruit and Tree lab, Byron, GA 31008.

Meloidogyne mayaguensis is a highly virulent root-knot nematode recently reported infecting various horticultural and ornamental plants in Florida. It is been reported that *M. mayaguensis* is able to break root-knot nematode resistance genes on tomato, sweet potato, and soybean and also induce more severe disease on many economically important crops compared to other common root-knot nematodes. Our objective was to determine the pathogenicity of this nematode relative to other common species found in Florida, namely *M. arenaria*, *M. incognita* and *M. javanica*. Microplots were inoculated with two levels of each nematode (low = one second-stage juvenile (J2) or egg/g of soil, high = three J2 or eggs/g of soil). 'Solar set' tomato was grown in the microplots for 82 days. The plants were removed and nematode densities in soil, percentages of roots galled, eggs, and plant fresh weights and heights were recorded. *M. mayaguensis* had a greater number of eggs than all other *Meloidogyne* spp. with the exception of *M. arenaria* ($P = 0.05$). *M. mayaguensis* also had a greater percentage of root galling at both inoculum levels, 95% and 99% for low and high, respectively. The size of galls induced by *M. mayaguensis* was unusually large compared with the other three species. *M. arenaria* had a greater number of J2 in soil than all other *Meloidogyne* spp. Fresh weight and height of plants were not different among treatments.

INNOVATIVE APPROACHES FOR SOIL FUMIGATION IN VEGETABLE PRODUCTION SYSTEMS. Chellemi D. O.¹ and J. Mirusso.² ¹USDA, ARS, U.S. Horticultural Research Laboratory, 2001 South Rock Rd., Fort Pierce, FL 34945, and ²Mirusso Enterprises, Delray Beach, FL.

An apparatus was developed for injecting soil fumigants beneath raised planting beds covered by plastic mulch without disturbing the integrity of the beds. Soil fumigation using a mixture of 1,3-dichloropropene (1,3-D) and chloropicrin (Pic) was combined with abbreviated soil solarization periods, virtually impermeable films (VIF) and different application sequences and evaluated for their effects on soilborne pests and marketable yield of fresh market tomato and pepper. Worker exposure to fumigants was mitigated by separating land preparation activities from the fumigant application process. Growers without access to drip irrigation systems were provided with an option for fumigating soil beneath established plastic mulched beds. Fumigation under VIF dramatically improved retention of 1,3-D and Pic in the soil, reduced the survival of plant pathogenic fungi, controlled nutsedge and achieved marketable yields equivalent to adjacent areas fumigated with methyl bromide:chloropicrin. Under VIF, pest control was achieved at reduced fumigant application rates. Combining soil solarization with soil fumigation improved control of soilborne diseases. Control of nutsedge species was improved when the fumigant application was delayed for 7 days after the plastic mulched beds were formed. Chemical and nonchemical soil disinfestation methods can be combined with novel application technology and procedures to improve the spectrum of pest control and reduce fumigant application rates.

A SNP TEST FOR CystX RESISTANCE TO SOYBEAN CYST NEMATODE. Chen, H.,¹ R. A. Vierling,¹ J. Faghihi,² and V. R. Ferris.² Departments of ¹Agronomy and ²Entomology, Purdue University, West Lafayette, IN 47907, and ¹Indiana Crop Improvement Association, Lafayette, IN 47909.

Our PUSCN14 soybean germplasm confers complete and broad-based resistance against soybean cyst nematode (SCN) and has been licensed under the name CystX to breeding companies for incorporation into high yielding cultivars. Breeders

using CystX germplasm have been eager for a quick laboratory test to aid in identifying CystX and non-CystX plants. A genetic test for the presence or absence of a particular QTL will greatly accelerate the breeding of new CystX cultivars. Our initial goal was to associate a SNP with the major SCN-resistance QTL on linkage group B. By comparing different single nucleotide polymorphism allele states (SNP) with greenhouse bioassay data from lines that differ in their SCN resistance, we have identified a SNP allele state that is correlated with the major genetic component of the CystX resistance. The test is nondestructive and leaf tissue from field grown material can be genotyped, which will allow for results to be known prior to flowering or harvest.

POST-REPRODUCTIVE SURVIVAL IN *CAENORHABDITIS ELEGANS* LIFE SPAN EXTENSION MUTANTS. **Chen, J., and E. P. Caswell-Chen.** Department of Nematology, University of California, Davis, CA 95616.

The post-reproductive life span of selfed hermaphrodite *Caenorhabditis elegans* was studied using large cohorts of individuals of wild type, N2, and two longevity mutants, *daf-2* DR1572 and *clk-1* MQ130. The post-reproductive life span as a proportion of total life span ranged from 39% to 69% for *C. elegans*, compared to the post-reproductive life span of ca. 36%, 51%, and 0% in humans, lab mice, and *Drosophila*, respectively. The *daf-2* and *clk-1* worms have a mean post-reproductive life span of ca. 21 and 10 days, respectively, an increase of 15.5 and 4 days greater than that in the wild-type. The longevity extension mutants did not show an increase in the duration of the reproductive period. Although each cohort and genotype was grown under the same environmental conditions, the post-reproductive life spans of the individual isogenic worms were not the same. The variation in post-reproductive life extension of *C. elegans* suggests stochastic influences on genetic control of life span.

EFFECTS OF CROPPING SYSTEMS ON PARASITISM OF *HETERODERA GLYCINES* BY *HIRSUTELLA RHOSSILIENSIS* AND *H. MINNESOTENSIS*. **Chen, Senyu.** University of Minnesota, Southern Research and Outreach Center, Waseca, MN 56093.

Hirsutella rhossiliensis and *Hirsutella minnesotensis* are two fungal species producing adhesive conidia to infect vermiform nematodes. These fungi have been commonly found on the second-stage juveniles (J2) of *Heterodera glycines* in Minnesota, South Dakota, and Michigan in the United States, and in China. The fungi appeared to be highly pathogenic to *H. glycines* and parasitized a high percentage of J2 in some fields. Effects of tillage and crop sequence on the fungal parasitism were investigated in Minnesota fields. The tillage effect was inconsistent. A higher percentage of J2 parasitized by the fungi in no-till than conventional tillage was observed only at one out of five sites in 1 year during 2002-2004 investigation. Nevertheless, in a no-till, monoculture soybean field, *H. rhossiliensis* parasitized a high percentage of J2, and the soil was suppressive to *H. glycines* populations. Crop sequence affected the fungal parasitism. Compared with susceptible cultivars, *H. glycines*-resistant soybean cultivars in the preceding year reduced the percentage of J2 parasitized by *Hirsutella* the following season. The percentage of J2 parasitized by *Hirsutella* was higher in soybean fields than in corn fields, and the percentage of J2 parasitized in midseason was generally higher than at planting and at harvest. After 2 or 3 years of corn, the fungal parasitism was generally undetectable. In plots of first-year soybean following 5 years of corn, fungal parasitism increased from undetectable level at planting to 2% to 4% of J2 parasitized by the end of the season. Fungal parasitism was similar in plots of second- through fifth-year soybean after 5 years of corn and in plots of soybean monoculture. The effect of crop sequence on the fungal parasitism of J2 may be attributed to a density-dependent relationship between the parasite and its host.

PASTEURIA PENETRANS: ECOLOGICAL INTERACTIONS WITH ROOT-KNOT NEMATODES. **Chen, Z. X.** Entomology Department, Michigan State University, East Lansing, MI 48824.

Pasteuria penetrans is a mycelial, endospore-forming, bacterial parasite of *Meloidogyne* spp. Endospores of *P. penetrans* are resistant to various environmental fluctuations, chemicals, and could survive dormant in soil for decades. Endospores attach to the cuticle of second-stage juveniles (J2) when they move through soil in search of hosts. Although the mechanism is unknown, as soon as J2 establish their feeding sites and start feeding the endospores germinate and a germ tube emerges from each penetrating the nematode body wall. After entering the nematode pseudocoelom, the germ tube develops into a cauliflower-like mycelium that multiplies and eventually forms endospores. Endospores are released into soil when plant roots along with the parasitized females decompose. Although *P. penetrans* is viable only in the host pseudocoelom, ecological interactions exist far and beyond the parasitic phase. J2 with endospore-attached have reduced mobility in soil and their ability to search for hosts is hindered as well. After entering roots, endospore encumbered J2 take longer to establish feeding sites and develop into females than healthy J2. While parasitized females continue to feed, they do not produce eggs. However, the loss in fecundity is not the major factor contributing to nematode suppression. The biological control potential of *P. penetrans* relies on an orchestrated cascade of nematode responses, growth manipulations, population interactions, and spatio-temporal population dynamics. The ecological interactions of *P. penetrans* with *Meloidogyne* spp. at all different levels, both inside roots and outside in soil, are discussed with an emphasis on relation to nematode suppression.

DRENCHING AND DIPPING TREATMENTS FOR NEMATODE-INFECTED PLANTS. **Chinnasri, B., B. S. Sipes, and K. T. Sewake.** Department of Plant and Environmental Protection Sciences, University of Hawaii, Honolulu, HI 96822.

Radopholus similis and *Aphelenchoides fragariae* are common in Hawaii's anthurium and orchid production. These nematodes affect flower quality and quantity. There are few effective controls. The effect of NaOCl and Zerotol, a product that generates hydrogen peroxide, on *R. similis* in tissue was elucidated. NaOCl at 0%, 2%, 3%, 4%, 5% and Zerotol at 0, 1:500, 1:200, 1:100, 1:50 (Zerotol product : water) were freshly prepared and 30 ml drenched onto an 11-cm-d clay pot filled with cinder media containing *R. similis* infected roots. Thirty minutes after exposure, nematodes were extracted by the Baermann funnel technique. NaOCl and Zerotol did not reduce the population of *R. similis* in plant roots. From 92 to 97% of the nematodes retrieved were active and moving in water 7 days after extraction. The efficacy of Avid® at 0.09 ml/L and Pylon® at 0.35 ml/L was tested against *A. fragariae*. Solutions were prepared and infected orchid immersed. One month later, nematode numbers were reduced by 95% by both the Avid and Pylon treatments relative to the untreated control. While NaOCl and Zerotol may be sterilants, Avid and Pylon, with their limited penetration of the plant tissue, have proven to be better alternatives in eliminating *R. similis* and *A. fragariae* from plant tissue. Plant tissues that inhibit direct contact between nematodes and chemicals reduce their efficacy.

CHANGES IN VIRULENCE OF *HETERODERA GLYCINES* FIELD POPULATIONS IN ILLINOIS. **Colgrove, A. L., and T. L. Niblack.** Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

The soybean cyst nematode, *Heterodera glycines*, is the most economically important soybean pathogen in Illinois. Yield losses due to *H. glycines* are limited through nonhost rotation and the use of resistant cultivars, more than 90% of which carry resistance from a single source, Plant Introduction (PI) 88788. In a 1991 survey, most of the *H. glycines* populations were classified as race 3, defined by Female Indices (FI) of less than 10 on all four race differential soybean lines, including PI 88788. ($FI = N^1/N^2 \times 100$, where N^1 = the mean number of females on a differential and N^2 = the mean number of females on a standard susceptible). Recent tests showed an increase in the number of *H. glycines* populations that are virulent on PI 88788. From 2001 through 2004, HG Type and IL SCN Type tests were conducted on *H. glycines* populations from Illinois fields. The HG Type test currently includes seven resistant soybean indicator lines. The IL SCN Type test, a truncated version of the HG Type test, consists of only those sources of resistance available to Illinois growers: PI 548402, 88788, and 437654. Of 110 tests, 74% had FI greater than 10 on PI 88788. In 2004, screening of 636 *H. glycines*-resistant cultivars showed that about half of those labeled as "resistant to race 3" expressed low or no resistance to three different *H. glycines* populations that fit the definition of race 3. Improved labeling and testing, of both soybean cultivars and *H. glycines* populations, are necessary to protect the usefulness of resistant cultivars to control losses due to the nematode.

MANAGEMENT OF THE FUSARIUM WILT/ROOT-KNOT NEMATODE DISEASE COMPLEX IN COTTON. **Colyer, P.D.,¹ and T. L. Kirkpatrick.²** ¹LSU AgCenter, Red River Research Station, Bossier City, LA 71113, and ²University of Arkansas, Southwest Research and Extension Center, Hope, AR 71801.

This presentation will focus on the current status of managing the Fusarium wilt/root-knot nematode disease complex in cotton. The Fusarium wilt/root-knot nematode complex of cotton was first described by Atkinson in 1892. Despite over 100 years of research, management of the disease complex remains difficult. Considerable progress has been made in developing resistance to Fusarium wilt in commercial cultivars, but developing commercial lines with high levels of resistance to the root knot nematode has not been as successful. Three commercial cotton cultivars with only moderate resistance to the root-knot nematode have been developed. In lieu of resistance, contact nematicides and soil fumigants have provided some success in reducing the severity of the disease complex, but their cost and availability in the future may limit their use. Management using crop rotations, which are limited, have not been widely adapted because of farm program limitations and reduced profits with alternative crops. Recent concern about the possible introduction of a virulent strain of the pathogen from Australia has resulted in the identification of several races not previously isolated in the USA. The identification of these races may alter the effectiveness of some of the management strategies.

NEMATODE CONTROL IN ESTABLISHED TURFGRASS. **Crow, W. T.** Entomology and Nematology Department, University of Florida, PO Box 110620, Gainesville, FL 32611.

Fenamiphos has been the primary nematicide used on turfgrasses in the United States for the past 30 years. However, production of this nematicide will cease in 2007. This has created a critical need to identify innovative new nematode management options for use on established turf. Therefore, during the past 5 years, a great deal of effort in Florida has gone toward nematode management research on turfgrasses. One of these innovations has been the application of low rates of soil fumigants such as 1,3-dichloropropene and metam sodium, which are now registered for this use in certain regions of the USA. These fumigants have proved efficacious against some plant-parasitic nematodes infecting turf, and damage to the grass is typically within acceptable levels. The development and use of new chemistry for use on turfgrass, while not

abundant, has been ongoing. Other approaches have been the use of botanically derived nematicides, various root stimulants, and other "alternative" products. Many of these alternative products have not been efficacious in research trials, while a few show potential. Biological control of plant-parasitic nematodes with various bacterial and fungal pathogens has been explored, and some have shown promise. Application of entomopathogenic nematodes to suppress plant-parasitic nematodes in Florida has not been successful, but may benefit other regions. Finally, breeding efforts to identify and use nematode resistant or tolerant turfgrass cultivars have been initiated. The practicality and challenges of each of these tactics will be discussed, as well as their use in an integrated pest management program. Previously published and unpublished data will be presented.

SURFACE SWIMMING BEHAVIOR OF INFECTIOUS JUVENILES OF *ROMANOMERMIS CULICIVORAX*. Dang, G. T. T., R. Perez-Pacheco, and E. Platzer. Department of Nematology, University of California, Riverside, CA 92521.

Romanomermis culicivora is a mermithid nematode parasitizing a wide range of mosquito species. Previous investigators have reported thigmotactic behavior of these nematodes. Better understanding of J2-stage thigmotactic behavior assist in generating a more efficient distribution method that would yield better results as a biocide. Thus, the surface-swimming behaviors of 33 *R. culicivora* J2-stage nematodes were quantified. A 24-well plate with polyethylene "stick" partitions cemented into the center of each well simulated partitions/debris in a water habitat likely encountered by *R. culicivora* juveniles. De-ionized water was pipetted into each well and one J2 juvenile was introduced into each well. Plates were maintained in a stable environment with constant ambient temperature and lighting for a 300 second observation period. Times at the center partition, intermittent space, well wall, and total resting time (consisting of the sums of the resting times at all three aforementioned areas) were recorded and averaged between all 33 subjects. The average times in seconds + SD spent at the simulated center partition, intermittent space (away from any partitions), and well wall were 16+21s, 194+69s, and 85+76s, respectively. The average total resting time was 85+60s. Based solely on these observations, it can be concluded that the thigmotactic behaviors of *R. culicivora* J2 juveniles, as previously reported, are common, although the juveniles seem favor the open environment as compared to partitions encountered in their environment. Hence, a more beneficial strategy for distribution of *R. culicivora* as a biocide would be to distribute the infectious J2 juveniles in the open areas of water where mosquitoes may be encountered.

SYMPTOM DEVELOPMENT OF *MELOIDOGYNE CHITWOODI* ON FIVE POTATO VARIETIES IN THE PACIFIC NORTHWEST. David, N. L., R. E. Ingham, K. J. Merrifield, and N. M. Wade. Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR 93771.

Columbia root-knot nematode, (*Meloidogyne chitwoodi*, CRKN) is a serious pathogen of potato (*Solanum tuberosum*) in many areas of the western United States. CRKN reduces tuber quality by inducing surface galling and/or internal brown spots. Earlier research by Pinkerton et al. (1991) suggested that CRKN infected Russet Burbank tubers between 940 and 1000 soil degree-days base 5C (DD_{5C}) after planting, but other varieties have not been examined. Furthermore, appearance of external and internal symptoms in relation to degree-day accumulation has not been established for any variety. In the present trial, the epidemiologies of internal and external tuber symptoms of CRKN were studied in five varieties to better understand host/pathogen dynamics. Alturas, Russet Burbank, Russet Norkotah, Ranger Russet, and Yukon Gold were planted in a field infested with CRKN. Tuber samples were collected weekly beginning 69 (870 DD_{5C}) days after planting and evaluated for incidence of external damage, as well as incidence and severity of internal damage. External symptoms were first observed on all varieties 85 days (1050 DD_{5C}) after planting, while internal symptoms were not observed until 92 days (1170 DD_{5C}) after planting. Although initial internal symptoms were delayed compared to external symptoms, final incidence of internal and external tuber symptoms was similar. Visual observation of infection sites in tubers confirmed that internal symptoms were not expressed until egg masses were present. Subtraction of the 540-640 DD_{5C} required from infection to egg laying (Pinkerton et al. 1991) from the 1170 DD_{5C} at which egg masses and internal symptoms were observed (this study) suggests that CRKN may infect tubers as early as 700 DD_{5C}) after planting.

INFLUENCE OF SOIL TEMPERATURES UNDER POLYETHYLENE MULCH AND BARE SOIL ON ROOT-KNOT NEMATODE EGG LAYING. Davila, M., L. H. Allen, and D. W. Dickson. Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611-0620.

The first egg laying by *Meloidogyne arenaria* race 1, *M. javanica*, and *M. incognita* race 4 was determined on four squash (*Cucurbita moschata*) crops grown on raised black polyethylene mulched drip irrigated beds or bare beds during two spring and two fall seasons in Florida. All beds were fumigated before planting in spring with methyl bromide 67-33 at 392 kg/ha. Nematode eggs were inoculated into each plant-hole at 6,000/plant-hole and then squash was seeded into the plant-holes. A second crop followed in the fall and the experiment was repeated the following year. Plots were arranged in a randomized complete block with four reps. Soil temperature was recorded at five depths (8, 23, 38, 53, 69 cm) using a temperature logger. Root systems were sampled every 2 days, stained and dissected to determine different developmental stages until appearance of egg laying females. Soil covered with mulch showed a higher mean soil temperature, and the

number of days required for egg laying was decreased compared to bare beds. Time and degree days (DD) above base temperatures 8.8, 10.2, and 9.3 °C for egg laying in mulched beds was ca. 16 days and 343 DD, 16 days and 320 DD, and 14 days and 307 DD for *M. arenaria*, *M. javanica*, and *M. incognita*, respectively. Egg laying was delayed about 2 days on bare soil compared with mulched soil for all nematode species and the number of DD accumulated increased due to the increase in days. All three *Meloidogyne* spp. evaluated showed similar DD requirements for egg laying in a range of 307-343 DD. This information may be useful for predicting egg mass formation by *M. arenaria*, *M. javanica*, and *M. incognita* under different climatic conditions, and also for estimation of numbers of generations completed.

WATERMELON YIELD IS SUPPRESSED BY *MELOIDOGYNE INCOGNITA*. Davis, R. F. USDA-ARS, Crop Protection and Management Research Unit, P.O. Box 748, Tifton, GA 31793.

Watermelon (*Citrullus lanatus*) is an excellent host for the southern root-knot nematode (*Meloidogyne incognita*), which is often found in fields of watermelon in the southeastern US. Surprisingly, the effect of *M. incognita* on yield of watermelon has not been documented previously. The effect of *M. incognita* race 1 on the number of melons produced, the weight of individual melons, and the total weight of all melons was evaluated in a field trial with 24 replications of methyl bromide-fumigated and non-fumigated plots. Fumigated plots had very low nematode pressure during the study, whereas non-fumigated plots had high nematode pressure. Mature fruit were collected during two harvests, with the second harvest 8 days after the first. Thirty percent fewer fruit were collected from non-fumigated plots than from fumigated plots during the first harvest. The total weight of fruit also was reduced by 30%, but the mean weight per fruit was not affected. The number and weight of fruit from the second harvest did not differ between fumigated and non-fumigated plots. When number and weight of fruit were combined for the two harvests, the number of fruit collected was 19% lower and the weight of fruit was 23% lower in non-fumigated plots than in fumigated plots, but the mean weight per fruit was not affected. Following the second harvest, the amount of root galling on all plants was assessed on a 0-10 scale based on percentage of the root system galled, and the mean ratings were 1.0 for fumigated plots and 5.3 for non-fumigated plots. *Meloidogyne incognita* reduced the yield of watermelon by reducing the number of fruit produced and delaying the maturity of those fruit, but the weight of individual fruits was not reduced. This documents that *M. incognita* is a significant pathogen of watermelon, and that nematode management is necessary.

NEMATODE COMMUNITIES FROM TWO VERNAL POOLS IN SANTA ROSA PLATEAU ECOLOGICAL RESERVE, CALIFORNIA. De Ley, P.,¹ I. T. De Ley,¹ M. Yoder,¹ S. Esfahani,¹ K. Carter,¹ J. Abolafia.² ¹Department of Nematology, University of California, Riverside, CA 92521, and ² Departamento de Biología Animal, Biología Vegetal y Ecología, Universidad de Jaén, Jaén, 23071, Spain

Vernal pools are ephemeral wetlands, typically with a diverse and highly adapted flora and fauna. We conducted the first nematode survey on record for this ecologically important habitat. Soil samples were collected on three dates from four locations in and around each of two vernal pool basins in the Santa Rosa Plateau Ecological Reserve. For eight of the twelve samples, nematodes from plant roots and debris were extracted in a mist chamber separately from the rest of the soil, which was sieved. An estimated fifty-one nematode genera were isolated, including at least sixty-two species. Soils from the two pools were substantially different in the composition and dynamics of their nematode communities. Nematode abundances were analyzed using the nonparametric Friedman test. Significant differences were observed between/among exact locations, sample dates and extracted sample fractions. Differences in abundance patterns were also significant across nematode genera. During the dry summer phase, roots and plant debris appear to play an important role in both pools as refuges for omnivorous, bacterivorous and phytoparasitic nematodes. Adults of large nematodes, in genera such as *Dorylaimus* and *Labronemella* congregate preferentially in dead plant material, perhaps not only in response to drought but also because of the fine texture of the underlying soil. "Aquatic" genera such as *Tobrilus* persist throughout the dry phase, especially in plant debris. We recommend that future nematode surveys of alternately inundated and desiccated soil habitats also include methods for extraction of roots and plant debris.

FIRST RESULTS FROM A COMBINED MORPHOLOGICAL AND MOLECULAR INVENTORY OF MARINE NEMATODES FROM THE GULF OF CALIFORNIA. De Ley, Paul,¹ I. Tandingan De Ley,¹ K. Morris,² E. Abebe,² M. Mundo,¹ M. Yoder,¹ J. Heras,¹ D. Waumann,³ A. Rocha-Olivares,⁴ A. H. Jay Burr,⁵ J. G. Baldwin,¹ and W. K. Thomas.² ¹Department of Nematology, University of California, Riverside CA 92521; ²Hubbard Center for Genome Studies, University of New Hampshire, 35 Colovos Rd., Durham, NH 03824; ³Universidad Autónoma de Baja California, Facultad de Ciencias Marinas, Apdo. Postal 453, Ensenada, Baja California, C.P. 22800, México; ⁴Centro de Investigación Científica y de Educación Superior de Ensenada, Apdo. Postal 2732, Ensenada, Baja California., CP 22860, México; and ⁵Department of Biological Sciences, Simon Fraser University, Burnaby, BC V5A 1S6, Canada.

Thirty-seven individual intertidal nematodes from the Gulf of California and from the California coastline were subjected to multifocal imaging by Video Capture and Editing followed by Polymerase Chain Reaction (VCE+PCR) of the D2D3

region of large subunit rDNA. The resulting video clips and sequence data are made available online in the database NematOL (<http://nematol.unh.edu/>). Sequence analysis suggests that these 37 individuals represent at least 32 species, none of which matches available D2D3 sequences in public databases. The recorded multifocal vouchers allowed us to provisionally identify each specimen to genus, and are currently being used to match specimens with ongoing identifications and descriptions of preserved material. We also successfully applied our methods to individuals of some other meiofaunal phyla such as gastrotrichs and tardigrades. Combining molecular barcodes with multifocal voucher archives through VCE+PCR and NematOL is part of a wider effort at structuring and changing the process of taxonomic discovery. We argue that data-rich surveys and phylogenetic tools for analysis of barcode sequences are essential components of the exploration of micro- and meiofaunal biodiversity.

SYSTEMATICS OF *MESOCRICONEMA XENOPLAX* REVISITED: COMBINED ANALYSIS OF MORPHOLOGICAL AND MOLECULAR MARKERS. De Ley, T. I.,¹ Q. Li,¹ J. Abolafia-Cobaleda,² M. McKenry,¹ I. Kaloshian,¹ and P. De Ley.¹ ¹Department of Nematology, University of California, Riverside CA 92521, and ²Departamento de Biología Animal, Biología Vegetal y Ecología, Universidad de Jaén, Jaén 23071, Spain.

The ring nematode *Mesocriconema xenoplax* is a widespread species whose systematics is complicated by numerous taxonomic and diagnostic problems at species and genus level. Field data suggest that California populations of *M. xenoplax* respond differently on identical grape rootstock varieties. In order to test these populations for morphological and/or molecular differences, we analyzed individual nematodes with Video Capture and Editing followed by Polymerase Chain Reaction (VCE+PCR) of the rDNA Internal Transcribed Spacer region (ITS) and large subunit D2D3 domain. Sequence analysis revealed the existence of two prevalent variants, differing from one another by eight substitutions in ITS and corresponding to a coastal haplotype versus a Central Valley haplotype. The latter included a population from the type locality of *M. xenoplax*. A few of the analyzed individuals exhibited single nucleotide polymorphisms matching the character state of either haplotype for the corresponding nucleotide positions. Both ITS and D2D3 phylogenetic analyses suggested that the 2 coastal populations formed a cluster within a clade that also included the 3 central valley populations. Observed genetic differences corresponded with a phenotypic difference in stylet length, with the coastal populations possessing smaller stylets compared to the Central Valley populations. We present morphological data from the analyzed individuals and discuss some implications for the taxonomy, identification and management of *M. xenoplax*.

IMPACT OF EARLY SOYBEAN PRODUCTION SYSTEM ON *HETERODERA GLYCINES* REPRODUCTION. Donald, P. A.¹ and G. Shannon.² ¹ARS USDA, Crop Genetics and Production Research Unit, Jackson, TN 38301, and ²University of Missouri, Delta Center, Portageville, MO 63873.

The emerging trend of matching relative maturity of soybean cultivars with annual precipitation patterns for increased and stable yield is gaining popularity in the Mid-South. Early-maturing soybean cultivars are planted in mid- to late April to ensure adequate water availability and avoid summer moisture deficits during seed fill, in contrast to the current practice of planting maturity group (MG) V soybean cultivars in May, resulting in inconsistent yields and seed quality due to the effect of drought stress during seed fill and wet conditions at seed maturation. Soybean cyst nematode (SCN)-susceptible and resistant soybeans (glyphosate-resistant) from MG 0 through IV were grown at two locations in Missouri and one in Tennessee for two years to measure the effect of early maturing cultivars on SCN reproduction. Cultivars were planted in mid to late April in 2002 and 2003 and matured in late July to early August. The Pi for the MO loam site was 415 and 196, the MO sandy loam site was 1120 and 1335, and the TN sandy loam site was 3560 and 1146 eggs/120 cm³ of soil in 2002 and 2003, respectively. SCN Pf/Pi ranged from 0.01 to 0.7 with a mean of 0.3 in the MO loam site, from 0.3 to 3.0 with a mean of 0.85 in the MO sandy loam site, and from 0.3 to 3.1 with a mean of 0.9 in TN. Soybean yield was highest with MG IV cultivars.

SOYBEAN CYST NEMATODE-RESISTANT AND SUSCEPTIBLE CULTIVAR YIELD IN INFESTED SOIL IN NORTH-CENTRAL USA. Donald, P. A.,¹ P. E. Pierson,² S. K. St. Martin,³ P. R. Sellers,⁴ G. R. Noel,⁵ A. E. MacGuidwin,⁶ J. Faghihi,⁷ V. R. Ferris,⁸ C. R. Grau,⁶ D. J. Jardine,⁹ H. Melakeberhan,¹⁰ T. L. Niblack,¹¹ W. C. Stienstra,¹² G. L. Tylka,¹³ T. A. Wheeler,¹⁴ and D. S. Wysong.¹⁵ ¹Research Plant Pathologist, USDA ARS Crop Genetics and Production Unit, Jackson, TN 38301; ²Technology Development Manager, Monsanto, St Louis, MO 63167; ³Professor, Department of Horticulture and Crop Science, The Ohio State University, Columbus, OH 43210; ⁴Director, Master Gardener Program, Department Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907; ⁵Research Plant Pathologist, USDA, ARS Soybean/Maize Germplasm, Pathology, Genetics Research Unit, and Professor, Department of Crop Sciences, University of Illinois, Urbana, IL 61801; ⁶Professor, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706; ⁷Research Nematologist, Department of Entomology, Purdue University, West Lafayette, IN 47907; ⁸Professor, Department of Entomology, Purdue University, West Lafayette, IN 47907; ⁹Professor, Extension Plant Pathology, Department of Plant Pathology, Kansas State University, Manhattan, KS 66506; ¹⁰As-

sociate Professor, Department of Entomology, Michigan State University, East Lansing, MI 48824; ¹¹Professor, Department Crop Sciences, University of Illinois, Urbana, IL 61801; ¹²Professor Emeritus, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108; ¹³Professor, Department of Plant Pathology, Iowa State University, Ames, IA 50011; ¹⁴Associate Professor, Texas Agricultural Experimental Station, Rt. 3, Box 219, Lubbock, TX 79401; and ¹⁵Professor Emeritus, Department of Plant Pathology, University of Nebraska, Lincoln, NE 68583.

