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ABSTRACTS

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NEMATICIDAL ACTIVITY OF PHYTOCHEMICALS FROM SOME ARID LAND PLANTS. **Abdel-Rahman, F., and M. A. Saleh**. Departments of Biology and Chemistry, Texas Southern University, Houston, TX 77004.

Nematicidal activities of phytochemicals from twenty two arid-land plants were evaluated. The tested plants were; *Artemesia herba alba; Artemisia cinae; Astragalus vogelli; Acacia raddiana; Aschillia millifolium; Conzya bonariensis; Clerodendrom inerma; Cassia acutlifolia; Eugina aromatica; Zygophyllum album; Senicio desfontainei; Ruta graveolens; thymus vulgaris; schinum malle; Tamarix aphylla; Tamarix nilotica; monsonia nivea; hyoscgamus nuticus; Launaea cassiniana; nigella sativa; Pulicaria crispa;* and *ruta graveolens.* Organic and water extracts were fractioned and their activity was evaluated separately. Water screening bioassay was used to determine the effect of each tested plant extract on the activity of the second-stage larvae of *Meloidogyne incognita.* Second-stage larvae of *M. incognita* were incubated in each organic and water extract for 48 hours, then inoculated to healthy tomato seedlings to determine the effect of each phytochemical fraction on the pathogenicity of *M. incognita* as it was determined from gall and egg-mass counts. The organic fractions wet-screen bioassay and the post infection development tests revealed that *Tamarix aphylla* was very effective caused 80% mortality, and 19% root galls; *Astragallus vogelli* caused 63% mortality and 15% galls; *launaea cassiniana* caused 62% mortality and 6% galls. The most effective water extracts were *Artemisia monosperma* caused 78% mortality and 16% galls; and *Eugina aromatica* caused 65% mortality and 15% root galls. Organic and Aqueous fractions for some selected plant extracts with different concentrations, 100 ppm, 500 ppm, and 1000 ppm were tested in wet-screen bioassay using the second-stage larvae of *M. incognita* to determine the lowest crude organic and / or aqueous plant extract concentrations which cause mortality of second-stage larvae of *M. incognita*. *Eugina aromatica* aqueous fraction was the most effective in causing mortality of second-stage larvae at different concentrations. Organic fraction of *Tamarix nilotica* was the most effective at 1000 ppm concentration, causing high nematode mortality. Other organic plant fractions caused moderate to high mortality using 500 ppm and 1000 ppm concentrations in wet-screen bioassay.

RESPONSE OF TWO CARROT (*DAUCUS CAROTA* L.) CULTIVARS TO VARYING LEVELS OF ROOT-KNOT NEMATODE (*MELOIDOGYNE HAPLA*) INOCULUM. **Abuan, M. M., L. M. Villanueva, and T. D. Masangcay.** Horticulture Research and Training Institute, Benguet State University, La Trinidad, Benguet 2601, Philippines.

The effect of *Meloidogyne hapla* parasitism on the growth and yield of two carrot cultivars, new Improved Kuroda and Tokita Kuroda were studied under greenhouse conditions at Benguet State University, La Trinidad, Benguet, Philippines. The plants were individually inoculated with 0, 50, 100, 500, 1000 and 5000 juveniles per pot. Data on the fresh root weight, gall index, number of galls on secondary roots, number of galls or damaged areas on tap root, number of egg masses and nematodes in roots and yield were obtained at the completion of the trial. A statistical design of randomized blocks with treatments arranged according to a 2 × 6 factorial model (cultivars × Pi) was established and each experimental unit (one plant growing per pot) was replicated five times with two plants per replicate. Fresh top weight was not significantly affected by *M. hapla* in both cultivars tested. However, root gall index, number of galls on secondary roots, number of galled or damaged areas on tap roots, number of eggmasses and nematodes in the roots increased significantly as the Pi was increased in both cultivars. The tap root quality was significantly affected by nematode inoculation in New Improved Kuroda compared to Tokita Kuroda. In the former cultivar, all the inoculated plants were not able to produce marketable yield. At lower Pi, Tokita Kuroda was able to produce marketable roots but not at 1000 and 5000 juveniles per pot.

COMPARISON OF METHODS FOR IDENTIFICATION OF *MELOIDOGYNE ARENARIA* FROM FIELD SOIL SAMPLES WITH MIXED *M. ARENARIA* AND *M. INCOGNITA* POPULATIONS. **Agudelo, P., S. A. Lewis, and B. A. Fortnum.** Department of Entomology, Soils, and Plant Sciences, Clemson University, Clemson, SC 29634.

A real-time PCR assay, using species-specific primers and SYBR Green I Dye, for identification of *Meloidogyne arenaria* is described. The specificity of the reaction was confirmed by testing for amplification of DNA from other *Meloidogyne* species and from *M. arenaria* populations of different geographic origins. Comparisons with other identification methods using mature females and juveniles were made. The real-time PCR assay provided a sensitive means for the rapid identification of juveniles from soil samples with mixed populations of *M. incognita* and *M. arenaria*.

INDUCTION OF SYSTEMIC ACQUIRED RESISTANCE AND SUSCEPTIBILITY IN TOMATO BY TWO *MELOIDOGYNE INCOGNITA* POPULATIONS. **Anwar, S. A.,[1] and M. V. McKenry.[2]** [1]Department of Plant Pathology, University of Agriculture, Faisalabad, Pakistan and [2]Department of Nematology, University of California, Riverside, CA 92521.

The effects of virulent San Quentin and avirulent Beltran *Meloidogyne incognita* populations on the host suitability of tomato cv. Favorita were investigated on a split-root system in controlled environment at 30OC. Initially one-half of the split-root system was inoculated with 5000 eggs of virulent San Quentin or avirulent Beltran M. incognita populations as inducer inoculum (prior). Later at 0, 7, 13, 21 and 27 days 5000 eggs from virulent or avirulent populations were applied to the other half of the root as challenge inoculum. Each challenge inoculation had a corresponding check in which the same population was applied to only one-half of a split-root system. The rate of reproduction [Final egg population (Pf)/initial egg population (Pi = 5000)] was determined 63 days after challenge inoculation. Induced inoculation with the Beltran population of *M. incognita* significantly suppressed reproduction of the San Quentin population applied as a challenge inoculation. The Pf/Pi of challenge *M. incognita* was 30, 24, 12, 6 and 5, whereas for the corresponding checks it was 27, 29, 25, 25 and 12. Concomitant or sequential inoculations of either inducer or challenger populations did not alter host suitability. However, if the inducer inoculation was virulent it significantly enhanced reproduction of the challenging avirulent population compared to the check. Pf/Pi of challenge avirulent population was 3, 5, 18, 25, and 20 whereas for the corresponding check it was 1, 2, 7, 9 and 11. These results demonstrate that prior infection of tomato with virulent or avirulent populations could induce system resistance or susceptibility to subsequent infections.

USE OF THE PARASITIC NEMATODE *THRIPINEMA NICKLEWOODI* AS A BIOLOGICAL CONTROL AGENT FOR WESTERN FLOWER THRIPS INFESTING CHRYSANTHEMUM. **Arthurs, S.,[1] and K. M. Heinz.[2]** [1]USDA-ARS, Yakima Agricultural Research Laboratory, Wapato, WA 98951, [2]Department of Entomology, Texas A&M University, College Station, TX 77843-2475.

The parasitic nematode *Thripinema nicklewoodi Siddiqi* (*Tylenchida: Allantonematidae*) naturally infects feeding stages of western flower thrips *Frankliniella occidentalis Pergande* (*Thysanoptera: Thripidae*) residing within the buds, open flowers and foliar terminals of flowering plants. The association is not lethal, but the embryos of infected female hosts degenerate leading to sterility. No mass culture methods are available for *T. nicklewoodi* but an in vivo approach was successfully used to produce a continuous supply of the infective stages. Both fertilization and horizontal transmission of *T. nicklewoodi* is achieved in 1.5 ml microcentrifuge tubes containing thrips larvae (infection arenas), in the presence of 100% humidity, a temporary food source and preferably a damp substrate. Infected thrips are reared on bean *Phaseolus vulgaris L.* for two weeks at 25°C to allow the reproduction and development of a single generation of nematodes within the host's abdominal cavity. The nematode was evaluated as a thrips biological control agent in chrysanthemum. The inoculative releases of two parasitized hosts per plant enabled *T. nicklewoodi* to become established within existing flower thrips populations under greenhouse conditions. However, horizontal transmission rates on plants were density dependent and low level inoculations were not effective over a single crop cycle. Further studies showed that transmission of *T. nicklewoodi* persisted for 9 host generations, infected up to 83% of adult thrips and provided long-term suppression of discrete caged populations, but only after uneconomically high thrips densities had been reached. Introduction of *T. nicklewoodi* or other allantonematid nematodes may be more valuable to suppress pest outbreaks in perennial crops with higher damage thresholds.

POLYMORPHISM OF THE ITS-rRNA GENE CLUSTER WITHIN AND AMONG POPULATIONS OF *HETERODERA GLYCINES* FROM CHINA, JAPAN, AND THE UNITED STATES. **Atibalentja, N.,[1] and G. R. Noel.[2]** [1]Department of Crop Sciences, University of Illinois at Urbana-Champaign, 1102 South Goodwin Avenue, Urbana, IL 61801 and [2]USDA, ARS, Urbana, IL 61801.

For many years researchers have known that populations of *Heterodera glycines* are variable, but exactly how much polymorphism is present within and among *H. glycines* populations has not yet been quantified. At the molecular level, the extent of polymorphism within and among populations is measured by the nucleotide diversity (π) and nucleotide divergence (d_A) indices, respectively. To estimate these two parameters for the ITS-rRNA gene region, we used the PCR RFLP technique to genotype 10 randomly selected virgin females from each of 19 *H. glycines* populations from China (7), Japan (5), and US (7). Populations of *H. glycines* from the US were more polymorphic with 0.014 nucleotide substitutions per site, compared to 0.010 and 0.012 for China and Japan, respectively. The US populations were more closely related to Chinese populations (d_A = 0.00096) than they were to the Japanese populations (d_A = 0.000775). Nucleotide divergence between Chinese and Japanese populations was 0.000537.

INTRASPECIFIC ITS1 POLYMORPHISM IN THE COLOMBIA LANCE NEMATODE *HOPLOLAIMUS COLUMBUS*. **Bae, Chang-Hwan,[1] A. L. Szalanski,[2] and R. T. Robbins.[1]** [1]Department of Plant Pathology and [2]Entomology, University of Arkansas, Fayetteville, AR 72701.

Hoplolaimus columbus is a widespread and important agricultural pest in the Southeastern United States and it reproduces by parthenogenesis. The first internal transcribed spacer (ITS1) region of the ribosomal RNA of *H. columbus* was amplified and sequenced to assess genetic variation within and between populations and to explore phylogenetic relationships. The pattern of sequence variation suggests that three different ITS1 variants (Type I, Type II, Type III) exist within populations. To confirm that these ITS 1 variants exist in each population, three sets of type specific primers are used in 14 different populations and showed that Type II and Type III are presented in all populations of *H. columbus* and Type I occurred at a low frequency. The existence of several ITS1 variants complicates phylogenetic analysis if these variants come from nonorthologous ITS1 types.

EFFECTS OF THE CELL DEATH PROTEIN CED-4 ON *CAENORHABDITIS ELEGANS* AND *MELOIDOGYNE INCOGNITA*. **Bahaji**, **A., T. Padukkavidana**, **C. Tristan**, **G. W. Polack, and Al. Calderón-Urrea**. Department of Biology, California State University, Fresno, Fresno, CA 93711.

Caspases, cysteine proteases that cleave at aspartate residues, are directly involved in eliciting PCD, and, in *C. elegans*, are activated by the product of the gene *ced-4*. Here we present evidence that direct exposure of *C. elegans* or *Meloidogyne incognita* to the CED-4 protein can efficiently reduce fecundity or gall formation respectively. Since *in vitro* exposure of *C. elegans* to CED-4 decreased fecundity, we decided to test if the exposure of a *M. incognita* to CED-4 would have a similar effect. We generated transgenic tobacco plants containing the *Ced-4* gene and exposed homozygous plants to *M. incognita* J2 worms. We observed that among 7 lines tested, five lines showed a statistically significant reduction in the number of galls formed. This phenotype correlated with high levels of *ced-4* RNA detected as established by a competitive RT-PCR assay. We concluded that high RNA expression of *Ced-4* is correlated with a low galls formation especially in the lines L14 and L17, which showed the highest *Ced-4* expression level. Although anti-CED-4 antibodies could not detect CED-4 in plant tissue, we used a functional assay to detect the activity of the plant-expressed CED-4. Since caspase 9 is the orthologue of CED-3, and can be activated by CED-4, we tested protein extracted from transgenic *Ced-4* plants for its ability to activate caspase 9 in an *in vitro* assay. We observed high activity with *C. elegans* wild-type protein extracts, compared to the mutant nematodes. Furthermore, we observed that the protein extracts from transgenic tobacco plants complemented the activity in the protein extracted from mutant nematodes to levels detected in wild-type nematodes. We concluded that this complementation was due to the presence of CED-4 in the transgenic tobacco plants expressing *ced-4* RNA. We discuss the feasibility of using cell death proteins to control parasitic nematodes.

OSMOLARITY AND THE STABILITY OF PRIMARY PHASE IN *PHOTORHABDUS LUMINESCENS*. **Bailey G. C.,[1] T. Bliss,[1] K. C. Krasmil-Osterfeld,[2] and B. J. Adams.[1]** [1]Department of Microbiology and Molecular Biology, Brigham Young University, 775 WIDB, Provo, UT 84602; [2]Monsanto Company, 700 Chesterfield Parkway W, Chesterfield, MO 63017.

Photorhabdus bacteria have been observed to exist in both a primary and secondary phase. *Photorhabdus* supports the growth of its symbiotic, nematode partner *Heterorhabditis*, but only if the bacteria is expressing the primary phase phenotype. Subculturing *Photorhabdus* in broth with low osmolarity has been observed to be a significant factor in promoting the bacteria to switch from primary to secondary phase. Broth osmolarities higher than 400 mOsmols seem to stabilize primary phase bacteria, while osmolarities lower than 60 mOsmols appear to encourage a switch to secondary phase. Factors such as temperature, light intensity, pH, and access to oxygen also have been reported to play a role in directing the phase of the bacteria, but with highly variable responses. Variation in broth osmolarity and culture temperature was observed for effect on phase shift. Primary phase bacteria were cultured in broths of osmolarity levels ranging from 25 to 800 mOsmols. Each level of osmolarity was exposed to temperatures ranging from 10 to 40 degrees Celsius. The colonies were subcultured every 48 hours for six days. The capacity of high osmolarity broths to stabilize primary phase bacteria despite stressful temperatures is discussed.

MITIGATION OF ROOT HEALTH PROBLEMS IN SAUDI ARABIAN WHEAT PRODUCTION. **Becker, J. O.,[1] M. A. Braim,[2] and J. King.[3]** [1]Department of Nematology, University of California, Riverside, CA 92521; [2]Corpslanding, Hutton Cranswick, Driffield, UK; [3]Todhia Arable Farms, KSA.

Irrigated wheat (cv. *Yecoro Rojo*) was grown in rotation with Rhodes grass (*Chloris gayana*) on a 3250 ha farm in central Saudi Arabia. The wheat was direct-drilled into glyphosate-killed grass stubble. After several years, this practice resulted in unthrifty wheat crop from the third leaf stage on. At mid-season, 50–100 lesion nematodes (*Pratylenchus* spp.) per g of wheat roots and 200–1000 stunt nematodes (*Merlinius brevidens*) per 100 cc rhizosphere soil were extracted. The brownish-discolored roots frequently harbored *Pythium* spp. and *Rhizoctonia solani*. In pot tests, drench application of either oxamyl, metalaxyl or pencycuron improved wheat growth and tillering over the non-treated check. In two field trials, soil application of metalaxyl and oxamyl resulted in yield increases of 21% and 18%, respectively in trial 1, and 33% to 48%, respectively in trial 2. An experimental nematicidal seed treatment (1.58 g oxamyl/kg seed) reduced lesion nematodes

seven weeks after seeding by 80%, resulted in 60% more tillers and approximately 1.6 t/ha yield increase compared to the non-treated check.