The soybean cyst nematode (*Heterodera glycines* Ichinohe) (SCN) is distributed throughout soybean production regions of the United States and is of major economic importance. To assess the impact on yield of planting SCN-resistant and susceptible soybean cultivars in SCN-infested or -noninfested fields on soybean yield, research sites were established in 1994 and 1995 in 63 fields that were either SCN-infested or noninfested in 10 states in the north central USA. Eight SCN-resistant and eight SCN-susceptible public soybean cultivars representing maturity groups (MG) I to IV were planted at each site. Soil samples were taken at planting to determine initial nematode population density, SCN race, and soil classification. Soybean yields ranged from 658 to 3840 kg/ha overall. SCN-resistant cultivars yielded more than susceptibles in SCN-infested sites but not at noninfested sites. Highly significant interactions were measured among initial nematode population, cultivar, and location. Owing to the significance of location, no region-wide predictive equations could be developed for yield loss based on initial nematode population densities in the soil. Yield loss due to SCN was confounded by other stress factors which included temperature and moisture extremes.

TROPHIC CASCADES AND NON-TARGET EFFECTS OF AUGMENTING ENTOMOPATHOGENIC NEMATODE COMMUNITIES. **Duncan, L. W.,¹ J. H. Graham,¹ F. El-Borai,¹ and D. L. Porazinska.²** University of Florida, IFAS, ¹Citrus Research and Education Center, Lake Alfred, FL 33850, and ²3205 College Ave., Fort Lauderdale, FL 33314-7799.

Augmentative biological control is employed to increase the mortality of a pest by temporarily increasing the numbers of a biocontrol agent beyond its equilibrium density. Implicit in this tactic is the expectation that numbers of the biocontrol agent will eventually decline to the equilibrium state, due in part to the effects of natural enemies. Trophic cascades resulting from EPN augmentation could affect pest control variously, depending on the strength of specific density dependent responses in the food web. Two weeks after augmenting the entomopathogenic nematodes (EPN) beneath citrus trees to control soilborne larvae of the weevil *Diaprepes abbreviatus*, there were significant increases in mortality of sentinel weevil larvae, prevalence of the nematophagous fungi *Arthrobotrys* sp. and *Dactylaria* sp., and prevalence of free-living bacterivorous nematodes in cadavers of sentinel weevils. Six weeks following EPN augmentation, significantly fewer sentinel weevil larvae died in augmented compared to non-augmented plots. Application of composted manure as a mulch layer beneath trees decreased the prevalence of nematophagous fungi, increased the prevalence of endemic EPN, and increased the mortality of sentinel weevil larvae. In the laboratory, addition of *S. riobrave* to soil from the orchard increased the mortality of both *S. riobrave* and *S. diaprepesi* that were added to the soil seven days later. At the end of two weeks, greater numbers of nematophagous fungi and fewer EPN remained in soil that was augmented at the beginning of weeks one and two, than in soil that was augmented only in week two. These effects did not occur in soil that was air-dried to disrupt fungal activity prior to the experiment. *Heterorhabditis zealandica* was significantly less affected by pre-augmentation than were the steinernematid species. Apparently, effects of the post-application biology of EPN on biological control can be modulated in important ways by selecting appropriate species and cultural practices.

COLONIZATION OF PINE BOLTS, CANTS, SLABS, AND BOARDS WITH BARK BY *BURSAPHELENCHUS XYLOPHILUS* AND ITS *MONOCHAMUS* VECTORS. **Dwinell, L. D.** Southern Research Station, USDA Forest Service, Athens, GA 30602.

Wood packing material (pallets, packing cases, dunnage, etc.) (WPM) is a pathway for the introduction of invasive pests. The International Plant Protection Convention's ISPM-15 guidelines for treatment of WPM do not require treated wood to be bark-free. Some IPPC members, however, have unilaterally moved to include a bark-free requirement for WPM. The primary objective of this study was to determine if pine sawyers (*Monochamus* spp.) and the pinewood nematode (*Bursaphelenchus xylophilus*) (PWN) colonize heat-treated loblolly pine (*Pinus taeda*) slabs with surface bark and boards with edge bark. A second objective was to determine if these pests colonize heat-treated pine bolts. The experiment was established on October 1, 2004. Loblolly pines were felled and immediately sawn into 20 bolts, 6 cants, 32 boards and 32 slabs. Half of the bolts, slabs and boards were heated in an 85°C oven to a core wood temperature of 60°C. The samples were placed outdoors on two concrete pads for six weeks and then moved into a greenhouse. Colonization of the samples by pine sawyers and nematodes was assessed after 3 months. Pine sawyers and the PWN colonized all of the heat-treated and control bolts. *Pissodes nemorensis* (deodar weevil) colonized the control bolts, but not those that had been heat-treated. Some 6.25% of the slabs were attacked by sawyers and colonized by the PWN (one heat-treated and one control). The cants and boards with edge bark were not colonized by pine sawyers and/or the PWN. This study confirms that heat-treating fresh logs to a core temperature of 60°C does not prevent their colonization by pine sawyers and the PWN. Results of this study

also indicate that properly heat-treated lumber with edge bark used for wood packing material does not represent a significant risk from the PWN and its *Monochamus* vectors.

INTERACTIONS WITHIN THE *BURSAPHELENCHUS*–*MONOCHAMUS* COMPLEX. Dwinell, L. D. Southern Research Station, USDA Forest Service, Athens, GA 30602.

Many nominal species of *Bursaphelenchus* are phoretic on species of *Monochamus* (Cerambycidae). The evolution of phoresy, which is a symbiotic relationship in which one organism transports another of a different species, has given rise to several parasite-host interactions such as insect x conifer host, nematode x insect host, nematode x fungal host, and nematode x conifer host. *Pinus*, *Abies*, and *Picea* are common hosts of species of *Monochamus*. The nematode, which is carried in the insects' tracheal system, is transmitted to recently dead or dying trees or freshly felled logs when the insect oviposits (a.k.a. secondary transmission). This is the most common means of transgenerational transfer of species of *Bursaphelenchus*. The nematodes are secondary colonizers and feed on fungi in the wood. Blue-stain, caused by mycangial fungi, is an indicator of the tree or log also being colonized by bark beetles. Primary transmission occurs when *B. xylophilus* dauer larvae are transferred while the adult insect feeds on shoots of healthy, noninfested pines and the parasite-host interaction may result in pine wilt. Transmission may also occur when the vector insect feeds on exposed phloem/cambial tissue on freshly felled logs. *Bursaphelenchus xylophilus*, which is native to North America, and *B. cocophilus*, however, are the only two species within the genus generally considered to be pathogenic. *Bursaphelenchus cocophilus*, vectored by *Rhynchophorus palmerum*, is an obligate parasite, and causes red ring disease of palms. *Bursaphelenchus mucronatus* is the most common aboriginal species of *Bursaphelenchus* phoretic on Asian and Euro-Siberian species of *Monochamus*. *Bursaphelenchus xylophilus* and *B. mucronatus* have been intercepted in transported coniferous wood (i.e., logs, chips, lumber, and WPM). An understanding of the interactions within the *Bursaphelenchus*-*Monochamus* complex has contributed to the development of wood management systems and mitigation procedures that significantly reduce the risk of transported wood being a pathway for these pests.

***HISTIOSTOMA EGYPTI*, A NEW SPECIES OF HISTIOSTOMATID MITE ASSOCIATED WITH ENTOMOPATHOGENIC NEMATODES (HISTIOSTOMATIDAE, ACARID MITE, ACARI). El Bishlawy, S. M. O., and S. F. M. Allam.** Department of Agricultural Zoology and Nematology, Faculty of Agriculture, Cairo University, El-Gamaa St., Giza 12613, Egypt.

New species of histiostomatid mite was isolated from in vivo cultures of entomopathogenic nematodes in Egypt. This species of histiostomatid mite is distinguished by the anterior apodemes 3 extending around the apex of trochanter 3 and directed anteriomedially, medial apex is not closely with posterior median apodeme, genu of leg 3 with one CG setae was longer than MG setae, tibia of leg 3 with one Ö setae, genu and tibia 4 was the same as of leg 3 but with shorter setae in deutonymph (hypopus), the female has filiform dorsal setae. This mite species is associated with entomopathogenic nematodes (EPN) belong to the genera *Heterorhabditis* and *Steinernema*, it punctures the bodies of dauer stages and feeds on the internal and external bacteria and microorganisms associated with the in vivo cultures of these beneficial nematodes. When the population of this mite increases, it usually cause great damage to the EPN in vivo cultures.

VARIABLE-RATE APPLICATIONS OF TELONE II ON COTTON FOR RENIFORM NEMATODE MANAGEMENT. Ellis, G. R.,¹ G. W. Lawrence,¹ S. Samson,² W. A. Givens,² and K. S. Lawrence.³ ¹Department of Entomology and Plant Pathology and ²Department of GeoResources Institute, Mississippi State University, Mississippi State, MS 39762. ³Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849.

An experiment was conducted in a cotton field naturally infested with reniform nematodes. The field was located near Natchez, MS, and was approximately 20 acres in size. Soil samples were collected on a one-quarter acre grid to determine initial nematode populations. The populations ranged from 774 to 11,352 nematodes/500 cm³ soil. A distribution map was generated from the population levels, which served as the prescription map for the variable rate applications. Three threshold classes were developed from the interpolated map, which included low (700 to 3,000 nematodes), medium (3,000 to 6,000 nematodes), and high (6,000 to 12,000 nematodes). These thresholds were used to indicate which level of nematicide would be applied to the different zones. Telone II was applied at uniform rates of 1.5, 3, and 5 gallons/acre and a variable rate application of 1.5 to 5.0 gallons/acre depending on the degree of nematode infestation. Temik 15G was also applied at a rate of 4 pounds/acre for the control treatment. The conventional rate of Telone II at 5 gallons/acre produced the greatest seed cotton yield at 2,235 pounds per acre. The conventional rate of 3 gallons/acre and the variable rate applications were very similar, yielding 2,105 and 2,091 pounds/acre, respectively. The variable rate application rate averaged 3 gallons/acre. This could explain the similarity in the yields between the two treatments. The variable rate applications were successful because rates as high as 5 gallons/acre were applied and net returns were comparable to the 3 gallons/acre conventional rate application. Further research is being performed in order to maximize the efficiency of variable rate nematicide applications.

THE USE OF *S. FELTIAE*-INFECTED *GALLERIA MELLONELLA* FOR THE SUPPRESSION OF *MELOIDOGYNE JAVANICA*. **Fallon, D. J.,¹ H. K. Kaya,² and B. S. Sipes.¹** ¹Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822, and ²Department of Nematology, University of California, Davis, CA 95616.

Steinernema feltiae MG-14 juveniles and *S. feltiae*-infected *Galleria mellonella* were tested for their efficacy against *Meloidogyne javanica* root penetration and development. Application of *S. feltiae* juveniles reduced root penetration by 54% after 3 days ($P < 0.05$), but the suppressive effect was lost when egg production was measured after 35 days (n.s.). However, application of *S. feltiae*-infected *G. mellonella* reduced *i* penetration by 65% after 3 days ($P < 0.05$), and reduced egg production by 57% after 35 days ($P < 0.05$) when compared to control treatments. Biomass of *S. feltiae*-infected *G. mellonella* treated plants inoculated with *M. javanica* was 29% higher than water treated plants, compared to a 5% increase in *Steinernema feltiae* MG-14 juvenile treated plants after 35 days (n.s.). In a separate experiment, *S. feltiae*-infected *G. mellonella* applied to soil at different ages post-infection, reduced *M. javanica* penetration in the range of 27–59% (n.s.), but failed to reduce egg production after 35 days. Biomass of *S. feltiae*-infected *G. mellonella* treated plants hosting *i* increased 28% to 61% compared to water-treated plants (n.s.) at 35 days. Persistent suppression of *M. javanica* by entomopathogenic nematodes remains elusive, although direct application of *S. feltiae*-infected insects does hold some promise for further development.

PHYLOGENETIC RELATIONSHIPS AMONG SIX MERMITHID NEMATODE SPECIES INFERRED FROM 18S RDNA SEQUENCES. **Fen, X. U.,¹ E. G. Platzer,² and G. Wang.¹** ¹College of Life Science, Central China Normal University, Wuhan 430079, China, and ²Department of Nematology, University of California, Riverside, CA 92521.

This is the first report to infer the phylogenetic relationships among Mermithidae primarily from China. Nucleotide sequences of 18S rDNA (772 base pairs) were compared among six species. A molecular phylogenetic tree was constructed by using maximum parsimony (MP), neighbor joining (NJ) and minimum evolution (ME) methodologies. The sequence data revealed considerable variation in 193 nucleotide sites among the sequences analyzed, with transitions outnumbering transversions. All three phylogenetic analyses generated similar affinities, with six mermithid species grouped into two distinct clusters. The congeners *Romanomermis culicivora* and *R. wuchangensis* (mosquito parasites) clustered with *Amphimermis* sp. (Homoptera parasites). A second cluster was formed by *Ovomermis sinensis* (parasite of Lepidoptera), *Agamermis changshaensis* (parasite in Homoptera) and *Octomyomermis* sp. (parasitic in mosquito larvae). The genetic distances of these six species revealed closer intrageneric distances (0.27% nucleotide dissimilarity) relative to intergeneric comparisons (3.41 to 12.98%). Furthermore, inter-specific cross experiments revealed no reproductive isolation between *R. culicivora* and *R. wuchangensis*. These results suggest that *R. culicivora* and *R. wuchangensis* may be subspecies. In addition, an unidentified mermithid from a grasshopper in Hubei differed from *Ovomermis sinensis* by 0.67% suggesting a possible assignment to the genus *Ovomermis*.

CARBON CHANNELS AND FOOD WEB STRUCTURE. **Ferris, Howard.** Department of Nematology, University of California, Davis, CA 95616.

The services performed by soil food webs are determined by the identity, abundance and interactions of the component organisms. Food webs are shaped by resource availability, predation and environmental conditions. Resources enter the web through detritivore and herbivore channels which merge through the activities of generalist predators and omnivores. Biomass of omnivores and predators often does not correlate directly with that of lower trophic levels due to differing temporal and spatial dynamics. However, the abundance and diversity of predator and omnivore nematodes, and the inferred abundance of other organisms of similar sensitivity, must provide some indication of soil suppressiveness. The functional redundancy of a diverse food web suggests resilience of function to temporal population cycling or conditions unfavorable for some species. Functional complementarity, an amplification of functional redundancy, recognizes a positive relationship between level of a service and diversity of niche and attributes among organisms of a functional group. One determinant of the biomass of predator and omnivore nematodes may be the nature of the prey. Differences in metabolic rates of organisms in the bacterial and fungal channels lead to the speculation that conservation of C, and transfer to higher trophic levels, may be greater through the fungal than the bacterial channel. However, higher metabolic rates may be offset by higher rates of organism turnover. Expressing the biomass of herbivores and that of fungivores and bacterivores as a percentage of biomass of all nematodes reveals whether resources for higher trophic levels are supplied through channels in which negative or positive ecosystem services are performed; it may suggest need for reduction of the herbivore channel and amplification of the detritivore channels or the alteration of environmental conditions.

THE ROLE OF RESEARCH IN THE CRITICAL USE EXEMPTION PROCESS. **Finman, H.** U.S. Environmental Protection Agency, Office of Air and Radiation, Washington DC 20460.

The underpinning of the critical use exemption program is the scientific research, conducted by nematologists and others, on the technical and economic efficacy of alternatives to methyl bromide. The *Montreal Protocol on Substances that*

Deplete the Ozone Layer and the U.S. Clean Air Act require the phase out of methyl bromide but provide for certain, limited exemptions. Decision IX/6 agreed to by Parties to the Protocol allows for the continued production and import of methyl bromide for those critical uses that do not have technically and economically feasible alternatives available to them. U.S. government analysts and international experts rely on data generated by the scientific community to determine if a particular methyl bromide use pattern can adopt alternatives that will effectively control the pest/pathogen complex. Information on an alternatives' ability to control a particular pest must then be translated into economic considerations such as effect on yield and crop quality. This paper explains the critical use exemption process, elaborates on the methodology employed in critical use exemption program, and then describes the data elements that government experts need in order to make a determination on the technical and economic viability of a methyl bromide replacement.

SOIL ORGANIC MATTER INPUTS, SOIL HEALTH, AND ENHANCED ACTIVITY OF NON-SPECIFIC ANTAGONISTS OF PLANT-PARASITIC NEMATODES. **Forge, T. A.** Agriculture and Agri-Food Canada, Pacific Agri-Food Research Centre, Agassiz, British Columbia, Canada V0M 1A0.

The input of organic matter is a primary determinant of the structure and function of soil biological communities, including natural enemies (antagonists) of plant-parasitic nematodes. Over 65 years ago Linford and co-workers described suppression of root-knot nematodes resulting from the incorporation of pineapple crop residues, and attributed the suppression to enhanced activity of generalist microbial and invertebrate antagonists. Since then, research has shown that chitin-based amendments can suppress some nematode species via enhanced activity of chitinolytic bacteria and actinomycetes. Also, specific types of green manures and animal manure slurries are now known to release nemato-toxic compounds in soil and function as bio-fumigants. However, for agronomic reasons, the organic matter inputs most likely to be used in low-input, sustainable or organic cropping systems include relatively low-N or stable materials, such as senescent crop residues, mature composts, and solid manures applied at moderate N rates. I will present research showing that these types of organic matter inputs improve most indicators of soil health and increase indicators of soil food web structure, including populations of predacious nematodes and micro-arthropods. However, these inputs do not consistently result in suppression of plant-parasitic nematodes. I will discuss some possible reasons for the observed variable responses to composts and solid manures, and suggest some alternative approaches to understanding the relationships between enhanced food web structure, abundance of natural enemies, and regulation of populations of plant-parasitic nematodes. In particular, I propose that future research would benefit from increased focus on long-term regulation of nematode population dynamics rather than relatively short-term control, and better knowledge of the temporal dynamics of decomposition, soil food web structure and populations of the target species of plant-parasites.

INTERACTIONS BETWEEN THE SOYBEAN CYST NEMATODE, *HETERODERA GLYCINES*, AND *FUSARIUM SOLANI* F. SP. *GLYCINES* IN SOYBEAN ROOTS IN GREENHOUSE TRIALS. **Gao, X.,¹ T. A. Jackson,¹ K. N. Lambert,¹ S. Li,¹ G. L. Hartman,² and T. L. Niblack.¹** ¹Department of Crop Sciences, University of Illinois at Urbana and Champaign, Urbana, IL 61801, and ²USDA-ARS, 1101 W. Peabody, Urbana, IL 61801.

The soybean cyst nematode (SCN, *Heterodera glycines*) and *Fusarium solani* f. sp. *glycines* (Fsg), the causal organism of soybean sudden death syndrome (SDS) are major pathogens of soybean in the United States. There have been inconsistent reports on the nature of the interactions between SCN and Fsg in the past two decades. We investigated the interactions between SCN and Fsg in greenhouse trials. Experiments consisted of different inoculum levels of SCN and Fsg used simultaneously in factorial combinations on soybean cultivar Williams 82, susceptible to both SCN and SDS. Measurements included root and foliar symptoms, shoot height, flower numbers, root dry weight, shoot dry weight, area under the disease progress curve (AUDPC), and SCN reproduction. The trials were harvested 6 weeks after planting, and repeated in two greenhouses. Because environmental conditions of each greenhouse varied, statistical analyses considered the greenhouse effect. Both SCN and Fsg had an effect on the growth of soybeans. The high inoculum level of Fsg suppressed SCN reproduction, whereas the low Fsg inoculum level had no impact. SCN did not significantly increase AUDPC for SDS. The overall impact of SCN and Fsg on soybean growth and development was mostly additive.

ROLE OF A MAIZE LIPOXYGENASE IN THE INTERACTION OF MAIZE AND *MELOIDOGYNE INCOGNITA*. **Gao, X., J. L. Starr, and M. Kolomiets.** Department of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843-2132.

LOX genes encode for lipoxygenases that oxidize polyunsaturated fatty acids to produce an array of structurally and functionally diverse oxylipins. There are two general classes of LOX isoforms, 13- or 9-LOXs, based on the particular carbon atom that is oxidized by a particular isoform. Products of some 13-LOXs (i.e., jasmonates and C6 volatiles) function in plant defenses against pests and pathogens. The function of 9-LOX is not well understood. Here we report for the first time on the involvement of a 9-LOX gene in resistance to root-knot nematode. Northern blots and semi-quantitative RT-PCR analysis of the 9-LOX gene, *ZmLOX3*, from maize revealed that its transcripts increased steadily in roots inoculated with *Meloidogyne incognita*, with a peak in concentration at 7 to 10 days after inoculation. To further investigate

the potential role of oxylipins and *ZmLOX3* in the interaction of maize and *M. incognita*, a knockout mutant has been generated and its response to nematodes characterized.

FERGUSOBIA QUINQUENERVIAE/FERGUSONINA TURNERI COMPLEX GENOTYPES ARE NOT CORRELATED WITH MELALEUCA QUINQUENERVIA CHEMOTYPES. Giblin-Davis, R. M.,¹ W. Ye,^{1,6} K. A. Davies,² S. J. Scheffer,³ G. S. Taylor,² M. F. Purcell,⁴ G. Wheeler,⁵ T. D. Center,⁵ and W. K. Thomas.⁶ ¹FLREC, University of Florida/IFAS, Davie, FL 33314; ²University of Adelaide, Glen Osmond, SA 5064, Australia; ³USDA-ARS, Beltsville, MD 20705; ⁴USDA ABCL, Indooroopilly, QLD 4068, Australia; ⁵USDA-ARS, Invasive Plant Research Lab, Davie, FL 33314; and ⁶Hubbard Center for Genome Studies, University of New Hampshire, Durham, NH 03824.

A survey was conducted from July 1-22, 2004 from Sydney, New South Wales to Cairns, Queensland in Australia throughout much of the known range of *Melaleuca quinquenervia* for galls that were induced by the *Fergusonina turneri/Fergusonia quinquenerviae* complex. All *M. quinquenervia* trees found with galls were sampled for leaves (collected into ethanol) for subsequent GC-MS analysis to confirm the sesquiterpene chemotype (Viridiflorol vs Nerolidol) of the tree. GPS coordinate data were taken for each sampled tree and galls were dissected, if young and therefore less likely to yield mature flies, or set up in rearing bags to await fly emergence. Dissected galls were used to collect DNA samples of nematodes (into 1M NaCl) and larval flies (into 95% ethanol) and morphological voucher specimens were fixed in formalin-glycerin for processing to permanent slides. Overall, we collected 23 new samples each of flies and nematodes from individual shoot bud galls from chemotyped *M. quinquenervia* from 5 different locations in New South Wales and Queensland from areas that had both chemotypes. Twenty-six percent of these were from confirmed Nerolidol trees versus 74% from confirmed Viridiflorol trees. Nematodes and flies were sequenced for a partial fragment of mtCOI and compared. No consistent genotypic patterns that correlated with host chemotype were observed suggesting that the complex can be released for biological control against both chemotypes of the invasive tree existing in Florida without concern for selective advantages.

THE VAGARIES OF ALTERNATIVES TO METHYL BROMIDE FOR NUTSEDGE CONTROL. Gilreath, J. P., B. M. Santos, and J. W. Noling. University of Florida, Gulf Coast Research and Education Center, 14625 CR 672, Wimauma, FL 33598.

Control of yellow and purple nutsedge (*Cyperus esculentus* and *C. rotundus*, respectively) has been the most difficult task for all alternatives to methyl bromide tested to date. Nutsedges are difficult to control due to the rapid formation of tuber chains with apical dominance, the dense nature of those tubers, tuber dormancy factors, and the ability to survive under long term adverse conditions. Research focused on herbicides to be used in combination with alternative fumigants when it was determined that none of the alternatives provided consistent control. Pebulate (Tillam herbicide) in combination with 1,3-D + chloropicrin was selected as the most likely replacement for methyl bromide for mulched tomato production. Unfortunately, pebulate was not re-registered, thus setting back the search for alternatives for tomato. For other mulched vegetables and cut flowers, there are even fewer herbicide options to consider and manufacturers are not enthusiastic about pursuing registration of herbicides for minor use crops. Additionally, herbicide residues can be a problem and plant back restrictions exist on some labels. Current emphasis is on screening new fumigants and attempting to overcome the limitations of existing fumigants. In particular, we have investigated ways to improve upon fumigant delivery, either with new equipment, such as the Mirusso-Yetter Avenger coultter rig, or refinements in use of existing procedures, such as improved understanding of water and fumigant movement when delivered by drip irrigation. Another approach has been to re-examine rates for some fumigants; suggested rates for propylene oxide were 30 to 40 gal/acre but good control of nutsedge required 80 gal/acre. Studies with virtually impermeable films (VIF) and a high barrier metalized film indicate that rates either can be reduced to as low as 25% of normal for methyl bromide or standard rates with these barrier films can impart considerable improvement in nutsedge control (e.g. iodomethane and 1,3-D + chloropicrin). Most tomato producers in Florida will be trying a one-half rate of methyl bromide in combination with metalized film on part of their acreage this fall with some producers committing more than one-half of their crop to this technique. Lower rates are technically feasible, but difficult for growers to deliver with the degree of uniformity required for consistent efficacy.

EFFICACY OF USING AT-PLANT AND POST-PLANT APPLICATION OF ALDICARB FOR CONTROL OF HETERODERA SCHACHTII ON SUGAR BEET. Hafez, S. L. and P. Sundararaj. University of Idaho, Parma Research and Extension Center, 29603 U of I Ln, Parma, Idaho 83660.

Two field experiments were conducted at the Parma Research and Extension Center, Parma, Idaho to evaluate the efficacy of Aldicarb applied at planting or split application compared to Counter on the sugar beet cyst nematode management. The experiments were laid out in a randomized complete block design with seven treatments for the first experiment and ten treatments for the second experiment each with five replications. In the first experiment there was a significance difference in the sugar beet characters due to the application of Aldicarb treatment. Maximum beet yield and the highest beet vigor were recorded in the treatment with Aldicarb applied at the rate of 33 lbs at planting followed by

Aldicarb applied as planting/post (20+13). Minimum beet yield and the lowest vigor were recorded in the plot treated with Counter. In the second experiment application of Aldicarb at planting or split application eight weeks after planting significantly increased the beet yield as compared to the untreated control. Percent yield increase (25.9%) was maximum in the treatment with Aldicarb applied at planting and post application. Lowest level of yield increase was observed in the plots treated with Counter alone while Aldicarb applied along with Counter as post application significantly increased the yield. Aldicarb performed better than Counter in terms of beet yield and percent sugar.

CHEMICAL CONTROL OF *MELOIDOGYNE CHITWOODI* ON POTATO IN IDAHO. Hafez, S. L., and P. Sundararaj. University of Idaho, Parma Research and Extension Center, 29603 U of I Ln, Parma, Idaho 83660.

Two field experiments were conducted at the University of Idaho, Idaho, USA to study the efficacy of chemical nematicides for control of Columbia root knot nematode, *Meloidogyne chitwoodi* in potato. Both experiments were laid out in a randomized complete block design with thirteen and seven treatments each with seven replications in a sandy loam field heavily infested with *M. chitwoodi*. In the first experiment Vapam HL and Telone II were applied combinations with Temik 15G or Mocap EC or Vydate L (fall or spring) or Fosthiazate. In the second experiment efficacy of Mocap EC and Vapam (tank mix) were compared to Admire, Telone II, and Mocap alone for their efficacy on Columbia root knot nematode. Russet Burbank seed pieces were planted in rows three feet apart and five months after planting, the tubers were hand-harvested and weighed, graded and evaluated for nematode infection. Yield of tubers from different treatments of both studies indicated that there was a significant reduction in the percent nematode infection followed by an increase in marketable yield and total yield in different combinations of all treatments as compared to control plot. In the first experiment percent of tubers with nematode infection in treated plots ranged from 11 to 99.6. In the second experiment lowest level of nematode infection was recorded in the fall or spring application of Mocap + Vapam (tank mix) than other treatments. Percent of tubers with nematode infection in treated plots ranged from 8.6 to 91.9.

EFFECTS OF REDUCED RATES OF SOIL FUMIGANTS UNDER LOW DENSITY AND VIRTUALLY IMPERMEABLE FILMS FOR PEST-PATHOGEN CONTROL. Hamill, J. E., and D. W. Dickson. Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611-0620.

The efficacy of methyl bromide 67:33 (393, 295, and 196 kg/ha) and Telone C35 (327, 243, and 164 liters/ha) in conjunction with virtually impermeable film (VIF) technology were evaluated on tomato. Treatments were arranged in a split-plot design with four replicates. Fumigant treatments and an untreated control were the whole plots, and mulch type [VIF and low density polyethylene mulch (PE)] were the sub plots. Data collected included: (i) number of nutsedge plants 4 weeks after planting, (ii) marketable fruit yield, and (iii) subjective gall ratings. The interaction between mulch type and treatment effects on nutsedge counts and galling percentages were significant ($P < 0.001$) thus statistical comparisons were made within mulch type. All rates of methyl bromide provided control of nutsedges except at 196 kg/ha under PE mulch. Telone C35 at 327 and 243 liters/ha under VIF film provided moderate control of nutsedges. Telone C35 at 327 liters/ha under PE film gave moderate reduction of nutsedges compared to untreated plots. The interaction between mulch type and treatment effects on marketable yields were not significant thus data for the two mulch types were combined. All treatments had higher marketable yields than untreated plots ($P < 0.05$). Reduced rates of Telone C35 resulted in lower marketable yields than Telone C35 at 327 liters/ha and all methyl bromide treatments. Gall ratings for all methyl bromide treatments were lower than the untreated control, whereas for Telone C35 gall ratings were lower than in untreated plots only at 327 liters/ha under VIF ($P < 0.05$).

BIOFUMIGATION – AN ALTERNATIVE APPROACH TO METHYL BROMIDE FOR THE MANAGEMENT OF *MELOIDOGYNE INCOGNITA* ON CUCUMBER IN LEBANON. Haroutunian, Garo,¹ Frédéric Marion-Poll,² Saad L. Hafez,³ Dominique Kayser,⁴ and Berj Hatjian.⁵ ¹MeBr. Alternatives Project/Ministry of Environment-UNDP, Lebanon; ²Institut National Agronomique Paris-Grignon, France; ³University of Idaho, Parma Research and Extension Center; ⁴United Nations Development Program; and ⁵Ministry of Environment, Lebanon.

An experiment was conducted in a commercial green house to find out the efficacy of oil radish (*Raphanus sativus* spp. Var. Boss) for the control of *Meloidogyne incognita* in greenhouse grown cucumber. The experiment was in a completely randomized block design with four treatments and four replications each including an untreated control. The treatments were fallow, methyl bromide (1,000 kg/Ha), biofumigation crop (oil radish) with and without plastic cover. Oil radish seeds were sown in the greenhouse at the rate of 3 kg/dunum (1 dunum = 1000 m²) and 6 weeks after when the height of the green manure crop reached 35-40 cm, it was incorporated to the soil. Half of the area was covered with polyethylene films (50 µm), and the other half was left uncovered. On the same day, methyl bromide treatments were applied at the rate of 100 kg/dunum, covered with the polyethylene film. After 2 weeks, plastic films were removed, soil was aerated and 2-week-old cucumber seedlings were planted. Ten plants were marked in the middle of each treatment area and plant vigor, fruit yield were recorded at appropriate time. Root gall index of individual treatments were recorded at harvest. Lowest gall index was observed in the methyl bromide treated plots (0.9) followed by the green manure plots covered with polyethylene

films (1.7), green manure plots without cover (2.0), and fallow control (7.1). Highest fruit yield was from the green manure covered with polyethylene (2,733 g) and methyl bromide plots (2,785 g) followed by uncovered green manure plots (2,033 g). There was no significant difference in yield between methyl bromide treated and biofumigation plots.

COMPARATIVE STUDIES OF BODY AXIS DETERMINATION IN NEMATODE EMBRYOGENESIS. **Hasegawa, K.,^{1,3} K. Futai,³ S. Miwa,¹ and J. Miwa.^{1,2}** ¹Institute for Biological Function, and ²College of Bioscience and Biotechnology, Chubu University, 1200 Matsumoto, Kasugai, 487-8501 Japan; ³Laboratory of Environmental Mycology, Graduate School of Agriculture, Kyoto University, Sakyo-ku, Kyoto 606-8175 Japan.

The anterior-posterior axis determination of the pinewood nematode *Bursaphelenchus xylophilus* is opposite to that of the free living soil nematode *Caenorhabditis elegans*; we found that the presumptive region of sperm entry into the *B. xylophilus* oocyte becomes the future anterior portion of the embryo¹. To understand the evolution of nematode developmental systems, we cloned the following full length genes, *Bx-par-1* (Ser/Thr kinase), *Bx-mex-3* (RNA binding protein), *Bx-tbb* (beta tubulin), and *Bx-daf-21/hsp90* (heat shock protein), from *B. xylophilus* cDNA and analyzed their structure and function. They are all the homologues of *C. elegans* genes, whose expression is necessary for early embryogenesis. To apply the reverse genetics to *B. xylophilus*, double strand RNA for each of the four *B. xylophilus* genes was synthesized by in vitro transcription, and both *B. xylophilus* and *C. elegans* were soaked in dsRNA for RNAi. The animals were then checked for visible phenotypes. Both nematodes could suck up the dsRNA buffer, and we could detect the abnormal phenotypes caused by *B. xylophilus* dsRNA in *C. elegans*, but not in *B. xylophilus*. We are trying to establish other methods for functional analysis of *B. xylophilus* genes to understand the evolution of developmental systems by studying similarities and differences between these two nematodes.

LIGHT MICROSCOPE COMPARISONS OF THE PLEIOMORPHISM OF *PASTEURIA PENETRANS* AND *P. USGAE* IN *IN-VITRO* CULTURE. **Hewlett, T., S. Griswold, and K. Smith.** Pasteuria Bioscience, LLC, Alachua, FL 32615.

Pasteuria Bioscience has demonstrated that *Pasteuria penetrans*, a parasite of *Meloidogyne* spp. can be grown *in-vitro* in liquid culture media. Recent experiments have shown that the medium used for log phase growth of *P. penetrans* cells is suitable for culture and sporulation of *P. usage*, a parasite of *Belonolaimus longicaudatus*. This allows, for the first time, observation and comparison of cells and sporulation structures of *P. usage* with *P. penetrans* with an inverted microscope. *B. longicaudatus* males and females encumbered with endospores of *P. usage* were extracted from soil collected at a golf course green. *Meloidogyne arenaria*, artificially encumbered with *P. penetrans* endospores, was cultured on tomatoes in a green-house. Infected nematodes were surface sterilized and ruptured into culture media in six-well plates and examined with an inverted microscope over a two-week period. The *in-vitro* life cycle of *P. usage* is similar to *P. penetrans*. Cells are similar in size and shape but mycelial structures produced by *P. usage* are thicker and larger than *P. penetrans*. Sporulation structures were similar between the two species. *P. usage* produced larger thalli and spores as reported from scanning electron studies by Bekal, et al. 2001.

HAPLOTYPE HYPERVARIATION AND RAMPANT GENE REARRANGEMENT SHAPE MERMITHID NEMATODE MITOCHONDRIAL GENOME ORGANIZATION. **Hyman, B. C.,^{1,2} S. Tang,³ Z. Wu,⁴ E. G. Platzer,^{1,2} and R. Pacheco.²** Departments of ¹Biology and ²Nematology; Interdepartmental Graduate Programs in ³Genetics and ⁴Cell, Molecular, and Developmental Biology, University of California, Riverside, CA 92521.

Rare syntenic conservation, sequence duplication, and the use of both DNA strands to encode genes are signature architectural features defining mitochondrial genomes of Enoplea. To address the frequency of gene rearrangement within nematode mitochondrial DNA (mtDNA), we are studying mitochondrial genome variation within a more confined taxonomic unit, the family Mermithidae. The complete nucleotide sequences of the mosquito parasitic nematodes *Romanomermis culicivorax*, *R. nielsenii*, and *R. iyengarii* mtDNA have been determined. Duplicated expanses encompassing different regions of the mitochondrial genomes were found in each of these congeners. These mtDNA shared only four to seven rRNA and protein gene junctions, indicating extensive gene rearrangement within the *Romanomermis* lineage. The few conserved gene clusters are likely ancestral and facilitate prediction of progenitor gene orders and genetic mechanisms leading to the complex, contemporary transcriptional organization of *Romanomermis* mtDNA. Rapid structural changes are also observed at the conspecific level. Rolling circle amplification was used to isolate complete mitochondrial genomes from individuals in local populations of *Thaumamermis cosgrovei*, a terrestrial isopod parasite. Mitochondrial DNA length variants ranging from 19 to 34 kb are observed. The complete nucleotide sequences of two haplotypes have been determined, revealing a constant region encoding most mitochondrial genes and a hypervariable segment between the ATPase 6 and the 16S rRNA coding sequences. The hypervariable locus contains intact and truncated copies of several mitochondrial genes, duplicated to different copy numbers, resulting in mtDNA size variation. Unique haplotypes propagated at geographically distant sampling locations suggests that constant mutation generates new *T. cosgrovei* mtDNA forms.

THE EFFECT OF POTATO VARIETY ON COLUMBIA ROOT-KNOT NEMATODE (*MELOIDOGYNE CHITWOODI*) POPULATION DYNAMICS. **Ingham, R. E., N. L. David, N. M. Wade, and K. J. Merrifield.** Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR 93771.

Columbia root-knot nematode (*Meloidogyne chitwoodi*, CRKN) is a serious pathogen of potato (*Solanum tuberosum*) in many areas of the western United States. Research on the epidemiology of CRKN on potato has been conducted primarily on Russet Burbank, but little is known on other varieties. Previous research has indicated that the first generation of CRKN invades potato roots soon after they are present, and the second generation hatches from roots between 940 and 1000 soil degree-days base 5C (DD_{5C}) after planting. Based on these results, in-season nematicide programs to control CRKN with oxamyl have begun at 950 DD_{5C} after planting. Trials at Hermiston, Oregon were established to study CRKN population dynamics on five popular potato varieties grown in the western United States. Alturas, Ranger Russet, Russet Burbank, Russet Norkotah, and Yukon Gold were planted on 12 April, 2004 in a field infested with CRKN. Soil was sampled weekly from planting until 14 September. Regardless of cultivar grown, soil populations of CRKN declined rapidly after planting, and the second generation of CRKN began hatching from potato roots (indicated by rapid increase in J2 recovered from soil) between 900 and 1000 DD_{5C} after planting. This suggests that timing of nematicide applications based on CRKN population dynamics would be the same for all varieties tested. When population dynamics were evaluated based on density of CRKN at planting, second generation J2 were recovered from soil earlier when initial populations were high than when initial populations were low. These results suggest that in-season nematicide applications should begin earlier when initial CRKN densities are high.

EFFECT OF SOIL VOLUMES AND CONTAINER MATERIALS IN *ROTYLENCHULUS RENIFORMIS* AND *MELOIDOGYNE INCOGNITA* RACE 3 REPRODUCTION. **Jones, J. R.,¹ K. S. Lawrence,¹ E. Van Santen,² and S. R. Usery, Jr.¹** ¹Department of Entomology and Plant Pathology, ²Agronomy and Soils Department, Auburn University, Auburn, AL, 36849.

An experiment was conducted to evaluate optimum soil volumes and container types on *Rotylenchulus reniformis* and *Meloidogyne incognita* race 3 reproductions in the greenhouse. In all experiments, the treatments, arranged in a 3 x 5 factorial, were: (i) clay, plastic, or polystyrene containers and (ii) soil volumes of 90, 250, 500, 750 or 1,000 cm³. Pots were inoculated with 1,000 *R. reniformis* juveniles and vermiform adults or 1,000 *M. incognita* eggs. Each test was harvested 60 days after inoculation and repeated twice. In test 1, *R. reniformis* reproduction per cm³ of soil was greater ($P < 0.05$) in polystyrene pots at each different soil volume when compared to all other treatments. Egg reproduction was greater in 500 cm³ through the 1,000 cm³ polystyrene pots. In test 2, *R. reniformis* vermiform and egg populations were highest ($P < 0.05$) in the 90, 250, 500, 750 cm³ polystyrene pots. *Meloidogyne incognita* egg and J2 populations were greater ($P < 0.05$) for all polystyrene pots except for 1,000 cm³ in test 3. In test 4, *M. incognita* J2 populations were higher in clay pots with soil volumes of 250, 500, 750 and 1000 cm³, as compared plastic and polystyrene pots. Egg reproduction was greater ($P < 0.05$) in the 500 and 1,000 cm³ clay pots whereas the 250 cm³ plastic was highest and 90 cm³ and 750 cm³ polystyrene pots were higher.

TERMITE-ASSOCIATED NEMATODES IN SOUTH FLORIDA. **Kanzaki, N.,^{1,2} R. H. Scheffrahn,² and R. M. Giblin-Davis.²** ¹JSPS Postdoctoral Research Fellow, Laboratory of Forest Protection, Faculty of Agriculture, Kagoshima University, 1-21-24 Korimoto, Kagoshima, 890-0065, Japan, and ²Fort Lauderdale Research and Education Center, University of Florida/IFAS, 3205 College Ave., Davie, FL 33314.

Only a few nematode species have been reported as phoretic or parasitic associates of termites. To elucidate nematode-termite associations more clearly, common termite species in South Florida were surveyed for nematode associates. We examined eight species of "laboratory populations", which had been collected from various locations in South Florida and maintained in the laboratory and eight species of "field populations" collected at Secret Woods County Park, FL, on 21 February 2005. Individual termites (n = 10-43) from each colony were carefully dissected to examine for nematodes. Nematodes were observed from two species of laboratory populations and five species of field populations. Preliminary morphological observations of the nematodes were made with a microscope and samples were stored on Whatman FTA paper or in nematode lysis buffer for future sequencing efforts. A sub sample of the nematodes was inoculated onto Tryptic Soy Broth agar to establish cultures for identification and description. Most isolated nematodes were either dauer or parasitic juveniles of rhabditid species. A few diplogasterid dauer juveniles were observed. The number of nematodes isolated from individual termites was generally less than five. Therefore, in most cases, we could not clarify the termite organ harboring the nematodes and/or establish a voucher nematode culture. So far, we have established four rhabditid nematode cultures. Morphological and molecular identification and description of these cultured-species and intensive survey for the re-isolation of the other species are necessary to elucidate the diversity and natural history of these nematode-termite associates.

THE FIRST OBSERVED ASSOCIATION BETWEEN NEMATODES AND CARPENTER BEES. **Kanzaki, N.** JSPS Postdoctoral Research Fellow, Laboratory of Forest Protection, Faculty of Agriculture, Kagoshima University, 1-21-24 Korimoto, Kagoshima, 890-0065, Japan.

Several *Bursaphelenchus* species are known to have phoretic relationship with soil dwelling bees in North America. To test for this type of relationship in East Asia, the large Japanese carpenter bee, *Xylocopa appendiculata circumvolans* was surveyed for nematodes. Seventeen female adults of the bee were caught by net at Kamigamo Experimental Station of Kyoto University Forest, Kyoto, Japan, 31 August 2002, and were carefully dissected to examine for nematode infection. Forty-six percent (8 individuals) of the bee harbored third stage dauer juveniles of a species of nematode in their oviducts, and the average number \pm SD of nematodes isolated from the bees was 42 ± 97 (0-288). The nematode juveniles were cultured on mycelia of *Botrytis cinerea* on potato dextrose agar (PDA) and identified as a species of *Aphelenchoides* based on morphological traits of the adults. Cultured nematodes were used for sequencing the partial (ca. 1 kb) cytochrome oxidase subunit I of mitochondrial DNA and partial (ca. 900 bp) small subunit ribosomal DNA for comparisons with the sequences deposited at GenBank. The DNA sequences of the *Aphelenchoides* sp. were close to those of other *Aphelenchoides* species deposited in the database, but the precise phylogenetic status of the bee-associated species could not be determined because of limited sequences available for *Aphelenchoides* species. This *Aphelenchoides* sp. may be an undescribed species because of the isolation afforded by its unique association with a carpenter bee in wood. However, further morphological and molecular studies are needed to describe and determine the phylogenetic status of this species.

TRANSFER AND BUILDUP OF SOIL SUPPRESSIVENESS TO PEANUT ROOT-KNOT NEMATODE USING *PASTEURIA PENETRANS*. **Kariuki, G. M., D. W. Dickson, and M. L. Mendes.** Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611-0620.

Suppressiveness to the peanut root-knot nematode is caused by *Pasteuria penetrans*. To determine whether the suppressive agent could be moved, established, and managed we transferred *P. penetrans* and *Meloidogyne arenaria* race 1 to a noninfested site located at the Plant Science Unit, Citra, FL. Host crops for *M. arenaria* are grown in summer (peanut) and winter (wheat, vetch). At the end of the each cropping seasons the baseline population density of *M. arenaria* and *P. penetrans* were determined. *P. penetrans* increased from an average of 1.3 endospore/second-stage juvenile (J2) to an average of 5.0 endospore/J2 over one season. Three treatments, nontreated control, 1, 3-dichloropropene at 168 liters/ha and chloropicrin at 263 kg/ha (both broadcast) were imposed in spring of 2004. At the end of the summer crop 49% of nematode females in the nontreated control were infected by *P. penetrans*, followed by 1, 3-D with 33%, and chloropicrin with 22%. The number of *P. penetrans* endospores attached per J2 was 39, 34, and 16 in 1.3-D, nontreated, and chloropicrin treated plots, respectively. These treatment effects were different ($P < 0.05$). The study affirms that *P. penetrans* induced suppressiveness can be transferred, the soil fumigant 1, 3-D reduces numbers of J2 without decreasing *P. penetrans*, and that chloropicrin is detrimental to *P. penetrans*.

POCHONIA CHLAMYDOSPORIA, A BIOLOGICAL CONTROL AGENT FOR USE AGAINST ROOT-KNOT NEMATODES (*MELOIDOGYNE* SPP.) IN INTENSIVE VEGETABLE PRODUCTION. **Kerry, B.,¹ I. Esteves,¹ S. D. Atkins,¹ B. Peteira,² Ana Puertas,² and L. Hidalgo-Diaz.²** ¹Nematode Interactions Unit, Rothamsted Research, Harpenden, Herts, UK, and ²Centro Nacional de Sanidad Agropecuaria, Havana, Cuba.

Pochonia chlamydosporia is a diverse fungus and individual isolates require careful selection for their potential as biological control agents. Selected isolates have proved stable in mass culture and produce $>10^7$ chlamydospores on a cracked rice grain medium. Spore production and viability, virulence, enzyme production and rhizosphere colonization were similar in fresh material and in in vitro cultures maintained for >60 subcultures in the laboratory. The fungus may be applied as a soil amendment of colonized rice grain or as a chlamydospore powder at a rate of 5,000 chlamydospore g^{-1} soil. *Pochonia chlamydosporia* must be applied to soil before crops that support its growth in the rhizosphere but it is able to establish in soil from a single application and in appropriate crop rotations the fungus remained active throughout a cropping cycle of five crops over a 15 month period. In one cropping cycle of tomato–tomato–cabbage–bean–tomato, the infection of root-knot nematode eggs increased from 30% to $>80\%$ in the initial and final tomato crops, respectively, and the numbers of second-stage juveniles in soil were decreased by $>90\%$ in treated soil compared to untreated soil. The time taken for the fungus to provide significant levels of infection requires careful management. Real time PCR has been used to monitor and quantify specific isolates after their application. Toxicology tests for the fungus have been completed to EPA standards. The fungus is being commercially developed as the biological control agent, KlamiC.

EFFECT OF TRANSGENIC TOBACCO EXPRESSING MODIFIED RICE CYSTEINE PROTEASE INHIBITOR TO MIGRATORY ENDOPARASITIC NEMATODES. **Khaitong, T.,¹ K. Obsuwan,² B. S. Sipes,¹ and A. R. Kuehnle.²** Departments of ¹Plant and Environmental Protection Sciences and ²Tropical Plant and Soil Sciences, University of Hawaii at Manoa, 3190 Maile Way, Honolulu, HI 96822.

Rice cysteine protease inhibitor (rice cystatin) interferes with many plant-parasitic nematode digestive systems resulting in reduced nematode growth and development. Modified rice cysteine protease inhibitor (OclD86) gene, a more

effective mutant, was transformed into *Nicotiana tabacum* cv. Xanthi using *Agrobacterium* - mediated transformation. DNA from tobacco leaves rooted in selective media was subjected to PCR analysis and putatively transformed tobacco lines were analyzed by RT-PCR to confirm expression of the transgene. Approximately 12 transformed tobacco lines were selected and challenged with *Pratylenchus penetrans*. The effect of the transgenic tobacco plants on growth and development of the nematode was determined. Preliminary results showed no statistically differences in nematode final population, plant height, shoot fresh weight between transgenic tobacco lines and wild-type control 6 weeks after inoculation. However, shoot dry weight and root fresh weight in all transgenic lines were lower than the wild-type control. *Pratylenchus penetrans* did not develop well in the experiment.

TWO NEW SPECIES OF XYALA FROM THE SEA OF CORTEZ, MEXICO. King, Ian W., M. Mundo-Ocampo, J. Baldwin, and P. De Ley. Department of Nematology, University of California, Riverside, CA 92521.

This is the first in a projected series of studies on the free-living marine nematodes of the Sea of Cortez. Two new species in the genus *Xyala* Cobb, 1920 (Nematoda: Xyalidae) are described with light microscopy and scanning electron microscopy. Specimens were collected from an intertidal region near Santa Clara, Sonora State, Mexico. Both new species most closely resemble *X. striata* Cobb, 1920, a species to which a group of very variable morphotypes has been assigned. Despite the wide range of values for morphological characters which has resulted from these assignments, both of these new species differ from *X. striata* in having shorter spicules, fewer longitudinal striae and a relatively shorter pharynx and tail (as indicated by higher values for b and c, respectively). The two new species differ from each other in the diameter of the amphid (both absolute and as a percentage of the corresponding body diameter), the length of the cephalic setae, the relative position of the vulva, the number of longitudinal striae, and the length of the tail (both absolute and as a proportion of the total body length). Type specimens of both species will be illustrated as multifocal vouchers, which can be previewed online at <http://faculty.ucr.edu/~pdeley/vce.html>. A tabular key of *Xyala* will also be presented and briefly discussed.

CURRENT STATUS AND FUTURE DIRECTIONS OF METHYL BROMIDE ALTERNATIVES FOR PEPPER. Kokalis-Burelle, N., D. O. Chellemi, and E. N. Roskopf. USDA, ARS, U.S. Horticultural Research Laboratory, Fort Pierce, FL 34945.

Florida is one of the largest pepper producers in the U.S. with approximately 7,000 ha harvested in 2001 and 2002. Weeds are considered the most important pests in Florida pepper production followed by soilborne pathogens and nematodes. The most problematic weed species are nutsedge (*Cyperus rotundus* and *C. esculentus*), nightshade (*Solanum* spp.), white clover (*Trifolium repens*), and ragweed (*Ambrosia artemisiifolia*). Soilborne pathogens causing significant yield loss include Phytophthora blight (*Phytophthora capsici*), damping-off (*Rhizoctonia solani*), and *Pythium* spp.) root-rot (*Pythium* spp.), and white mold (*Sclerotinia sclerotiorum*). Root-knot nematode (*Meloidogyne* spp) is the most important plant-parasitic nematode affecting pepper. Currently, the best available alternative to methyl bromide for pepper production in Florida is a combination of Telone C-35 (1,3-dichloropropene + 35% chloropicrin) at 187 to 221 liters/ha broadcast 3 to 5 weeks before planting, followed by chloropicrin at 78 to 116 kg/ha shanked into the bed, and a herbicide tank mix of Clomazone at 1.1 kg a.i./ha and s-metolachlor at 0.72 to 1.1 kg a.i./ha over the bed top at plastic laying. Problems with currently identified alternatives include additional costs for broadcast fumigation and herbicides, increased field maintenance costs, and inadequate efficacy and potential phytotoxicity of herbicides. Also, uncertainty surrounding regulatory issues and registration status of existing and developing alternatives make production planning difficult for growers. Methyl bromide alternative research programs in Florida are concentrating on addressing issues important to growers, and on insuring the future viability of the pepper industry. Areas of research focus on development of alternative application technology, improving performance of existing chemical fumigants, evaluation of existing and new fumigants using virtually impermeable films, development of new chemical products, and integration of nonchemical strategies into commercial production systems.

EFFECTS OF NEMATODE MANAGEMENT STRATEGIES ON RHIZOSPHERE ECOLOGY. Kokalis-Burelle, N.,¹ R. McSorley,² and K.-H. Wang.² ¹USDA, ARS, U.S. Horticultural Research Laboratory, Fort Pierce, FL 34945, and ²Entomology and Nematology Department, University of Florida, Gainesville, FL, 32611-0620.

Effective nematode management strategies are closely related to crop management practices that improve soil fertility and the ability of plants to tolerate stress from plant-parasitic nematodes. Use of nematode-suppressive cover and green manure crops often reduce nematode activity and improve crop health by increasing soil organic matter, releasing allelopathic compounds, and by supporting beneficial rhizosphere microbial populations. These beneficial soil microorganisms improve crop health by increasing the availability and uptake of mineral nutrients, serving as antagonists to pathogens, and inducing systemic resistance. Up to 10% of rhizobacteria isolated from crops are antagonistic or suppressive toward plant-parasitic nematodes. The most predominant genera of beneficial bacteria are *Pseudomonas* spp. and *Bacillus* spp., both of which are considered important pathogen antagonists in most soil ecosystems. Establishment of beneficial rhizosphere bacteria using cultural practices such as the use of rotation, cover, and green manure crops provides an

opportunity to improve nematode management by integration of cultural practices with more sustainable nitrogen management techniques.

DAMAGE BY *MELOIDOGYNE* SPP. TO SOYBEAN IN INDIANA. **Kruger, G.,¹ J. B. Santini,² and A. Westphal.¹** Departments of ¹Botany & Plant Pathology and ²Agronomy, Purdue University, West Lafayette, IN 47907.

Root knot nematodes, *Meloidogyne* spp. (RKN), are damaging plant parasites of soybean in the southern U.S., but little is known about their effect on soybean in Indiana. We quantified the damage caused by RKN in a commercial soybean field known to be infested with RKN. Three patches in the field with stunted soybean plants were evaluated. A transect of 30 m was placed through each stunted area, starting with healthy plants, extending across the damaged area, and back into healthy plants. At every 1.5 m along each transect for a total of 21 samples per transect, 60 cm of row were sampled for top fresh weight. Roots were dug and rated for nematode-induced galling and soil samples were taken. The soil samples were used in a greenhouse bioassay. Motile nematodes were extracted with Baermann funnels. Soil fertility and soil texture analyses were conducted. RKN-induced galling was the greatest factor affecting top plant weight ($r^2 = 0.40$). Soil nutrient levels explained much less of the variation in top weights ($r^2 = 0.20$). RKN can damage soybean in the Midwest and deserves more attention once the distribution of *Meloidogyne* spp. is established.

PHYLOGENETIC RELATIONSHIPS OF HETERORHABDITID NEMATODES AND THEIR SYMBIOTIC PHOTORHABDUS BACTERIA. **Kuwata, R.,¹ T. Yoshiga,¹ M. Yoshida,² and E. Kondo.¹** ¹Laboratory of Nematology, Faculty of Agriculture, Saga University; ²National Institute of Agro-Environmental Sciences, 1-Honjo, Saga, Japan.

Entomopathogenic nematodes in the genus *Heterorhabditis* have specific mutualistic association with *Photorhabdus* bacteria. To elucidate a phylogenetic relationship between the nematodes and the bacteria, we analyzed the partial sequences of mitochondrial CO1 gene and of 16S rRNA gene from 16 nematode-bacterial complexes including six Japanese complexes. The 16 nematode isolates examined were classified into three species groups of *H. indica*, *H. megidis* and *H. bacteriophora* with showing a sister relationship between the last two nematode groups. The classification of the bacteria did not always coincide with that of the nematodes. The bacteria from *H. indica* and *H. bacteriophora* were referred to be a sister species: the bacteria from these two nematodes were different from all the bacteria from *H. megidis*. Based on these results we considered that the heterorhabditid nematodes and their symbiotic bacteria evolved similarly in some combinations and differently in the other combinations.

MANAGEMENT OF THE ROOT-KNOT NEMATODE, *MELOIDOGYNE INCOGNITA*, ON SUNFLOWER BY A FORMULATED SOIL AMENDMENT. **Korayem, A. M.,¹ A. Wahab,² and M. Mohamed.¹** ¹Department of Plant Pathology, National Research Center, Cairo, Egypt, and ²Department of Zoology and Nematology, Al-Azhar University, Cairo, Egypt.

The effect of different rates of application from a formulated compost containing plant and animal wastes and other natural compounds on the growth and yield of sunflower plants and on the root-knot nematode (*Meloidogyne incognita*) development was studied under field conditions. All tested rates of compost significantly reduced the root galls, females, egg masses, and *M. incognita* juveniles in soil. Correlation between the rate of application and nematode reproduction was positive. No significant increase in the plant length and disc weight was found at all tested compost rates, yet the fresh weight and seed yield were significantly increased. Improving the plant growth parameters was better at the medium application rate (2.5 kg/m²) than both the lowest (1.25 kg/m²) and the highest rate (3.75 kg/m²).

A BIOCHEMICAL AND MOLECULAR ANALYSIS OF SOYBEAN ROOTS EXPRESSING ROOT-KNOT NEMATODE CHORISMATE MUTASES. **Lambert, Kris N., S. Bekal, V. V. Lozovaya, and A. V. Lygin.** Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

Meloidogyne javanica chorismate mutases (*Mj-cm*) are proteins secreted by plant parasitic nematodes into plant tissues as the parasite establishes its association with its host. CM are enzymes in the shikimate pathway, a primary plant metabolic route that produces aromatic amino acids and numerous secondary phenolic compounds that control critical plant processes. The shikimate pathway does not exist in animals, but some plant parasitic nematodes secrete CM into the plant tissue presumably to manipulate the regulation of the plant pathway to benefit the nematode. Nematode CM could alter plant cell development, cell wall structure, or they could suppress host plant resistance responses. Transgenic soybean hairy roots expressing *Mj-cm* were analyzed for their composition of phenolic compounds using HPLC. In hairy roots expressing *Mj-cm* a general suppression of phenolic metabolism was observed; however, one phenolic compound increased dramatically upon *Mj-cm* induction. This metabolic profiling data suggests that the *Mj-cm* enzyme suppresses the synthesis of phenolics in plants, possibly by diverting the flow of the shikimate pathway through a different metabolic route. The implications of this shikimate pathway suppression are broad, but one consequence should be an increase in susceptibility to disease by plants expressing *Mj-cm*. To test this hypothesis, we infiltrated tomato leaves with *Agrobacterium* with or without *Mj-cm* in a plant binary vector. Tomato leaves infiltrated with *Agrobacterium* alone showed a rapid cell death

response, but *Agrobacterium* carrying *Mj-cm*, did not elicit a cell death response. This result suggests *Mj-cm* is capable of suppressing a pathogen induced plant defense response, implying *Mj-cm-1* could play a similar role in plant/nematode interactions.

THE EFFECT OF LESION NEMATODE MANAGEMENT BY ROTATION OR GREEN MANURE CROPS ON POTATO EARLY DYING. **LaMondia, J. A.** The Connecticut Agricultural Experiment Station Valley Laboratory, Windsor, CT 06095.