NEMATICIDAL SEED COATING REDUCES ROOT-KNOT NEMATODE-ENHANCED *RHIZOCTONIA* DAMPING-OFF IN COTTON. **Becker, J. O., and J. Smith Becker**. Department of Nematology, University of California, Riverside, CA 92521.

Cotton damping-off, caused by *Rhizoctonia solani*, can result in substantial stand loss and sublethal attacks of the fungus may cause crop stunting. Root-knot nematodes (*Meloidogyne incognita* race 3) are known to enhance damping-off, even at low population densities. In growth chamber and small plot trials with *Gossypium hirsutum* cv *Sierra RR Acala*, the potential benefits of a nematicidal seed coating were evaluated in the presence of both the fungus and the nematodes. In *R. solani*- and *M. incognita* race 3-infested sandy soil, seed coating with 0.15 mg abamectin/seed mitigated early *M. incognita* attack and increased root length compared to non-protected seedlings. When applied with a fungicides/insecticide combination (1.66 g azoxystrobin, 0.28 g fludioxonil, 0.83 g mefenoxam per 100 kg seed and 0.32 mg thiamethoxam per seed), the abamectin-containing seed coating resulted in the best seedling stand and largest root systems compared to the non-treated check or to a fungicides/insecticide seed coating without the nematicide. These results demonstrate that potential benefits of a nematicidal seed coating are not limited to crop protection against the target nematode pests.

A GENETIC APPROACH TO UNDERSTANDING *HETERODERA GLYCINES* VIRULENCE. **Bekal, S., T. L. Niblack, M. E. Hudson, L. L. Domier, and K. N. Lambert**. Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

Heterodera glycines, the soybean cyst nematode (SCN), is the most devastating pathogen of soybean in the USA. SCN is a very diverse and adaptable nematode capable of reproducing on what would normally be considered SCN resistant soybean. This phenomenon is also known as resistance breaking and such nematodes are referred to as virulent. The build up of virulent SCN populations is an ever-growing threat to soybean production since it renders the major method of SCN control, nematode resistant plants, ineffective. If the underlying genetics of SCN virulence could be understood, then virulent SCN populations could be monitored and crop rotation strategies could be devised to prevent the spread and accumulation of virulent SCN. To this end, we have initiated a project to map-base clone a recessive SCN virulence gene that allows the nematode to grow on the resistant soybean cultivar Hartwig. In this project we are identifying DNA sequence polymorphisms between virulent and avirulent inbred SCN biotypes. These polymorphisms are being converted into molecular markers, which in turn are being mapped to generate a high resolution SCN genetic linkage map and to find markers linked to the SCN virulence loci. Some of our molecular markers are being generated from bacterial artificial chromosome end sequences, thus this project will also facilitate the ultimate integration of our SCN genetic map to a future SCN physical map.

OCCURRENCE AND DISTRIBUTION OF *MELOIDOGYNE* SPP. IN NEW ZEALAND. **Bell, N. L.[1], K. W. L. Knight[2], C. F. Mercer[3], F. Shah[4], D. Sturhan[5], L. T. Aalders[1], G. W. Yeates[6], R. N. Watson[1], J. W. Marshall[4] and G. D. Page.[7]**
[1]AgResearch Ltd, Ruakura Research Centre, Private Bag 3123, Hamilton, New Zealand; [2]Biosecurity New Zealand, Gerald Street, P.O. Box 24, Lincoln, New Zealand; [3]AgResearch Grasslands, Tennent Drive, Private Bag 11008, Palmerston North, New Zealand; [4]Crop and Food Research, Canterbury Agriculture & Science Centre, Private Bag 4704, Christchurch, New Zealand; [5]c/o Biologische Bundesanstalt, Toppheideweg 88, 48161 Münster, Germany; [6]Landcare Research, Private Bag 11052, Palmerston North, New Zealand; [7]AgriQuality, PO Box 41, Auckland, New Zealand.

Information on the nationwide distribution of plant parasitic nematode species can be used to identify habitat, host preference and climatic limitations; and to predict spread and potential pest problems. Much of the knowledge about the identity and distribution of economically important nematodes in New Zealand is either spread across a number of publications or unpublished in individual agencies, making an overview difficult. This poster presents an inter-agency project which brings together information on the New Zealand-wide distribution of all the *Meloidogyne* species reported and confirmed from New Zealand in the nearly 100 years since they were first recorded in this country. The species considered to occur in New Zealand are: *M. ardenensis, M. arenaria, M. fallax, M. hapla, M. incognita, M. javanica, M. naasi, M. trifoliophila*, along with three currently undefined species. Morphology of J2 specimens, root galls and molecular techniques have been used to determine species. *Meloidogyne* spp. are widely distributed throughout the country; they are common in arable and other cultivated soil but are also present in non-agricultural soils. Data from published papers are incorporated with unpublished data from previous surveys of, and studies conducted in, agricultural sites. In recent surveys *M. naasi* was found in 25% of the pasture and 40% of the barley fields sampled, *M. fallax* in 20% of potato crops sampled (with a much greater prevalence in North than South Island sites); and *Meloidogyne* spp. from 25% of non-agricultural sites sampled. The undescribed species were found from forests, where only 7% of samples contained *Meloidogyne* nematodes. As well as the location of *Meloidogyne* species, data will be collated on hosts and soil type at each location, where these are known.

NEMATODE INVASIONS: COURTESY OF NATURE OR HUMANITY? **Bernard, E. C.** Entomology and Plant Pathology, Ellington Plant Science, The University of Tennessee, Knoxville, TN 37996-4560.

Human-aided movement of nematodes accelerates the dispersal of nematodes to previously unoccupied locations, but does not override the basic ability of individual species to become established in a particular site. Most crop-damaging plant-parasitic nematodes are highly favored by soil turmoil for monocultured crops, which likely reduces nematode predators and parasites and promotes nematode reproduction, but these nematodes do not do well in stable sites. For instance, *Meloidogyne incognita* and *M. hapla* can be devastating in monocultured crop fields, but do not occur in adjacent forest. Instead, obscure root-knot species, some undescribed, are found there in low densities on perennial woody plants. Also, Hawaiian crop parasites have not been detected in surveys of nematodes in Hawaiian native forests. Thus, the origins of major plant-pathogenic nematodes may be savannahs or riparian zones where herds of ungulates periodically churn the soil; most of these nematodes would be dependent on accidental transportation to reach distant sites. Other nematodes have found ways to disperse long distances, apparently on their own. Evidence from newly vegetated sites (volcanic areas, glacial retreats) suggests that criconematid nematodes pioneer such sites, and thus must have much greater dispersal ability than previously recognized. *Paratylenchus* spp. are the most typical plant parasites found in the rhizospheres of sparse vegetation growing around Kilauea crater, on the upper slopes of Mauna Loa, and on newly exposed land in front of Columbia Glacier. Similarly, *Criconema longulum* has been reported from mountaintops in Switzerland, the Adirondacks, and Alaska. However, despite the demonstrated dispersability of some taxa, there remain many anomalies. For instance, *Merlinius adakensis* is reported from Alaska and Pakistan, and *Helicotylenchus pseudorobustus* is distributed in North America and Nigeria. Although the taxonomic characters of these populations may "fit," their distributions do not make sense biogeographically. Diagnostic molecular tools are sensitive enough to provide valuable information on the relatedness of distant populations, and can help solve many knotty questions regarding speciation, dispersal, and distributions.

NEMATODE COMMUNITIES OF SMALL POOLS IN AN AGRICULTURAL LANDSCAPE. **Bert, W., M. Messiaen, F. Hendrickx, J. Manhout and G. Borgonie.** Department of Biology, Ghent University, Gent, Belgium.

A total of 14 pools from 5 regions in North-West of Belgium were studied; three pools within each region were selected along a maximal gradient of surrounding agricultural land-use intensity. The total nematode density (9–411 ind./10 cm^2 per pool), and especially the number of species (4–12 species per pool) was low in this study. In total, seventeen genera of free-living benthic nematodes, belonging to 15 families, were identified. *Tobrilus gracilis* (Bastian, 1865) and *Eumonhystera filiformis* (Bastian, 1865) were the most common species; they were found in respectively 13 and 12 of the 14 sampled water bodies. The genera *Tobrilus* and *Eumonhystera* jointly comprise 77% of the total nematofauna. Consequently, the investigated water bodies were dominated by deposit feeding *Monhysteridae* and/or by chewing *Tobrilidae. Diplogasteridae* and *Rhabditidae*, normally related with eutrophic habitats, were virtually absent. A straightforward pattern of environmental variables and nematode composition versus agricultural land use was not observed. Sets of environmental variables were statistically selected that best explained the among pool variation of total density, diversity, the feeding-types composition and the individual density of the six most important species. It was demonstrated that morphologically very similar species can show highly different ecological traits. However, relating the presence or density of individual nematode species to environmental factors remains difficult. The overall high level of eutrophication, the presence of a substantial mud layer and a low sediment oxygen level are put forward as the main factors explaining the observed low density and diversity.

TYLENCHINA (NEMATODA) PHYLOGENY: EVIDENCE FROM MOLECULES AND GONODUCT MORPHOLOGY. **Bert, W., P. Weekers, and G. Borgonie.** Department of Biology, Ghent University, Gent, Belgium.

Although the parasitic biology of certain plant parasitic *Tylenchina* is already well documented, a broader understanding of the evolution of the mode and direction of plant parasitism has been largely speculative. Plant parasitic nematodes share functional similarities regarding feeding, but many similarities in feeding and associated body structures result from parallel evolution. Although molecular-based phylogenies are advancing, *Tylenchina* phylogenies based on 18S rRNA are not fully resolved (Baldwin et al., 2004). A more comprehensive sampling and exploration of additional informative morphological characters is required. For the morphological part of this study, we have investigated the cellular architecture of expelled gonoducts; this was done for more than 100 different *Tylenchina* species. Molecular data of 40 new and 50 known 18S rDNA sequences, were analyzed by the Akaike Information Criterion (Modeltest), Maximum Likelihood and Maximum Parsimony methods (PAUP*), and Bayesian Inference (MrBayes). Long-branch attraction is recognized as a major problem, and its possible influence on previous phylogenies is discussed. Our results support common ancestry for the morphological disjunct *Tylenchomorpha* and *Cephalobomorpha*; the morphology of oviduct and spermatheca of the *Cephalobomorpha* is apparently similar to what is known for the *Tylenchoidea*. Several well resolved more terminal clades are also maintained by gonoduct data, e.g. the *Meloidogyne* spermatheca is distinctive from any other nematode genus, and

the cellular architecture of the criconematid uterus is clearly different from that of the other tylenchs. On the other hand, our molecular analyses indicate the polyphyletic nature of the families *Tylenchidae, Belonolaimidae* and *Pratylenchidae*. It is concluded that the cellular gonoduct architecture, besides being diagnostically informative, provides useful additional characteristics to infer phylogenetic relationships. Yet, additional taxa, genes and morphological data are necessary to resolve particular deep branches within the *Tylenchomorpha*.

BIOLOGICAL CONTROL POTENTIAL OF A DIPLOGASTERID PREDATOR, *MONONCHOIDES GAUGLERI*. **Bilgrami, A. L., C. Brey and R. Gaugler**. Department of Entomology, Rutgers University, New Brunswick, NJ 08901.

Diplogasterid predators possess several beneficial biological control traits. They reduced plant-parasitic nematode population significantly under laboratory and in pot experiments. However, their efficacy has not been tested against plant-parasitic nematodes under field conditions. Present study was performed to assess biological control potential of *Mononchoides gaugleri* against naturally occurring nematodes in a turf grass field. Experiments were made in 10 cm in diameter and 10 cm deep micro plots. Two thousand predators were inoculated per micro plot. Plots without predators served as negative control. Experiments were replicated 13 times. Observations were made after 30 days. Random sampling was conducted to isolate and identify nematode species occurring in the field. *Tylenchorhynchus*, *Ditylenchus*, *Hoplolaimus*, *Helicotylenchus*, *Tylenchorhynchus*, *Aphelenchoides*, *Dorylaimus*, *Mesodorylaimus*, *Eudorylaimus*, *Acrobeloides*, *Mononchus*, *Monhystera*, *Thornenema* and *Pratylenchus* were identified and their populations recorded. *Mononchoides gaugleri* killed significant number of tylenchid nematodes reducing them from 537/microplot to 382/microplot, a significant reduction of 28.86%. Bacterial feeders also declined from 61.0% to 41.5% at the end of 30 days. There was little reduction in dorylaim nematode population as a result of predation. Predatory nematodes reduced to 15–25%/microplot at the end of 30 days. Study suggests that predatory nematodes reduced plant-parasitic and bacterial feeding nematodes but failed to persist for long durations under field conditions. Reduction in parasitic and bacterial feeding nematodes could be achieved with the application of predatory nematodes in the field. However, further studies on the persistence and mass culturing of predatory nematodes are needed.

CONSERVATION OF LUMINESCENCE GENES IN *PHOTORHABDUS LUMINESCENS*. **Blackburn, D.[1], J. M. Chaston[2], and B. J. Adams.[1]** [1]Department of Microbiology and Molecular Biology, Brigham Young University, Provo, UT 84602; [2]Department of Bacteriology, University of Wisconsin-Madison, Madison, WI 53706.

Photorhabdus luminescens, a bacterial endosymbiont of the insect pathogenic nematode *Heterorhabditis bacteriophora*, is the only known terrestrial bioluminescent bacterium. Though it is unknown why a soil bacterium would need to produce light, it has been suggested that bioluminescence may be used to attract insects or to rid the bacterium of toxic oxygen radicals through the luminescent reaction. This project looks at possible selective mechanisms for the conservation of genes involved in *Photorhabdus* bioluminescence. Random gene knockouts were done to find genes that are associated with variation in luminescence and to obtain mutant strains for use in testing whether inactivation or reduction in luminescence affects the ability of the bacterium to symbiose with its nematode host. Comparisons of light emittance between wild type *P. luminescens* and selected mutants were performed using a flow cytometer. Results show a significant decrease in luminescence of the mutant strains. The area surrounding each disrupted gene was sequenced and blasted against the existing genome sequence to determine which genes were interrupted. In vitro growth assays show that bacteria lacking the luminescence genes do not promote nematode growth. In vivo symbiosis assays shed additional light on the relationship between bioluminescence and symbiosis.

CHARACTERIZATION OF SOIL MICROBIAL COMMUNITIES USING 16S rDNA RIBOSOMAL SEQUENCE TAGS. **Blas, A. L.[1] Q. Yu,[1] B. Sipes,[2] S. C. Miyasaka[3] and R. Ming.[4]** Departments of [1]Molecular Biosciences and Bioengineering, [2]Plant and Environmental Protection Sciences and [3]Tropical Plant and Soil Sciences, University of Hawaii at Manoa, Honolulu, HI 96822; [4]Department of Plant Biology, University of Illinois at Urbana-Champaign, Urbana, IL 61801.

Development of culture-independent, microbial community analysis over the last two decades has greatly enhanced our knowledge of the complexity of soil microbial communities. Assessment of the impact of pathogen control systems on soil microbial communities is requisite for the appropriate evaluation of such systems. In this study, we use Serial Analysis of Ribosomal Sequence Tags (SARST) to characterize changes in the soil microbial community in a field subjected to green manure treatment for the control of root-knot nematode (*Meloidogyne javanica*) in taro (*Colocasia esculenta*). Total soil microbial DNA was isolated from 1 or 10 g soil samples and verified for PCR suitability using primers (P8f and P1492r) targeting the bacterial 16S rDNA gene cluster. PCR amplicons were cloned into TOPO-TA vector and forty-eight randomly selected clones were sequenced. Resulting BLAST searches returned sequence homology primarily to uncultured soil bacteria. Utilizing published genomic sequence data of the hypervariable V1 region of the bacterial small subunit rRNA gene (SSU rDNA) we will determine the changes in soil microbial diversity and abundance of a taro field in Hilo, Hawaii.