The effects of green manure rotation crops on early dying of potato caused by *Verticillium dahliae* and *Pratylenchus penetrans* were evaluated in microplots. Superior potato, Dwarf Essex rapeseed, Saia oat, Haychow sorghosudangrass, Trudan 8 sudangrass, Piper sudangrass, Pearl millet 101, *Rudbeckia hirta*, Polynema marigold, or Gary oat were grown for a full season (2002) in 12 replicate plots and shoots were either removed or tilled into soil. Lesion nematodes were extracted from oat cover crop roots in the fall. In the following year (2003), potatoes were grown, leaves symptomatic of early dying were counted weekly and area under the disease progress curve (AUDPC) was determined. Tuber yield was measured from each microplot. Nematodes were again extracted from fall oat cover crop roots. All of the rotation crops except Gary oat resulted in lower fall 2002 lesion nematode densities than potato. There were no differences in lesion nematode populations between plots in which shoots had been incorporated or removed. In the 2003 potato crop, after a single year of rotation, there were no differences in the AUDPC after different rotation crops, perhaps due to rapid senescence of the crop under wet conditions, but tuber yields were higher after *Rudbeckia hirta*, Pearl millet and Polynema marigold than for other rotation crops or continuous potato. Nematode recovery from the oat cover crop following potatoes in 2003 was greatest after oat, rapeseed, Trudan 8 sudangrass, and potato. Populations were still reduced after previous rotation to *R. hirta*, Polynema marigold, Pearl millet, Piper sudan, and Haychow sorgho-sudangrass. These data indicate that a single year of rotation can reduce *P. penetrans* populations and may aid in management of potato early dying.

EFFECT OF METAM SODIUM APPLICATIONS ON COTTON FOR THE MANAGEMENT OF THE RENIFORM NEMATODE IN MISSISSIPPI. **Lawrence, G. W.,¹ and K. S. Lawrence.²** ¹Department of Entomology and Plant Pathology, Mississippi State University, Mississippi State, MS 39762, and ²Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849.

Metam sodium, (Vapam), an isothiocyanate, was examined in Mississippi for its benefits in cotton production for management of the reniform nematode. The test was conducted in an established cotton production location and naturally infested with the reniform nematode (*Rotylenchulus reniformis*). Metam sodium (11.4, 47.3, and 75.5 liters/ha) was compared to with aldicarb (0.85 kg a.i./ha). Metam sodium was applied with a modified John Deere ripper hipper. A CO₂ charged system was used to propel the fumigant through flow regulators mounted on stainless steel delivery tubes attached to the trailing edge of forward swept chisels. Metam sodium was injected 45.7 cm deep 18 days prior to planting with one chisel per row. Rows were immediately hippered with disk hillers to seal and prevent rapid loss of the fumigant. Cotton plants were taller, had more open bolls and high cotton weights in the metam sodium and aldicarb treatments compared with the control. Seed cotton yields in the metam sodium plots were increased an average of 314 and 938 kg/ha over aldicarb and the control. Reniform nematode numbers averaged across the season were 4,362, 5,390, 4,979 reniform nematodes/250 cm³ soil in metam sodium, aldicarb, and the control plots, respectively.

EVALUATION OF TILLAGE AND EXPERIMENTAL NEMATICIDES ON *MELOIDOGYNE INCOGNITA* ON COTTON IN ALABAMA. **Lawrence, K. S.** Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849.

The effect of tillage and two experimental nematicides on *Meloidogyne incognita* populations and cotton plant growth, development and yield were observed in a cotton trial conducted at the Auburn University, E. V. Smith Research Station, in 2003 and 2004. The plot area had been a tillage system cover crop test for the previous 10 years. The two tillage systems were conventional tillage and strip tillage. Nematicide treatments consisted of an at plant application of aldicarb, thiamethoxam, imidacloprid, and a non-treated control. The numbers of thrips and the resulting insect damage index were reduced ($P < 0.05$) by all the nematicide treatments in both tillage regimes as compared to the control. Plants were taller ($P < 0.05$) in the strip tillage plots as compared to the conventional tillage plots at 21 and 35 days after planting (DAP). The numbers of *M. incognita* were reduced by all nematicides at 60 and 90 DAP in the conventional tillage plots. No reductions in numbers of nematodes were observed in the strip tillage treatments. Gall ratings averaged 3.5 in conventional tillage plots as compared to 4.1 in strip tillage plots; however, aldicarb reduced ($P < 0.05$) gall ratings when compared to thiamethoxam and imidacloprid. Seed cotton yield was increased ($P < 0.05$) by aldicarb under conventional tillage as compared to the experimentals and the control; however, under the strip tillage system imidacloprid increased ($P < 0.05$) yields over all other treatments.

ELEVATED CHOLESTEROL LEVEL IN A *CAENORHABDITIS ELEGANS* LINE IS CORRELATED WITH INCREASED LONGEVITY AND STRESS RESISTANCE. **Lee, E.-Y.,¹ Y.-H. Shim,² D. J. Chitwood,³ S. B. Hwang,⁴ J. Lee,⁴ and Y.-K. Paik.¹** ¹Department of Biochemistry, Bioproducts Research Center and Yonsei Proteome Research Center, Yonsei University, Seoul 120-749, Republic of Korea; ²Department of Biological Sciences, Konkuk University, Seoul 143-701, Republic of Korea; ³Nematology Laboratory, USDA, ARS, Beltsville, MD 20705; ⁴Department of Biological Sciences, Seoul National University, Seoul 151-742, Republic of Korea.

Like all nematodes, *Caenorhabditis elegans* has a nutritional requirement for sterol because of the lack of several enzymes in the de novo sterol biosynthesis pathway. Similar to most of the plant-parasitic nematodes investigated thus far, *C. elegans* converts plant sterols to cholesterol. Unlike phytoparasitic nematodes, *C. elegans* introduces an additional double bond at the carbon-7 position of the cholesterol nucleus to form 7-dehydrocholesterol, which typically is the major sterol of *C. elegans*. In animals that biosynthesize cholesterol de novo, the enzyme 7-dehydrocholesterol reductase (DHCR) catalyzes the reverse of this reaction, i.e., the reduction of 7-dehydrocholesterol to cholesterol. Analysis of sterols from *C. elegans* fed 7-dehydrocholesterol indicates that the DHCR-catalyzed reaction does not occur in *C. elegans*. To determine the effects of alterations in sterol composition on *C. elegans*, a mammalian DHCR was expressed in *C. elegans*. A DHCR expression vector was injected and then chromosomally incorporated by gamma irradiation. The transgenic *C. elegans* contained 80% more cholesterol than wild-type nematodes. The two lines were compared in sterol composition, number of offspring, life span, UV resistance, and thermotolerance. The transgenic strain produced 40% fewer offspring than the wild type, although growth rates were not significantly different. The mean life span of the transgenic strain was 31% longer in sterol-deficient medium than the wild type. The mean survival rate of the transgenic *C. elegans* was longer than that of the wild type when nematodes were exposed to ultraviolet irradiation or temperature stress (35°C).

EVALUATION OF ENTOMOPATHOGENIC NEMATODES AND GREEN MANURES FOR CONTROL OF CABBAGE MAGGOT, *DELIA RADICUM*. **Leger, C., and E. Riga.** Department of Plant Pathology, Washington State University, Pullman, WA 99163; and IAREC, Washington State University, Prosser, WA 99350.

Cabbage maggot (CM) is a major pest of the genus Brassica. Overwintering pupae become adult flies and the female flies lay eggs in the soil near the plant stems. The emerging CM larvae feed on roots and stems of plants and infested seedlings may wither or die. Up to now, CM control management relied on synthetic insecticides. Therefore, CM developed resistance against several insecticides which leads us to the search for alternative management strategies. For this purpose, the susceptibility of the CM first instar larvae to two entomopathogenic nematode species (EPN) (*Steinernema carpocapsae* and *Steinernema feltiae*) and two marigold species (*Tagetes erecta* and *Tagetes patula*) was evaluated in laboratory and glasshouse (GH) bioassays. Ten petri dishes containing 5 CM larvae were treated as follows: (i) three EPN concentrations (100, 300 and 500 infective juveniles/larva); (ii) three fresh tissue extracts of marigolds (leaves, leaves and flowers, and roots); and (iii) a combination of EPN and marigold. The petri dishes were incubated for 72 hr at 20°C and then larvae mortality was evaluated; each treatment was repeated three times. The preliminary results show that the most effective EPN species is *S. feltiae* causing up to 69 % mortality at a concentration of 300 IJ/CM larva. Similar experiment is in progress under glasshouse conditions using pots containing cabbage plants. Marigold fresh tissue extract assays under laboratory and greenhouse conditions are currently being evaluated.

HOST STATUS OF SEVERAL PERENNIAL ORNAMENTAL PLANTS TO FOUR ROOT-KNOT NEMATODE SPECIES IN GROWTH ROOM AND GREENHOUSE EXPERIMENTS. **Levin, R.,¹ J. A. Brito,² W. T. Crow,¹ and R. K. Schoellhorn.¹** ¹University of Florida, Gainesville, FL 32611, and ²Division of Plant Industry, Gainesville, FL 32608.

The host status of *Liriopne muscari* cv. Evergreen Giant, *Pittosporum tobira* cv. Variegata, *Salvia leucantha*, *Odontonema cuspidatum*, *Musa acuminata* ssp. *zebrina* cv. Rowe Red, and *Codiaeum variegatum* cv. Gold Dust to *Meloidogyne incognita* Race 2, *M. javanica*, *M. arenaria* Race 1, and *M. mayaguensis* was evaluated, twice in a growth room and once in a greenhouse, in separate experiments for each plant species. Each experiment consisted of five treatments (four *Meloidogyne* spp. and a control), which utilized 30 plants that were arranged in a randomized complete block design, with six replications in the growth room trials and three replications in the greenhouse experiments. Each plant was inoculated with 5,000 eggs and juveniles of a respective *Meloidogyne* spp., or remained noninoculated. The plants were maintained for at least 60 days in a temperature and light-controlled growth room, or for at least 154 days in a greenhouse, watered daily, and fertilized weekly. Plants were then harvested by replication. Root galling index, number of *Meloidogyne* eggs and juveniles, dry shoot weights, and fresh root weights were compared among nematode species. Results suggest that *L. muscari* cv. Evergreen Giant is a good host to all of the *Meloidogyne* spp. isolates tested except for *M. arenaria* Race 1, for which it is a poor host. *Salvia leucantha* and *M. acuminata* ssp. *zebrina* cv. Rowe Red were good hosts to the *Meloidogyne* spp. isolates tested, and *P. tobira* cv. Variegata, *O. cuspidatum*, and *C. variegatum* cv. Gold Dust were nonhosts to the *Meloidogyne* spp. isolates tested.

SMALL GRAINS AND ERECT EARS OF RICE CAUSED BY WHITE-TIP NEMATODE *APHELENCHOIDES BESSEYI* IN CHINA. **Lin, M.,¹ Ding, X.,¹ Wang, Z.,² Zhou, F.,² and Lin, N.¹** ¹Plant Protection College, Nanjing Agricultural University, Nanjing 210095, China, and ²Dahua Seeds Company Ltd. of Jiangsu Province, Nanjing 210018, China.

The small grains and erect ears of rice were found in a large scale in China. Typical symptoms were length of rice ears shorter than normal, number of grains was decreased. grain was little and black-brown in color, some kernels were distorted. The flag leaf was normal. An *Aphelenchoides* sp. was isolated from small grains and erect ears of rice cultivars Wuyujing 7. There were 2,014 nematodes in 100 infected kernels, and up to 74 in single grain, 92% of the grains tested had nematodes in the infested panicles. The nematodes in middle part of spike and shrunken grain were more than other parts and normal. The diagnosis characters of nematode were lateral fields about one-fourth as wide as body, with four incisures. The tail terminus bearing a mucro of diverse shape with three to four pointed processes. The female post-vulval uterine sac extending less than 50% of distance vulva to anus, no sperm present. Oocytes usually in two to four rows. The male spicules had a middle rostrum. Morphological character and measurements were conspecific with *Aphelenchoides besseyi* Christie, 1942.

AN INNOVATIVE METHOD FOR COUNTING CYSTS OF SOYBEAN CYST NEMATODES USING FLUORESCENCE IMAGING TECHNOLOGY. **Lu, H., J. Tallman, X. Hu, E. Anderson, M. Chamberlin, and G. Lu.** Pioneer Hi-Bred International Inc., A DuPont Company, Johnston, IA 50131.

Use of resistant cultivars is one of the major tactics for combating soybean cyst nematode (SCN), *Heterodera glycines*, which is the most destructive pathogen affecting soybean seed production. However, development of SCN-resistant soybean cultivars is a very labor-intensive process, partially due to the lack of a high-throughput method for counting the SCN females (cysts) that develop on soybean roots. We have developed a fluorescent image-based system for counting cysts on excised seedling roots cultured on artificial media in Petri dishes. In this system, the SCN cysts fluoresced when exposed to a wavelength of 570 nm. The fluorescent images were captured by a digital camera, transferred to a computer, and displayed on a monitor. Instead of counting the cysts on the roots using a microscope or an amplifier portion-by-portion, the cyst-image spots of the entire samples were viewed at once and counted manually. This system significantly improved the efficiency and accuracy of counting cysts developed on cultured seedling roots compared to a microscope counting method, and has potential for applications in high-throughput screening of nematode resistant crops.

EFFECTS OF *BELONOLAIMUS LONGICAUDATUS* MANAGEMENT AND NITROGEN FERTILITY ON TURF QUALITY OF GOLF COURSE FAIRWAYS. **Luc, John E.,¹ W. T. Crow,² R. M. Giblin-Davis,³ J. B. Sartain,⁴ and J. L. Stimac.²** ¹Clemson University, Pee Dee Research and Education Center, Florence, SC 29506; ²Entomology and Nematology Department, University of Florida, Gainesville, FL 32611; ³University of Florida, Fort Lauderdale Research and Education Center, Fort Lauderdale, FL 33314; ⁴Soil and Water Science Department, University of Florida, Gainesville, FL 32611.

Field experiments evaluated the effects of nematicide and fertility on turf quality of 'Tifway 419' bermudagrass parasitized by the sting nematode *Belonolaimus longicaudatus*. Treatments were: untreated, and nematicide (1,3-dichloropropene) treated combined with different N fertility levels. Effects of treatments on numbers of *B. longicaudatus* and turf quality were compared. Nematicide consistently reduced numbers of *B. longicaudatus*, but fertility level had no effect on *B. longicaudatus*. Turf quality of nematicide treated plots was improved compared with untreated plots during both experiments. Increasing N fertility improved turf quality in nematicide treated plots in some cases, but had no effect on turf quality in untreated plots in either experiment. Results suggest that N fertility may not improve turf quality at sites infested with *B. longicaudatus* unless nematode management tactics are effective in reducing nematode densities.

MANIPULATION OF HATCHING BEHAVIOR IN *HETERODERA GLYCINES* THROUGH EXPOSURE TO LOW TEMPERATURE. **Masler, E. P.,¹ S. Sardanelli,² and I. A. Zasada.¹** ¹Nematology Laboratory, USDA, ARS, Beltsville, MD 20705, and ²Plant Nematology Laboratory, University of Maryland, College Park, MD 20742.

We have previously shown that laboratory cultures of *Heterodera glycines* produced eggs that segregated into two groups defined by hatching behavior. Group I eggs readily hatched in water (27°C) in a linear pattern through 12 days, with 60 to 70 total percent hatch (TPH). The 30 to 40% of eggs that did not hatch in water, did hatch when exposed to soybean plants. These Group II eggs resembled *H. glycines* eggs collected from the field in their hatching behavior. Physiological differences between Group I and II eggs can provide a means to investigate developmental and hatching mechanisms in this species. In initial experiments, we used refrigeration (5°C) to mimic one environmental parameter encountered in the field. Refrigerated eggs, subsequently returned to 27°C, exhibited decreased TPH with increased refrigeration length (e.g., ~70 TPH, no refrigeration; 30 TPH after 2 weeks refrigeration, 20 TPH after 4 weeks, <10 TPH after 6 weeks). Regardless of refrigeration length, hatch rates were linear through 12 days, hatch curves had similar shapes, and mortality was the same for both non-refrigerated control eggs and refrigerated groups. Eggs refrigerated for more than 8 weeks did not hatch in water but hatched when exposed to soybean plants. We argue that refrigeration affects one or more critical stages in

embryogenesis, and developmental pathways that render the pre-infective juvenile ready to hatch were arrested. This developmental arrest was overcome by some outside stimuli (e.g., host plant) but not by others (e.g., temperature). Data are provided to illustrate the effect of refrigeration on hatching kinetics, and models are presented for investigating the developmental mechanisms affected.

NEW CHEMISTRY FOR NEMATODE MANAGEMENT. McCarter, J. P. Divergence, Inc., St. Louis, MO 63141.

Divergence Inc. (www.divergence.com) is developing safe and effective nematode control technologies with novel modes of action to combat nematode diseases of plants, animals, and humans. Extensive sequence data is now available from parasitic nematode sequencing projects (Nature Genetics, 36:1259, 2004). Using a genomic filtering approach (Trends in Parasitology, 20:462, 2004) we have selected a series of genes that are essential to nematodes yet distinct from the gene complement of other animals and plants, thus providing the basis for selective and safe modes of action. A number of the protein products of these genes are amenable to chemical inhibition and are currently being developed as nematicide targets. Efficacy data against *Meloidogyne incognita* and *Heterodera glycines* for chemistry developed from this approach will be presented.

INTERACTION OF NEMATODES WITH OTHER AGRICULTURAL PEST SPECIES. McGawley, E. C., M. J. Pontif, C. Overstreet, and J. B. Sumner. Department of Plant Pathology and Crop Physiology, LSU AgCenter, Baton Rouge, LA 70803.

Over the last 20 years, field and microplot trials have been conducted to explore the interactions between nematodes (soybean cyst, root-knot, reniform, and a variety of less pathogenic genera) and fungi (wilt, stem-canker, charcoal rot, pod and stem blight, stem rot, and anthracnose), insects (three-cornered alfalfa hopper and soybean looper), and weeds (hemp sesbania, morning glory, and johnsongrass). In most cases, interactions have been antagonistic with respect to reproduction of the pathogens and additive with respect to effects on plant growth. In some instances, however, synergistic interactions affecting plant growth limitation and pathogen reproduction have been observed. Data from studies such as these are essential if we are to eventually develop multi-pest injury thresholds that are based upon the total pest spectrum present at a given time. The underlying hypothesis to be tested across multiple crops is whether or not significant economic loss results from complexes of pest species present at levels usually considered individually to be below damage thresholds.

CHEMICAL MANAGEMENT OF NEMATODES IN LOUISIANA: 2000-2004 FIELD AND MICROPLOT TRIALS WITH RICE, SOYBEAN, SUGARCANE, COTTON AND ASSORTED VEGETABLES. McGawley, E. C., M. J. Pontif, C. Overstreet, and J. B. Sumner Department of Plant Pathology and Crop Physiology, LSU AgCenter, Baton Rouge, LA 70803.

Between 2000 and 2004, trials with rice, soybean, cotton and sugarcane were conducted to evaluate the efficacy of several labeled (Telone II [T--45.3 and 90.6 kg a.i./ha] and Methyl Bromide [MB-preplant--130.9 kg a.i./ha]) and one experimental (Agri-Terra, a colloidal suspension containing 1% monobasic sodium phosphate as the active ingredient [AgT-9.5 l/ha]) nematicide against indigenous nematode communities. Compared with non-treated controls at harvest, labeled materials provided acceptable reniform and cyst nematode control (population reductions of 28% to 55%) and yield response (increases of 8% to 30%). However, application of AgT produced reductions in nematode populations and increases in yield that were equal to and in most cases greater than these. Soil used in microplot trials was steam pasteurized and infested with nematodes in 2000 and 2004 and non-treated, naturally infested field soil was used in 2001 and 2002. Chemical treatments and controls were the same as those described for field trials with rates adjusted to 15 or 35 kg capacities of microplots. In all trials, AgT was equal to or better than labeled materials.

EFFECTS OF WINTER OVERSEEDING ON PLANT-PARASITIC NEMATODES ON GOLF COURSES IN FLORIDA. McGroary, P. C., and W. T. Crow. Entomology and Nematology Department, University of Florida, PO Box 110620, Gainesville, FL, 32611.

Winter overseeding is a cultural practice used to improve the aesthetics of golf courses in the southern United States during times of bermudagrass dormancy. Perennial ryegrass (*Lolium perenne*) and annual ryegrass (*Lolium multiflorum*) are frequently used for this purpose. One disadvantage of using these grasses for winter overseeding is that they might be suitable hosts for phytoparasitic nematodes. With soil temperatures remaining sufficiently high, phytoparasitic nematodes may increase by feeding on root biomass provided by the winter overseed; which could be problematic to bermudagrass transition in the spring. The objective of this study was to determine if winter overseeding with perennial ryegrass or annual ryegrass might increase nematode problems on bermudagrass in northern Florida. Two trials were established on fairway stands of 'Common' bermudagrass during the fall of 2003/2004. Plots in both trials were either overseeded with a blend of annual rye grass and perennial ryegrass or not overseeded. Population densities of *Belonolaimus longicaudatus* and *Hoplolaimus galeatus* were monitored throughout until bermudagrass came out of dormancy in the spring. In one trial numbers of *B. longicaudatus* increased ($P < 0.05$) in overseeded plots, more than doubling population densities compared to non overseeded. Numbers of *H. galeatus* were not different between overseeded and non-overseeded plots in either trial.

Additional experiments to evaluate the host status of perennial and annual ryegrass cultivars to *B. longicaudatus* under controlled environment conditions are being conducted.

IN-VIVO DAMAGE THRESHOLD FOR PLANTING JUGLANS SPECIES IN THE PRESENCE OF PRATYLENCHUS VULNUS. McKenry, M., T. Buzo, and S. Kaku. Department of Nematology, University of California, Riverside, CA 92521.

Growth of high vigor (HV) and medium vigor (MV) Paradox trees was monitored in the presence of various inoculation levels of *Pratylenchus vulnus*. This population had been disassociated from perennial crops for a decade so the replant problem was absent. Inoculum consisted of 1 m³ of soil placed in a field completely surrounding the roots of 4 mo-old trees. Using a serial dilution the 1 m³ of soil provided inoculum at 500, 20, 1, and 0 *P. vulnus* / 250cm³ of soil. These *Juglans* trees were clones. Trees of HV grew 30% taller than MV whether in 500 *P. vulnus*/250cm³ or none ($P = 0.05$). At the highest inoculum level both selections supported 500 to 2500 *P. vulnus* 8 mo later. Trees of HV exposed to 1 *P. vulnus*/250cm³ reached 0 to 7,500 *P. vulnus*/250cm³ with a mean population of 2,750. At inoculum levels of 500/250cm³ the soil counts around HV ranged from 500 to 2,500 with a mean of 1,425. Regardless of nematode inoculum level HV always produced significantly larger trunk diameters than MV. The HV trees without nematodes grew significantly larger trunks than those with 1 or more *P. vulnus*/ 250 cm³. The MV trees without nematodes grew significantly larger trunks than those with 20 or more *P. vulnus*. One vermiform *P. vulnus*/250cm³ of soil can quickly find roots of *Juglans* species and within 8 mo achieve overall population levels comparable to soils where 500 vermiform *P. vulnus* occur. The damage threshold level of *P. vulnus* to walnut is less than 1/250 cm³ soil.

OPTIMIZING PERFORMANCE OF SOIL SOLARIZATION FOR MULTIPLE PEST MANAGEMENT. McSorley, R.,¹ K.-H. Wang,¹ and R. J. McGovern.² Departments of ¹Entomology and Nematology and ²Plant Pathology, University of Florida, Gainesville, FL 32611.

Solarization provides a means of utilizing readily available solar energy directly for pest management. Solarization has been used successfully in Florida for managing a variety of nematode, disease, and weed problems. For example in one test in south central Florida, solarization suppressed *Meloidogyne incognita* and *Dolichodorus heterocephalus* throughout a 3-month crop of impatiens (*Impatiens x wallerana*), resulting in yields in solarized plots that were more than twice those of nonsolarized plots. Current work focuses on optimizing solarization conditions, such as duration, time of the year, and bed orientation. Encroachment of weeds into edges of raised beds can be a concern. Efficacy of solarization on edges of beds was examined in beds oriented in a north-south (NS) direction and in beds oriented in an east-west (EW) direction. During a 6-wk solarization period, soil temperatures >45°C were achieved on more days in edges of beds oriented NS (17 to 25 days) than in beds oriented EW (4 to 6 days). Invasion of weeds into bed edges was greater in beds oriented EW (70% to 77% of bed edge covered) than in beds oriented NS (9% to 22% of bed edge covered), and nematode population levels in bed edges followed similar trends. Orienting beds in a NS direction could produce an improvement in solarization if invasion of bed edges by weeds or nematodes is a concern. These and other findings can help to optimize the performance of solarization for management of a variety of soilborne problems.

CURRENT STATUS AND FUTURE DIRECTIONS OF METHYL BROMIDE ALTERNATIVES FOR FLOWER CROPS. McSorley, R., and K.-H. Wang. Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611.

Ornamental flower growers have relied on soil fumigation with methyl bromide (MB) to manage nematodes, soilborne diseases, and weeds. Nematode management in cut flowers is especially problematic because the nematode problems and their management are not well known for many flower crops. Additionally, alternative chemical products must not show phytotoxicity across a range of different plant species, and use must be compatible with an urbanizing production area. In recent tests in Florida, the alternative fumigants metam sodium (MS) and MS + chloropicrin performed similarly to MB in management of weeds and nematodes in snapdragon (*Antirrhinum majus*) production. When included, solarization performed similarly to these alternative fumigants and MB, especially in weed management. However, these tests were performed in sites with history of prior MB fumigation, so efficacy of multiple years of MS or solarization is unknown. Regardless of soil fumigation treatment, sites can become contaminated by soil and water movement resulting from heavy rainfall events and hurricanes. Management of field borders and water flow can be important in such sites. Since many growers produce a variety of different flower crops, rotation could be an option if susceptibilities of flower cultivars to nematodes and other pests were known. In greenhouse studies, several cultivars of lisianthus (*Eustoma grandiflorum*) were relatively resistant to *Meloidogyne incognita*, larkspur (*Consolida ajacis*) was intermediate, and snapdragon was highly susceptible. However, much more information of this type is needed before rotations to minimize impact from root-knot nematodes could be planned. Furthermore, successful management of one key pest, such as *M. incognita*, is an inadequate alternative to MB; multiple soilborne problems, including weeds and diseases, must be successfully managed as well.

POSSIBLE EFFECT OF ECOLOGICAL ADAPTATION ON *MELOIDOGYNE HAPLA* PATHOGENICITY. **Melakeberhan, H.,¹ S. Mennan,² and S. Chen.³** ¹Department of Entomology, Michigan State University, East Lansing, MI 48824; ²TUBITAK- NATO Visiting Scholar from Ondokuz Mayıs University, Samsun, Turkey, 55139; ³ University of Minnesota, Southern Research and Outreach Center, Waseca, MN 56093.

In the absence of resistant cultivars and the pending loss of methyl bromide (MBr), the vegetable and nursery industries in temperate climates face many challenges in managing *Meloidogyne hapla*. The challenges include identifying and developing sustainable MBr alternatives such as nutrition, and organic and mustard biofumigant types of soil amendments, which, in turn, require some understanding of the structurally depleted and chemically polluted soil environments where the nematode causes problems. As part of a project dealing with developing MBr alternatives, this study was conducted to determine if *M. hapla* populations from varying production systems differ in their response to MBr alternatives. Three Michigan nurseries (1, 2 and 3) and one (4) vegetable field were characterized for soil physical properties and nematode community structure and *M. hapla* isolates' pathogenicity. Fields 1, 2, 3, and 4 had loamy sand, sandy, sandy, and muck soils with pHs of 7.15, 6.56, 7.43, and 6.30, respectively, and significantly different from one another. The nursery fields showed significant imbalance in P, Ca and Mg while the vegetable field was nutritionally richer than the nursery fields. More herbivores and fungivores and less bacteriovores were found in Field 1 than in Field 4, indicating ecological differences between the two soil types. No parasitism of *M. hapla* populations by *Hirsutella* was observed in either field. When the four *M. hapla* isolates were subjected to nutrition-based N-Viro Soil (recycled municipal biosolid) amendments in two greenhouse experiments ($28 \pm 2^\circ\text{C}$) for a month using tomatoes and sandy loam soil, the population from the vegetable field was the least pathogenic. The preliminary results suggest that *M. hapla* populations may not respond similarly to MBr alternatives, challenging the "one-size-fits-all management approach". Possible relationships among soil physio-chemical-driven ecological changes, nematode adaptation and the challenges to developing appropriate management options and decision-making processes will be discussed.

THE RELATIONSHIPS AMONG SOIL CONDITIONS, FERTILIZER, *HETERODERA GLYCINES* POPULATION DENSITY, AND SOYBEAN YIELD. **Melakeberhan, H.** Department of Entomology, Michigan State University, East Lansing, MI 48824.

Soil fertility is among the options considered for managing *Heterodera glycines* and other stress-induced crop yield losses in many soil types and conditions. The objective of this two-year study was to determine the effects of fertilizer application on *H. glycines* population dynamics and soybean yield under low and high *H. glycines* and other unidentified soil stress conditions. In a split-plot design, *H. glycines*-resistant (Jack) and two susceptible (CX 252 and Kenwood-94) soybean cultivars were planted in the high and low stress portions of the field and treated with either none (control) or standard fertilizer rates with and without nitrogen. Nematode population dynamics in soil and roots were determined at approximately monthly intervals until harvest in 1998 and 1999. Preplant soil nutrient and soil pH analysis showed no difference among plots. Generally N, P, and K were higher in leaves from the low stress than from the high stress area, but the reverse was true for Ca, Mg, Mn, Fe and Al, suggesting soil-driven nutrient uptake imbalance. Jack accumulated less Fe and Al than either CX 252 or Kenwood-94, indicating differences in nutrient metabolism and in dealing with soil-driven stresses among the cultivars. All developmental stages and white, yellow, brown and dark colored cysts were recovered at all sampling times and were unaffected by nutrient treatment, indicating multiple generations per year. Seed yield was higher in the low stress compared with the high stress condition. While decreasing nodulation, application of fertilizer with nitrogen resulted in approximately 22% yield increase compared with the no fertilizer or fertilizer without nitrogen. These results suggest that supplementary nitrogen may be necessary to compensate for *H. glycines*-induced damage. The confounding factors and the agronomic challenges to adopting nitrogen supplement in legumes under nematode infestation will be discussed.