DEVELOPMENT AND ISOLATION OF A STABLE PRIMARY PHASE VARIANT OF *PHOTORHBDUS LUMINESCENS.* **Bliss T.,[1] D. Blackburn,[1] C. Bailey,[1] A. Smith,[1] J. M Chaston,[2] and B. J. Adams.[1]** Department of Microbiology and Molecular Biology, Brigham Young University, Provo, UT 84602; [2]Department of Bacteriology, University of Wisconsin-Madison, Madison, WI 53706.

The entomopathogenic nematode *Heterorhabditis bacteriophora* (with its bacterial symbiont, *Photorhabdus luminescens*) has become an effective pest control agent used extensively throughout the world. Entomopathogenic efficacy is dependent on the life-stage of the nematode. Preliminary observations in our lab suggest the phases of the bacterial symbiont and the life-stages of the nematode are correlated. When the bacteria are in primary phase, associated nematodes are in a potent infective juvenile stage—able to invade insects, develop and reproduce. When the bacteria switch to the secondary phase, the nematodes are unable to recover from the dauer stage and cannot reproduce. This phenomenon is problematic for the industrial production of *H. bacteriophora* because nematodes associated with secondary phase bacteria cannot be sold as effective bio-insecticides. Moreover, scientific studies using *H. bacteriophora* or *P. luminescens* are often hindered by the sudden, irreversible switching of the bacteria from primary to secondary phase. Global, irreversible, and reproducible switching to secondary phase has only been achieved after four 96-hour subcultures in low osmolarity broth. Here we aim to isolate a stable primary phase mutant of *P. luminescens* TT01 by random single gene knockout with a suicide plasmid from *E. coli* S17–1 pLOF Km. Spontaneous rifampin resistant strains of TT01 were incubated with the plasmid carrying *E. coli* and allowed to conjugate. Mutants were screened for plasmid uptake and then subjected to prolonged low-osmolarity sub-culturing. The presence (primary phase) or absence (secondary phase) of luminescence was used as a preliminary screen for phase variance. Of 1,698 mutants, <100 exhibited luminescence after sub-culturing. The six brightest mutants were assayed to determine effectiveness in conferring pathogenicity to axenic *H. bacteriophora* juveniles. More extensive screening definitively characterized phase phenotype. Genetic studies currently underway aim to elucidate the phase variation regulatory element(s) in these six mutants.

USING ENTOMOPATHOGENIC NEMATODES AS MODEL ORGANISMS IN HIGH SCHOOL EDUCATION MODULES AND THE BENEFITS OF FIELD TESTING. **Bliss T.,[1] M. K. Anderson,[2] M. Jett,[2] D. Yourick,[2] and B. J. Adams.[1]** [1]Department of Microbiology and Molecular Biology, Brigham Young University, Provo, UT 84602; [2]Division of Pathology, The Walter Reed Army Institute of Research, Silver Spring, MD 20910.

Heterorhabditis bacteriophora is a model organism that can be effectively manipulated by high school students in a laboratory module setting to help students learn and apply the scientific method and elements of good experimental design. In addition, field testing of modules is effective at creating early student-scientist interaction and establishing collaborations between high school science teachers and professional scientists as a way of addressing problems in science education in America today. In 2004, for example, more than 25% of 7^{th}–12^{th} grade science teachers did not hold a major or minor degree in science. With the rising numbers of less-qualified science teachers in our school systems, it is increasingly important that scientists strive to become active educational resources. Field testing of scientific modules in local schools is an excellent medium for creating such scientist-teacher interaction. Moreover, the use of nematodes as a model organism works toward the Society of Nematologists' goal to "advance the science of nematology in both its fundamental and economic aspects." We have developed a nematode-based experimental module with the objectives of helping students (1) improve the ability to think systematically through a problem by applying the scientific method; (2) learn the basic methods of working with entomopathogenic nematodes (EPN) in the laboratory; (3) apply simple concepts of experimental design and data analysis; (4) understand the biological, ecological and economical importance of EPN; and (5) improve skills in research presentation. Preliminary field testing at the Walter Reed Army Institute of Research Summer Internship Program shows students can successfully formulate hypothetical questions regarding EPN and then design experimental protocols to answer those questions. Further field testing in Utah Valley high schools confirms the module's ability to accomplish the five objectives listed above.

EGG BIOLOGY, REPRODUCTION AND ECOLOGY OF *ROTYLENCHULUS RENIFORMIS* ISOLATES. **Bruce, J. M. and E. C. McGawley**. Department of Plant Pathology and Crop Physiology, Louisiana State University, Baton Rouge, LA 70803.

Rotylenchulus reniformis (Rr) is a serious nematode pathogen. Differences in reproduction, cultivar response and host preference among geographic populations of the nematode have led to the hypothesis that distinct pathotypes exist in nature. In 2004, new populations of the nematode were obtained from AL, AR, GA, HI, LA, MS, PR, and TX, and 10–20 single egg mass (SEM) isolates 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 SEM isolates from the same geographic population. SEM isolates AL, GA, LA1, LA2, MS, PR, and TX were used to evaluate numbers of egg masses per 0.1 g root, numbers of eggs per egg mass, egg mass diameter and egg size. After 39 days at 24°C, significant differences were seen in numbers of egg masses per 0.1 g dry root. MS, AL and LA1 had greater numbers of egg masses (34.5, 26.5, and 26.2, respectively) than GA, TX2 and PR (5.1, 3.6 and 2.7). There were no significant differences in egg mass sizes

among isolates but there were significant differences in egg length and width. A microplot trial to further characterize the differences between geographic populations was conducted with Deltapine 434 cotton using SEM isolates AL, AR, GA, HI, LA1, LA2, MS, PR, and TX. The 161-day test corroborated the results found in the lab: again, MS, AL and LA1 had more egg masses per 3 g of root (413, 231, and 229) than TX2, GA, LA2 and PR (206, 178, 159, and 33). Two additional isolates used, AR and HI, fell near the top and bottom, with 284 and 109 egg masses per 3.0g root sample. A follow-up study using six populations (AL, GA, LA1, MS, PR, and TX) from the microplot was conducted in the lab to evaluate infectivity over time. Significant differences in infectivity rate were seen after 15 days, but not after 5 or 10 days. Further studies are in progress to characterize the effects of temperature on reproduction and infectivity.

ENGINEERING *COFFEA ARABICA* FOR RESISTANCE TO *MELOIDOGYNE KONAENSIS* USING CYSTEINE AND SERINE PROTEINASE INHIBITORS. **Cabos, Roxana,[1] B. S. Sipes,[1] D. P. Schmitt,[1] H. J. Atkinson,[2] and C. Nagai.[3]** [1]Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI, USA, [2]Centre for Plant Biochemistry and Biotechnology, University of Leeds, Leeds, UK, [3]Hawaii Agriculture Research Center, Aiea, HI, USA.

Coffea arabica cv. Typica was transformed with a modified cystatin gene from rice, OcIΔD86, which successfully reduced the population of plant-parasitic nematodes when engineered in other crops such as banana and potato. This gene, alone or paired with a cowpea trypsin inhibitor, was introduced into *C. arabica* somatic embryos using *Agrobacterium tumefaciens* or particle bombardment. The resistance genes were placed under the control of the constitutive CaMV35S promoter or the tubulin promoter which expresses preferentially in root tissue. Over 2,500 secondary somatic embryo lines remained after a 7-month selection period on 30 mg/L geneticin sulfate. Regeneration occurred in 44 of the selected lines. Genomic DNA analyzed by PCR confirmed the existence of the inserted gene in 18 of the regenerated lines. RT-PCR of roots and leaves demonstrated that the OcIΔD86 transcript was being produced in 12 lines. A biological assay was performed in the laboratory to test the effectiveness of the proteinase inhibitors against *Meloidogyne konaensis*, a highly pathogenic root-knot nematode found in the Kona and Ka'u regions of the Big Island of Hawaii. The experiment was carried out in 15.5-cm-diameter clay pots containing sterilized soil:silica sand mix for a period of 347 days. Depending on the size of the plant, each pot was inoculated with 4,000, 2,000, or 1,000 *M. konaensis* eggs. The nematode reproduction factor (Rf) of 91 plants representing 41 transformed lines was compared to the Rf of the wild-type controls. The quantity of cystatin mRNA produced in the roots during infection by *M. konaensis*, as determined by qRT-PCR, was compared to plant growth and nematode reproduction on the transgenic lines. This management system may be effective for the control of *M. konaensis* on coffee.

ASSESSING THE FITNESS CONSEQUENCES OF PARASITE INFECTION DECISIONS. **Campbell, J. F.** USDA ARS GMPRC, Manhattan KS 66502.

Important 'decisions' confront all animals as they search for and assess resources, and the outcomes of these 'decisions' may be strong agents of selection on life history strategies. The relative costs and benefits have been studied in many systems, and have led to the development of some basic theories in behavioral ecology. For example, considerable theoretical and empirical research has been done on insect parasitoid host acceptance. However, there are likely to be substantial differences between the costs and benefits of infection for a parasitoid female depositing eggs, compared to a parasite infective stage. In the last decade, there has also been a considerable amount of research done on how parasite infection influences host behavior, and whether or not these changes are adaptive for the parasite. Receiving less attention, however, has been the influence of variation in host quality on the infection behavior of parasite infective stages and the fitness consequences of host preference for the parasite. As the talks in this session have illustrated, parasite infection biology is an interesting and challenging avenue of research, which also has a great deal of potential applied application. In this presentation, research on parasitic nematode infection behavior will be reviewed and placed in a broader behavioral ecology context. Emphasis will be placed on the insights gained from the integration of mechanistic and functional approaches to understanding host infection.

EFFICACY OF ABERMECTIN ON *ROTYLENCHULUS RENIFORMIS* NEMATODE DEVELOPMENT ON COTTON IN VARIOUS SOIL TYPES. **Carpenter, D., K. S. Lawrence, and T. V. Boozer**. Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849.

Variations in the efficacy of the seed treatment abermectin have been observed in *R. reniformis* infested cotton fields across the southeast. This study examines the rate of development of the reniform nematode over time as effected by abermectin treated and non treated cotton seed planted in difference soil types. Two replicated greenhouse trials were conducted at the Plant Science Research Center at Auburn University. Root samples were processed at 3, 6, 9, 14, 19, 25, and 35 days after inoculation (DAI) to determine life stage development, number of nematodes per gram of root and

numbers of egg masses. The life stages attained by *R. reniformis* were slowed by the abermectin seed treatment as compared to the untreated control in the Decatur silty loam soil. Forty-seven percent of the population of the reniform in the untreated control treatment advanced to stage B, the open C shape with an enlarged vulva region at 6 DAI as compared to only 19% in the abermectin. The typical reniform shaped females without egg masses were observed at 9 DAI in the untreated control but not until 14 DAI in the abermectin treatment. The mature reniform-shaped females with egg masses were observed at 25 DAI in both treatments. The highest percentage of mature egg-laying females and secondary infections were observed in both abermectin and control treatments at 35 DAI. The initial life stage development of the reniform nematode appears to be delayed in the presence of abermectin.

LOW TEMPERATURE SEM AND MOLECULAR PHYLOGENETICS OF THREE DIPLOSCAPTER POPULATIONS. **Carta, L.,[1] A. Skantar,[1,2] and E. Erbe.[3]** USDA, ARS, [1]Nematology Laboratory, [2]Molecular Plant Pathology Laboratory, and [3]Soybean Genomics and Improvement Laboratory, Beltsville, MD 20705.

Two genetically distinguishable soil populations of *Diploscapter cf. lycostoma* that lack males are described and compared with a population of *Diploscapter coronatus*. Low temperature SEM observations showed for the first time lateral lip flaps classically known as laciniae with filopodia-like extensions. These appeared to be independently deformable and either contacted the cuticle of other nematodes, stretched inwardly into the stoma opening, or interdigitated with one another. The face views of lateral stoma walls of *D. cf. lycostoma* populations are centrally incurved relative to *D. coronatus* , and internal hamuli margins above the stoma have distinct profiles in the two morphospecies. All populations had enlarged tail annules, and sperm within offset spermathecae. Procorpus length, body width/basal bulb width, gonad lengths, and phasmid position on the tail showed slight differences between the two nominal species. Small subunit (SSU) rDNA parsimony and Bayesian phylogenetic trees failed to demonstrate separation of these two Diploscapter species, and their position relative to *Caenorhabditis* or *Oscheius myriophila (Rhabditidae)* was not resolved. However Hsp90 trees resolved Diploscapter independently of a highly supported clade of *Caenorhabditis* and *Oscheius* . Intron number in this Hsp90 segment varied from one to five, with the first and fourth intron showing some positional homology across taxa. Diploscapter and parasitic *Tylenchida* had very similar intron positions. The phylogenetic significance of Hsp90 and its chaperone function for cytoskeletal proteins are briefly discussed.

HOW A GASTROINTESTINAL NEMATODE REGULATES THE SPATIO-TEMPORAL DYNAMICS OF ITS HOST. **Cattadori, I. M.,[1] P. J. Hudson,[1] and D. Haydon.[2]** [1]Center for Infectious Disease Dynamics, Penn State University, University Park, PA 16802; [2]Division of Environmental and Evolutionary Biology, University of Glasgow, Glasgow G12 8QQ Scotland.

The intensity of gastrointestinal parasites within a host population is the result of two important components, host susceptibility and host exposure to infective stages. Susceptibility is modulated by host immunity, hormones or physiological conditions, while exposure is influenced by the survival, development and transmission of infective stages, which in turn are affected by environmental conditions. Consequently, weather changes have a profound and direct influence on parasite transmission but they can also act indirectly on parasites through their influence on host susceptibility, by altering food availability, host growth rate and reproduction. We have investigated the dynamics of the nematode *Trichsostrongylus tenuis* in red grouse in England using time series on host and parasite abundance from more than 100 host populations with details on habitat and climatic variables for every population. Our aim was to examine how *T. tenuis* interacts with the host and affects the dynamics of red grouse temporally and spatially. We have shown that *T. tenuis* reduces female breeding success and destabilizes population dynamics, which results in cyclic fluctuations in abundance of 3 to 8 years period. We have also shown that extreme, unpredictable weather conditions influence the development and transmission of the nematode, and in so doing synchronize fluctuations in grouse populations over wide areas. These findings illustrate that parasite transmission is linked over large geographical areas by extreme weather events that can potentially lead to large scale infection outbreaks and dramatic declines in host abundance.

CAENORHABDITIS ELEGANS AND *HELIX ASPERSA*. **Chen J., E. E. Lewis, J. R. Carey, and E. P. Caswell-Chen.** Departments of Nematology and Entomology, University of California, Davis, CA 95616.

We have recovered *C. elegans* from the garden snail *Helix aspersa*. A behavioral assay, based on an attraction index, was used to quantify attraction of *C. elegans* dauer juveniles to the snail. The treatments included: a dead snail, an *E. coli* OP50 food plug, or a blank agar plug. The bioassay arena was a Petri plate filled with NGM and sectors delineated to allow monitoring of worm movement. The treatment was placed at the edge of the plate, worms at the center, and over time the numbers of worms in each sector counted. A higher positive index number indicates more movement toward the stimulus, and a negative index number indicates movement away. The attraction index was calculated at 9 time intervals, approximately 3 min each. The attraction indices for snail, *E. coli*, and blank agar were 18.8, 3.5, and 0.7 at Interval 3, respectively, and 25.3, 14.1, and -0.5 at Interval 9. When *C. elegans* dauers were placed in snail food and fed to the snails, the worm was extracted from the feces of the snails. The natural life history of *C. elegans* is discussed.