HOST STATUS OF PETUNIA CULTIVARS TO ROOT-KNOT NEMATODES. **Mendes, M. L.,¹ D. W. Dickson,¹ R. Schoellhorn,² R. Cetintas,¹ and J. A. Brito.³** ¹University of Florida, Entomology and Nematology Department, Gainesville, FL 32608-0620; ²University of Florida, Environmental Horticulture, Gainesville, FL 32611-0670; ³Division of Plant Industry, Gainesville, FL 32618-7100.

The host status of 19 petunia (*Petunia hybrid*) cultivars and one marigold cultivar to *Meloidogyne mayaguensis* and *M. incognita* race 4 were evaluated in a completely randomized design with six replicates in a greenhouse. 'Rutgers' tomato was included as a susceptible host to both nematode species. The inoculum consisted of 3,000 eggs and (or) second-stage juveniles/10 cm pot. The plants were harvested and the number of galls and egg masses per root system were recorded 64 days after inoculation. All petunia cultivars were highly susceptible to both nematodes. Marigold (*Tagetes erecta*) 'Jamie Spry' was a non-host to both nematodes. The size of galls induced by *M. mayaguensis* was unusually large compared with those induced by *M. incognita*. The average gall diameter of *M. mayaguensis* ranged from 0.10 cm to 0.60 cm on petunia roots and 0.10 cm to 1.1 cm on tomato roots, whereas the gall size of *M. incognita* on petunia roots was usually very

inconspicuous, ranging from 0.05 cm to 0.30 cm, and from 0.05 cm to 0.60 cm on tomato roots. There were no differences in susceptibility of petunia cultivars between the two nematode species ($P > 0.05$).

MELOIDOGYNE JAVANICA RACE 4 ON PEANUT IN FLORIDA. Mendes, M. L.,¹ D. W. Dickson,¹ R. Cetintas,¹ and J. A. Brito.² ¹University of Florida, Entomology and Nematology Department, Gainesville, FL 32608-0620; ²Division of Plant Industry, Gainesville, FL 32618-7100.

Meloidogyne javanica is one of the most common root-knot nematodes found in Florida. Recently it was reported infecting peanut in a commercial field near Williston, FL. The nematode species was identified based on perineal pattern and confirmed based on esterase phenotype. A single egg mass population was developed. Differential host test showed that this population reproduces on both peanut and pepper, thus the isolate is identified as *M. javanica* race 4.

SUPPRESSION OF MELOIDOGYNE HAPLA POPULATIONS BY HIRSUTELLA MINNESOTENSIS AND N-VIRO SOIL. Mennan, S.,^{1,3} S. Chen,² and H. Melakeberhan.¹ ¹Department of Entomology, Michigan State University, East Lansing, MI 48824; ²University of Minnesota, Southern Research and Outreach Center, Waseca, MN 56093; ³TUBITAK-NATO Visiting Scholar from Ondokuz Mayıs University, Samsun, 55139, Turkey.

The effects of the nematophagous fungus *Hirsutella minnesotensis* (Hm) and the recycled municipal biosolid N-Viro Soil (NVS) on *Meloidogyne hapla* populations from Rhode Island (RI), Connecticut (CT), Geneva, New York (NYG), Lyndonville, New York (NYL), Michigan (MI), and Wisconsin (WI) were studied in two greenhouse experiments. In Experiment 1, tomato seedlings were inoculated with 0 or 600 eggs of each nematode population separately mixed with 0, 0.02, or 0.1 g fresh Hm mycelium/100 cm³ soil in pots containing 500 cm³ soil and maintained at 25±2°C for two months. In Experiment 2, 0.1 g fresh Hm mycelium and 1 g NVS/100 cm³ soil were used alone or together following the same procedures in Experiment 1. Both Hm and NVS effectively suppressed *M. hapla* populations, but the degree to which each population was affected varied slightly. In Experiment 1, the fungus at 0.02 and 0.1 g reduced the total number of nematodes in roots by 61% to 98% for individual nematode populations, with the highest for NYG and RI, intermediate for NYL and CT, and lowest for MI and WI populations. In Experiment 2, Hm at 0.1 g reduced nematode number by 31% to 83% for individual nematode populations in one test, but only slightly reduced densities of NYG and CT populations in another test. NVS reduced nematode number by 33% to 92% for individual populations in two repeated tests. The combination of the two agents resulted in greater nematode reduction compared with Hm alone, but not as compared with NVS alone. Across all fungal and NVS treatments, reduction of nematode number was generally greater in NYG, CT, and NYL than in MI and NYL populations. This study demonstrated that Hm and NVS may be used as a potential suppressor of *M. hapla* in vegetable production systems in the Great Lakes Region.

CODON USAGE PATTERNS IN NEMATODEA: ANALYSIS BASED UPON 26 MISSION CODONS IN 32 SPECIES. Mitreva, M.,¹ M. C. Wendl,¹ J. Martin,¹ T. Wylie,¹ J. Parkinson,² M. Blaxter,³ R. H. Waterston,⁴ and J. P. McCarter.^{1,5} ¹Genome Sequencing Center, Washington University, St. Louis, MO 63108; ²Hospital for Sick Children, Toronto Canada; ³University of Edinburgh, UK; ⁴University of Washington, WA; and ⁵Divergence, Inc., MO.

Codon usage has direct utility in molecular characterization of a species and also serves as a marker for molecular evolution. In order to better understand codon usage within the diverse phylum Nematoda, we analyzed a total of 265,494 ESTs in 93,645 clusters from 30 nematode species. Putative translations, obtainable for 75% of clusters, were based upon homology to known or predicted proteins. The full genomes of *Caenorhabditis elegans* and *C. briggsae* were also examined. A total of 25,871,325 codons were analyzed and a definitive codon usage table for all species was generated. Similarity in codon usage can be quantified by the chi-square statistic. Related nematodes have previously been observed to have similar codon usage but the evolutionary distances at which conservation diminishes had not been established. We show that codon usage similarity in Nematoda is a short-range phenomenon, generally persisting over the breadth of a genus but then rapidly diminishing within each clade. A second focus was the underlying factors that bias codon usage. In comparing species, we find a strong correlation between the overall AT/GC content of the genome and similarity in codon usage. Surprisingly, differences exist among species and clades in the degree of codon-usage bias as measured by effective number of codons, indicating potential differences in selective pressures or population dynamics.

RENIFORM NEMATODE (ROTYLENCHULUS RENIFORMIS) SPREAD WITHIN A SOUTHEASTERN ARKANSAS COTTON FIELD OVER A THREE-YEAR PERIOD. Monfort, W. S.,¹ and T. L. Kirkpatrick.² ¹University of Arkansas, Plant Pathology Department, Fayetteville, AR 72701, and ²University of Arkansas, Fayetteville, SWREC, Hope, AR 71801.

Reniform nematode was first observed in May 2001 in a cotton field in Ashley County, Arkansas that was part of a variable-rate nematicide study utilizing grid-sampling. Intensive sampling of the 6 ha field was conducted in the spring and fall for four years (2001 to 2003) from 512 grids (30.5 meters × 4 meters) and nematode populations were determined. Reniform nematode population density maps were constructed utilizing Global Positioning Systems and Geographic

Information Systems. Reniform nematodes were initially observed in only 1 of the 512 plots in May 2001 with a population density of 6,364 nematodes/500cm³ soil. By the end of the first year the nematode was found in 17 of the 512 plots with population density ranging from 682 to 10,909 nematodes/500 cm³ soil. Over the course of the 3-year period reniform nematode incidence increased to 107 of 512 plots with population density ranging from 227 to 32,727 nematodes/500 cm³ soil. Reniform nematode spread could be explained by the direction of tillage and water flow in the low end of the field. Highest population densities were observed in areas of the field with soil types ranging from 54% to 60% silt.

THE EFFECTS OF ENVIRONMENTAL CONDITIONS ON EMBRYONIC DEVELOPMENT AND EGG HATCH OF THE SOUTHERN ROOT-KNOT NEMATODE, *MELIODOGYNE INCOGNITA*. Nagle, K., C. White, R. Powell, M. Rivera, L. Vansant, and M. Schroeder. DuPont Crop Protection, Newark, DE 19714.

Crop losses due to nematode attack are estimated to be between 80 and 140 billion dollars annually. Regulatory pressure on current chemical nematicides, both fumigants and non-fumigants, has provided a challenge to researchers in both industry and academia to find new chemical management tools that are safe and effective. Chemical discovery programs that involve the evaluation of the nematicidal activity of large numbers of novel compounds require the ability to provide millions of healthy test specimens on a weekly basis. Methods to maximize the efficiency of *Meloidogyne* J2 production from a laboratory culture are an integral component of the evaluation process. We previously reported on the use of salt to synchronize embryonic development and improve hatch of root knot nematode eggs. Here we extend those studies to include the effects of temperature and light on J2 production.

XIPHINEMA KRUGI: SPECIES COMPLEX OR A COMPLEX SPECIES? Neilson, R. Scottish Crop Research Institute, Dundee, DD2 5DA Scotland, UK.

The underlying basis of biodiversity is manifested directly and indirectly, through studies of taxonomic relationships, and of growth, form, adaptation and function. With a decreasing taxonomic resource base, at some point the delineation of a "species", based solely on molecular data will have to be addressed. Such decisions in the future will have to consider the impact of intra- and inter-population variability. Although recognized in classical taxonomy, few if any molecular studies have dealt with this issue. As a result of inter-population morphological and morphometric heterogeneity, the taxonomic status of *Xiphinema krugi* (Nematoda: Longidoridae) has previously been questioned. Thus 14 morphologically putative *X. krugi* populations were used as a model and subjected to both taxonomic and molecular analyses to compare and contrast inter-population variability. The molecular and classical taxonomic data yielded four congruent groupings. The study of *X. krugi* presented here clearly demonstrates the potential problems ahead for molecular based biodiversity/ecological studies. We have demonstrated the possibility that *X. krugi*, currently believed to be a morphologically variable single species is in fact, either a species complex comprising of four distinct genotypes or a complex of cryptic species. The use of emerging technologies in ecological studies not only presents opportunities to the ecologist but also challenges of interpretation and understanding at a resolution hitherto unknown.

CHARACTERIZATION OF A BACTERIAL ENDOSYMBIONT OF SOYBEAN CYST NEMATODE, *HETERODERA GLYCINES*. Noel, G. R.,^{1,2} and N. Atibalentja.² ¹USDA, Agricultural Research Service, Urbana, IL 61801, and ²Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

The existence of bacterium-like endosymbionts of cyst nematodes has been known for almost 30 years, having been observed during TEM of *Globodera rostochiensis* from Bolivia, *Heterodera goettingiana* from the UK, and *H. glycines* from the US. These endosymbionts were studied in females of *G. rostochiensis* and *H. goettingiana* and in second-stage juveniles of *H. glycines*. Our TEM studies also found the endosymbiont in various tissues of adult *H. glycines* including oocytes and sperm cells. Attempts to culture the endosymbiont have not been successful. In order to identify the organism we extracted DNA from surface-sterilized second-stage juveniles and used universal bacterial primers to PCR-amplify, clone, and sequence the near full length of the 16S rRNA and gyrase B (*gyrB*) genes. A BLAST search of DNA databases revealed a 93% similarity to the 16S rDNA and an 81% similarity to *gyrB* of the 'Bacterioidetes' symbiont, 'Candidatus Cardinium hertigii'. This endosymbiont, which parasitizes wasps of the genus *Encarsia*, was described in 2004. The bacterial endosymbiont of *H. glycines* has brush-like arrays of microfilament-like structures characteristic of the genus *Candidatus Cardinium*. However, a dissimilarity of 7% for the 16S rDNA and 19% for *gyrB* of the *H. glycines*-infecting bacterium with *Candidatus C. hertigii* indicate that the bacterial endosymbiont of *H. glycines* represents a new genus.

ALTERNATIVES TO METHYL BROMIDE USING AN INTEGRATED APPROACH. Noling, J. W.,¹ and J. P. Gilreath.² University of Florida, IFAS, ¹CREC, Lake Alfred, FL 33850, and ²GCREC, Baum, FL, 33598.

This symposia presentation will provide a basic overview of Florida research efforts to develop a more integrated crop and pest management program to replace methyl bromide in south Florida fruit and vegetable crops. These field research efforts initially focused on an integrated, co-application approach of different fumigants, herbicides, and other alternative tactics to achieve pest control efficacy and crop yield response similar to that of methyl bromide. This research has

repeatedly demonstrated that the pest control efficacy of the available fumigant alternatives is generally more highly dependent upon uniform delivery and distribution within the field. To address problems of application uniformity and treatment performance inconsistency, significant refinements have been made in soil application technologies of the alternative fumigants via standard shank, mechanical incorporation, and with plastic mulch technologies to reduce overall field application rates and soil emissions. Significant advancements have been made in our understanding of the dynamics of gas phase movement of fumigants in soil and of optimized drip irrigation delivery of agrochemical. New pest management strategies include integration of new tillage operations and irrigation application practices to enhance the performance of chemigated fumigant alternatives. A variety of studies have demonstrated the importance of weeds to serve as excellent hosts to nematodes (notably *Meloidogyne* spp.), and consequently, the need for integrating weed and nematode management. In summary, the basic elements and outcomes of a Florida research program to will be presented, including discussion of the importance of edaphic and environmental conditions, multiple pest interactions, new IPM field monitoring protocols for root-knot nematodes, and biologically and culturally integrated pest management strategies.

GROWER CONDUCTED ROOT-GALL SURVEYS FOR FIELD DIAGNOSIS AND SAMPLING FOR ROOT-KNOT NEMATODE. Noling, J. W.,¹ L. W. Duncan,¹ and J. P. Gilreath.² University of Florida, IFAS, ¹CREC, Lake Alfred, FL 33850, and ²GCREC, Baum, FL, 33598.

With methyl bromide soil fumigation, grower decisions regarding nematode monitoring or management were unnecessary because of the broad spectrum efficacy of the fumigant. With less effective pest management alternatives, new monitoring systems are needed, particularly in multiple cropping systems where crops are grown sequentially. Eight commercial vegetable fields were comprehensively surveyed using root gall assessments, rather than soil sampling, to characterize root-knot nematode (*Meloidogyne* spp.) spatial patterns and mean gall severity. Plants were systematically removed at increments of 15 m within rows sequentially across the field after final crop harvest. Basic sampling units consisted of six-row (198 m long) blocks. Mean galling was estimated for a range of sample sizes and targeted field locations for each of 96 blocks. An aggregated distribution was observed in most blocks. Random sampling from the data set revealed that sampling precision increased significantly when overall root gall severity was greater than 5 (scale 0 to 10) in a field block. At this level of root galling, a minimum of 4 to 6 plants/six-row block must be inspected to achieve acceptable precision. When the nematode problem is less severe and overall root gall severity less than 5, as many as 2 to 10 more plants must be inspected per block to assess nematode problems with the same sampling precision. These studies demonstrate that use of crop plants as bioindicators of nematode problems can be used to accurately characterize root-knot nematode infestation level and spatial pattern. The new sampling procedure is user friendly and will enhance nematode scouting and problem diagnosis capabilities of growers.

NEMATODE INTERACTIONS ON PEACH AND PECAN. Nyczepir, A. P., C. C. Reilly, and B. W. Wood. USDA-ARS, SE Fruit & Tree Nut Research Laboratory, 21 Dunbar Rd., Byron, GA 31008.

The association of peach-tree-short-life (PTSL) with old peach land has led to the hypothesis that some predisposing factors persist in old orchard sites that make new trees more susceptible to cold injury, bacterial canker (*Pseudomonas syringae*) or both. It has been demonstrated that old sites are not a prerequisite for PTSL to occur and that the presence of *Mesocriconema xenoplax* is a critical biotic component for PTSL development. Recent results indicate that PTSL development in subsequent peach orchards was dependent upon the cumulative population exposure of trees to *M. xenoplax*. In the real world, however, *M. xenoplax* is not the only nematode pest present in peach orchards in the southeastern United States. Other pests include *Meloidogyne* spp. and *Pratylenchus vulnus*. The question that arises is the interactive role these other nematodes have with *M. xenoplax* in predisposing trees to PTSL. In two separate field microplot studies with *M. xenoplax*, evidence indicates that no trees growing in either *M. incognita* or *P. vulnus* infested soil died from PTSL. However, tree growth was reduced more in the presence of *M. xenoplax* + *M. incognita* than either of these nematodes alone. In pecan, the interaction between *M. xenoplax* and *M. partityla* was suspected in trees exhibiting above-ground symptoms that included dead branches in the upper canopy, stunted growth, and (or) mouse-ear leaf symptoms. In a field microplot study, trees growing in *M. partityla* and *M. partityla* + *M. xenoplax* infested soil exhibited greater mouse-ear symptoms than in the noninoculated plots. Understanding the interaction between nematode pests and their associated disease complex is important in developing the appropriate nematode management strategy.

SUSTAINABLE CONTROL OF SOILBORNE PATHOGENS IN DRYLAND TARO. Ortiz, A.,¹ B. S. Sipes,¹ J. Cho,¹ J. Y. Uchida,¹ and S. Miyasaka.² Departments of ¹Plant and Environmental Protection Sciences and ²Tropical Plants and Soil Sciences, University of Hawaii at Manoa, Honolulu, HI 96822.

Taro (*Colocasia esculenta*) germplasm collections Thaipalm and Cho 2000 totaling 54 accessions, were screened for resistance, tolerance and susceptibility to *Meloidogyne javanica* a major pathogen of taro grown in dryland cropping systems. Susceptibility was observed in the germplasm, with nematode reproduction factors (Rf) in the range of 1.3 - 84.58. Resistance to *M. javanica* may be present within *C. esculenta*. Cultivars which hold promise as poor hosts to *M. javanica*

include Thailand #259, Nepal # CL83027, Thailand #6, Thailand #164, Thailand #2, GXH #173, HXP #140, SXP #171 and IXPNG #18. Susceptibility of 10 green manure crops to *M. javanica* were evaluated. Green manure species were selected based on known inhibitory compounds toward soil-borne pathogens. Host susceptibility and biomass production of green manure crops were assayed. Marigolds, and sorghum X Sudan grass hybrids were poor hosts to *M. javanica* with Rf values ranging from 0 to 0.61. The sorghum X Sudan grass hybrids produced the greatest amounts of biomass, averaging 5.2 g dry weight/plant. The interactive effects between *M. javanica* and a *Pythium* sp. on seven *Tagetes*, four *Brassica*, seven sorghum X Sudan grass, and four other green manure crops were evaluated. The *Brassica* spp. and four of the marigold cultivars were killed by the combined infections. Sunn hemp and all of the sorghum X Sudan grass cultivars were unaffected by either pathogen. The sorghum X Sudan grass cultivars continued to yield the highest biomass. Sorghum X Sudan grass was a poor host to *M. javanica*, produced copious biomass, and is an excellent green manure candidate for use in dryland taro cropping systems. In conjunction with resistant varieties, green manures are an important component of a sustainable program to control soil-borne pathogens. This research provides necessary information to develop a sustainable method to control soil-borne pathogens in dryland taro cropping systems.

THE RESPONSE OF TELONE AGAINST THE SOUTHERN ROOT-KNOT NEMATODE IN COTTON ACROSS SOIL ELECTRICAL CONDUCTIVITY ZONES. Overstreet, C.,¹ M. C. Wolcott,¹ E. Burris,² G. B. Padgett,³ D. Cook,² and D. L. Sullivan.² LSU Agricultural Center, ¹Department of Plant Pathology and Crop Physiology, Baton Rouge, LA 70803; ²Northeast Research Station, St. Joseph, LA 71366; and ³Northeast Research Station-Macon Ridge, Winnsboro, LA 71295.

Soil electrical conductivity (SEC) can be used as an effective surrogate for soil texture in the Mississippi alluvial soils of Louisiana. Four fields were mapped for SEC and were divided into a series of zones based on an average range or value for each zone. All of these fields had high to extremely-high populations of the southern root-knot nematode. Two of the locations had the southern root-knot nematode in all zones in the test. The other two locations had root-knot nematode in only specific zones within the field. An additional response of the nematicide 1, 3-Dichloropropene (Telone II) applied at the rate of 28 l/ha beyond that of the grower standard of aldicarb (Temik 15%G at 0.59 kg/ha) was measured in three fields and Telone against an untreated in a fourth location. Cotton was harvested with a yield monitor at three of the locations and individual plots at the fourth. At all of the locations, the lowest SEC zones gave the greatest yield response from Telone (averaged 150 to 432 kg/ha of lint differences between treatments for zone 1). As the SEC levels increased up to 15 to 25 mS/m, responses were either not significantly or economically different. This study shows the potential for very site-specific application of a fumigant such as Telone when combining nematode populations, SEC, and yield response.

HYBRIDIZATION OF TWO WIDELY SEPARATED SPECIES OF *ROMANOMERMIS*. Pacheco-Perez, R.,¹ E. G. Platzer,² and B. C. Hyman.² ¹CIIDIR-Oaxaca del Instituto Politecnico Nacional, Oaxaca, Mexico; ²University of California, Riverside, CA 92521.

The genus *Romanomermis* is the most thoroughly studied group of mermithid nematodes parasitic in mosquitoes. A great deal of basic and applied research is still necessary to achieve the full potential of these organisms for the biological control of mosquitoes. A program to increase strain vigor through hybridization of *Romanomermis* species from widely separated geographic areas is in progress. *Romanomermis culicivorax* (Louisiana, USA) and *Romanomermis iyengari* (Pondicherry, India) were propagated in *Culex pipiens*. Emergent postparasites were isolated in individual wells of 12-well culture plates until sexually mature. Both intra- and interspecies reciprocal crosses between males and females were established. Interspecific matings were infrequent in contrast to intraspecific mating. Egg production from interspecific matings was sparse and development to infectious preparasites was rare. However, some preparasites have developed and infected larvae of *Aedes aegypti* and *C. pipiens*. Adult females and males have been frozen for genotyping and bioassays are in progress to define baseline values for infectivity and fecundity of parental and hybrid strains.

A NOVEL APPROACH TO IDENTIFY PLANT-PARASITIC NEMATODES USING MALDI-TOF MASS SPECTROMETRY. Perera, Modika R.,¹ V. A. Vanstone,² and M. G. K. Jones.¹ ¹Plant Biotechnology Research Group, WA State Agricultural Biotechnology Centre, School of Biological Sciences and Biotechnology, Murdoch University, Perth, Western Australia 6150, Australia, and ²Department of Agriculture WA, ³Baron-Hay Court, South Perth, Western Australia 6151, Australia.

The ability to accurately identify nematodes at the species level requires specific skills in nematode taxonomy. The most common approach used is microscopy, which takes time and requires specific training: molecular approaches to plant nematode identification can also be used. Matrix-Assisted Laser Desorption/Ionization Time of Flight Mass Spectrometry (MALDI-TOF MS) is being used as a new approach to identify microorganisms. Identification is based on the characteristic protein profile of the organism generated by MALDI-TOF MS. In MALDI-TOF MS, pulses from a UV laser are absorbed by a crystalline matrix to which macromolecules have been adsorbed, and this results in relatively gentle desorption and ionization of the macromolecules, which are then accelerated using a high voltage and travel along the flight tube. The time taken to reach the detector is directly related to the mass/charge ratio of the molecule. MALDI-TOF MS is a promising

approach for rapid identification of organisms because of the simple sample preparation and the rapidity of the technique. The aim of this work was to develop a rapid, simple method to identify plant parasitic nematodes, based on analysis of protein profiles of nematodes generated by MALDI-TOF MS. Two methods have been used: grinding and direct analysis of intact nematodes. Both methods were standardized using the wheat seed-gall nematode *Anguina tritici* as a model. The standardized methods were applied to analyze the seed-gall nematodes *A. tritici* and *A. funesta* and the root-knot nematode, *Meloidogyne javanica*, which infects many horticultural crops. Characteristic protein profiles and diagnostic peaks were identified for individual nematode species and for mixtures of these species. The results provide proof-of-concept that these nematode species can be identified by protein profiling using MALDI-TOF MS. This approach could be extended to identify other plant and non-plant parasitic nematodes by generating unique species-specific protein profiles.

MASS PRODUCTION OF MERMITHID NEMATODE PARASITES OF MOSQUITO LARVAE IN MEXICO. Pérez-Pacheco, R., and G. Flores. CHDIR-Oaxaca del Instituto Politécnico Nacional, Oaxaca, Mexico.

Mermithid nematodes are under evaluation for the biological control of mosquito larvae in Mexico. Mass production of the mermithid nematodes, *Romanomermis iyengari* and *Romanomermis culicivorax*, was accomplished in a dedicated facility with the use of 500 rearing trays containing 1000 larvae of *Culex quinquefasciatus* per tray as the host species. Infection of mosquitoes, development of the mermithid parasitic phase, and emergence of the postparasites was completed every ten days. The rearing system produced 360 cultures monthly that after maturation yielded 150 million infectious nematodes for application to mosquito habitats of 50,000 m² (at an estimated concentration of 3,000 nematodes/m²). From 1995 to 2004, 2,000 to 3,000 preparasitic nematodes per m² were applied to mosquito breeding sites in the vicinity of communities with endemic malaria on the coast of the state of Oaxaca, Mexico. Parasitism of mosquito larvae was evaluated three days post-infection. The level of parasitism in larvae of *Anopheles* varied from 46% to 100% depending on the characteristics of the habitat. Thus, the mermithids proved to effectively reduce the population of malaria-transmitting mosquitoes.

REPRODUCTIVE FITNESS OF ISOLATES OF MELOIDOGYNE GRAMINICOLA FROM NEPAL ON SELECTED RICE AND WHEAT VARIETIES. Pokharel, R. R.,¹ G. S. Abawi,¹ C. D. Smart,¹ and J. M. Duxbury.² ¹Department of Plant Pathology, NYSAES, Cornell University, Geneva, NY 1445; and ²Department of Crop and Soil Science, Cornell University, Ithaca, NY 14853.

The rice root-knot nematode (*Meloidogyne graminicola*, Mgr) is widespread and causes yield reduction losses to rice and wheat grown in Nepal and other countries in South and Southeast Asia. Reproduction and root-galling severity (RGS) of five isolates of Mgr (NP8, 12, 29, 37 and 43), obtained from different rice-wheat production fields in Nepal, were evaluated in greenhouse tests on five rice (Mala, Masuli, Bammorcha, Bonet and Labelle and four wheat (Brikuti, NL 792, BL 1473 and NY strain) varieties or germplasm. Seeds of rice and wheat (10 seeds/pot) were planted in 10 cm clay pots filled with pasteurized soil (30 minutes at 60°C) and inoculated with 5000 eggs of Mgr. The pots were maintained in a greenhouse at about 25°C, and fertilized once a week. After 60 days, roots were washed free of soil and root-galling severity was rated on a scale of 1 (no visible galls, healthy roots) to 9 (>80% of roots galled). Eggs were then extracted from roots by blending in 1% sodium hypochloride solution for 3.5 minutes and passed the suspension over 150-µm and 25-µm pore sieves. The 5 Nepalese isolates of Mgr differed significantly in their reproduction in RGS rating produced on the rice and wheat germplasm tested. Also, there was a significant interaction between the isolates of Mgr and both the rice and wheat tested. For example, the reproduction factor (RF) of isolate NP 8 on the rice varieties Mala and Bonet was 6.0 and 19.8, respectively.; whereas the RF of isolate NP 37 on the same varieties was 19.8 and 4.9, respectively. In addition, there was no close correlation between the reproduction factors and the RGS ratings induced by the same isolate of Mgr on each varieties/germplasm. These results will aid in selecting isolates for use in identifying resistance crop germplasm and in diversifying current rice-wheat production systems.

IMPACT OF THREE WEED SPECIES ON REPRODUCTION OF ROTYLENCHULUS RENIFORMIS ON COTTON AND SOYBEAN. Pontif, M. J., and E. C. McGawley. Department of Plant Pathology and Crop Physiology, LSU AgCenter, Baton Rouge, LA 70803.

From 1998-2002, microplot studies were conducted to determine the effects of cotton, soybean, and three endemic weed species, (morning glory-MG, hemp sesbania-HS and johnson grass-JG), on reproduction of *Rotylenchulus reniformis*. Over three trials, the co-culture of cotton with any of the three weed species suppressed reproduction of reniform nematode significantly. On cotton alone, nematode reproductive (R) values averaged 59.5 at harvest. For MG, HS, and JG when alone, R values averaged 53.3, 25.3, and 20.0, respectively. R values for the combination treatments with cotton averaged 44.1 for cotton-MG, 30.0 for cotton-HS, and 25.0 for cotton-JG. Data for soybean trials over 2001 and 2002 followed a trend similar to that observed for cotton. Suppression of reniform populations in these trials could have resulted either from physical or from the secretion of allelopathic compounds by weeds. A preliminary greenhouse study was conducted during the fall of 2000 to test the allelopathy hypothesis. Clay pots containing 2 kg steamed-sterilized soil were arranged as a

randomized block. Five replications of 11 treatments were established and soil was infested with 600 *R. reniformis* juveniles. Treatments 1-4 were cotton, MG, JG, and HS, each alone. Treatments 5, 6, and 7 included cotton co-cultured, respectively, with MG, JG, and HS. Treatments 8 to 10 were the same as 5 to 7, but were watered with leachates from individual weed species. A control in which plants received leachates from cotton was also included. R values showed significant reductions in reniform populations in the presence of leachates from two of the three weed species and lends support to the allelopathy hypothesis.