EFFECT OF COVER CROPS ALFALFA, RED CLOVER, PERENNIAL RYEGRASS, AND RYE ON SOYBEAN CYST NEMATODE POPULATION AND SOYBEAN AND CORN YIELDS IN MINNESOTA. **Chen, S.[1,2], D. L. Wyse,[3] G. A. Johnson,[2,3] P. M. Porter,[3] M. J. Haar,[4] S. R. Stetina,[1,4,5] D. R. Miller,[2] K. J. Betts,[3] and L. D. Klossner.[4]**
[1]Department of Plant Pathology, University of Minnesota, Saint Paul, MN 55108; [2]Southern Research and Outreach Center, University of Minnesota, Waseca, MN 56093; [3]Department of Agronomy and Plant Genetics, Saint Paul, MN 55108; [4]University of Minnesota Southwest Research and Outreach Center, Lamberton, MN 56152. Current address: [5]USDA ARS Crop Genetics and Production Research Unit, Stoneville, MS 38776.

A field experiment was carried out to evaluate alfalfa (*Medicago sativa*), red clover (*Trifolium pratense*), perennial ryegrass (*Lolium perenne*), and rye (*Secale cereale*) as cover crops for their effects on soybean cyst nematode (SCN, *Heterodera glycines*), and soybean (*Glycine max*) and corn (*Zea mays*) yields in Waseca, Lamberton, and Rosemount, MN. Alfalfa, red clover, and perennial ryegrass were interseeded in soybean at 0 or 2 wks after planting soybean in 2002 and 2004, and killed with herbicide prior to planting corn in 2003 and 2005. Rye was planted after harvesting corn and killed in the following spring. SCN-resistant and susceptible soybean cultivars were planted in 2002 and SCN-susceptible soybean was planted in all plots in 2004. Nematode egg population densities were determined at planting, midseason, and harvest in the soybean years, and at planting and harvest in the corn years. As expected, SCN-susceptible soybean supported greater egg population density than SCN-resistant soybean. Reduction of egg population density by red clover and alfalfa was observed in some occasions, probably mainly due to reduced soybean growth, but the effects were inconsistent. Perennial ryegrass did not affect SCN in most cases. There was no effect of rye on egg population density. SCN-resistant soybean produced higher yield than susceptible soybean at all sites in 2002. Compared with susceptible cultivar, the resistant cultivar in 2002 increased yield of soybean in 2004 at Rosemount, but not at the other two sites. The cover crops interseeded with soybean not only reduced soybean yield but also lowered corn yield in the following years in most cases. The results suggest that red clover and alfalfa as cover crops may reduce nematode populations, but to avoid yield loss due to competition an even later planting date for the cover crops may be more appropriate for use in the soybean-corn rotation in Minnesota.

TILLAGE AND CROP SEQUENCE EFFECTS ON *HETERODERA GLYCINES* AND SOYBEAN AND CORN YIELDS. **Chen**, **S.** Southern Research and Outreach Center, University of Minnesota, Waseca, MN 56093.

Conservation tillage has been increasingly used in the Midwest United States. A long-term (5–10 years) effect of tillage practices on the soybean cyst nematode (SCN), *Heterodera glycines*, and soybean and corn yields in the corn-soybean rotation was studied in two fields near New Richland and Waseca, Minnesota, USA. The experiments were arranged in a split plot design with tillage treatments as main plots and crop sequences as subplots. Tillage treatments were no-tillage (NT) and conventional tillage (CT), which was moldboard plowing after harvesting corn and chisel plowing after harvesting soybean in fall, and field cultivation in spring prior to planting. The crop sequences were six (Waseca) or nine (New Richland) combinations of SCN-susceptible 'Sturdy', SCN-resistant 'Freeborn' (PI 88788 resistance source) and 'Pioneer brand 9234' (Peking resistance source) soybean rotated annually with the nonhost corn. Tillage did not affect SCN egg population density at New Richland and had only minimum and inconsistent effects on SCN at Waseca, confirming that tillage is not an option solely for managing SCN population in the northern climate and soil conditions. However, CT resulted in up to 370 kg/ha greater soybean yield than NT at Waseca. Growing SCN-resistant cultivars was effective in the corn-soybean rotation for managing SCN and minimizing yield loss to SCN. Resistant soybean not only reduced SCN population density and increased soybean yield in the year when it was grown, but also resulted in a smaller inoculum population density and increased yield of susceptible and resistant soybean cultivars in the following soybean years. Pioneer brand 9234 was more effective than Freeborn in reducing SCN population density and increasing soybean yield in the following years. Corn yield was reduced (up to 2,221 kg/ha) by NT as compared with CT in Waseca, but there was no tillage effect on corn yield in New Richland.

TIME COURSE OF PR-1 GENE EXPRESSION IN PINEAPPLE FOLIAR APPLIED WITH ACIBENZOLAR-S-METHYL AND ITS EFFECTS TO REPRODUCTION OF RENIFORM NEMATODES. **Chinnasri, B.,[1] B. S. Sipes,[1] T. Borsics,[2] and D. A. Christopher.[2]** [1]Departments of Plant and Environmental Protection Sciences, [2]Molecular Biosciences and Bioengineering, University of Hawaii at Manoa, Honolulu, HI 96822.

Systemic acquired resistance (SAR) is an intrinsic ability of plants to defense themselves against pathogens and characterized by the coordinated expression of pathogenesis-related genes (PR-genes). In our study, the expression of PR-1 gene (SAR marker) 1, 7, 14, or 21 days following application by acibenzolar-s-methyl (a potent SAR inducer) and the effect of the SAR induction on *Rotylenchulus reniformis* were investigated. Pineapple crowns were planted onto 15-cm-diameter clay-pots. One month later, pineapple plants were foliar applied with a solution of 100 mg acibenzolar/L of water (10 ml per plant). One, 7, 14, or 21 days post application, pineapples were uprooted, washed, and RNA extracted. First strand cDNA was generated and a regular PCR was performed using the two primers specific to pineapple PR-1 gene. A 266 bp band, indicative of PR-1 induction, was present in acibenzolar-treated pineapple up to 21 days post treatment. To determine

the effect on nematodes, a separate set of pineapples were grown and treated with acibenzolar at 0, 50, 100 or 200 mg/L of water. One, 7, 14, or 21 days post application, 40,000 eggs of *R. reniformis* were inoculated onto pineapples. The result showed that reproduction of nematode on pineapples treated with 100 or 200 mg/L was 55% lower than that on pineapples treated with 0 or 50 mg acibenzolar/L. Nematode reproduction on pineapples treated with the same concentrations but inoculated at different times was not significantly different ($P > 0.05$). SAR was induced in pineapple as early as 24 hours after acibenzolar application, remained activated for at least 21 days post application, and reduced nematode reproduction at rates above 100 mg/L.

SUPPRESSION OF ROOT-KNOT NEMATODE-FUNGAL DISEASE INTERACTIONS ON TOMATO AND CANTALOUPE WITH THE USE OF ABAMECTIN SEED TREATMENT. **Cochran, A.,[1] L. Payan,[2] and D. Hofer.**[1] [1]Syngenta Crop Protection AG, Basel, Switzerland; [2]Syngenta Crop Protection, Western Regional Technical Center, Visalia, CA 93292.

Nematode-fungal disease interactions are known to occur in many agricultural crops including fruiting vegetables such as cantaloupe and tomato. Abamectin seed treatment, brand name Avicta (Syngenta Crop Protection AG), has shown good early season control of root-knot nematode, *Meloidogyne incognita*, on these crops. In addition, mefenoxam, fludioxonil, and azoxystrobin fungicide seed treatments have shown efficacy against many soil borne pathogens. Greenhouse studies conducted in 2004 with cantaloupe, and in 2005 with tomato showed that when nematode eggs were co-inoculated with a fungal disease causing agent (*Fusarium* spp.), the nematicide/fungicide seed treatment significantly improved the protection of these crops against Fusarium wilt of tomato and melon, (*Fusarium oxysporum* f. sp. *lycopersici* and *melonis*, respectively) when compared to the fungicide treatment alone. In the case of melons, abamectin, in combination with mefenoxam and fludioxonil seed treatments, significantly reduced Fusarium wilt incidence by over 80%. In the case of tomato, evaluations at crop harvest demonstrated a ten fold decrease in Fusarium wilt incidence for the best abamectin-fungicide seed treatment combination. For both studies, Abamectin/fungicide seed treatment increased crop height (tomato), crop vigor (tomato and melon), plant weight (tomato), and fruit weight (tomato) along with suppressing Fusarium wilt (tomato and melon).

A MOLECULAR AND HOST RANGE ANALYSIS OF INTERSPECIFIC CROSSES OF *HETERODERA* SPECIES. **Colgrove, A. C., U. Reuter-Carlson, and T. L. Niblack.** Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

Cyst nematode species in the *schachtii* group are considered closely related and interbreeding has been reported. "Hybridization" of economically important species could have significance in field situations and laboratory research. Investigations of host-parasite interactions, such as host resistance to *Heterodera glycines*, could be aided by host-hybrid cyst nematode combinations involving model organisms (e.g., Arabidopsis, a poor host of *H. glycines*, but a known host of *H. schachtii*). A PCR-RFLP protocol that uses Fok1-digested ITS1 fragments was developed to distinguish *H. glycines* and *H. schachtii* (Szalanski, et al., 1997, JON 29: 255–267). Fragments unique to each species included a 252-bp fragment for *H. glycines* and a 181-bp one for *H. schachtii*. Controlled crosses were used to develop *Heterodera glycines-H. schachtii* hybrids. *Heterodera glycines*-infected soybean (Glycine max) and *H. schachtii*-infected sugarbeet (Beta vulgaris) were placed in hydroponic culture 10 days after infestation (DAI). Virgin females were transferred from roots to agar plates at 25 DAI. Males of the opposite species were added, and mating was observed. Egg-filled females were transferred to pots of Lespedeza striata 'Kobe', a common host. Cysts that developed on lespedeza roots were harvested and subjected to PCR-RFLP. The presence of both diagnostic bands confirmed hybrid progeny. In a separate test, cysts from isolates originally collected by George Bird from Michigan fields infested with both *H. glycines* and *H. schachtii* were subjected to marker and host range analysis. Preliminary results indicate that a mixture of genotypes, including naturally occurring hybrids, exist in these fields. Additional molecular and host range analyses are being conducted to further investigate controlled and natural hybridizations of *H. glycines* and *H. schachtii*.

BIOINFORMATIC ANALYSIS OF THE SOYBEAN CYST NEMATODE. **Craig, J., K. N. Lambert, S. Bekal, M. E. Hudson, T. L. Niblack, and L. L. Domier.** Department of Physiological and Molecular Plant Biology and Crop Sciences, University of Illinois, Urbana, IL 61801.

The genome of the soybean cyst nematode (SCN), *Heterodera glycines*, has been recently sequenced using a highly parallel microbead system developed by 454LifeSciences. In order to assess the coverage obtained by this sequencing project, we looked at the number of expressed sequence tags (ESTs) with matches in the dataset. Initially, the ESTs contained on the Affymetrix soybean array were used. We found that 95% of the SCN ESTs have 90% or greater identity to our dataset. The ESTs that did not match the SCN genome were analyzed further and half were determined to be non-SCN genes. This brought our coverage to over 98%. This assessment is currently being expanded to include all known SCN ESTs. The number of SCN ESTs within Genbank presently stands at 24,438. The sequencing of the SCN genome creates the opportunity to develop a unigene set largely devoid of contaminants. Current analysis indicates that there are

approximately 8,500 unique ESTs known. To further expand upon the known genes, the 454 SCN genome sequences have been tested for homology to other known proteins using blastx. This allows for the mining of new sequences not represented by the SCN EST database. The discovery of novel genes is an important step towards understanding how this nematode manipulates its host and will help lead to new methods of control.

BIOLOGICAL SOIL CRUST CAN AFFECT THE SPATIAL DISTRIBUTION OF NEMATODE COMMUNITIES NEAR DESERT PLANTS. **Darby, B. J., and D. A. Neher.** Plant and Soil Science Department, University of Vermont, Burlington, VT 05405.

Free-living nematodes are functionally important components of soil food webs that aid litter decomposition in arid lands by grazing on saprophytic microflora. Biogeochemical cycling is spatially heterogeneous, resulting in patches of greater soil fertility adjacent than between desert shrubs. Biological soil crusts also influence the distribution of nutrients through photosynthesis, nitrogen fixation, and chelating minerals. Previously, we found that nematode communities are more abundant, diverse, and ecologically mature when associated with late than early successional crust flora without plants. In the present study, we enumerated nematode communities in a distance gradient away from desert grasses and shrubs, associated with either early or late successional crust flora, and at two locations in the Colorado Plateau. We found that nematode abundance was greater adjacent than between plants for both grasses and shrubs, and that this pattern differed between plants that were associated with late or early successional stage crust flora. Bacterivorous nematode genera shifted in composition away from plants, but this pattern was similar between plant types and crust types. This suggests that the microclimate of plant canopies can influence the composition of bacterivores desert soils. However, the relative shift in stylet-bearing (herbivorous, fungivorous, predaceous, and omnivorous) nematodes away from plants depended on crust stage and plant form. This suggests that food source (vascular and non-vascular flora) can affect the composition of these nematodes that circumscribe desert plants. Our study confirms that nematodes are not only receiving nutrients from the rhizosphere, but also from biological soil crusts between rhizospheres.

ESTIMATING GENUS-SPECIFIC ANHYDROBIOSIS OF DESERT NEMATODES FROM FIELD SOILS. **Darby, B. J., and D. A. Neher.** Plant and Soil Science Department, University of Vermont, Burlington, VT 05405.

The ecological functioning of nematodes in arid land soils is restricted to periods of moisture that allow nematodes to remain active and mobile. Adaptation of nematodes to extreme hot and dry soils requires the ability of nematodes to become metabolically inactive (anhydrobiotic), often physically coiling. We modify an existing technique of arresting the uncoiling of nematodes in the field by sorting coiled and uncoiled specimens onto a microslide for identification to genus. In contrast to previous reports, we could identify coiled and uncoiled nematodes to genus with some prior familiarity of well-preserved specimens for reference. We demonstrate an application of this technique to an experiment that determined the rehydration patterns of various desert nematode genera following a wetting event and the difference in activity states by depth. We found that most desert nematodes became active very quickly after wetting, usually within an hour. Although nematodes were more abundant in shallow (0–10 cm) than deep (10–20 cm) soil, more nematodes were active in deep soil rather than shallow soil the morning of a representative summer day. However, some genera, such as *Acromoldavicus, Aporcelaimellus, and Tylenchus*, had similar proportions of activity states in shallow and deep soil. Our study supports the existing literature that nematodes respond quickly to wetting events, but our work is unique by providing the opportunity to identify nematodes to genus when extracted from field soils in an anhydrobiotic state. In the future, we will apply this method to quantifying the response of desert nematode communities to changes in climate, such and altered precipitation and temperature patterns.

POST HARVEST SYMPTOM DEVELOPMENT OF *MELOIDOGYNE CHITWOODI* ON POTATO. **David, N. L.,[1] R. E. Ingham,[1] N. D. McKinley,[2] B. A. Charlton,[3] K. J. Merrifield,[1] and N. M. Wade[1].** [1]Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR 93771; [2]DuPont Agricultural Products, 4280 Montaigne Lane S., Salem OR 97302; [3]OSU Klamath Falls Experiment Station, 6941 Washburn Way, Klamath Falls, OR 97603.