IDENTIFICATION OF PRATYLENCHUS SPECIES USING PCR WITH SPECIES-SPECIFIC PRIMERS. Qiu, J., V. M. Williamson, and B. B. Westerdahl. Department of Nematology, University of California, Davis, CA 95616.

The two of the most important root lesion nematodes in California *Pratylenchus penetrans* and *P. vulnus* are difficult to distinguish by morphological characters. We have developed a PCR method with species-specific primers to identify and distinguish these species. Four pairs of species-specific primers were designed from ITS sequences of rDNA from *P. penetrans*, *P. vulnus*, *P. scribneri* and *P. thornei* that amplified 470, 368, 617, and 603 bp DNA fragments respectively. The same temperatures can be used in the PCR reaction with all four pairs of primers and different size of DNA are amplified by different primers. These allow us to carry out multiplex PCR to amplify DNA from target species or separate species in a mixed population. The primers also amplified target DNA from soil extracts of field samples at levels as low as a single lesion nematode among other free-living or plant parasitic nematodes.

ROLE OF ACOUSTICAL SIGNATURE MONITORING IN ECOSYSTEM ASSESSMENT. Quintanilla, M.,¹ A. Smucker,² S. Gage,¹ and G. Bird.¹ ¹Department of Entomology, ²Department of Crop and Soil Sciences, Michigan State University, E. Lansing, MI 48824.

Acoustical signature monitoring is a relatively new, innovative and non-intrusive methodology that appears to have potential for assessment of ecosystem structure, function and quality. MSU scientists developed a low cost and continuous acoustical monitoring system (combinations of relative sound intensities-decibels and KHz frequency levels) and used it to quantify and analyze the above ground acoustic signatures of 265 bird, 52 insect and 42 amphibian species. A team is currently evaluating the utility of the procedure for assessment of relationships between aboveground, surface and below-ground components of agricultural ecosystems; including nematode community structure and its relationship to soil quality. An initial step in this procedure was to identify acoustical properties of soil pore and water-stable aggregate stability. Characteristic acoustical signatures were observed in a comparison soil aggregates from three soil-management clades: (i) a never-tilled Kellogg Biological Station soil, (ii) a Wooster forest soil and (iii) a conventionally-tilled Hoytville soil. In water, the conventionally-tilled Hoytville soil had the greatest relative sound intensity at all KHz levels. The same was true for the three soil-management clades in CaSO₄. In NaHmP, however, the never-tilled Kellogg Biological Station soil had a greater relative sound intensity at the higher KHz levels, compared to the response in water or CaSO₄. Acoustical signature characterization for the nematode component of ecosystems is being done at both individual species and community structure levels.

GENE EXPRESSION OF EARLY STAGES OF GIANT CELLS INDUCED BY MELOIDOGYNE JAVANICA USING LASER CAPTURE MICRODISSECTION. Ramsay, K., Z. Wang, and M. G. K. Jones. Plant Biotechnology Research Group, Western Australian State Agricultural Biotechnology Centre, Murdoch University, Perth, WA 6150, Australia.

Root-knot nematodes (*Meloidogyne* spp.) are economically important plant pests that establish a specific relationship with host roots, and induce the formation of feeding cells known as 'giant cells'. These provide nutrients, and support nematode growth and reproduction. The induction of the feeding cells involves a series of developmental and biochemical changes, reflecting alterations in expression of many endogenous plant genes in response to nematode stimuli. Identification of key genes/products in the interaction is a major challenge in understanding how root development is modified during giant cell formation. We have used a micropipette system to obtain pure giant cell cytoplasm for molecular analysis, but it is difficult to obtain pure cytoplasm from early stages of the interaction. Laser Capture Microdissection (LCM) is a technique that was developed to isolate specific cells from heterogeneous tissues, and in this study LCM has been used to access the cytoplasm of 4 days post inoculation giant cells in tomato roots, from paraffin-embedded sections. Total RNA was isolated from laser dissected sections, and used in RT-PCR to investigate expression of some cell cycle genes (cyclins) in giant cells, and to confirm that full length cDNA fragments were present. Two D-type cyclin genes, LeCycD3;2 and LeCycD3;3, were expressed at higher levels in giant cells compared to other cell-cycle-related cyclin genes, suggesting that the induction of the G1 phase of the cell cycle may be triggered in response to stimulation by the infecting nematode. The isolated RNA was then amplified using in vitro transcription by T7 RNA polymerase. After two rounds of amplification, the amplified RNA was used to construct a giant cell cDNA library

CURRENT STATUS AND FUTURE DIRECTIONS OF METHYL BROMIDE ALTERNATIVES FOR TOMATOES. Rich, J. R. University of Florida, North Florida Research and Education Center, 155 Research Road, Quincy, FL 32351.

Fresh market tomato (*Lycopersicon esculentum*) production has greatly benefited from the development of the raised-bed, polyethylene mulch cultural system and concurrent soil fumigation with methyl bromide (Mbr). This chemical provided excellent control of weeds, diseases, insects and nematodes and proved to be a key component in the success of this production system. With the impending loss of Mbr, the search for alternatives first focused upon the short-term but necessary goal of finding practically useful chemical replacements. Many field trials were conducted to determine Mbr replacement chemicals, rates, chemical combinations and application methods for nematode and other soil-borne disease management. Chemical alternatives to Mbr are now available to manage soil-borne fungal and nematode diseases in fresh market tomato production. The most efficacious and widely recommended chemical alternatives to Mbr are combinations of 1,3-dichloropropene and chloropicrin. As these chemical alternatives were being developed, however, other longer term and more sustainable soil-borne disease management methods also were being assessed. Among these was deployment of tomato cultivars resistant to nematodes and soil-borne diseases and sod-based crop rotations, both of which were generally less needed when methyl bromide was used in fresh market tomato production. Use of these two effective disease management practices, with and without alternative chemicals, have been studied and proven successful in both university research and onfarm trials. The advantages and limitations of methyl bromide replacement chemicals, plant resistance, sod-based rotations, and their combined use will be discussed.

AVICTA COMPLETE PAK: COMPLETE PEST CONTROL IN COTTON. Rideout, S. L.,¹ and D. H. Long.² ¹Syngenta Crop Protection, Leland, MS 38756, and ²Syngenta Crop Protection, Greensboro, NC 27409.

Over the past three growing seasons, Syngenta Crop Protection has conducted field trials in over 60 locations across the cotton belt examining the efficacy of a novel seed treatment nematicide, AVICTA. The use of a seed treatment nematicide offers many obvious benefits to producers, including, controlled application environment, precision seed to seed loading, reduced soil loading, and ease of use. Data from these trials indicates that AVICTA (abamectin) reduced numbers of nematodes present in the soil and the plant, reduced galling, and increased plant vigor in areas infested with root-knot, Columbia lance, and reniform nematodes. Yields from Avicta-treated plots were similar if not better than Temik applied at 5 lb/A. Additionally, no issues with phytotoxicity or crop safety were observed in Avicta-treated plots. Syngenta plans to launch AVICTA Complete Pak, a promotional combination of AVICTA, Cruiser and Dynasty CST in cotton in 2006. AVICTA Complete Pak will be the first product that will effectively protect cotton plants from early season nematodes, insects and diseases.

DEVELOPMENT OF RAPID DIAGNOSTIC TOOLS TO IDENTIFY VIRULIFEROUS STUBBY ROOT NEMATODES *PARATRICHODORUS ALLIUS*. Riga, Ekaterini,¹ J. Crosslin,² R. Larsen,² and N. Guerra.¹ Washington State University, IAREC, Prosser, WA, 99350, and ²USDA-ARS, Prosser, WA 99350.

Stubby root nematodes are becoming of increasing importance to the potato industry in Washington State, as they are vectors of Tobacco Rattle Virus (TRV), the causal agent of Corky Ringspot disease. The current non-quantitative method of evaluating a field for the presence of *P. allius* takes several days while the detection of viruliferous *P. allius* requires a glasshouse bioassay that takes up to six weeks followed by an ELISA test. In order to reduce considerable resource inputs, a rapid and reliable method is required to identify stubby root nematode species. Few studies have utilized molecular techniques to identify viruliferous nematode species. Therefore, a rapid molecular assay has been developed using RT-PCR to identify viruliferous nematodes within 24 hours. The technique can detect TRV (453 bp) in as few as five nematodes.

THE REAL-WORLD PERSPECTIVE OF A GROWER. Roan, W. Six Ls Packing Co., Naples, FL.

Growers face a number of significant challenges in implementing alternatives to methyl bromide. This presentation will address: 1) Production region and field specific challenges/uniqueness; 2) nematode concerns and controls in a fresh plastic primary crop; 3) secondary cropping systems on re-used plastic/drip with associated nematode concerns and controls; 4) integrated chemical and natural control strategies; and 5) discovery and analysis of field distribution of nematode populations. Finally, a discussion of future directions will conclude the presentation.

REPRODUCTION OF RENIFORM NEMATODE ON SOYBEAN CULTIVARS IN 2004 TESTS. Robbins, R. T.,¹ E. Shipe,² P. Chen,³ L. Rakes,¹ L. E. Jackson,¹ E. E. Gbur,⁴ and D. G. Dombek.⁵ Departments of ¹Plant Pathology, ³Crop, Soil, and Environmental Sciences, ⁴Agricultural Statistics, ⁵Arkansas Crop Improvement Program, University of Arkansas, and ²Entomology, Soils, and Plant Sciences, Clemson University.

During 2004, 194 soybean varieties from the Arkansas variety testing program, 34 Clemson and 8 Arkansas breeding lines were tested in the greenhouse to determine their suitability as hosts for the reniform nematode (RN), *Rotylenchulus reniformis*. All treatments were inoculated with 1,200 vermiform RN. The resistant varieties Forrest and Hartwig, RN susceptible Braxton, and RN fallow soil served as controls. The mean number of vermiform nematodes extracted from the soil of each treatment was calculated, as were the reproductive indices (RI = Pf/Pi), PI/PI of Forrest (RF), and PI/PI of Hartwig (RH). Varieties with RF = s significantly greater than the RF on Forrest (1.00) were considered suitable hosts for

R. reniformis. Of the Arkansas test varieties 183 of 194 had more RN reproduction than Forrest whereas 189 lines, including Forrest, had more RN reproduction than Hartwig. The following varieties were not different than Forrest; Delta King XTJ54X9, S02-611RR, HBK R4945CX, Hartwig, S00-9970-08, S00-9970-09, Southern States RT5730N, DT98-7278, Southern States RT5602N, FFR 5663RR, Terral TVX56R405, and ARX E57104, whereas only Delta King XTJ54X9, S02-611RR, HBK R4945CX, S00-9970-08, S00-9970-09 were not different than Hartwig. On 22 of the 34 Clemson breeding lines RN reproduced more than on Forrest, while on 33 RN reproduced more than on Hartwig. On all of the Arkansas breeding lines RN reproduced more than on either Forrest or Hartwig.

INTROGRESSION OF RESISTANCE TO *ROTYLENCHULUS RENIFORMIS* INTO *GOSSYPIMUM HIRSUTUM* FROM *G. LONGICALYX*. **Robinson, A. F.,¹ A. A. Bell,¹ D. M. Stelly,² D. Dighe,² and M. A. Menz.²** ¹USDA-ARS, College Station, TX 77845; ²Department of Soil and Crop Sciences, Texas A&M University, College Station, TX 77843.

There is no significant resistance to *Rotylenchulus reniformis* in any primitive or agronomic genotype of Upland cotton (*G. hirsutum*). Introgression of immunity from *G. longicalyx* was undertaken by backcrossing *G. hirsutum* onto two triple-species hybrids obtained via interspecific crosses among *G. hirsutum*, *G. longicalyx*, and either *G. armourianum* (HLA hybrid) or *G. herbaceum* (HHL hybrid). Both hybrids are male sterile and vegetatively maintained but 1 to 5% of seed within hybrid flowers set when pollinated with *G. hirsutum*. After several thousand crosses, 700 heterogeneous first generation backcross progeny were obtained, from which 28 resistant fertile progeny were selected to generate 22 progeny lines from HLA and six from HHL. All lines were advanced with selection for resistance at each generation to the 4th, 5th, or 6th backcross; 1,400 progeny were evaluated. Resistance to *R. reniformis* was inherited via pollen as well as maternally and suppressed nematode populations 99% relative to susceptible controls. In 4th, 5th, and 6th backcross generations, plants were indistinguishable from agronomic cotton throughout growth and boll production in the greenhouse; resistant and susceptible plants occurred in a 1:1 ratio as expected for a single dominant gene; and cells had the normal 52-chromosome complement of *G. hirsutum*. All progeny from selfing the third backcross generation in some but not other lines had a normal *G. hirsutum* phenotype, indicating no deleterious recessive genes. Genome-specific in situ hybridization studies on chromosomes of selected advanced plants indicated a single, small alien segment from *G. longicalyx*. Development of markers for the seed industry is underway.

EMERGENCE PATTERNS, DISPERSAL AND HOST INVASION OF TWO *STEINERNEMA FELTIAE* ISOLATES. **Rolston, A. N., C. T. Griffin, and M. J. Downes.** Institute of Bioengineering and Agroecology, Department of Biology, National University of Ireland Maynooth, Co. Kildare, Ireland.

The dispersal of entomopathogenic nematode infective juveniles (IJs) from the natal cadaver has been a contentious issue with regards to dispersal of the sexes and IJ infectivity. Levels of sexual selection acting on male and female *Steinernema feltiae* can be important when investigating dispersal, host invasion and mating dynamics. Nematodes were allowed to emerge from cadavers infected with one of two *S. feltiae* isolates: the commercial UK76 and the Irish SB11. Two different emergence behaviours were observed: Non-Dispersed individuals that remained on or around the natal cadaver for up to 10 days; Dispersed individuals that actively moved away from the cadaver. Following immersion in 1% SDS solution for one hour, a greater proportion of Dispersed individuals were determined to be of IJ stage. This may explain why Dispersed individuals were significantly more infective than Non-Dispersed nematodes. Dispersed and Non-Dispersed individuals were allowed to develop into adults in either host *Galleria mellonella* insects or in hanging drops of haemolymph. The subsequent sex ratios varied between emergence patterns and isolates. *Steinernema feltiae* has never been shown to conform to the male colonization hypothesis, which proposes that males should invade and establish within host insects before females. Potentially, therefore, males should be capable of surviving alone in cadavers for longer periods than females. This was only true for the SB11 isolate after four and six weeks within the cadaver.

INVESTIGATION OF METAPOPOPULATION STRUCTURE IN *BURSAPHELENCHUS CONICAUDATUS* (NEMATODA: APHELENCHOIDEA) JAPANESE POPULATION. **Saeb, A. T. M.,² and P. S. Grewal.¹** Departments of ¹Entomology and ²Plant Pathology, The Ohio State University, Wooster, OH, 44691.

We investigated the population structure of the yellow spotted longicorn beetle, *Psacothoe hilaris*, associated nematode *Bursaphelenchus conicaudatus*. Our hypothesis is that the fragmentation of *B. conicaudatus* population throughout the Japanese Islands will lead to subspecies structure in the nematode population. But a limited gene flow will be reserved between the local populations by the aid of the insect vector, *P. hilaris*. The hypothesized conditions represent the classic metapopulation structure. Sequences of *B. conicaudatus* partial cytochrome oxidase subunit I in the mitochondrial DNA were acquired from the genbank. Sequences were aligned in 29 alignment groups with sequence similarities ranging from 89% to 100% and the pairwise differences ranging from 0% to 11%. The number of observed haplotypes was 24 with haplotype diversity, (H) of 0.9729. The AT content varied between 69% to 71.3%. Sequences characteristics for the observed haplotypes were also studied including monomorphic, polymorphic and parsimony informative sites. Codon usage, substitution patterns and nucleotide composition for the studied haplotypes were also analyzed. The average ratio

of the transitional and transversional changes ($R = si/sv$) was 1.7. R-values for the 1st, 2nd and 3rd positions of codons were also calculated. Phylogenetic, using neighbor joining (NJ), UPGMA, maximum parsimony (MP) and minimum evolution (ME), and population genetic analysis have proved subspecies structure in *B. conicaudatus* Japanese population. Genetic differentiation value was not significant. Limited gene flow has been observed. The value of *Nst* is 0.69679, *Fst* is 0.69153 and *Gst* is 0.15356. This is the first study to investigate metapopulation structure in insect associated nematodes. This study can produce useful information for the pine wood nematodes strategies.

ASSESSING NEMATODE COMMUNITIES IN DISTURBED SOILS; THE NECESSITY OF HIGHER RESOLUTION DIAGNOSTICS IN NEMATODE ECOLOGY. **Sánchez-Moreno, S.,^{1,2} H. Ferris,¹ and A. Navas.²** ¹Department of Nematology, University of California, Davis, CA 95616; ²Museo Nacional de Ciencias Naturales (CSIC), 28011 Madrid, Spain.

We studied the effects of heavy metal (Cu, Ni, Pb, Zn) soil pollution on the nematode community of the Guadiamar River banks (southern Spain). Abundance of nematode taxa, trophic structure, colonizer-persister groupings and functional guild structure were determined in relation to soil pH, residual heavy metal content and soil texture. *Acrobeles*, *Trichodorus*, *Boleodorus*, *Psilenchus* and *Acrobeloides* were the taxa most sensitive to metal pollution. *Tylenchorhynchus*, *Ditylenchus*, *Aphelenchoides* and *Rotylenchus* were most tolerant. Among the groupings, bacterial-feeders, omnivores, cp-4 and cp-5 groups were the most affected by heavy metals. By the end of the survey, as indicated by maturity and food web indices, the functional status of the soil community was nearly recovered. Using Canonical Correspondence Analysis, we compared the sensitivity of taxa and ecological groupings to heavy metals to determine the ecological homogeneity of the various groupings. Of the nematode groupings, functional guilds, which integrate feeding habits and life cycle characteristics, provide higher levels of functional resolution. Functional guilds had greater ecological homogeneity and provided more consistent information about the relationships between nematode community and levels of pollutants. Similarly, high-resolution analyses based in functional guilds, provided better understanding of community properties, soil food web characteristics and soil functions.

GENETIC AND MOLECULAR ANALYSIS OF INFECTIVE JUVENILE LONGEVITY IN THE ENTOMOPATHOGENIC NEMATODE *HETERORHABDITIS BACTERIOPHORA*. **Sandhu, S., and P. S. Grewal.** Department of Entomology, The Ohio State University, OARDC, Wooster, OH 44691.

Poor storage stability is a major obstacle to the expansion of entomopathogenic nematode use in biological control. One of the main reasons for the poor storage stability is the short longevity of the infective juveniles (IJs). We established inbred lines in *Heterorhabditis bacteriophora* GPS11 strain to determine genetic variability in longevity of the IJs. The IJ longevity was determined as LT_{90} in weeks by assessing the survival of IJs in water at 25°C over time followed by probit analysis. The results indicated that IJ longevity was significantly different among the inbred lines and it varied between 16 to 20 weeks. We then selected the long-lived line A6 and short-lived line A12, to determine the molecular mechanisms regulating longevity in the two lines. In a separate study of the cDNA library of the *H. bacteriophora* IJs we found homologs of *Caenorhabditis elegans* aging genes and selected four candidate genes to determine the differences in their expression in the two long and short lived lines. Two of these are members of the insulin-like signaling pathway (AKT/PKB kinases *akt-1* & *pdk-1*), which regulates growth, metabolism and longevity and the other two are stress resistance genes (superoxide dismutase, *sod-4* and heat shock protein, *hsp-4*). We hypothesized that the genes *akt-1* and *pdk-1* are upregulated in the short-lived inbred line and downregulated in the long-lived line based on their role in *C. elegans*. Similarly, we hypothesized that the genes *sod-4* and *hsp-4* are upregulated in the long-lived line and downregulated in the short-lived line. We followed real-time RT-PCR approach to quantify the levels of these transcripts in the two lineages and these experiments are now underway.

NEMATODE CONTROL AND OTHER BENEFITS OF RESISTANT CATCH CROPS. **Schlathölder, M. P. H.** Petersen Saatzucht Lundsgaard GmbH & Co. KG, D-24977 Grundhof, Germany.

Breeding of nematode resistant fodder radish and mustard varieties started more than 30 years ago when P. H. Petersen introduced the first fodder radish (*Raphanus sativus*) cultivar Pegletta that reduce the sugar beet cyst nematodes (*Heterodera schachtii*) population about 80%. From that time onwards several newer varieties of oilradish and mustard were developed for the control of potato and sugar beet nematodes. The nematode population reduction of the newest varieties is more than 90% and the use of resistant fodder radish like Colonel and Picobello or mustard (Accent) is the common way of controlling *Heterodera schachtii* in Germany and several other countries as the Netherlands and France even in conventional agriculture. P. H. Petersen selected from their fodder radish gene pool some varieties like Commodore and Defender that suppress multiplication of *Meloidogyne sp.* on several crops. In potato crop rotations oil radish varieties like Siltta and Nova that reduces stubby root nematodes (*Trichodorus sp.*), are widely used in Germany. In addition to reducing the nematode population catch crops supply more than one ton additional organic matter to the soil, decomposes quickly due to its narrow C : N ratio, increases soil's humus content, serves as an energy source for the beneficial micro organisms

in the rhizosphere, suppress weed and disease pests, reduce pesticide input, reduce soil erosion. The residues from these green manure crops add humus that improves soil tilth, water holding capacity, and nutrient availability.

ASSESSING ECONOMIC PERFORMANCE OF METHYL BROMIDE ALTERNATIVES. Schaub, J. Office of Risk Assessment and Cost-Benefit Analysis, U.S. Department of Agriculture, Washington, DC 20250.

Demonstrated technical efficacy against pests of concern is not sufficient to establish the viability of methyl bromide alternatives in agricultural production systems. Alternatives must also demonstrate acceptable economic performance. Interpreting and applying the economic criteria for exempting the critical uses of methyl bromide from phaseout under the Montreal Protocol remains problematic because of inadequate data and the lack of consensus on thresholds for economic feasibility. A conceptual framework for assessing economic performance is presented and the economic performance of alternatives is illustrated through analysis of United States Critical Use Nominations for methyl bromide for soil fumigation uses. In certain circumstances, alternatives resulting in lower crop yields may be economically acceptable. Factor prices and substitutability, market timing, and costs of capital contribute to economic performance. Adoption of alternatives depends on growers' confidence that technical and economic performance is transferable to their situation and will be sustainable.

CHALLENGES TO ADOPTION OF METHYL BROMIDE ALTERNATIVES. Schneider, S. M. USDA/ARS San Joaquin Valley Agricultural Sciences Center, Parlier, CA 93648.

Methyl bromide (MB) was phased out in the United States and other developed countries as of 1 January 2005 in accordance with the Montreal Protocol. The exceptions to the phase-out are growers with approved Critical Use Exemptions (CUE) for 2005 and Quarantine/Preshipment applications. Research to develop and implement acceptable MB alternatives has been conducted by university, government, and industry scientists since the MB phase-out was announced. Progress has been made, but many challenges remain. In the U.S. nomination submitted to United Nations Environmental Programme in 2005 for CUEs for MB use in 2007, approximately 72% of the preplant methyl bromide nominated was for use in southeastern U.S. cropping systems. Crops include tomato (48%), peppers (23%), cucurbits (12%), strawberry (10%), eggplant (2%), turf grass (2%), forest seedlings (2%), and ornamentals (1%). CUE applications are good indicators of cropping systems that are still dependent on methyl bromide. Barriers to adoption of alternatives include technical limitations (diagnostics, efficacy of control of key pests, consistency of control), regulatory constraints (registration, karst topography, buffer zones), and economic issues, as well as human nature and the amount of risk an individual grower is willing to accept. Potential solutions include expanded uses for currently registered materials, further development of experimental materials, improved efficacy of non-chemical management strategies, new application technologies, and integrated management using all available options. Methyl bromide alternatives will be adopted when acceptable, consistent efficacy is demonstrated under local, commercial conditions; alternatives are registered at all necessary levels; they are cost effective; product and application expertise is available; and growers have gained experience with the alternatives.

UPTAKE AND EFFECT OF FLUORESCEIN ISOTHIOCYANATE BY *HETERODERA GLYCINES*. Schroeder, N. E., and A. E. MacGuidwin. Department of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706.

Live second stage juveniles of the soybean cyst nematode were found to take up Fluorescein Isothiocyanate (FITC), a fluorescent probe. Eggs were incubated in hatching chambers with 2 ml of 3 mM ZnCl₂ and a final concentration of 0.1 mg/ml FITC dissolved in dimethylformamide (DMF). Hatched juveniles were then examined using a fluorescent stereomicroscope with a GFP filter. All hatched juveniles fluoresced beyond normal auto-fluorescence, primarily in the esophageal and intestinal regions. Dissection of animals as well as fluorescent quenching techniques indicated the stain was not simply bound to the cuticle. Stained nematodes remained active and did not lose their fluorescence even after 2 weeks at room temperature. Fluorescence was not stable through development. Males who developed from fluorescent juveniles did not retain the stain when examined. Hatching studies indicated that FITC and the DMF solvent reduced hatch. However, additional experiments showed that those individuals who successfully hatched remained viable and able to infect roots. This research has potential as both a tool for examining nematode behavior (e.g., "tracking" individual nematodes) as well as for studying chemical uptake.

ASSESSMENT OF RESISTANCE OF SOME WILD *BETA* GERMPLASM TO SUGARBEET CYST NEMATODE. Seddigheh, F. College of Agriculture, Azad University, Varamin, Iran.

Sugarbeet cyst nematode is one of the most damaging pathogens of sugar beet in Iran and at present none of the existing commercial sugar beet cultivars have resistance to the nematode. The reactions of different wild beet germplasm to *Heterodera schachtii*, were evaluated by measuring nematode hatching in response to root diffusates and rate of juvenile penetration into the roots. Petri dishes containing root diffusates and surface-sterilized cysts were maintained for 3 weeks at 20 and 25 C. Hatched juveniles were removed and root diffusates exchanged weekly. Thirty-three seedlings of each accession, transplanted into 36-cm³ tubes filled with fine sand, were inoculated with 300 freshly hatched *H. schachtii*

juveniles. Numbers of females that developed were counted after 1 month. Significant differences were detected in the effects of root diffusates on nematode hatch in vitro (13% to 29% at 20°C and 48% to 70% at 25°C) and in the rate of attraction and development of nematodes in roots of the different germplasm sources (0.8, 16, 24, 68 females on roots of resistant control, and lines 112, 124 and 668 respectively).

ESTIMATION, DIAGNOSIS, AND COMPARATIVE PROCEDURES IN NONLINEAR REGRESSION MODELS. Shafii, B., and W. J. Price. Statistical Programs, College of Agricultural and Life Sciences, University of Idaho, Moscow, ID 83844-2337.

Biological research data are often represented using nonlinear model specifications which contain parameter estimates with meaningful interpretations. Common examples include germination/emergence analysis, growth models, bioassay experiments, and dose-response functions. Estimation of nonlinear models are normally carried out using iterative least squares, maximum likelihood, or Bayesian techniques. Subsequent statistical inference on estimated model parameters is an important aspect of applied research and can range from simple confidence intervals on individual parameters to more complex comparisons of underlying models and treatment structures. The iterative estimation techniques used in nonlinear regression analysis, however, can often yield correlated parameter estimates and nonlinearity in the estimation situation, leading to unreliable statistical inferences. Thus, for nonlinear models, evaluation of the asymptotic properties of the parameter estimates should be carried out in addition to standard regression diagnostics. This presentation will describe relevant procedures for the estimation, diagnosis, and treatment comparison for nonlinear regression models. The techniques will be demonstrated using the SAS software system. Examples shown will utilize data from various agricultural research studies.

ULTRASTRUCTURE OF THE EARLY CELLULAR IMMUNE RESPONSES OF *ANOPHELES QUADRIMACULATUS* TO THE INVASION BY *ROMANOMERMIS CLICIVORAX* ROSS AND SMITH (NEMATODA: MERMITHIDAE). Shamseldean, M. M.,¹ and E. G. Platzer.² ¹Professor of Zoology and Nematology, Department of Agricultural Zoology and Nematology, Faculty of Agriculture, Cairo University, Giza 12613, Egypt; ²Professor of Biology and Nematology, Department of Nematology, University of California, Riverside, CA 92521.

Observations with the Transmission Electron Microscope were made to record interactions of *Romanomermis culicivorax* with hemocytes of *Anopheles quadrimaculatus* larvae. Within 5 minutes after penetration, Host granules and granulocytes were attached to the surface of the nematodes parasitic stages, meanwhile nematodes secreted an outer layer onto the cuticle. Plasmatocytes were also attached to the parasitoid surface as early as 12 hours post-penetration, As an evasion mechanism from the host immune system, the parasitic nematodes continuously shed a secretion coat. This study suggests that, the mermithid nematode *Romanomermis culicivorax* through coevolution has developed a unique system to avoid the defense mechanism of their mosquito hosts.

SCANNING ELECTRON MICROSCOPE OBSERVATIONS ON THE CELLULAR IMMUNE RESPONSES OF MOSQUITO LARVAE INFECTED WITH *ROMANOMERMIS CULICIVORAX* ROSS AND SMITH (NEMATODA: MERMITHIDAE). Shamseldean, M. M.,¹ and E. G. Platzer.² ¹Professor of Zoology and Nematology, Department of Agricultural Zoology and Nematology, Faculty of Agriculture, Cairo University, Giza 12613, Egypt; ²Professor of Biology and Nematology, Department of Nematology, University of California, Riverside, CA 92521.