Columbia root-knot nematode (*Meloidogyne chitwoodi,* CRKN) is a serious pathogen of potato in many areas of the western United States, reducing tuber quality by inducing surface galling and/or internal brown spots. Potatoes are shipped year round and many are stored for over 9 months in refrigerated cold storage at less than 5C. Because the effect of cold storage on post harvest symptom development of CRKN in potato was unknown, field trials were conducted in the Klamath Basin (KB) of Oregon and San Luis Valley (SLV) of Colorado during 2002 and 2003 to determine if internal and/or external CRKN symptoms increased following commercial storage or incubation at room temperature until tubers had accumulated over 500 degree-days base 5C. The percentage of tubers with external symptoms from the KB did not increase following incubation during 2002 and 2003 compared to the harvest evaluation. Similarly, external symptoms on potatoes from the SLV did not increase following commercial storage or incubation during 2002. However, the percentage of tubers

with external symptoms increased by 7 and 10% following storage and incubation respectively during 2003. Internal CRKN symptoms did not change following storage at either location, but significantly increased after incubation by 10% and 6% in the KB and by 19% and 13% in the SLV during 2002 and 2003, respectively. These studies indicate that no additional CRKN symptom development occurred when tubers were stored at 5C or less, but that internal symptoms may increase at higher temperatures.

ROW PLACEMENT IS INNEFFECTIVE AS A CULTURAL CONTROL PRACTICE FOR *MELOIDOGYNE INCOGNITA* IN COTTON. **Davis, R. F**. USDA-ARS, Crop Protection and Management Research Unit, Tifton, GA 31793.

The objective of this study was to determine if planting cotton into the space between the previous year's rows reduces crop loss due to nematodes compared to planting in the same row every year. In 2004, nematode counts were lower in plots planted in the previous year's rows on 4 Aug ($P = 0.064$) and 15 Oct ($P = 0.087$). Root galling was lower ($P \leq 0.05$) and yield was higher ($P = 0.081$) in fumigated plots, but galling and yield were not affected by row placement. On early sampling dates (24 May and 8 Jul) in 2005, nematode counts were higher ($P \leq 0.05$) in plots planted in the previous year's rows, but counts at the end of the season (31 Oct) were higher where the row placement had been moved. Root galling was lower ($P \leq 0.05$) and yield was higher ($P = 0.069$) in fumigated plots, but galling and yield were not affected by row placement. Changing the placement of rows reduced nematode pressure early in the growing season in only one of the two years studied, but end-of-season root galling and lint yield were not affected in either year by changing the placement of rows, nor was the effect of fumigation on yield influenced by row placement. Therefore, row placement is unlikely to contribute to *M. incognita* management in cotton.

INBREEDING AND TRAIT DETERIORATION IN ENTOMOPATHOGENIC NEMATODES. **Dillman A. R.,[1] J. M. Chaston,[2] D. I. Shapiro-Ilan,[3] A. Bilgrami,[4] R. Gaugler,[4] and B. J. Adams[1].** [1]Brigham Young University, Provo, UT 84602, [2]Univ. of Wisconsin-Madison, Madison, WI 53706, [3]SE Fruit and Tree Nut Research Unit, USDA-ARS, SAA, Byron, GA 31008, [4]Department of Entomology, Rutgers Univ., New Brunswick, NJ 08901.

Entomopathogenic nematodes (genera *Heterorhabditis* and *Steinernema*) are biocontrol agents that kill their invertebrate hosts with the aid of a mutualistic bacterium. Their use in field applications has achieved some success, although a lack of performance predictability has hindered their widespread acceptance in pest management programs. Because of the crucial nature of traits that affect the nematodes' ability to act as biological control agents (viz. virulence, heat tolerance, and fecundity), it is important to understand trait loss in laboratory culturing. Our study aims to discover the cause of avirulence generated through in vitro culturing of nematodes by considering the major factors that influence genetic changes under laboratory conditions: drift, selection, and inbreeding. We answer these questions by performing sexual crosses of inbred nematodes with each other and with the wild-type parental strain. Our results show a successful recovery of both fecundity and heat tolerance in *Steinernema carpocapsae*, revealing that the development of avirulence is genetic and occurs by inbreeding. We explore two theories that seek to explain the existence of inbreeding depression: the overdominance and partial dominance hypotheses. Based on predictions specific to each hypothesis, we conclude that our results support the theory of overdominance.

DEVELOPING A RESEARCH AND EXTENSION PROGRAM FOR CONTROL OF THE GUAVA WEEVIL IN BRAZIL USING ENTOMOPATHOGENIC NEMATODES. **Dolinski, C.** Universidade Estadual do Norte Fluminense Darcy Ribeiro/ CCTA/LPP. Av. Alberto Lamego, 2000, Pq. Califsrnia, Campos dos Goytacazes, RJ, 28015-620.

Guava is becoming one of the most important fruit crops in the State of Rio de Janeiro, Brazil, and the guava weevil, (*Conotrachelus psidii*), is considered a major pest because it directly damages the fruit and is difficult to control with chemical pesticides. In association with guava growers from Cachoeiras de Macacu, we are developing an integrated pest management (IPM) program for this pest using entomopathogenic nematodes (EPN). Weevil adults appear in September-October and remain in the field until March. Females lay their eggs on green fruit, and the oviposition site is marked by a characteristic dark depression. When the fruit ripens and falls to the ground, 4th instar larvae enter the soil and pupate after a variable time interval. Emerging adults return to the trees for copulation and oviposition. In Petri dish assays with sand and 100 IJ/larva, *Heterorhabditis indica* Hom1 and *H. baujardi* LPP7 caused 85 and 80% mortality, respectively. In sand columns with 500 IJ/larva, *H. baujardi* LPP7 was the most effective and killed 70% of the larvae. In a greenhouse study with guava trees in 20-L pots and 2000 IJ/pot, *H. baujardi* LPP7 caused 58% mortality. For field trials using *H. baujardi* LPP7, the guava growers paid for the necessary government registration and are rearing the nematodes themselves in *Galleria mellonella* larvae. The nematodes are applied in orchards as infected cadavers. Initial results indicate a decrease in adult weevils of 40–70% with an application of 20 cadavers per tree. In addition, the growers are using cultural control by removing all damaged fruit from their orchards to reduce pest innoculum for the following year. In the absence of insecticides, natural enemies (e.g., Coccinellidae and Chrysopidae) are also being seen more often. By eliminating pesticides, these strategies have effectively reduced production costs by 40%.

RESISTANCE IDENTIFICATION FOR SOYBEAN CYST NEMATODE: A COMPARISON OF RATING METHOD AND INOCULUM LEVELS. **Donald, P. A.,[1] and L. D. Young[2].** USDA ARS Crop Genetics and Production Research Unit, [1]Jackson, TN 38301; [2]Stoneville, MS 38776.

Soybean cyst nematode resistance in soybean germplasm is an important tool for management of the nematode. However, evaluation methods to identify resistance have not been standardized. Two techniques were compared to evaluate plant resistance using races 3 and 14 under greenhouse test using an inoculum level of 1,000 eggs/ml. A visual rating (VR) with five numeric categories was compared to microscopic enumeration of cysts on the roots and computation of a female index (FI) which has been suggested for use in four resistance categories. The study consisted of 673 entries from the Tennessee State Soybean Variety Test over three years. The VR had larger F values and lower coefficient of variation than the FI. There is a significant increase in the amount of time required to collect data for the FI compared to the VR however environmental conditions can greatly affect visualization of cysts in the VR method. Two inoculum levels, 1,000 and 4,000 eggs/ml, were tested with both races on the seven indicator lines plus susceptible check for the HG Type Test. No significant differences were seen in the FI at the two inoculum levels for race 3. The race 14 standard deviation FI was higher at the lower inoculum level than the higher inoculum level and there were significant differences between indicator lines at the higher inoculum level. The VR system is a less time consuming procedure than the FI in identifying resistance to soybean cyst nematode. Inoculum level can affect identification of resistant soybean lines in certain races.

SUPPRESSION OF *DIAPREPES ABBREVIATUS* IN POTTED CITRUS BY COMBINATIONS OF ENTOMOPATHOGENIC NEMATODES WITH DIFFERENT LIFESPANS. **El-Borai, F. E., and L. W. Duncan.** University of Florida, Citrus Research and Education Center, Lake Alfred, Florida 33850.

Two experiments were conducted to test the hypothesis that augmenting EPN communities with short-lived EPN species can cause an eventual reduction in biological control of insect larvae by partially displacing longer lived EPN species. Pots containing citrus seedlings growing in pasteurized soil mix (50:50/v:v Candler fine sand and shredded Canadian sphagnum peat moss) were infested with factorial combinations of *Steinernema riobrave* (Sr; short-lived) and *S. diaprepesi* (Sd; long-lived) in both experiments, and with Heterorhabditis zealandica (Hz; short-lived) alone or in combination with Sr or Sd in the second experiment. Larvae of the weevil *Diaprepes abbreviatus* were added to the pots periodically and plants were grown for up to 8.5 months in the first experiment and 13.5 months in the second. No interactions ($P \geq 0.05$) occurred between any of the treatments. The growth and survival of the citrus plants and the suppression of weevil larvae were greatest ($P \leq 0.05$) in all treatment combinations that contained Sd and intermediate for all Sr treatment combinations that lacked Sd. Augmenting pots with Hz did not significantly affect plant growth or insect suppression. S. diaprepesi was the only EPN species recovered by baiting soil with insect larvae at the end of each experiment. Neither experiment supported our hypothesis. Accordingly, we speculate that reported temporary reductions in EPN prevalence in citrus orchards following EPN augmentation is likely the result of population responses by EPN antagonists.

FIELD AND GREENHOUSE EVALUATIONS OF SELECTED FUNGICIDES ON DEVELOPMENT OF SOYBEAN CYST NEMATODE. **Faghihi, J.,[1] R. A. Vierling,[2] and V. R. Ferris[1].** [1]Department of Entomology, Purdue University, West Lafayette, IN 47907, [2]Indiana Crop Improvement Association and Department of Agronomy, Purdue University, West Lafayette, IN 47907.

Greenhouse screening of soybean seedlings with soybean cyst nematodes (SCN) takes several months and results can be confounded by fungal infection of the seedlings. We have found that *Rhizoctonia solani* infection has been a major problem and we had to resort to the use of fungicides. We assessed the effectiveness of several fungicides to control *R. solani*, and observed their effect on SCN. We discovered that an unusually low number of cysts developed on susceptible plants treated with Cleary 3336F fungicide in the greenhouse. Repeated greenhouse studies revealed the effectiveness of this fungicide as a possible management tool for SCN. Field studies in 2003 and 2004 showed an increase in yield in plots treated with Cleary 3336F. No yield increase was observed in 2005 field studies. The final population of SCN was not reduced by Cleary 3336F in any of the field tests.

ENHANCING *STEINERNEMA* SPP. SUPPRESSION OF *MELOIDOGYNE JAVANICA*. **Fallon, D. J.,[1] H. K. Kaya,[2] and B. S. Sipes.[1]** [1]Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822; [2]Department of Nematology, University of California, Davis, CA 95616.

Consistent control of plant-parasitic nematodes by biological agents has been difficult to achieve. Entomopathogenic nematodes have demonstrated varying degrees of success in suppressing a wide range of plant-parasitic nematode species in laboratory, greenhouse, and field studies. *Steinernema feltiae* MG-14- and *S. glaseri* NJ-infected *Galleria mellonella* were tested for their efficacy against *Meloidogyne javanica*. *Steinernema feltiae*- and *S. glaseri*-infected cadavers significantly reduced *M javanica* egg production in cowpea after 35 days. In combination with the nematode trapping fungi *Arthrobotrys oligospora*, a significant interaction was observed between the nematode trapping fungi and nematode-infected cadavers. Fewer eggs were recovered from *M. javanica* infested plants treated with *S. feltiae*-infected cadavers in

the presence of *A. oligospora* than plants treated with *S. feltiae*-infected cadavers in the absence of *A. oligospora*. But there was no such relationship when *S. glaseri* or frozen *G. mellonella* cadavers were used in conjunction with *A. oligospora*. Soil amendment with 10% v/v cowpea leaves in conjunction with entomopathogenic nematode-infected insect cadavers reduced *M. javanica* egg production, but the affect was not significant. Enhanced suppression of *M. javanica* by entomopathogenic nematodes is possible, but variability in the suppressive effect remains a concern.

QUANTIFICATION OF THE SOYBEAN CYST NEMATODE, *HETERODERA GLYCINES*, IN SOYBEAN ROOTS WITH REAL-TIME PCR. **Gao, X., A. L. Colgrove, K. N. Lambert and T. L. Niblack.** Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

The soybean cyst nematode (SCN), *Heterodera glycines*, is a destructive pathogen of soybean, and causes significant yield losses worldwide. Recently, interest has increased in the evaluation of seed treatments with novel nematicidal compounds to protect young plants of soybean and other crops from nematode infections. Enumeration of nematodes in plant tissues, which is necessary for this type of evaluation, requires time-consuming staining procedures and labor-intensive microscopic observations. In this study, we explored the use of a real-time quantitative polymerase chain reaction (QPCR) assay based on detection of the chorismate mutase gene (*Hg-cm-1*) for quantification of SCN. The QPCR protocol was sensitive and specific, and showed a high degree of correlation with the number of juveniles in roots. This assay will greatly reduce the amount of time and resources required to assess treatment effects on nematode infection in seedlings.

THE ROLE OF 2,4-DAPG PRODUCING PSEUDOMONADS IN SUPPRESSING SOILBORNE PATHOGENS **Gardener, B. McSpadden, D. Rotenberg, R. Joshi, F. Baysal, and M. S. Benitez.** Department of Plant Pathology, Ohio State University, OARDC, Wooster, OH 44691.

Native populations of *Pseudomonas* spp. that produce 2, 4-diacetyl-phloroglucinol (DAPG) are widely distributed in agricultural soils. Roots can harbor significant populations of these antibiotic-producing bacteria, and they can contribute to soilborne disease suppression in the field. However, the magnitude of that contribution is dependent on a variety of environmental factors that can also affect the relative abundance and activities of nematodes, soilborne pathogens, and their plant hosts. Farmers may choose different approaches to soilborne disease control that potentially influence the abundance and activities of DAPG-producers as well as the pathogens they can suppress. In the study of both, conventional and organic cropping systems, we found that different management practices can significantly alter patterns of rhizosphere colonization and the degree to which native and inoculated populations contribute to crop health. Specifically, we have found i) complex interactions between the effects of tillage and rotation on DAPG-producers in corn and soybean fields, ii) significant correlations between the abundance of DAPG producers and various soil fertility components, iii) enrichment of both pathogen pressure and antagonists in certain farming systems, and iv) significant correlations between soil disease suppression and multiple populations of rhizosphere bacteria. From these studies, we have developed a model that describes the relationship between the abundance of DAPG producers and plant productivity under conditions of varying plant disease pressure.

CHEMICAL NEMATICIDES EFFICACY FOR THE MANGEMENT OF *MELOIDOGYNE CHITWOODI* ON POTATO IN IDAHO. **Hafez, S. L., and S. Palanisamy.** University of Idaho, Parma Research and Extension Center, Idaho 83660.

Three field experiments were conducted at the University of Idaho, Parma Research and Extension Center, Parma, Idaho to demonstrate the effects of Telone II alone and in combination with Metam Sodium on *Meloidogyne chitwoodi* management and yield of potato. The experiment was laid out in a randomized complete block design with five treatments each with six replications in a sandy loam field. Mocap treatments were surface broadcast using a hand held plot sprayer with 800 Oz flat fan nozzles at 50 psi calibrated to deliver 34.5 gallons per acre. Telone II and Vapam were applied as broadcast by ripper and fumigation bar, respectively. Potato cv. Russet Burbank seed pieces were planted in rows three feet apart. Weeding and other normal cultural practices were followed. Five months after planting, the tubers were hand-harvested from 15 feet of the middle two rows of each plot and weighed. The tubers were graded and evaluated for nematode infection. Yield of tubers from different treatments indicated that there was an increase in marketable yield and total yield in different combinations of all treatments as compared to untreated control plots. Nematode infected tubers as well as percent of nematode infection were also significantly reduced by the treatments compared to untreated control plots. Percent of tubers with nematode infection in treated plots ranged from 0.9 to 37.0. Lowest level of nematode infection was recorded in the Telone 15 gal/A + Vapam 30 gal/A, with the maximum marketable yield than other treatments.

GREEN MANAURE CROPS EVALUATION FOR THE SUPPRESSION OF *MELOIDOGYNE CHITWOODI* AND *HETERODERA SCHACHTII*. **Hafez, S. L., and S. Palanisamy.** University of Idaho, Parma Research and Extension Center, Idaho 83660.

Two experiments were conducted in a green house to study the effect of green manure varieties on the population of Columbia root knot nematode *Meloidogyne chitwoodi* (Experiment–I) and sugar beet cyst nematode *Heterodera schachtii*

(Experiment-II). In the first experiment efficacy of new green manure varieties were compared for their effect on *M.chitwoodi* population. Five varieties of green manure were planted on May 7 in a completely randomized block design with five replications each. Crop was harvested on July 2 and data on fresh and dry weight of shoot and root along with the nematode population in the soil and root were recorded. Data indicated that among all varieties comet showed maximum fresh and dry weight of shoot and root. Lowest level of total *M.chitwoodi* population in the root and the maximum percent of reduction of root population (98.7) were observed in the Comet followed by Defender (94.9). In the second experiment efficacy of seven green manure varieties were compared for the effect of suppression of *Heterodera schachtii*. All these varieties were planted in individual pots filled with soil infested with sugar beet cyst nematode (4 eggs and larvae/cc soil) on August 5 in a completely randomized block design with five replications each. Crop was harvested on October 10 and data on eggs and larval population in the soil along with the viable cysts were recorded. Data indicated that all varieties (except Mustard blend) significantly reduced cyst, eggs and larval population compared to fallow.

EFFICACY OF ENTOMOPATHOGENIC NEMATODES IN CONTROLLING ORCHARD PESTS IN CHINA. **Han, R. C., L. Cao, and X. H. Qiu.** Guangdong Entomological Institute, Guangdong Academy of Sciences, Guangzhou 510260, China.

The apple fruit moth *Carposina nipponensis* is one of the most important pests, causing economic lose of 1.7 billion dollars per annum in 1 million ha of apple orchards in northern China. The banana stem borer *Odoiporus longicollis* and banana weevil borer *Cosmopolites sordidus* are most important pests in banana plantations. Litchi stem borer *Arbela dea* and litchi longhorn beetle *Aristobia testudo* are key litchi pests in the southern China. Entomopathogenic nematodes (EPN) have been used to control all these orchard pests in China. Field trails resulted in more than 90% larval mortality of the *C. nipponensis* larvae from the overwintered cocoons with 0.5–0.9 million infective juveniles (IJ) of *S. carpocapsae* A24 per square meter. The field results also indicated that 76–90% of the overwintered larvae, 68–92% of the pupae and 25–80% of the adults of the banana stem borer were controlled by spraying 3–6 million IJ of *S. carpocapsae* A24 nematodes into each residual stem base of the banana. Successful control of the litchi pests was achieved in 1,700 ha of litchi orchards in Guangdong. Over 86% *A. dea* mortality was obtained by spraying 1,000 IJ of *S. carpocapsae* A24 around each borer hole of the litchi, and over 73% *A. testudo* mortality following injection of 3000 IJ of the same nematode strain into the freshly bored holes in litchi. The advantages and disadvantages of using EPN for the control of these orchard pests are discussed.

MANAGEMENT OF *MELOIDOGYNE INCOGNITA* WITH *RAPHANUS SATIVUS OLEIFERA* AND *ERUCA VESICARIA SATIVA* ON CUCUMBER IN LEBANON. **Haroutunian, G.[1], S. L. Hafez,[2] and F. Marion-Poll[3].** [1]MeBr. Alternatives Project / Ministry of Environment-UNDP, Lebanon, [2]University of Idaho, Parma Research and Extension Center, USA, [3]Institut National Agronomique Paris-Grignon, France.

An experiment was conducted in a commercial green house to find out the efficacy of oil radish (*Raphanus sativus* spp. Var. Boss) and arugula (*Eruca vesicaria sativa*) for the control of *Meloidogyne incognita* in greenhouse grown cucumber. The experiment was in a completely randomized block design with five treatments and four replications each including an untreated control. The treatments were fallow, methyl bromide (1,000 kg/Ha), biofumigation crops (oil radish) with and without plastic cover and arugula with plastic cover. Oil radish and arugula seeds were sown in specific treatment plots inside the greenhouse at the rate of 30 kg per Ha, and the resulting crop was incorporated to the soil after six weeks, when plants reached 35–40 cm of height. Transparent polythene film of 50 microns thickness was used to cover some of the treated plots, while others were left uncovered. After two weeks polythene films were removed, the soil was aerated and cucumber seedlings were transplanted. Ten plants were marked in the middle of each treatment area, and plant vigor and fruit yield were recorded. Data indicated that after 12 weeks of harvest, the highest fruit yield resulted from the oil radish and arugula areas covered with polyethylene (respectively 1895 g and 1890 g per plant), followed by the methyl bromide treated plots (1824 g). Uncovered oil radish plots yielded 1758 g. while untreated fallow plots only yielded 1076 g. There was no significant difference in yield between methyl bromide treated and both oil radish and arugula areas covered with polyethylene.

USING ENTOMOPATHOGENIC NEMATODES AGAINST *OTIORHYNCHUS* SPP. IN FIELD GROWN STRAWBERRIES-THE GOOD AND THE BAD NEWS. **Haukeland, S.** Bioforsk, Norwegian Institute for Agricultural and Environmental Research, Plant Health & Plant Protection Division, Hogskoleveien 7, 1432 Aas, Norway.

Entomopathogenic nematodes (EPN) are used for biological control of the soil dwelling stages of the vine weevil, *Otiorhynchus sulcatus*, a serious pest of ornamentals and field grown strawberries. The nematodes *Heterorhabditis megidis, H. bacteriophora* and *Steinernema kraussei* are commercially available in Europe for control of the vine weevil. *S. kraussei* is sold as a cold active strain for use outdoors at low temperatures. In southern Norway, several field trials were conducted in strawberries to examine the effect of *S. kraussei* on vine weevil larvae at low temperatures. The trials were assessed one month after nematode treatments and showed rather poor effect at soil temperatures below 12°C. Further work is necessary

to investigate the long-term effect of the applied nematodes as well as biotic and abiotic factors that may affect nematode efficacy. For comparison, the nematode *H. megidis* was used in these same trials and the results showed that this species is effective at soil temperatures above 12°C, there was no indication that *H. megidis* had any effect at low temperatures.

DIRECT AND INDIRECT EFFECTS OF MUSTARD AMENDMENTS ON TWO ENTOMOPATHOGENIC NEMATODE GENERA, *STEINERNEMA* AND *HETERORHABDITIS*. **Henderson, D.,[1] R. Ramirez,[2] E. Riga,[1] and W. E. Snyder.[2]** Departments of Plant Pathology[1] and Entomological Sciences[2], Washington State University, Pullman, WA 99164.

The use of mustard green manure has become a common practice of integrated pest management for plant parasitic nematode control in Washington State, USA potato cropping systems. Mustard plants contain inert concentrations of glucosinolate compounds and a myrosinase enzyme which upon disruption of plant tissue, by practices such as tillage, allows glucosinolate to interact with the enzyme myrosinase. In the presence of water, glucosinolates are hydrolyzed into several isothiocyanate products, believed to be nematicidal. In addition to mustard green manure, entomopathogenic nematodes (EPN) have shown promise as biological control agents against both insect pests and plant parasitic nematodes. It is possible that mustard green manures, used for the control of plant parasitic nematodes, are also antagonistic to entomopathogenic nematodes. The objective of this research was to determine the effect of glucosinolate on the infectivity and mortality of two EPN genera, *Steinernema* and *Heterorhabditis*. Two cultivars of *Brassica juncea* with high or low glucosinolate levels were tested against seven EPN species: *Steinernema carpocapsae*, *S. glaseri, S. riobrave, S. feltiae, Heterorhabditis marelatus, H. bacteriophora*, and *H. megidis*. In a laboratory bioassay, the greater wax moth, Galleria melonella was used to determine the indirect effect of mustard cultivar on EPN infection. A subsequent Petri dish bioassay examined the direct effect of mustard cultivar on EPN mortality. These data suggest that mustard cultivars with high glucosinolate levels have a negative effect on EPN infection rate and mortality. However, the direct and indirect effects varied between the genera and between species.

NEMATICIDAL PROPERTIES OF FURFURAL AND THE DEVELOPMENT FOR NEMATODE CONTOL IN VARIOUS CROPS FOR THE UNITED STATES MARKETS. **Hensley, J.,[1] and G. Burger.[2]** [1]Agriguard Company LLC, Jackson, TN, and [2]Illovo Sugar Ltd. Durban, South Africa.

Furfural is a natural product derived from various plant sources. Sugar cane bagasse is an important raw material used in the production of furfural. Agriguard Company LLC, a US company jointly owned by Illovo Sugar Ltd, a South African sugar and furfural producer, and Harborchem, are developing and commercializing furfural as a nematicide. In South Africa, furfural is labeled for the control of nematodes in peanuts, potatoes, tomatoes, sugar cane, corn, onions, peppers, lettuce, turf, carrots and ornamentals. Additional registrations are being developed for tobacco, citrus, deciduous fruit, dry beans, cucurbits, and vine crops. In the United States, furfural is being investigated for the control of nematodes in turf, peanuts, vegetable crops, ornamentals and fruit and vine crops. Due to furfural's low phytotoxicity, applications can be made post plant as well as preplant to crops. This unique property allows for in-season applications to provide season long nematode control. Furfural is a contact nematicide and must be mechanically incorporated or moved into the soil profile with irrigation. Rates ranging from 53.5 kg ai/ha to 138.5 kg ai/ha are currently being tested and activity has been demonstrated on *Belonolaimus* spp (Sting), *Hoplolaimus galeatus* (Lance), *Criconemella xenoplax* (Ring), *Meloidogyne* spp (Root Knot) and *Paratrichodorus* minor (Stubby Root) nematodes. Preplant applications up to 448 kg/ha have been tested and shown to be safe on strawberries, tomatoes and peppers. Data will be presented demonstrating the control of nematodes in turf, vegetable crops, citrus and peanuts.

BIOLOGICAL CONTROL OF *MELOIDOGYNE INCOGNITA* USING IN-VITRO PRODUCED *PASTEURIA PENETRANS* IN A MICROPLOT STUDY. **Hewlett, T. E., S. T. Griswold, and K. S. Smith.** Pasteuria Bioscience LLC, FL 32615.

The efficacy of in-vitro produced endospores of *Pasteuria penetrans* was tested for control of *Meloidogyne incognita* on *Hostas* spp.. This test was conducted in a microplot trial at the Clemson University Pee Dee Research Station in South Carolina. Microplots were inoculated with nematodes with root pieces and soil collected from *M. incognita* green house pot cultures. *Hostas* spp. root systems were washed and transplanted into holes in microplots. Nematode-free soil was inoculated with 0 (untreated control), 104 or 105 in-vitro produced endospores/cc of soil. Treated soil mixtures, 350 cc, were placed around each plant root system. Treatments were replicated five times in plots and each plot received three plants. Plants were harvested for data collection after 60 days. Data collected included root weight, number of galls, number of juveniles in the soil, number of spores on juveniles, number of females infected with *P. penetrans*. There was a significant decrease in galling compared to the untreated control in plots with endospores densities of 105 spores/cc of soil. The nematode suppression at this endospore density is consistent with literature reports of suppression achieved with *P. penetrans* endospores produced in-vivo.

EVALUATING SCN SCREENING METHODS - INDUSTRY AND PUBLIC INSTITUTIONS. **Hicks, J.** Monsanto Corp., Ames, IA 50010.

A cooperative study was conducted by thirteen participants from public and private institutions to investigate the consistency of screening results for *Heterodera glycines*, soybean cyst nematode (SCN). Each institution screened three replications of 100 entries with a race 3 nematode population (Hg Type 0). Participants used their current lab procedures and nematode cultures. Final results were rated as resistant (R), moderately resistant (MR), moderately susceptible (MS), or susceptible (S) for ease in analysis. Significant differences were noted between participating institutions, among replications within a lab, and between labs and the known resistance of the entries to SCN race 3. Screening procedures are being compared for trends affecting the accuracy of the results. In a separate study, differentials used by ten of the participating labs were typed using eight SSR markers covering the genomic regions containing rhg1 and Rhg4 to determine whether there was variation in the checks compared to those obtained from the USDA. Markers indicated that variances were seen in the checks at five labs.

ENGINEERING RESISTANCE AGAINST PLANT-PARASITIC NEMATODES. **Huang, X., Yu Han, A. Wiig, J. McMillan, T. Jones, W. Talton, J. Dong, M. Offenheiser, P. Ren, R. Ascenzi, S. Hill, R. Zhen, and S. Chaudhuri.** BASF Plant Science L.L.C., RTP, NC 27709.

Plant parasitic nematodes are a major threat to all crops of economic importance with ~$100 billion annual crop losses worldwide. The soybean cyst nematode is by far the most important pathogen on soybean in the US. Therefore we are interested in genetically modifying soybean to achieve durable resistance and reduced yield loss. Genomic and molecular tools are utilized and discussed herein.

ANALYSIS OF CELL WALL SYNTHESIS IN FEEDING CELLS FORMED BY ROOT-KNOT NEMATODES. **Hudson, L. C[1], C. H Haigler[2], and E. L Davis[1].** [1]Department of Plant Pathology, Box 7903, [2]Department of Crop Science, North Carolina State University, Raleigh, North Carolina 27695.

Root knot nematodes (*Meloidogyne* sp.) are endoparasites that infect roots of many plant species and cause a large economic loss to many crops. Root-knot nematodes induce the differentiation of vascular parenchyma into multinucleate "giant-cells" that are enlarged and arise from repeated karyokinesis without cytokinesis. Giant-cells undergo extensive modifications of the cell wall architecture including cell wall thickening and the formation of ingrowths. The cell wall growth, expansion, and integrity of giant-cells are hypothesized to be mediated by both cell wall-loosening and cell wall biosynthetic enzymes of plant origin. The up-regulation of plant genes involved in wall loosening in giant-cells has been detected however the molecular basis of the thickened cell walls and ingrowths in giant-cells has not been fully explored. Expression patterns of the plant cellulose synthase (CesA) gene family, whose primary responsibility is cellulose anabolism, are being investigated in feeding site formation during plant-nematode interactions. Ten members of the CesA gene family from Arabidopsis thaliana are being analyzed for expression in giant-cells using RT-PCR. Transgenic Arabidopsis that contain different CesA gene promoter::GUS constructs are also being assayed to monitor CesA expression in giant-cells. Roots of available CesA mutants of Arabidopsis have been infected by root-knot nematodes for functional analyses of the effects of these mutations on the parasitic interaction. The data generated on cell wall deposition can subsequently provide not only a more comprehensive understanding of the wall architecture of giant-cells, but potentially to serve as a model of the fundamental biology of wall formation in plant cells.

CROP ROTATION FOR MANAGEMENT OF *MELOIDOGYNE CHITWOODI*. **Ingham[1], Russell E., N. L. David[1], D. A. Horneck[2], G. Clough[2], P. B. Hamm[2], and N. M. Wade[1].** [1]Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331, [2]Hermiston Agriculture Research and Extension Center, Oregon State University, Hermiston, OR 97838.