Interactions of the mermithid nematode, *Romanomermis culicivorax*, with hemocytes of *Anopheles quadrimaculatus* and *Culex quinquefasciatus* larvae were recorded with the Scanning Electron Microscope (SEM). In both mosquito species, host granules and granulocytes attached to the surface of parasitic nematodes. Nematodes secreted an outer layer onto the cuticle within 5 minutes after penetration. Plasmatocytes were also attached to the parasite surface by 72 hours after penetration in *Anopheles quadrimaculatus* and 120 hours in *Culex quinquefasciatus*. Molting nematodes were first observed 72 and 96 hours after penetration in *C. quinquefasciatus* and *A. quadrimaculatus*, respectively. Pits were observed on the ventral cuticle of the parasite following a molt. The parasitic nematodes continuously shed a secretion coat until 144 hours after penetration in *A. quadrimaculatus*, whereas in *C. quinquefasciatus* the nematodes shed the secretion coat throughout the entire parasitic stage during the course of parasitism. Mosquito hemocytes were attached to the surface of the nematodes as late as 120 hours in *C. quinquefasciatus* and 144 hours in *A. quadrimaculatus*. In some cases, nematodes became melanized in *A. quadrimaculatus*. This study suggests that, *R. culicivorax* has developed a unique system to avoid the defense mechanism of their mosquito hosts

IMPROVED STRAINS OF *STEINERNEMA CARPOCAPSAE* FOR CONTROL OF THE PECAN WEEVIL, *CURCULIO CARYAE*. Shapiro-Ilan, D. I.,¹ R. J. Stuart,² and C. W. McCoy.² ¹USDA-ARS, Southeast Fruit and Tree Nut Research Lab, Byron, GA 31008; ²Citrus Research and Education Center, IFAS, University of Florida, Lake Alfred, FL 33850.

The entomopathogenic nematode *Steinernema carpocapsae* has shown promise for control of adult stage pecan weevil, *Curculio caryae*, a key pest of pecan. In previous studies, the Italian strain of *S. carpocapsae* was found to be among the

most virulent to *C. caryae*, but possessed poor environmental tolerance. In contrast, the DD-136 strain exhibited high levels of environmental tolerance but low virulence. Our objective was to determine the feasibility of developing improved strains of *S. carpocapsae* by transferring the nematode's bacterial symbiont from the Italian strain to the DD-136 strain, and through hybridization between the two wild type nematodes. Three modified strains were created: one through bacterial transfer alone and two hybrids through controlled crosses. We hypothesized that the improvement approaches would result in strains possessing high levels of environmental tolerance similar to DD-136 and virulence similar to the Italian strain. The hypothesis was supported in two out of three modified strains. Heat and desiccation tolerance in the three modified strains was more than 2.5 fold greater than the Italian strain and not different from the DD-136 strain, except one hybrid had lower heat tolerance than DD-136. Mortality of adult weevils from the modified strains was greater than from DD-136 (and similar to the Italian strain) two or three days after treatment. Overall, the results indicate that bacterial transfer and hybridization could be a valuable tool in improving biocontrol efficacy of steinernematids.

DIVERSITY OF FREE-LIVING NEMATODES IN THE NORTHERN GULF OF MEXICO DEEP SEA. Sharma, J.,¹ R. Huettel,² J. Baguely,³ and P. Montagna.⁴ ¹University of Texas at San Antonio, San Antonio, TX 78249; ²Auburn University, AL 36849; ³University of South Carolina, Columbia, SC 29208; ⁴University of Texas Marine Science Institute, Port Aransas, TX 78373.

Meiofauna, a diverse group of small metazoans, is ubiquitous in northern Gulf of Mexico deep-sea sediments. Free-living nematodes are the dominant component of benthic meiofauna with high abundance, biomass, and diversity compared to the larger macrofauna. In general, regional species pools, dispersal capabilities, and process structuring communities on various scales is unknown for deep-sea environments; additionally, little is known about the distributions of organisms and how they respond to topographic and geochemical features in the deep sea. In this study the nematode fauna of the continental slope was studied in the northern Gulf of Mexico in relation to depth and longitude and other physico-chemical parameters. The effect of variations in topographical features from the Florida escarpment to the Texas/Louisiana slope on nematode assemblages is discussed. Abundance and diversity of nematodes was highest in sediments of the continental shelf and decreased along the continental slope to a depth of 4000 m. The relative abundance of nematodes was highest in the sediments near the Mississippi trough. Over 40 genera of nematodes were identified with the highest diversity in the continental shelf sediments. The non-selective feeders, Comesomatidae, were the dominant family in these sediments.

POST-PLANT NEMATODE CONTROL IN PINEAPPLES. Sipes, B. S.,¹ J. P. Mueller,² and D. P. Schmitt.¹ ¹Department of Plant and Environmental Protection Sciences, University of Hawaii, Honolulu, HI 96822; ²Dow AgroSciences, 9330 Zionsville Rd., Indianapolis, IN 46268.

For a time, post-plant nematode control in pineapple was achieved by a single application of di-bromo-chloro-propene (DBCP). With the removal of DBCP, post-plant nematode control in pineapple has relied predominately upon multiple applications of organophosphate nematicides. With the reformulation of 1,3-dichloropropene (1,3-D), the opportunity arises to again use fumigant-type nematicides for post-plant nematode control in pineapple. Evaluations of post-plant 1,3-D have been conducted in experimental plots and in multiple commercial pineapple plantation fields. Post-plant applications of 1,3-D as low as 500 ppm damaged pineapple roots. In field tests, this damage was evident as smaller average fruit sizes and lower marketable yields in some 1,3-D post-plant treatment regimes. Post-plant applications of 1,3-D however, reduced population densities of *Rotylenchulus reniformis* in pineapple with timing of application critical. Early or late applications of 1,3-D did not affect nematode populations adversely. In seasons with especially high nematode disease pressure, the nematode control achieved by application of 1,3-D outweighed the phytotoxicity of the fumigant. In fields with low nematode population densities or during seasons of limited nematode disease pressure, the yield loss caused by the 1,3-D application outweighed the nematode control achieved. The phytotoxicity and nematode control effects of post-plant 1,3-D applications must be balanced in an integrated nematode control program for pineapple.

APPLICATION OF MULTIPLE DISPLACEMENT AMPLIFICATION FOR IMPROVING PCR OF SINGLE COPY HSP90 AND FOR PRESERVATION OF GENOMIC DNA FROM LIMITED NEMATODE SPECIMENS. Skantar, A. M. and L. K. Carta. Nematology Laboratory, USDA-ARS, Plant Sciences Institute, Beltsville, MD 20705.

Because the quantity of nematode specimens available for molecular analysis is often limited, the number of analyses possible is constrained by the availability of DNA. Multiple displacement amplification (MDA) is a non-PCR method of whole genome amplification that employs phi29 DNA polymerase to amplify genomic DNA. Using four widespread species of root-knot nematode, we examined MDA DNA for PCR amplification of commonly used diagnostic markers for nematodes. MDA produced microgram quantities of template that resulted in successful amplification of the ribosomal internal transcribed spacer (ITS1) and 28S D2-D3 expansion regions, and gave PCR results that were comparable to template generated by the single nematode smash method. Hsp90 is a single-copy gene that is gaining in acceptance for plant-parasitic nematode diagnostics and phylogeny. MDA greatly improved amplicon yield in degenerate primer PCR of Hsp90, which we have found is more sensitive than multicopy ribosomal genes to limits on input template DNA. Therefore,

MDA should expand the number of molecular analyses possible for single nematodes. MDA will be widely useful for archiving DNA from valuable specimens and will provide a way for laboratories to share identical genetic material for molecular diagnostic applications.

OVERVIEW OF THE MONTREAL PROTOCOL. Smith, B. U.S. Department of Agriculture, Washington, DC.

The *Montreal Protocol on Substances that Deplete the Ozone Layer* serves as the basis for regulating chemical compounds that affect stratospheric ozone levels. In the United States, domestic authority to implement the Montreal Protocol is vested to the Environmental Protection Agency (USEPA) under the Clean Air Act. Amendments to the Clean Air Act in the 1990s included restrictions on the use of methyl bromide as a fumigant for control of soil and post harvest pests. The most recent manifestation of these controls on methyl bromide is contained in the Critical Use Exemption (CUE) process. This paper will describe the treaty and its impact on agricultural uses of methyl bromide in the United States, including ongoing Federal agency cooperation to reduce uses while maintaining viability in the sectors that have traditionally depended on methyl bromide for economic pest control.

SEEDLING PROTECTION OF PIMA COTTON AGAINST NEMATODE-INDUCED FUSARIUM WILT BY A NEMATOCIDE SEED COATING. Smith Becker, J., and J. O. Becker. Department of Nematology, University of California, Riverside, CA 92521.

Fusarium wilt is a destructive plant disease that occurs worldwide in most cotton production areas. Several races of the pathogen *Fusarium oxysporum* f. sp. *vasinfectum* (*Fov*) have been recognized and host response varies with cotton species and cultivars. In general, the severity of Fusarium wilt is increased when the plants are parasitized by root-knot nematodes. The cotton cultivar Pima S-7 is resistant to *Fov* race 1 but is a good host to *Meloidogyne incognita* race 3. In 6-week greenhouse trials, seed coating of Pima S-7 with the nematocide abamectin (0.1 mg a.i./seed) and a fungicide/insecticide combination Dynasty CST (25 g a.i./100 kg seed)/Cruiser (0.32 mg a.i./seed) provided good protection against *M. incognita* race 3 during the critical seedling stage. Moreover, in the presence of both the root-knot nematodes and *Fov* race 1, seedlings grown from abamectin coated seeds did not show any stunting, chlorosis and vascular discoloration that were typical for seedlings that were grown from seeds coated with only the fungicide/insecticide components. These trials demonstrated that the benefit of a nematocidal seed treatment might well extend beyond its efficacy against plant parasitic nematodes.

IS THE DAGGER NEMATODE A VECTOR OF BROME MOSAIC VIRUS IN ALABAMA? Srivatsavai, S. K., J. F. Murphy, and R. N. Huettel. Department of Entomology and Plant Pathology, Auburn University, AL 36849.

Brome Mosaic Virus (BMV) was first detected in Henry County, Alabama on wheat in 2002. It was detected in 3 more counties in 2003 and in eight counties in 2004, indicating it was becoming established in all wheat growing regions of the state. The vector for this virus has been reported to be the dagger nematode, *Xiphinema* spp. This study was conducted to determine if this nematode was associated with wheat production and the spread of the virus in Alabama. Soil samples were collected from one wheat variety trial in central Alabama twice a month from October to May and other fields throughout the state monthly during the growing season. Nematodes were extracted with sugar flotation and identified. The virus was identified using direct double antibody sandwich ELISA and compared to an isolate from Oklahoma. Even though plant parasitic nematodes were detected in all samples, no *Xiphinema* spp. were found in relation to any wheat field. However, *Xiphinema* sp. was found in adjacent fields associated with other crops in some locations. Therefore, in Alabama, *Xiphinema* spp. are not associated with nor the vector of BMV.

CAN HOST SHIFT OCCUR WHEN STEINERNEMA CARPOCAPSAE IS PRODUCED IN THE TERMITE, RETICULITERMES HESPERUS? Stefanovska, Tatyana,¹ E. E. Lewis,² Harry K. Kaya,² and Melissa Anderson.² ¹Department of Integrated Pest Management and Quarantine, National Agricultural University, Kiev, 03187, Ukraine; ²Departments of Nematology and Entomology, University of California, Davis, CA 95616.

Termites have characteristics that should make them susceptible to entomopathogenic nematode (EPN) infection; they are soil-dwelling, soft-bodied and live in a moisture-controlled colony. Yet, studies indicate that EPNs are not effective against termites. We tested the hypothesis that a host shift in *Steinernema carpocapsae* could be brought about by rearing it through the workers of the termite, *Reticulitermes hesperus*. We have examined the change in host shift of *S. carpocapsae* by alternating *R. hesperus* and *Galleria mellonella* as hosts. We compared *S. carpocapsae* infective juveniles (IJs) from the following treatments: stock IJs (only reared in *Galleria*), 2) IJs through termites, *Galleria* and termites again; 3) IJs through termites, *Galleria*, termites, *Galleria*, and termites. We measured the effects of the selection regime on *S. carpocapsae* by recording % termite cadavers producing IJs, IJ production per individual termite, the number of days between infection and emergence, and a host recognition assay. Significant differences between selected and control

nematodes in emergence per cadaver and the average number of days between infection and emergence were recorded. The percent of cadavers that produced IJs after the second cycle through termites (23.1%) was higher than in the first cycle (13.1%). The average total number of IJs per cadaver was also significantly higher after the second selection cycle (837) than average total number in first cycle (158). The average number days between infection and emergence in second cycle (13.5) were significantly lower than in first (15.8). A significant positive correlation between termite weight and the number of IJs emerging from the first reproductive cycle ($R=0.538$) was observed. However, there was no significant correlation between weight and emerged IJs in the second cycle. Reproductive selection will continue. Host recognition assays are currently under way. We suggest that host recognition plays a significant role in infectivity.

COMPARISON OF FEMALE- AND EGG-BASED SCREENING METHODS FOR RENIFORM NEMATODE RESISTANCE IN COTTON. **Stetina, S. R., and L. D. Young.** USDA ARS Crop Genetics and Production Research Unit, Stoneville, MS 38776.

Development of cotton germplasm with reniform nematode resistance requires evaluation of large numbers of plants. Shortening the time involved in the screening process is one way to increase the number of progeny that can be examined each year. In this study, the hypothesis that enumerating swollen females per gram of root is as efficient in identifying resistant genotypes as measuring eggs per gram of root was tested. The experiment was conducted four times between March 22 and December 20, 2004. Six cotton genotypes were used; three (Deltapine 33B, Deltapine 16, Stoneville 4892 BR) were susceptible and three (LA RN 4-4, LA RN 910, LA RN 1032) were resistant to reniform nematode. Twelve replicates of each genotype were planted in the greenhouse in 7.6-cm-diam. clay pots filled with soil mix (2 sand:1 infested field soil; Pi ranged from 4,425 to 23,615 reniform nematodes per 200 cm³ soil mix). Six replicates of each genotype were harvested 25 days after planting, the roots stained with red food coloring, and the number of females counted. The remaining plants were harvested 35 days after planting, eggs extracted from roots by stirring in a 0.615% NaOCl solution, and the number of eggs counted. Root fresh weights were recorded for all plants. Time required for growing plants, preparing samples, and counting was recorded. Contrasts were used to determine if the difference between a genotype and Deltapine 33B based on females per gram of root was equivalent to the difference based on eggs per gram of root. Assessments based on females were equivalent to assessments based on eggs in all four runs of the experiment ($P > 0.05$), which indicates that either life stage can be used. The time required for sample processing and counting was similar for both assessment methods, but an additional 10 days per cycle in the greenhouse were required for egg-based assessment. On a yearly basis, 40% more plants can be screened using a female-based assessment instead of an egg-based assessment.

PHYLOGENETIC RELATIONSHIPS OF NEMATODES FROM THE FAMILY HOPLOLAİMİDAE AS INFERRED FROM ANALYSIS OF THE D2-D3 EXPANSION REGION OF THE LSU rRNA GENE SEQUENCES. **Subbotin, S. A.,¹ J. T. Tamber,² D. Sturhan,³ N. Vovlas,⁴ P. Castillo,⁵ M. Moens,² and J. G. Baldwin.¹** ¹Department of Nematology, University of California, Riverside, CA 92521; ²CLO-Department for Crop Protection, 9820 Merelbeke, Belgium; ³Institut für Nematologie und Wirbeltierkunde, BBA, 48161 Mü, Germany; ⁴Istituto per la Protezione delle Piante, CNR, Sezione di Bari Via Amendola, 165/A, 70126 Bari, Italy; ⁵Instituto de Agricultura Sostenible, Consejo Superior de Investigaciones Científicas, Apdo 4084, 1480 Cordoba, Spain.

The Hoplolaimidae have special significance as an apparent sister taxon with the cyst-forming nematodes (Heteroderidae) and both clades include a large number of pests of great agricultural importance. The D2 and D3 expansion regions of the 28S ribosomal RNA gene were amplified and sequenced from 21 samples representing seven species of the genus *Helicotylenchus*, five species of the genus *Rotylenchus*, single isolates representing *Scutellonema brachyurus*, *Hoplolaimus seinhorsti*, *Aorolaimus perscitus* and two outgroup taxa, *Radopholus* sp. and *Aglenchus agricola*. Alignment was manually corrected using a secondary structure model for this gene fragment, and analyzed using maximum parsimony, maximum likelihood and Bayesian inference approaches. All phylogenetic analyses yielded trees with similar topology. The DNA dataset does not fully support monophyly of the genus *Helicotylenchus*. Sequences of the D2 and D3 regions for populations of *Helicotylenchus pseudorobustus* collected in Mü, Germany, Beijing, China and Fresno, CA, USA were identical. Nucleotide polymorphisms were observed within an Italian population of *Helicotylenchus vulgaris*.

EGG BIOLOGY AND REPRODUCTION OF *ROTYLENCHULUS RENIFORMIS* ISOLATES FROM THE SOUTHERN UNITED STATES. **Sumner, J. B. and E. C. McGawley.** LSU AgCenter: Department of Plant Pathology and Crop Physiology, Baton Rouge, LA 70803.

Rotylenchulus reniformis (Rr) is a serious nematode pathogen in the United States. Differences in reproduction, cultivar response and host preference among geographic isolates of the nematode have led to the hypothesis that distinct pathotypes exist in nature. Populations of Rr from LA, MS, AR, TX, and HI were used to evaluate egg hatch in water and in soil in the presence and absence of host roots, as well as numbers of egg masses per root system and numbers of eggs per egg

mass. Over the course of two preliminary trials, hatch of eggs of Rr isolates from 10 major cotton-producing parishes of LA were determined in soil and in water. In the first trial, the percent egg hatch in water and soil, respectively, averaged 90% and 93% for the Catahoula parish isolate and 58% and 45% for the Avoyelles isolate. In trial two, the Opelousas isolate had the highest percent egg hatch, which was 91% in water and 94% in soil. The Evangeline isolate had the lowest percent egg hatch in water, which averaged 58. The Avoyelles isolate had the lowest percent egg hatch in soil, which averaged 57. Additional egg hatch trials with isolates of Rr from LA, TX, and other southern states were conducted using a temperature range from 23° to 32°C. In 2004, new isolates of the nematode were obtained, and 10 to 20 single egg mass cultures of each were established in the greenhouse for evaluation of within-isolate differences in reproduction and pathogenicity. Results of these studies show minimal variation among single egg mass cultures from the same geographic isolate.

THE DISTRIBUTION OF THE PINWOOD NEMATODE IN PINE WOOD. Takeuchi, Y.,¹ N. Kanzaki,² and K. Futai.¹ ¹Graduate School of Agriculture, Kyoto University, Kyoto, Japan 606-8502; ²JSPS Postdoctoral Research Fellow, Faculty of Agriculture, Kagoshima University, Kagoshima, Japan 890-0065.³

The distribution of pinewood nematode (PWN) *Bursaphelenchus xylophilus*, a causal agent of pine wilt disease, in pine wood was investigated using species-specific PCR amplification. Several 2-year-old potted seedlings of 3 *Pinus* spp. with different susceptibility to the PWN were inoculated with the PWN, and DNA was extracted from the seedlings using commonly accepted CTAB method. The DNA extract served as template for PCR amplification with the PWN-specific primers which were designed for amplifying the ITS region in rDNA of the nematode. The results showed that this method was sensitive enough to detect the PWN even though the DNA content derived from PWN in the template was extremely small. This method revealed that the PWN could migrate in host tissues prior to dysfunction of resin secretion in the host seedling, regardless of host species. We also applied this method to detect an avirulent fungus, and found that the fungus could not disperse in pine seedlings by contraries. Thus, the method should be proved to be useful for diagnosing PWN infection before external or internal symptoms appear, and also applicable to a simple and quick detection of wide-ranging pathogens inhabiting in wood.

RESISTANCE OF SWEETPOTATO GENOTYPES TO ROOT-KNOT NEMATODES. Thies, J. A. U.S. Vegetable Laboratory, USDA, ARS, Charleston, SC 29414.

Thirteen sweetpotato (*Ipomoea batatas*) genotypes were evaluated for resistance to *Meloidogyne incognita*, *M. javanica*, *M. hapla*, and *M. arenaria* races 1 and 2 in greenhouse tests. The sweetpotato genotypes evaluated were: U.S. Plant Introduction (PI) 399163, Sumor, NemaGold, Excel, Tinian, Hernandez, Jewel, Regal, Porto Rico, Centennial, Georgia Jet, Sulfur, and Beauregard. *Meloidogyne incognita* was most pathogenic to sweetpotato of the four *Meloidogyne* spp. evaluated in these studies. The U.S. PI 399163 and Sumor were resistant (reproductive index < 1.00) to *M. incognita* in all tests. Only two genotypes, Beauregard and Porto Rico, were susceptible to *M. javanica*. All genotypes evaluated were resistant to *M. hapla*, *M. arenaria* race 1, and *M. arenaria* race 2. Sumor, U.S. PI 399163, and NemaGold were most resistant to the four *Meloidogyne* spp. used in these studies. *Meloidogyne incognita* is the most commonly occurring root-knot nematode species in sweetpotato growing areas of the southern U.S. and is pathogenic to most of the commonly grown sweetpotato cultivars. Thus, efforts to develop resistant cultivars with desirable horticultural characteristics for the U.S. market should focus on screening against *M. incognita*.

THE NEMATODE BRANCH OF THE TREE OF LIFE. Thomas, W. K.,¹ E. Abebe,¹ K. Morris,¹ F. Liu,¹ J. Baldwin,² M. Mundo,² E. Ragsdale,² D. Bumbarger,² S. Subbotin,² P. De Ley,² I. T. De Ley,² M. Yoder,² I. W. King,² D. Fitch,³ K. Kiontke,³ N. Gavin,³ E. Kolychkina,³ H. Chun,³ R. Ng,³ S.-Y. Chiou,³ T. Hadi,³ S. Nadler,⁴ A. Smythe,⁴ J. Ullberg,⁴ and C. Pagan.⁴ ¹HCGS, University of New Hampshire; ²Department of Nematology, University of California-Riverside; ³Department of Biology, New York University; and ⁴Department of Nematology, University of California, Davis.

Nematodes are arguably the most abundant and diverse metazoans on earth. Phylogenetic studies of this phylum have been impeded by their diversity, microscopic size, and few existing nematode systematists. We are advancing the phylogenetic understanding of the phylum by bringing together the multidisciplinary expertise of the world's leading specialists in nematode systematics to achieve two specific aims: (i) to generate a broadly representative evolutionary framework for the phylum based on 18S rRNA sequence data for 1,000 nematode species; (ii) to use the comprehensive 18S tree as a basis for selecting a subset of the nematode species for analysis with additional orthologous loci. The products of this project are a coordinated series of phylogenetic analyses to address and resolve the evolutionary history of one of the major branches of the Tree of Life, and are designed to establish the foundations for a coherent and sustainable worldwide-research program on nematode diversity and phylogenomics. Broader impacts include linking molecular and morphological

data through an online database, (NemAToL) supporting users of nematode systematics and the training of numerous graduate and undergraduate students, as well as postdoctoral associates.

SEASONAL POPULATION DYNAMICS AND HOST SUITABILITY OF *MELOIDOGYNE PARTITYLA* ON PECAN IN NEW MEXICO. **Thomas, S. H., and J. M. Trojan.** Department of Entomology, Plant Pathology and Weed Science, New Mexico State University, Las Cruces, NM 88003.

Meloidogyne partityla was first reported on pecan in the United States in 1996 in Texas. Populations have since been confirmed in AZ, FL, GA, NM, OK and additional sites in TX. Infected trees typically show canopy die-back, chlorotic leaves, and galled feeder roots. All known hosts of *M. partityla* are members of the Juglandaceae (pecan, walnut, hickory). In 2001 we began twice-monthly sampling of pecan roots from an infested, flood-irrigated, orchard near Las Cruces to determine the seasonal population dynamics of *M. partityla*. For four years, nematode eggs were recovered from roots of two randomly-selected trees at each sampling using 10% bleach extraction. Egg recovery remained low from May through August, but began increasing each year in September, coinciding with late-summer root growth. Egg recovery remained high through the winter, but declined during spring. The predominant pecan rootstocks in NM were evaluated for host suitability to the five root-knot nematode species that occur in the state. Pecan seedlings inoculated with 10,000 eggs of *M. partityla*, *M. incognita*, *M. javanica*, *M. hapla*, or *M. chitwoodi* were grown for 270 days in the greenhouse. All three rootstocks were excellent hosts for *M. partityla*, poor hosts for *M. hapla*, and nonhosts of *M. chitwoodi*, *M. incognita*, or *M. javanica*. Yellow nutsedge (*Cyperus esculentus*), purple nutsedge (*C. rotundus*), and grape (*Vitis vinifera*) were also nonhosts for *M. partityla*, and different alfalfa (*Medicago sativa*) cultivars provided ambiguous results. The predominance of *M. partityla* reproduction on pecan during fall and winter decreases the likelihood that insecticides/nematicides applied between May and August will reduce *M. partityla* injury to pecan.

EFFECT OF CROPPING SYSTEMS ON ABUNDANCE OF *PASTEURIA PENETRANS*. **Timper, P.** USDA-ARS, P.O. Box 748, Tifton, GA 31793.

Few studies have examined the impact of agricultural practices on biological control of nematodes. We have been conducting ongoing studies in Georgia to determine the effect of crop rotation and nematicides on abundance of *Pasteuria penetrans*. This bacterium is an obligate parasite of root-knot nematodes and is naturally present at low levels in most peanut fields in Georgia. In a 9-year field study, crop rotation had a dramatic effect on densities of *Pasteuria* endospores in soil. In the continuous peanut plots, endospore densities were extremely high, and *Pasteuria* suppressed reproduction of *Meloidogyne arenaria* by 91%; whereas in a cotton-peanut rotation, endospore densities were very low. Endospore densities in other rotations corresponded to how frequently a host for *M. arenaria* was grown. Reproduction of the nematode was suppressed by 76% in plots where a host plant was present 2 out of 3 years in a sequence. Frequent planting of peanut to increase the level of *Pasteuria* is unwise because it will lead to an increase risk of other soil-borne diseases. The question we are currently trying to address is: can we utilize an agronomically sound cropping system to increase the abundance of *Pasteuria* and create nematode-suppressive soils in growers fields? During the transition from nematode-conducive to nematode-suppressive soil, nematode populations may need to be held in check by nematicides. Previously, we found the application of aldicarb at planting did not influence densities of *Pasteuria* endospores; however, other nematicides and fumigants need to be evaluated.

STEINERNEMA CARPOCAPSAE EXUDATES SUPPRESS THE CELLULAR NON-SELF RESPONSES OF THE PEST INSECT *GALLERIA MELLONELLA*. **Walter, N. T.,¹ G. B. Dunphy,¹ and C. Mandato.²** Department of ¹NRS, MacDonald Campus of McGill University 21,111 Lakeshore Rd., Ste Anne de Bellevue, H9X 3V9, Department of ²Anatomy and Cell Biology, McGill Campus, Montreal, Canada.

The insect pathogenic nematode *S. carpocapsae* develops in insect blood (hemolymph) without being attacked by the insect blood cells (hemocytes). Since the nematodes release metabolites into the hemolymph, we sought to know if the exudates suppress the immediate hemocytic non-self responses to glass slides and bacteria. Dead nematodes in the insects lowered total hemocyte counts due to hemocytes adhering to nematodes (cellular encapsulation). Live nematodes were not encapsulated and did not affect hemocyte counts. Hemocytes did not change in insects with live nematodes whereas granular cells were lowered in larvae with dead nematodes. Thus live nematodes are either not recognized as non-self or suppress hemocyte responses by 24 h. Exudates from nematodes in phosphate buffer saline elevated the total viable hemocyte counts and granular cell levels by 8 h post-injection. This may represent hemocyte dissociation from tissues since the exudate dissociated hemocytes from slides. Suppression of the removal of *Bacillus subtilis* (Gram-positive bacterium) from the hemolymph was more immediate, 15 min post-injection and thereafter remained constant. *In vitro* the material also impaired the attachment of granular cells only to slides. The immunosuppressive material was detected by 6h and did not increase or decrease thereafter. The immunosuppressant was heat labile (destroyed at 65°C). Treatment of the exudate

with trypsin establishes the immunosuppressant as a protein. That it bound to mildly hydrophobic resin beads shows that the protein is very hydrophobic. Characterization of its mode of action continues.

INDIRECT EFFECTS OF ROOT HERBIVORY BY INSECT LARVAE ON THE STRUCTURE OF THE SOIL FOOD WEB. Treonis, A.,¹ and I. A. Zasada.² ¹Department of Biology, University of Richmond, VA 23173; ²USDA-ARS Nematology Laboratory, Beltsville, MD 20705.

Below ground insect herbivores have negative impacts on plant productivity, but less is understood about the indirect effects of these organisms on the detrital-based soil food web (e.g., the soil microbial biomass and microbial grazers). We used a corn (*Zea mays*) and corn rootworm (*Diabrotica virgifera*) model system to study the effects of increasing root herbivore density on soil food web structure. In a greenhouse, pots (11-cm diameter) of soil were established containing rootworm eggs (0, 100, 500, or 1,000 eggs pot⁻¹) and a single corn plant each. The experiment was destructively harvested 22 d after planting and soil microbial biomass and activity, protozoan density, and nematode abundance and community structure were analyzed. Plant root and shoot biomass declined with increasing rootworm density. Soil nematode abundance increased with rootworm abundance, averaging 8,098 kg⁻¹ soil at the highest density of rootworms and 3,878 kg⁻¹ in the control pots. Soil protozoa (ciliates, flagellates, naked amoebae) responded similarly, with 27,232 cells g⁻¹ in pots with the highest density of rootworms and 3,990 cells g⁻¹ in the control pots. Soil microbial biomass and respiration were unaffected by rootworm herbivory. Rootworm larvae convert living plant material into dead tissue and feces and likely enhance rhizodeposition. The response of microbial grazers in this study suggests that the soil food web is very sensitive to these inputs.

HISTOLOGICAL EXAMINATION OF YELLOW NUTSEDGE AND PURPLE NUTSEDGE TUBERS FOR THE PRESENCE OF MELOIDOGYNE INCOGNITA. Trojan, J. M.,¹ S. H. Thomas,¹ J. Schroeder,¹ and L. W. Murray.² ¹Department of Entomology, Plant Pathology, and Weed Science and ²University Statistics Center, New Mexico State University, Las Cruces, NM 88003.