Columbia root-knot nematode (*Meloidogyne chitwoodi,* CRKN) infects potato tubers and reduces quality to unmarketable levels at populations as low as 1/250-g soil. Most crops grown in rotation with potato increase CRKN, requiring fumigation or multiple applications of oxamyl. A four-year rotation experiment that began with high CRKN populations, 1,550/250-g soil in spring of 2001, demonstrated that CRKN could be managed with crops currently grown in the Columbia Basin of OR and WA if excellent host crops were grown early rather than late in the rotation. In a conventional rotation of wheat (W)-field corn (FC)-FC, CRKN densities were high (2,000/250-g soil) when potato was planted in spring of 2004 and 100% of harvested tubers were culled due to excessive CRKN infection. Reversing the order of these crops (FC-FC-W) allowed time for a mustard green manure crop (MGM) in fall of 2003 and reduced CRKN to 56/250-g soil and culls to 32%. A FC-W-supersweet corn (SSC)-MGM rotation reduced CRKN to 14/250-g soil and culls to 27%. A FC-SSC-green pea-lima bean (GP-LB)-MGM and a FC-W-GP-LB-MGM sequence reduced CRKN to 3/250-g soil and culls to 9% and 2%, respectively. Seed crops of bluegrass, perennial ryegrass and tall fescue followed by MGM reduced CRKN to 19, 0 and 0/250-g soil, respectively, and culls to 48%, 13% and 2%, respectively. All 11 alternative rotations substantially

reduced CRKN and had significantly fewer culls than the standard. Growers assumed alternative rotations would be less profitable but all alternative rotations had higher net value than W-FC-FC-potato.

SOIL AMENDMENT WITH A STREPTOMYCETE AND CHITIN AGAINST THE NORTHERN ROOT-KNOT NEMATODE (*MELOIDOGYNE HAPLA*). **Jobin, G., and G. Bélair.** Agriculture and Agri-Food Canada, St-Jean-sur-Richelieu, Quebec, Canada.

Nematicidal potency of soil amendments with two commercial formulation of a streptomycete strain and chitin were first assessed in cups filled with a pasteurized sandy soil, inoculated with 5 egg masses (average of 950 *M. hapla* eggs per cup) 24 h before application of treatments and left in the dark at 25°C during 30 days. Efficacy of treatments was assessed by counting the number of juveniles (IJ) recovered using the Baermann pan method. The best treatment was obtained with the joint application of Actinovate (10^5 cfu g^{-1} dry soil) and chitin at 1.0% (w/w) which reduced by 95% the mean number of IJ per cup (5 IJ/cup) when compared to the positive (untreated) control (102 IJ/cup). The next best treatments were the sole application of Actinovate and chitin 1.0% with a reduction of 86 (14 IJ/cup) and 79% (21 IJ/cup) respectively. The application of Actino-Iron formulation alone did not provide *M. hapla* control but its joint application with chitin 1.0% gave 85% control (15 IJ/cup). In a greenhouse trial, Actinovate and chitin were further tested on tomato (cv. Rutgers) plants grown in a composite rooting medium made from a nematode-infested organic soil and a pasteurized sandy soil (60:40, w/w) containing 54 *M. hapla* IJ per 100 g soil. After a 60-day period, the joint application of Actinovate (10^5 cfu g^{-1}dry soil) and chitin 1.0% provide the best control with a 73% control level (786 IJ/100 g soil) when compared to the positive (untreated) control (2915 IJ/100 g soil). This treatment also reduced the nematode galling by 86% (247 galls/g dry root) when compared to the positive control (1793 galls/g dry root).

ELIMINATION OF METHYL BROMIDE EMISSIONS FROM FUMIGATED SOIL. **Joyce, P. J. [1], R. Bielski,[1] and J. M. Halbrendt[2].** [1]Value Recovery, Inc., Bridgeport, NJ, 08014; [2] Penn State Fruit Research and Extension Center, 290 University Drive, Biglerville, PA 17307.

Methyl bromide (MB) is an effective soil fumigant. The global phase out of MB for agricultural use is aimed at eliminating atmospheric emissions that destroy the ozone layer. However, the Montreal Protocol makes provisions for MB use that is destroyed or recycled and does not contribute to ozone depletion. This work reports on the progress of developing a method to capture and destroy MB used for soil fumigation. The technology is based on a scrubber system invented by Value Recover, Inc. that irreversibly converts MB to non-hazardous byproducts at ambient temperatures using an inexpensive and environmentally benign reagent (ammonium thiosulfate). The initial experiments were performed in sealed soil cylinders (1.3 m × 0.3 m dia.) filled with sandy loam soil (moisture 0.032 vol%, temp. 13–20°C). One × seven tenths gram (267.85 Kg/Ha) MB was injected into the cylinder at 30 cm and equilibrated for 18 hr. Methyl bromide emissions were captured by sweeping the headspace with a slow volumetric flow-rate of fresh air to mimic convective removal of MB from farmland sealed with Virtually Impermeable Film (VIF). Vented MB was measured every 20 seconds and concentrations recorded. A bioassay using nematodes in nylon mesh bags buried at 15 and 45 cm within the cylinder was used to verify efficacy of the treatment. Results showed that 99.96% of the nematodes had been killed and 85% of the MB decomposed. Large scale (1.2 Ha) field trials using a trailer-mounted scrubber and VIF to capture MB are in preparation.

DEVELOPMENT AND REPRODUCTION OF RADOPHOLUS SIMILIS ON EXCISED *ANTHURIUM ANDRAEANUM* ROOT CULTURE. **Khaithong, T.,[1] B. S. Sipes,[1] and A. R. Kuehnle.[2]** Departments of [1]Plant and Environmental Protection Sciences and [2]Tropical Plant and Soil Sciences, University of Hawaii at Manoa, Honolulu, HI 96822.

Development and reproduction of *Radopholus similis* on an excised anthurium root in tissue culture was studied to determine if a single root of *Anthurium andraeanum* could be used for resistant screening in vitro. A single gravid female of *R. similis* could reproduce on Marian Seefurth, Paradise Pink, and Anuenue excised roots grown in half-strength MS medium supplemented with 150 ml/l coconut water. In Marian Seefurth, mature males and females were recovered within 25 days after inoculation (DAI) and gravid females were recovered within 30 DAI. When a single excised root of Marian Seefurth was inoculated with 1, 5, 10 and 20 gravid females, nematode reproduction rate (Final population/Initial population, Pf/Pi) decreased as inoculum level increased. We demonstrated that a single excised anthurium root can be used for resistant screening for *R. similis*, reducing the time and space needed for screenings. Using gravid females allowed the measurement of both nematode development and nematode reproduction.

DEVELOPMENT OF TRANSGENIC ANTHURIUM EXPRESSING MODIFIED RICE CYSTEINE PROTEASE INHIBITOR. **Khaithong, T.,[1] B. S. Sipes,[1] and A. R. Kuehnle.[2]** Departments [1]Plant and Environmental Protection Sciences and [2]Tropical Plant and Soil Sciences, University of Hawaii at Manoa, Honolulu, HI 96822.

Development of transgenic *Anthurium andraeanum*, an ornamental flowering plant, was conducted for resistance to *Radopholus similis* since resistant anthurium varieties were unavailable. Modified rice cysteine protease D86), which is

known to interfere with many plant-parasiticΔinhibitor (OcI nematode digestive systems resulting in reduced nematode growth and development, was transformed into two anthurium varieties: 'Marian Seefurth' and 'Paradise Pink', via Agrobacterium-mediated transformation of explants of etiolated shoot or leaf. Selective media (½-strength MS containing 50 mg/l kanamycin and 500 mg/l carbenicillin) containing two different combinations of hormones, 0.4 mg/l 2, 4-D and 0.2 mg/l BA or 0.2 mg/l IBA and 0.1 mg/l TDZ, were compared for callus induction. Faster callus induction and higher number of callused explants were obtained from etiolated shoot explants cultured on selective media supplemented with 0.2 mg/l IBA and 0.1 mg/l TDZ.

EFFECTS OF MULTIGUARD™ PROTECT ON ROOT-KNOT NEMATODE POPULATIONS AND DISEASE OF TOMATO AND PEPPER. **Kokalis-Burelle, N.** USDA, ARS, U.S. Horticultural Research Lab, Fort Pierce, FL 34945.

The effects of Multiguard Protect, a commercial formulation of furfural, on plant growth, nematode populations in roots and soil, and galling caused by *Meloidogyne spp.* were assessed in two greenhouse trials. 'Tiny Tim' tomato (*Lycopersinon esculentum*) and 'Capistrano' bell pepper (*Capsicum annuum*) were transplanted into pots containing field soil naturally infested with a mixed population of root-knot nematodes, as well as other pathogenic and microbivorous nematodes, and soil microorganisms. Multiguard treatments were applied as soil drenches at: 1) 452 kg/ha 7 days preplant; 2) 452 kg/ha 7 days preplant + 28 kg/ha 2 weeks post-plant; 3) 85 kg/ha 7 days preplant; 4) 85 kg/ha 7 days preplant + 28 kg/ha 2 weeks post-plant; 5) 28 kg/ha 2 weeks post-plant; 6) untreated control. The experiments were performed twice, once in fall 2005 and repeated winter 2006. No phytotoxicity or root disease, other than galling, was observed on either crop with any treatment. Multiguard significantly increased root-knot and other parasitic nematode populations in both roots and soil at 452 kg/ha in tomatoes and peppers in the fall trials compared to the untreated controls and lower rates Multiguard. However, galling was still significantly reduced on tomato at the 452 kg/ha rate in both trials. Galling on pepper was significantly lower than tomato and was not reduced by Multiguard treatments in either experiment. At high rates, Multiguard appears to have an effect on either the host plant or the nematode that inhibits gall formation in tomato.

ENTOMOPATHOGENIC NEMATODES FOR CONTROL OF CODLING MOTH IN APPLES AND PEARS: OVERCOMING OBSTACLES OF ENVIRONMENT AND ATTITUDE. **Lacey, L. A.** Yakima Agricultural Research Laboratory, USDA-ARS, WA 98951.

Codling moth (CM), a serious pest of apple and pear in most countries where these fruits are grown. Control of overwintering CM larvae would reduce or eliminate damage to fruit early in the following growing season. Entomopathogenic nematodes have shown promise as biological control agents of cocooned CM larvae in the Pacific Northwest, but several factors warrant investigation to provide growers with practical control options. Field trials with *Steinernema carpocapsae* and *S. feltiae* were conducted in apple and pear orchards to determine the effects of seasonal temperatures, post-application irrigation and method of application on larval mortality. Trials in late summer, fall and early spring, using application rates 10^6 infective juveniles (IJ)/tree plus supplemental wetting to aid survival of IJ revealed that the best control (94–95% mortality in sentinel CM larvae) when mean temperatures above 20°C. When temperatures dropped to a mean of 12–13°C control by *S. feltiae* was still effective (90% mortality), but *S. carpocapsae* was significantly less effective (58% mortality). Application of *S. carpocapsae* and *S. feltiae* (2.0×10^6 IJ/tree) with an airblast sprayer, provided results that were comparable to that of treatment of individual trees with a hand-held lance applicator. Tests in the fall with *S. carpocapsae* and *S. feltiae* applied with an airblast sprayer ($1 - 2.5 \times 10^9$ IJ/ha) in 4-year-old trellised apple and established Bartlett pear orchards provided effective control when orchards were kept wet for several hours. Compared with bare ground, wood chip mulch enhances control by providing cocooning sites for CM larvae and a substrate that is easy to treat, maintains moisture, and enhances nematode activity. Applications of *S. carpocapsae* and *S. feltiae* at a rate of 2.5×10^9 IJ/ha to bare and wood chip-mulched plots followed by 1 h of irrigation resulted in 21 and 65% reduction in sentinel larvae in bare plots compared to 93 and 85% in mulched plots, respectively.

WHOLE GENOME ANALYSIS OF *HETERODERA GLYCINES*, THE SOYBEAN CYST NEMATODE. **Lambert, K., S. Bekal, T. L. Niblack, M. E. Hudson and L. L. Domier.** Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

Heterodera glycines, the soybean cyst nematode (SCN), is the most devastating pathogen of soybean in the Midwestern United States, causing hundreds of millions of dollars in damage annually. To implement any genomics-based method of nematode control, DNA sequence information on the target organism is required. To this end, we have recently sequenced the SCN genome to a three-fold redundancy using a massively parallel microbead sequencing procedure. In this project, we are functionally classifying predicted SCN genes based on their homology to genes of other organisms. To correlate expression with function, we are using quantitative RT-PCR on a subset of new genes that appear to play critical roles in the life cycle of SCN. These new SCN genes are being assessed for expression in all developmental stages of the SCN life cycle. In addition to our gene expression analysis, we are developing resources for genome analysis that will allow us to conduct statistical genetics of important SCN traits. To accomplish this, we are identifying thousands of DNA sequence

polymorphisms between SCN biotypes that differ in virulence and host range. These DNA polymorphisms, such as single nucleotide polymorphisms (SNP) and simple sequence repeats (SSR), are being converted into easy to use molecular markers that will allow the identification of loci that control the ability of the nematode to grow on "SCN-resistant" soybean, as well as other important SCN phenotypes.

TIMING OF BROADLEAF TOBACCO POST-HARVEST TILLAGE AFFECTS TOBACCO CYST NEMATODE POPULATION DEVELOPMENT. **LaMondia, J. A.** The Connecticut Agricultural Experiment Station Valley Laboratory, Windsor, CT 06095.

Tobacco cyst nematode (TCN), *Globodera tabacum tabacum*, is a damaging pathogen of broadleaf cigar wrapper tobacco. Broadleaf is a stalk-cut tobacco type typically harvested 55 to 65 days after transplanting. Growers commonly stagger planting over a period of several weeks and harvest over a similar, staggered time frame. A common practice has been to leave cut stalks in the field until an entire field or farm is harvested. The fields are then tilled and planted to a winter cover crop. We performed experiments over several years to determine cyst nematode populations after immediate tillage to break up roots compared to leaving cut stalks and roots in the field from 2 to 6 wks after harvest before tillage and planting to a cover crop. Plots tilled immediately after harvest had consistently lower final TCN populations ($P < 0.02$ to 0.0003) and Pf/Pi ratios when compared to plots with delayed tillage 2 to 6 wk after harvest. Final TCN densities in soil after immediate tillage ranged from 64% to 18.5% of densities after tillage 2 wk or 6 wk after harvest, respectively. A factorial experiment with Nemacur or no Nemacur and early (immediately after harvest) or late tillage (5 wk later) demonstrated that both nematicide and tillage timing affected TCN final populations in soil. There were no significant interactions. When tillage of stalks and roots is delayed until harvest is complete on an entire field or farm, it allows late first generation or early second generation females to develop. Timely destruction of roots slows the increase of *G. t. tabacum* to damaging levels in soil (Pf/Pi ratios ranged from 0.65 to 2.21 after immediate tillage compared to 1.0 to 6.78 for later tillage) and reduces the need for chemical management to avoid yield loss.

AVICTA COMPLETE PAK AND TEMIK 15G FOR RENIFORM NEMATODE MANAGEMENT ON COTTON. **Lawrence, G. W. [1], K. S. Lawrence,[2] and J. Caceres[1].** [1]Department of Entomology and Plant Pathology, Mississippi State University, Mississippi State, MS 39762 and [2]Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849.

Avicta Complete Pak and Temik 15G were examined in Mississippi and Alabama for their effects on cotton production. The fields were located near Glen Allan, MS, and Belle Mina, AL and were naturally infested with the reniform nematode. Avicta Complete Pak is a seed treatment consisting of Avicta 4.17 FS (abamectin), Dynasty CST 125 FS, and Cruiser 5 FS applied by the manufacturer. Temik 15G (aldicarb) was applied in the seed furrow at the time of planting at a rate of either 4.0 or 5.7 kg formulated product/hectare. Cotton growth measurements were collected at 28 and 60 days after planting (DAP). There were no significant differences for plant height or nodes produced between treatments at either location. Reniform numbers were numerically lower in the Avicta and most of the Temik 15G treatments than the control at 28, 60, and 90 DAP in MS and at 30 DAP in AL. Fewer reniform nematodes were recovered in the Temik 15G (5.7 kg/ha) treatment 155 DAP. Seed cotton yields were significantly improved over the control with the addition of Avicta Complete Pak in Mississippi. Cotton yields were 3566, 3365 and 3078 kg seed cotton/ha in the Avicta Complete Pak, Temik 15G and control treatments, respectively in MS. Similar yield increases were observed in AL with 3965, 3908, and 3779 kg seed cotton/ha for the Avicta Complete Pak, Temik 15G and control treatments, respectively.