Yellow nutsedge (*Cyperus esculentus*, YNS) and purple nutsedge (*C. rotundus*, PNS) tubers protect *M. incognita* from the soil fumigant 1,3-dichloropropene, and can serve as a source of *M. incognita* inoculum infecting crops. Tubers surface-sterilized in 10% bleach for a sufficient length of time to kill nematodes and eggs on the tuber surface were also capable of transferring *M. incognita* to peppers. Histopathological studies were conducted to locate *M. incognita* within YNS and PNS tubers, identify the stage of nematode development, and determine which tuber tissues are infected. Nutsedge tubers produced in *M. incognita*-infested soil were fixed with formaldehyde, embedded in paraffin, sectioned, stained and examined under a light microscope. In the YNS tuber sections examined, juveniles and associated well-developed *M. incognita* giant cells were readily apparent. Giant cell formation occurred in vascular bundles of both the inner ground tissue and outer cortical tissue of tubers, distorting cellular structure and organization of the tuber vasculature. No eggs or egg masses have been observed within tubers. Despite extensive efforts, no indication of *M. incognita* has been detected in PNS tubers. Previous research established that surface-sterilized PNS tubers result in a greater transference of *M. incognita* inoculum to adjacent plants than occurs with surface-sterilized YNS tubers; however, the location and developmental stage of *M. incognita* within PNS tubers remains uncertain. This is the first histological study of the host-parasite relationship between *Meloidogyne incognita* and tubers of YNS or PNS. Both YNS and PNS tubers are important hosts for maintaining *M. incognita* populations in the field.

EVALUATING THE EFFICACY OF ORGANIC AMENDMENTS WITH STREPTOMYCES SARACETICUS ON CONTROLLING PLANT-PARASITIC NEMATODES. Tsay, T. T.,¹ W. S. Wu.² ¹National Chung Hsing University, Taichung 402, Taiwan; ²Department of Plant Pathology and Microbiology, National Taiwan University, Taipei 102, Taiwan.

Incorporating *Streptomyces saraceticus* alone or mixed with 0.2% crab shell powder to the soil would lower the egg mass and juvenile numbers of *Meloidogyne incognita* on water spinach in the pot tests, and the fresh weight of above-ground water spinach was also increased. Another set of greenhouse trails also showed that *S. saraceticus* was able to reduce the infection rates and populations of *Tylenchulus semipenetrans*, *Pratylenchus coffeae* and *Paratylenchus curviturus* significantly ($P = 0.05$). To supply nutrients for introducing *S. saraceticus* as a biological control agent, 14 organic wastes were tested for amendment. Among them, crab shell powder, soybean powder, rice bran and molasses were able to increase the population of *S. saraceticus*. When mixed with a combination of crab shell powder, castor pomace, soybean powder, marine algae powder and molasses gave the most significant controlling efficacy and this formula was named Lively Tiller Mixture (LT-M). Compared to the non-treated ones, application of LT-M at the rate of 2000 kg/ha in vineyards once a year, would lower the root-knot index 47.5% and 42.8% in summer and winter growing seasons respectively. The grape cluster weight and sugar content of golden Muscat grape increased 57, 76 grams and 3, 5 Brix° at the two harvest seasons respectively. LT-M in soil reduced 94.1% of *T. semipenetrans* and 63.6% of *P. coffeae* populations, compared to the non-treated control; when compared with nematicide treatments, LT-M not only had as good control results on *T. semipenetrans*, declined citrus plants also showed more vigorous growth at the end of the season.

DOES SOYBEAN CYST NEMATODE AFFECT SPECIALTY SOYBEAN GRAIN COMPOSITION AND YIELD?
Tylka, G. L., and F. Avendaño. Department of Plant Pathology, Iowa State University, Ames, IA 50011.

Farmers are beginning to grow specialty soybean varieties with altered grain qualities (modified protein and oil content) to obtain increased value and to satisfy the demand of emerging market niches. A common problem with soybean production is quantitative yield loss caused by *Heterodera glycines*, the soybean cyst nematode. Recent data indicate that *H. glycines* infection also may affect soybean grain protein and oil content. If production of specialty soybeans is to be successful in the Midwestern U.S., it must be determined whether *H. glycines* infection affects overall soybean grain quality and/or specific components of specialty soybean grain. A field microplot experiment was conducted to determine how *H. glycines* affects yield quantity and grain composition of several specialty soybeans. Treatments consisted of seven soybean varieties grown in soil with and without *H. glycines*. The specialty soybean varieties studied were IA2064 (low linolenic acid) and Vinton 81, IA1007, IA2041 and IA3011 (high protein). Two commodity soybean varieties, Jack (SCN resistant) and Corsoy 79 (SCN susceptible), also were included in the experiment. Each treatment combination was replicated four times. Plants in *H. glycines*-infested microplots were stunted and more chlorotic than those in non-infested microplots for almost every variety studied. Quantity of grain obtained from plants in *H. glycines*-infested microplots was numerically less than that in non-infested microplots for every variety, but the difference was significant ($P = 0.05$) only for IA2041 and Vinton 81. Total grain protein content ranged from 40.6% (Jack) to 45.9% (IA3011) and oil content from 17.2% (IA3011) to 19.8% (IA2064). Grain from Vinton 81 grown in microplots infested with *H. glycines* had significantly ($P = 0.05$) greater protein and less oil than grain from non-infested microplots. Levels of specific amino acids and fatty acids in the grain are being determined to determine if *H. glycines* affected specific proportions of these grain components.

MANAGEMENT OF *ROTYLENCHULUS RENIFORMIS* USING POULTRY LITTER AS A SOIL AMENDMENT.
Usery, S. R.,¹ K. S. Lawrence, C. H. Burmester,² and G. W. Lawrence.³ ¹Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849; ²Agronomy and Soils Department, Auburn University, Auburn, AL 36849; ³Department of Entomology and Plant Pathology, Mississippi State University, Mississippi State, MS 37959.

Evaluations were conducted in a greenhouse and in field plots to determine the effect of poultry litter on *Rotylenchulus reniformis* populations, bacterial and fungal populations, and seed cotton yields. Two types of poultry litter, house litter, and compost litter were evaluated at three rates. Results obtained from greenhouse evaluations indicate that poultry litter may have an antagonistic effect on *R. reniformis* populations. House litter produced 42% fewer *R. reniformis* vermiforms compared to its inorganic fertilizer equivalent. The application of house litter reduced ($P < 0.05$) *R. reniformis* eggs per gram of root by 279.5%, while compost litter applications had 107.4% fewer ($P < 0.05$) compared to the inorganic fertilizer equivalents. Increases in bacterial populations were observed in the compost litter treatments in the greenhouse evaluations. Compared to the inorganic fertilizer equivalents, a 210.5% and 231.3% increase in bacterial populations ($P < 0.05$) was observed at 60 DAP in the 1.0% and 2.0% house litter treatments, respectively. Fungal populations were not influenced by litter applications. Poultry litter applications in field trials did not effect ($P < .05$) *R. reniformis* populations at mid-season and harvest or seed cotton yields over two years of testing. Greenhouse evaluations confirm that poultry litter has the potential to reduce *R. reniformis* in cotton; however, field trials indicate that future long-term evaluations with poultry litter should be conducted before recommendations are made.

EFFECT OF ROTATION CROPS ON *HETERODERA GLYCINES* POPULATION DENSITIES IN A GREENHOUSE SCREENING STUDY. **Vetter, S. A.,¹ S. Chen,² D. L. Wyse,³ G. A. Johnson,² and P. M. Porter.³** ¹Department of Plant Pathology, University of Minnesota, Saint Paul, MN 55108; ²Southern Research and Outreach Center, University of Minnesota, Waseca, MN 56093; and ³Department of Agronomy and Plant Genetics, Saint Paul, MN 55108.

A greenhouse study was carried out to determine the effect of 46 field or cover crops on population densities of the soybean cyst nematode (SCN), *Heterodera glycines*, in soil collected in fall 2003 and spring 2004 from a soybean field in Minnesota. The crops were planted in the SCN-infested soil in 16-cm-diameter clay pots, which were arranged in randomized blocks with six replicates. A no-crop treatment was included as the control. Two and half months after planting, the crops were cut at the soil level, and a soil sample was taken. The aboveground plant tissues were chopped and mixed into the remaining soil for the next phase of the experiment. The soil samples were processed to determine nematode egg counts. After 8 weeks, soil samples were taken again for egg counts. The SCN-susceptible soybean "Sturdy" was then planted in a portion of the remaining soil in 7-cm-diameter pots as a bioassay to determine the viability of the eggs. Thirty-one days after soybean planting, mature females were washed from the soybean roots and counted. Egg population densities and bioassay female counts in the no-crop control treatment did not differ significantly from those of most crop treatments. However, the *Brassica campestris* and hairy vetch (*Vicia villosa*) treatments resulted in higher SCN egg population densities than the no-crop control treatment for some measurements. Sunn hemp (*Crotalaria juncea*), pea (*Pisum sativum*), lablab bean (*Lablab purpureus*), crimson clover (*Trifolium incarnatum*), mung bean (*Vigna radiata*), bundle flower (*Desmanthus illinoensis*), alfalfa (*Medicago sativa*), sunflower (*Helianthus annuus*), and SCN-resistant soybean "Pioneer 9234" treatments resulted in lower egg or female population densities than the no-crop control treatment for some

measurements. Sunn hemp most consistently showed the lowest numbers of eggs and females. As a group, legume crops resulted in lower egg population densities than monocots and Brassica species.

A NEW *ZELDIA* SPECIES FROM THE MOJAVE DESERT, CALIFORNIA. Waceke J. W.,¹ D. J. Bumbarger,² M. Mundo-Ocampo,² J. G. Baldwin,² and S. Subbotin.² ¹Department of Biological Sciences, Kenyatta University, P. O. Box 43844-00100, Nairobi, Kenya; ²Department of Nematology, University of California, Riverside, CA 92521.

A new *Zeldia* species is described from the Mojave Desert in California, USA. The new species shares with other species of *Zeldia* a single guard process in each primary axil and asymmetrical triangular-shaped lips. However, the new species differs from all the other species of the genus by the elongate spanner-shaped, deeply bifurcate probolae; the probolae of other *Zeldia* species are typically low, rounded and bicornuate. In addition, the new species has five lateral incisures (versus three in other species of the genus), the cuticle is tessellate (versus no tessellation in the other species) and unlike some other *Zeldia*, this new species lacks punctations. With respect to the high bifurcate probolae, the new species resembles *Chiloplacus* or *Stegelleta*. Although the new species resembles some *Nothacrobeles* with respect to overall asymmetry and triangular shape of the lips and the presence of tines on lips, molecular phylogenetic analyses excludes *Nothacrobeles* as a sister to the new species. Though generally morphologically consistent with *Zeldia* spp, the new species is genetically distant to *Z. punctata*. The morphological characters of the new species are the basis for a broader emended diagnosis of *Zeldia*, and suggest novel implications for evolution of Cephalobidae.

SSR AND AFLP-DERIVED MARKERS FOR ROOT-KNOT NEMATODE RESISTANCE IN COTTON. Wang, C., and P. A. Roberts. Department of Nematology, University of California, Riverside, CA 92521.

Root-knot nematode (*Meloidogyne incognita*) is a major pest of cotton, and host-plant resistance is an economical, safe and effective method for managing this nematode problem. Three cotton cultivars (two of *Gossypium hirsutum* and one of *G. barbadense*) were characterized for the phenotype of reactions to *M. incognita*, and for AFLP and SSR (microsatellite) molecular marker polymorphisms. Based on genetic and molecular analysis of F₁, F₂, F₃, BC₁F₁ and F₇ recombinant inbred lines from the cross of resistant cv. NemX (*G. hirsutum*) with susceptible cv. SJ-2 (*G. hirsutum*), four AFLP markers and two SSR markers were found tightly linked to a major resistance gene in NemX. One of the AFLP markers was sequenced and DNA primers were designed for specific amplification of both parents. Both NemX and SJ-2 generated same-sized bands. Comparison between the sequences generated from the bands in resistant and susceptible plants revealed a single base substitution associated with resistance that provided a recognition site for cleavage by the restriction endonuclease *Nla* III. Amplification with the designed primers and cleavage with *Nla* III yielded a cleaved amplified polymorphic sequence (CAPS) marker that segregated with the nematode resistance. Analysis of segregating progenies from the cross of NemX with susceptible *G. barbadense* cv. Pima S-7 and the test-cross NemX x F₁ (Pima S-7 x SJ-2) indicated that Pima S-7 contributed a separate factor involved in transgressive segregation for resistance. Additional SSR markers are being screened for localization and fine mapping of the resistance genes using the Pima S-7 x NemX and NemX x SJ-2 progenies.

USE OF ORGANIC AMENDMENTS TO CONSERVE NATURAL BIOLOGICAL CONTROL: NEMATODE-ANTAGONISTIC FUNGI AND PREDATORY NEMATODES. Wang, K.-H., and R. McSorley. Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611.

Use of organic amendments to enhance nematode-antagonistic fungi has been controversial. Therefore, results from using nematode-antagonistic fungi in augmentative biological control have been unpredictable, and the development of conservation biological control for nematode management has been hindered. This presentation provides a brief review of research, suggesting possible ecological explanations for this inconsistency. Some of the questions raised include: Could the response to organic matter depend on nematophagous fungi species? On the quality and quantity of the organic matter? On nematode density? On nematode community structure? Or on field history? Most importantly, could the enhancement of nematode-antagonistic fungi by organic matter lead to the enhancement of nematode-trapping activities? Increasing numbers of field sites with nematode suppressive soil are being reported, reinforcing the need to study cultural practices that can conserve naturally occurring nematode biological control agents. Some results have indicated that adding organic matter to the soil also increased the abundance of predatory nematodes. While the efficiency of plant-parasitic nematode suppressiveness by these beneficial soil organisms is variable, the combination of these organisms adds another dimension to the conservation of natural biological control of nematodes.

EFFECTS OF PRE-PLANT SOIL TREATMENTS ON NEMATODE COMMUNITIES. Wang, K.-H.,¹ R. McSorley,¹ and N. Kokalis-Burelle.² ¹Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611; ²USDA, ARS, U.S. Horticultural Research Laboratory, Fort Pierce, FL 34945.

While nematodes play important roles in soil nutrient cycling, many pre-plant soil management practices act as perturbations to nematode communities. A 2-year field trial was conducted to examine nematode communities in soil treated with methyl bromide (MB), 6-week solarization (S), cowpea cover cropping for 3 months (CP), a combination of

solarization and cowpea cover crop (S+CP), and a weed fallow control. At termination of pre-plant treatments, MB reduced ($P < 0.05$) total abundance of bacterivores, fungivores, herbivores, and predators, as well as fungivore/bacterivore ratio (F/B), richness and channel index compared to the control in year 1. Except for total predatory nematodes, these indices were also affected in year 2. In addition, MB also reduced diversity, maturity index (MI) and structure index (SI), but increased ($P < 0.05$) enrichment index (EI) in year 2. Although solarization had great impact on nematode communities in year 1, it only reduced ($P < 0.05$) richness and diversity in year 2. Cowpea cover crop alone reduced ($P < 0.05$) total abundance of fungivores and herbivores, F/B, and CI, but increased total abundance of omnivores and SI compared to the control in year 1, whereas it increased total abundance of bacterivores, fungivores, omnivores, and predators and EI, but decreased diversity in year 2. Combining solarization with cowpea had a similar effect on nematode communities as S in year 1, but increased ($P < 0.05$) total abundance of omnivores in year 2. Three months after pepper (*Capsicum annuum*) crop was planted, total abundance of each trophic group did not differ from the control in most treatments, except in the MB in year 1. However, F/B, richness, and diversity were lower ($P < 0.05$) in all these pre-plant treatments than the control. In year 2, 3 months after peppers were planted, total abundance of bacterivores, and fungivores, as well as richness in plots receiving pre-plant treatments were lower than the control. Differences in response of nematode communities to treatments in year 2 may be due to a severe hurricane season, resulting in proliferation of fungal pathogens at the experimental site.

ENTOMOPATHOGENIC NEMATODE-BACTERIAL SYMBIONT METABOLITES FOR APPLIED BENEFIT. Webster, J. M.,¹ J. Li,² and G. Chen.² Department of ¹Biological Sciences, Simon Fraser University, Burnaby, Vancouver, V5A 1S6; ²Welichem Biotech Inc., Burnaby, Vancouver, V5G 3L1.

Entomopathogenic nematodes (EPNs) and their bacterial symbionts parasitize insects, and have been commercialized for insect pest control. However, the lethal, holistic endpoint of the host-parasite interaction is not the only point of interest, rather some of the steps in the infection process merit particular focus. Following their release into the insect haemocoel the symbiotic bacteria grow rapidly and by 96h nematode development also has commenced. Some of the metabolites produced by these bacteria are antibiotics, and one of them is a non-steroidal, small molecule that, when extracted and purified, has significant anti-inflammatory effects when tested in mouse model experiments. In phorbol-12-myristate-13 acetate (0.01%w/v) induced edema in mouse ears the synthesized bacterial metabolite, as a 1.0% cream, significantly decreased ear inflammation in mice. This anti-inflammatory effect was supported *in vitro* by significant inhibitory effects of the compound on inflammation related activities of cytokines such as TNF alpha and IFN gamma. These effects on the inflammation component of the innate immune defence system in mammals parallels that occurring in the immune defence system in insects. The physio-biochemical activities of the products of the symbiotic partnership of EPNs and their bacteria in insects suggest that some of the small molecule metabolites could have pharmaceutical application.

EFFECTS OF TILLAGE INTENSITY ON POPULATION DENSITIES OF *HETERODERA GLYCINES* IN CROP SEQUENCES OF CORN AND SOYBEAN. Westphal, A.,¹ and T. J. Vyn.² Departments of ¹Botany and Plant Pathology and ²Agronomy, Purdue University, West Lafayette, IN 47907.

Heterodera glycines is one of the most important yield-reducing parasites of soybean. Management among other strategies uses resistant cultivars, but alternative agronomic practices that use additional modes of population reduction are urgently needed. The role of tillage intensity on population density development of *H. glycines* was investigated in long-term tillage plots, established in 1975 at the Agricultural Center for Research and Education, Purdue University. In this long-term experiment, main plots were crop sequence treatments: (i) continuous corn, (ii) corn-soybean, (iii) soybean-corn, and (iv) continuous soybean. Subplots in a split-plot arrangement were tillage treatments: (A) moldboard plow and secondary tillage, (B) chisel and secondary tillage, (C) ridge tillage, and (D) no tillage. In earlier years, the soybean cultivars used in the experiment had some resistance to *H. glycines*. Starting in 2003, a strip in each soybean subplot was planted to the *H. glycines*-susceptible cultivar Williams 82 to allow nematodes to reproduce. In spring and fall of 2003 and 2004, soil samples were collected to a depth of 30 cm to monitor nematode population densities. In the combined analysis of rotational and soybean monoculture plots of both years, fall population densities of *H. glycines* decreased with decreasing intensity of tillage in rotational soils, whereas population densities were not significantly different among tillage treatments in the monoculture soils. Tillage has effects on the soil environment that warrant further study to determine the impact on population densities of *H. glycines*.

DEVELOPMENT OF SUDDEN DEATH SYNDROME IN TWO SOYBEAN LINES IN FIELD MICROPLOTS INFESTED WITH *FUSARIUM SOLANI* F. SP. *GLYCINES* AND *HETERODERA GLYCINES*. Xing, L., and A. Westphal. Department of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907-2054.

Sudden death syndrome (SDS) of soybean, caused by *Fusarium solani* f. sp. *glycines*, is an important soil-borne disease in the Midwest. Since the putative interaction of the fungal pathogen and *Heterodera glycines* (SCN) under field conditions remains unclear, a four-factor factorial test was established in field microplots to investigate their interaction. Treatments were arranged in a randomized complete block with four replications. The four factors were: (i) methyl bromide-fumigation

at 450 kg/ha; (ii) inoculation with *F. solani* f. sp. *glycines*, (two strains, provided by USDA-ARS, IN); (iii) inoculation with *H. glycines*; and (iv) planting to Williams 82 (SCN-susceptible) or Cyst X20-18 (SCN-resistant). The inocula of the two *F. solani* f. sp. *glycines* isolates and of *H. glycines* were mixed into the upper 15 cm of soil at 3.9×10^5 macrospores per ml of soil or 1509 cysts with 1.18×10^5 eggs per 500 g of soil. Beginning at growth stages V6/R1, all plots were irrigated at least once a week to keep soil moisture levels >50 mbar. During September at R5/R7, we assessed foliar SDS severity on a scale of 0 to 9 and percentage defoliation. At R6/R8, we rated external necrosis on roots and recorded the presence of Diaporthe stem rot. Grain yields and soil population densities of *H. glycines* were determined at harvest. SDS severity was increased by the presence of *H. glycines*. Cyst-X20-18 had minimal SDS severity compared to Williams 82 when both were co-inoculated *H. glycines* and *F. solani* f. sp. *glycines*. Results suggest that determining population densities of *H. glycines* is imperative when evaluating SDS in commercial fields or research tests.

THE EFFECT OF SOIL ENVIRONMENT ON SOYBEAN CYST NEMATODE (*HETERODERA GLYCINES*) EGG HATCH. **Xu, Y.,¹ Y. Chen,² Z. Si,¹ Z. Li,¹ C. Li,¹ G. Wen.¹** ¹Northeast Institute of Geography and Agricultural Ecology, Chinese Academy of Sciences, Harbin 150040, China; ²Northeast Agricultural University, Harbin 150030, China.

Soybean cyst nematode, *Heterodera glycines* Ichnohe, is one of the major pathogens in soybean continuous cropping. To study the relationship between soil environment and the nematode damage on soybean crop, we treated the nematode eggs with soil fluids extracted from different crop rotation systems. The results show that the soil fluid from soybean field induces significantly higher egg-hatching rate than the fluid from the field of corn, sugar beet, wheat, and flax. The nematode damages accessed from yearly rotation field study are consistent with the egg-hatching experiment. Our results indicate that the root exudates from different crops play an important role in affecting the nematode active population. Our results also show that the soil extract from multi-year continuous cropping of soybean showed suppression on the nematode egg hatch. This indicates that the concentration of the putative factors may play a role in regulating the nematode egg hatch.

CHARACTERIZATION OF AN UNDESCRIBED *APHELENCHOIDES* SP. ON AQUATIC PLANTS. **Yang, T. S., T. T. Tsay, and P. C. Chen.** Department of Plant Pathology, National Chung Hsing University, Taichung 402, Taiwan.

An unknown *Aphelenchoides* species was isolated from aquatic plants on a commercial farm in Taichung, Taiwan. Nematodes were observed to accumulate in the buds of *Ceratophyllum demersum*, *Cabomba* spp., and in the axillar part of *Hydrilla verticillata*. This *Aphelenchoides* population had a body length ranging from 0.69-0.97mm, with slender body shape tapering to a multi-papillate tail tip. The stylet averaged 10 μ m in length with slight basal swellings. The lateral field was approximately 0.18 times body width with four incisures. Females had a single row of oocytes and an obvious post-uterine sac. Males were characterized by the rosethorne-shaped spicules. Chemotaxis tests indicated that this nematode could be attracted by apex and stem tissue sections of *Hydrilla verticillata* and was later found inside the tissue by staining. Among five fungus species tested as a food source, *Botrytis cinerae* could sustain the largest population of nematodes. Comparing morphometrics with the 40 described *Aphelenchoides* species found no congruent species. The morphometrical measurements were very similar to *A. fragariae* but differed in the number of incisures. Work is underway to describe this population of *Aphelenchoides* found on aquatic plants in Taiwan as a new species.

MORPHOLOGICAL AND MOLECULAR INVESTIGATION OF NEMATODES FROM FIG FRUITS IN AUSTRALIA. **Ye, W.,¹ R. M. Giblin-Davis,¹ K. A. Davies,² and W. K. Thomas.³** ¹FLREC, University of Florida/IFAS, Davie, FL 33314; ²University of Adelaide, Waite Campus, Glen Osmond, SA 5064, Australia; ³Hubbard Center for Genome Studies, University of New Hampshire, Durham, NH 03824.

Fig sycones (phases B-D) from 17 species of *Ficus* from South Australia, New South Wales, and Queensland, Australia were dissected over the course of six years and observed for nematodes. Samples were processed for morphological and molecular analyses. All nematodes found within the figs were assumed to be carried in by their respective fig wasp (Agaoninae) pollinators, this being confirmed in cases where pollinators were present. *Schistonchus* was the most commonly encountered nematode in endemic (subgenus *Urostigma*, section Malvanthera) and naturalized *Ficus* species from the Pacific. Phylogenetic analysis was performed using DNA sequences of the nuclear small and large subunit ribosomal RNA gene. In several cases, there was evidence of multiple species of *Schistonchus* from a single fig host species. In other cases, the same *Schistonchus* species was recovered from multiple sympatrically occurring fig species. Both cases are suggestive of host switching. *Parasitodiplogaster* was only recovered from fig wasps and figs of *F. virens* (subgenus *Urostigma*, section *Urostigma*). However, in addition to *Schistonchus*, a very unusual diplogasterid with molecular affinities to *Parasitodiplogaster* and a *Mononchoides* sp. was recovered from fig wasps and figs of *F. racemosa* (subgenus *Sycomorus*, section *Sycomorus*). A new species in the Hexatylna was recovered from *F. congesta* (subgenus *Ficus*, section *Sycocarpus*).

A PROTOTYPE ONLINE KEY FOR RAPID AND JARGON-FREE IDENTIFICATION OF NEMATODE GENERA, BASED ON THE PRINCIPLES OF SCALEFREE NETWORKS. **Yoder, M., K. Carter, I. King, I. T. De Ley, and P. De Ley.** Department of Nematology, University of California, Riverside, CA 92521.

Traditional dichotomous or polytomous keys are based on specific morphological character states that require knowledge of specialized terminology. Diagnostic morphology can be difficult to interpret with light microscopy, and in some cases measurements are required for correct identification. However, this formalized and labor-intensive approach does not reflect the actual process of identification in the minds of experienced specialists. Typically, for the first step in identification taxonomists rely on visual matches with memorized images and past observations of genera. The specialist then turns to formal keys when there is no recollection of having previously seen a specific genus in print or on slides. In order to simplify genus identification along similar lines for novice nematologists and ecologists, a prototype pictorial online key was designed using novel principles. The key can be accessed at <http://faculty.ucr.edu/~pdeley/scalefree.html> and consists of sets of clickable images that are navigated by visual matching or which can be inspected in more detail as multifocal images. For each genus, an adult female and male (where present) were video captured and edited (VCE) at five informative body regions to highlight key morphological characters for identification. Image sets of visually similar nematode genera were linked according to the principles of scale-free networks, ensuring that navigation through the key will not become significantly slower as more genera are added. Currently, the total number of genera included is 70, consisting of 55 terrestrial and 15 marine nematode genera.

RE-THINKING A RYE COVER CROP AS A PLANT-PARASITIC NEMATODE MANAGEMENT TOOL. **Zasada, I. A.,¹ S. L. F. Meyer,¹ and C. Rice.²** ¹USDA, ARS Nematology Laboratory, Beltsville, MD 20705; ²USDA, ARS Environmental Quality Laboratory, Beltsville, MD 20705.

A rye cover crop has been shown to suppress plant-parasitic nematodes, with variable results. To successfully utilize cover crops for plant-parasitic nematode management, it is necessary to understand the mechanism(s) of nematode suppression associated with these plants. We have demonstrated that hydroxamic acids, secondary plant metabolites found in the Poaceae, are toxic to *Meloidogyne incognita* *in vitro*. Degrees of toxicity varied, with DIBOA (2,4-dihydroxy-(2*H*)-1,4-benzoxazin-3(4*H*)-one) more toxic than its degradation product BOA (benzoxazolin-2(3*H*)-one), and MBOA (6-methoxy-benzoxazolin-2(3*H*)-one) more toxic than its parent compound DIMBOA (2,4-hydroxy-7-methoxy-(2*H*)-1,4-benzoxazin-3(4*H*)-one). However, use of this information to develop an effective and practical nematode management strategy, based upon the incorporation of rye cover crops, is likely to be complicated by genetic variability of rye, seasonal influences, and the complex nature of the soil environment. To begin addressing these problems, several cultivars including Wrens Abruzzi, Elbon, Merced, Aroostock, Wheeler, and Oklon were screened for the attributes of host suitability to *M. incognita* and hydroxamic acid content. Variation in these attributes among rye cultivars is discussed.

VIRULENCE PHENOTYPES OF *HETERODERA GLYCINES* POPULATIONS IN MINNESOTA. **Zheng, J., Y. Li, and S. Chen.** University of Minnesota, Southern Research and Outreach Center, Waseca, MN 56093.

The soybean cyst nematode (SCN), *Heterodera glycines*, is a major yield-limiting factor of soybean production in Minnesota. Knowledge of the virulence phenotypes of SCN populations is important in choosing appropriate resistant sources for breeding resistant cultivars and for managing the nematode. The SCN virulence phenotypes have been classified as "races" and recently as "HG Types". We investigated races of 59 SCN populations collected in 1997/1998, and races and HG Types of 46 populations collected in 2002 from soybean fields across SCN-infested counties in southern and central Minnesota. In the 1997/1998 samples, race 3 was predominant and represented 78% of the populations. The remaining populations were 11.9% race 1, 1.7% race 4, 6.8% race 6, and 1.7% race 14. In the 2002 samples, the populations were classified to 20.9% race 1, 69.8% race 3, 4.7% race 5, and 4.7% race 6. Percentages of 2002 populations with female indices (FI) higher than 10 on the HG Type indicator soybean lines PI 88788, PI 209332, and PI 548316 were 26.1% (highest FI 32), 15.2% (highest FI 35), and 47.8% (highest FI 36), respectively. No 2002 population produced FI > 5; 10 on the other indicator lines PI 548402, PI 90763, PI 437654, and PI 89772. The line PI 548316 was relatively susceptible to the Minnesota SCN populations and may not be recommended for breeding resistant cultivars in the state. More populations collected in 2002 are being tested and the data of 2002 populations will be compared with 1997-1998 populations to determine if there were changes of virulence phenotypes in response to the use of resistant cultivars during 1997-2002 in Minnesota.