POULTRY LITTER AS AN ORGANIC MEANS OF *ROTYLENCHULUS RENIFORMIS* MANAGEMENT ON COTTON. **Lawrence, K. S.,[1] G. W. Lawrence,[2] and S. R. Usery.[1]** [1]Department of Entomology & Plant Pathology, Auburn University, Auburn, AL 36849 and [2]Department of Entomology & Plant Pathology, Mississippi State University, Mississippi State, MS 39762.

Evaluations were conducted in a greenhouse, microplots and in the field to determine the effect of poultry litter on *R. reniformis* populations, soil microflora, and cotton yields. In the greenhouse, poultry litter was examined in test 1 at rates of 1, 2, 3, 4, 5, and 6 tons per acre and in test 2, two types of poultry litter; house litter and compost litter were evaluated. Microplots and field plots were also amended with poultry litter at rates of 1, 2, 3, 4, 5, and 6 and 1, 3, and 6 tons per acre respectively and compared to aldicarb (5.6 kg/ha). Results obtained from greenhouse evaluations indicate that poultry litter has a negative effect on *R. reniformis* population development. Numbers of R. reniformis were reduced by 55% and 72% at the lowest and highest poultry litter amendment levels, respectively. The application of house litter reduced *R. reniformis* eggs per gram of root by 279.5% and compost litter reduced population 107.4% as compared to the inorganic fertilizer equivalents. Bacterial populations were increased by poultry litter by an average of 220%. In microplots, poultry litter applications at increasing rates of 2 to 6 tons per acre were similar to aldicarb in reducing *R. reniformis* population levels and increased seed cotton yields. The addition of poultry litter to *R. reniformis* infested field plots significantly improved

seed cotton yields over the standard aldicarb and untreated control. These evaluations confirm that poultry litter has the potential to reduce *R. reniformis* in cotton.

OCCURRENCE OF ROOT-LESION NEMATODES, *PRATYLENCHUS* SPP., FROM CHRYSANTHEMUMIN KOREA. **Lee, Jae-Kook,[1] Dong-Ro Choi,[1] Hye-Rim Han,[2] Byeong-Yong Park,[3] and Dong-Geun Kim.[4]** [1]Applied Entomology Division, National Institute of Agricultural Science and Technology, RDA, Suwon; [2]Korea Forest Research Institute, Seoul; [3]National Institute of Crop Science, RDA, Suwon, Korea; [4]Gyeongbuk Agricultural Technology Adminstration, Daegu, Korea.

Plant-parasitic nematodes were surveyed on 50 major chrysanthemum greenhouses from different regions of Korea from May to Jun 2005. The most commonly encountered genera were *Pratylenchus* and *Paratylenchus* species. *Pratylenchus* spp. were detected from 43 samples among the 50 greenhouses and the average density was 1,095 nematodes/200 g soils+1 g root. The *Pratylenchus* spp. were identified as *P. penetrans, P. coffeae*, and *P. vulnus*. When spray chrysanthemum cv 'Chopin' was transplanted in September 2004 in a greenhouse infested with *Pratylenchus vulnus*, the nematode density increased from 854 nematodes/100 g soil in planting time to 14,985 nematodes/100 g soils+1 g root after 10 weeks at harvest time. About 87% of population of *P. vulnus* distributed to the depth of 0–30 cm. Shoot weight and shoot height decreased 24.8% and 8%, respectively, compared with plant grown in non-infested soil. *Pratylenchus* is the major plant-parasitic nematode in chrysanthemum grown under greenhouse in Korea.

WORMBASE: AN INFORMATION RESOURCE FOR RESEARCH OF *CAENORHABDITIS ELEGANS* AND RELATED NEMATODES. **Lee, R., and The WormBase Consortium.** Division of Biology, California Institute of Technology, Pasadena, CA 91125.

The goal of WormBase is to provide a knowledge environment to facilitate further research with *C. elegans* and related nematodes. We maintain a database, extract information from the literature, and disseminate integrated knowledge via the WWW. Information includes complete genome sequences, developmental cell lineages, neural circuit maps, and extensive genetic analyses that make functional connections between the genome, development, anatomy, and physiology. To allow rapid and easy access of available content, WormBase provides Web pages that focus on special topics such as Gene page, which contains summaries of everything that is known about a gene. Also, users can synthesize complex queries using tools such as WormMart, an interface based on BioMart. For power users, we offer FTP downloads of all of our data and software, and we provide directions for building local mirrors. To keep up with the rapid growth of research literature, we update our database every three weeks. But to ensure comparability between large-scale analyses, we maintain a stable version every ten releases. Completing its functionality as a knowledge portal, WormBase integrates with related services: Textpresso, a primary literature search engine, and WormBook, a collection of expert-written chapters about *C. elegans* biology. Since its inception in 2000, WormBase has become an indispensable resource for *C. elegans* researchers. Going forward, a new focus will be on comparative genomics as several other nematode genomes are being sequenced.

GENETIC LINKAGE MAP OF ROOT-KNOT NEMATODE, *MELOIDOGYNE HAPLA*. **Liu, Qingli, and V. M. Williamson.** Department of Nematology, University of California Davis, CA 95616.

Meloidogyne hapla has emerged as a model organism for studying plant and root-knot nematode interactions. Six strains of this species were characterized with regard to host specificity and DNA polymorphisms visualized as AFLP markers. Two of these strains, VW8 and VW9, differing in their pathogenicity on wild potato, SB-22, and common bean cv. NemaSnap were chosen for genetic crosses. By monitoring crosses with DNA markers, 183 F2 lines were produced. Heterozygous *M. hapla* females produce progeny upon parthenogenetic reproduction that segregate 1:1 for DNA markers rather than the 1:2:1 ratio typical of a Mendelian cross. This phenomenon can be explained since, in the absence of males, the diploid state is restored by reuniting sister chromatids in a single meiosis. In this model, meiotic parthenogenesis would result in rapid genomic homozygosity. Analysis of marker segregation has identified linkage groups and construction of a genetic linkage map is in progress. The current goal of this project is to construct a genetic linkage map with approximately 200 DNA markers. In parallel with the ongoing genome sequencing of *M. hapla*, this map will provide the framework for mapping phenotypic markers and candidate genes involved in infection and/or parasitism.

AVICTA COMPLETE PAK: SEED-DELIVERED TECHNOLOGY FOR THE MANAGEMENT OF EARLY-SEASON DISEASES, INSECTS AND NEMATODES IN COTTON. **Long, D.,[1] A. Cochran,[2] and D. Hofer[2].** Syngenta Seed Care, [2]Greensboro, NC 27419 and [2]Basle, Switzerland.

AVICTA Complete Pak is a promotional combination of three separately registered products: AVICTA seed treatment nematicide, Cruiser seed treatment insecticide and Dynasty CST seed-delivered fungicide, that offers cotton growers a convenient and innovative management tool against early season diseases, insects and nematodes. AVICTA, active ingredient abamectin, was registered on cotton in 2005 and is considered one of the most potent contact, non-systemic nematicides ever discovered. Over the past several years, AVICTA Complete Pak has undergone extensive in-house and

independent research comparing the seed treatment nematicide (AVICTA) to the standard in-furrow treatment of aldicarb (TEMIK 15 G), on root-knot, reniform and Columbia lance nematodes. In trials conducted from 2000 to 2005 by university researchers and independent crop consultants, AVICTA Complete Pak was shown to produce yields equal to or greater than Temik (5 lbs/A) 80 percent of the time.

PARASITISM GENES IDENTIFIED IN THE POTATO CYST NEMATODE, *GLOBODERA ROSTOCHIENSIS*, USING A COMPARATIVE GENOMIC APPROACH. **Lu, Shunwen, D. Tian, X. Deng, D. M. Thurston, and X. Wang.** Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Stylet secretions are encoded by parasitism genes expressed in the esophageal gland cells of plant-parasitic nematodes. These are the primary signals that facilitate nematode migration in host roots and control the formation of the elaborate feeding cells necessary for the development and reproduction of the nematode. Cyst nematodes of genera *Heterodera* (e.g. the soybean cyst nematode, SCN, *H. glycines*) and *Globodera* (e.g. the potato cyst nematode, PCN, *G. rostochiensis*) are devastating pests of agriculturally important crops in the US. Both SCN and PCN induce syncytia in their host roots for feeding. BLAST searches using fifty parasitism protein sequences of SCN against approximately 6,000 PCN ESTs have led to the discovery of ten new putative parasitism genes in PCN. Seven of them were further cloned including *CLE*-like gene (*GrCLE1*), venom allergen-like gene (*GrVAP1*), ubiquitin extension gene (*GrUBI1*), *SKP1*-like gene (*GrSKP1*), chorismate mutase gene (*GrCM1*), and two pioneer genes (*Gr4E02* and *Gr33E05*) corresponding to *4E02* and *33E05* of SCN. Southern blot analysis confirmed the presence of these genes in PCN. *GrCLE1*, *GrCM1* and *GrUBI1* were found to be expressed exclusively within esophageal gland cells of the nematode using mRNA in situ hybridization analysis. In addition, all seven genes except for *Gr4E02* were also expressed in parasitic stages, suggesting an important role in nematode parasitism of hosts. Further identification and functional studies of PCN parasitism genes will lead to a better understanding of the PCN-potato interaction and may provide targets for developing novel strategies to engineer PCN resistance in potato.

BIOLOGY OF A SEXUALLY TRANSMITTED NEMATODE (*MEHDINEMA ALII*) IN THE INDIAN HOUSE CRICKET (*GRYLLODES SIGILLATUS*). **Luong, L.** Department of Biological Sciences, University of Cinncinatti, Cinncinatti, OH 45221.

Sexually transmitted diseases (STD) are common throughout the animal kingdom. While most vertebrate STD are viruses and bacteria, the majority of invertebrate STD are macroparasites, such as mites, fungi, protists, and nematodes. Sexually transmitted nematodes are ubiquitous among insects and exhibit life histories that range from purely phoretic to entirely parasitic. These nematodes provide a good example of how parasitic nematodes might have evolved from phoretic ancestors. The two most well studied nematode STD are the ectoparasitic *Noctuidema guyanese*, found on noctuid moths, and the endoparasitic *Mehdinema alii*, which infects decorated crickets. Each nematode exhibits varying degrees of prevalence and pathogenicity. For example, *M. alii* only infect male hosts; female crickets serve exclusively as vectors for transmission to susceptible males. Field surveys and laboratory experiments show that female crickets are refractory to infection. As a result, host pathology is restricted to males, which produce smaller spermatophores than uninfected males. There are minimal effects on host mortality, which is consistent with theoretical predictions for STD in general. Moreover, STD are expected to have different ecological and evolutionary implications than other infectious diseases. Specialization for sexual transmission may account for this nematode's unusual morphology as well.

RESPONSE OF *PRATYLENCHUS PENETRANS* TO DECOMPOSING PLANTS: FROM LAB TO FIELD. **MacGuidwin, A. E.,[1] W. L. Petersen,[1] and D. L. Sexson.[2]** [1]Department of Plant Pathology, [2]Horticulture Department, University of Wisconsin-Madison, Madison, WI 53706.

Volatiles released from decomposing residue of Brassica spp are known to be nematicidal. We studied rapeseed (*Brassica napus*) and four other plant species, representing a range of bioactive compounds, for their effect on *Pratylenchus penetrans*; sorghum-sudangrass, African marigold, forage pearl millet, and corn. Laboratory experiments to test the impact of plant volatiles on nematodes used air-tight containers filled with 3.75 to 60 g fresh leaves mixed into 300 g soil. Nematodes in water-filled dishes were placed on the surface of the soil for seven days. All plant amendments induced greater mortality than the soil-only control for at least one level of residue. Rapeseed was the most effective in killing nematodes. The five cover crops were planted in a commercial field in May and incorporated into soil in July. The experiment was a split-plot with crop as the main plot and covering soil after plant incorporation (+/-) in July as subplots. The covers remained in place for two weeks. Potato was planted the following year. In the fall after planting cover crops there were significant main effects of crop and covering soil. Population densities of *P. penetrans* were lower in millet and marigold plots than in sorghum-sudangrass and rapeseed plots. Covered plots had fewer nematodes than plots left uncovered. The differences among the cover crop plots were not detectable in the spring when potatoes were planted, but there were fewer nematodes in the plots that had been covered. At the time of potato harvest, the effects of the previous year's cover crop were still discernible; population densities were lowest in millet plots. The discrepancy in the efficacy

of rapeseed between laboratory and field and the low efficacy of rapeseed relative to other cover crops in the field suggest that factors in addition to plant volatiles are responsible for reducing population densities of *P. penetrans* in the field.

CAUSES AND (SOME) CONSEQUENCES OF BIOLOGICAL INVASIONS. **Mack, R. N.** School of Biological Sciences, Washington State University, Pullman, WA 99164.

Biological invasions are those immigrations of organisms to areas outside the species' native range that result in the persistence and eventual proliferation and spread of the immigrants' descendants. The consequences of these invasive alien species (IAS) for the native biota, the environment and even the economy in the new range are often profound and chronic. Despite the accelerating frequency of emigrations of organisms worldwide, invasions remain comparatively rare: the number of invasions to any area is orders of magnitude smaller than the numbers of immigrations. This comparative rarity has long prompted the pursuit of evidence by which the identity future invaders could be predicted, thereby greatly facilitating steps to control or even prevent them altogether. At least 5 non-mutually exclusive lines of evidence have been investigated to shape predictions: commonality of traits or attributes among invaders worldwide, similarity of the climate in native and invaded ranges, lack of one of more forms of biotic resistance to the immigrant in the new range (e.g. competitors, grazers, predators and parasites), novel disturbance in the new range that is tolerated by the immigrants but not by the natives, and tolerance/avoidance of environmental stochasticity. Each line of evidence has support, yet each lacks a sufficiently wide range of examples to be deemed inclusive and the sole predictor. These investigations are warranted because the collective damage of IAS is enormous. In the United States the financial bill totals >140 billion dollars/year and is growing. The intrinsic costs to the nation's natural biodiversity are more worrying, as the conservation of many native species is threatened by aggressive invasive species in all biomes (freshwater, terrestrial and marine). Furthermore, biological invasions will likely provide bewildering new complications as invasive species react to global atmospheric changes and in some cases even contribute directly to these changes.

PREVALENCE AND SPECIES IDENTIFICATION OF ROOT-LESION NEMATODE IN POTATO SOILS OF MANITOBA, CANADA. **Mahran, A.,[1,3] M. Tenuta,[1] T. Shinners-Carnelley,[2] and F. Daayf.[3]** [1]Department of Soil Science, [2]Manitoba Agriculture, Food, and Rural Initiatives, [3]Department of Plant Science, University of Manitoba, Winnipeg MB Canada R3T 2N2.

The root-lesion nematode, *Pratylenchus* spp. (Cobb) Chitwood and Oteifa is a serious pest of potato (*Solanum tuberosum* L.*)* throughout most potato producing areas of North America. Yield reduction to potato varies with species of the root lesion nematode. The species of *Pratylenchus* dominant in potato fields in the eastern provinces of Canada (Prince Edward Island, New Brunswick, Quebec, Ontario) is *P. penetrans*. In Manitoba, Canada's second largest potato producing province, surveys conducted in 1991, and 2002 to 2004 showed that *Pratylenchus* spp. was present in 33% of a total of 135 fields sampled with populations ranging from 4 to 5,300 nematodes/kg soil. None of the surveys identified the species of *Pratylenchus* present in Manitoba. The objective of this study was to determine the prevalence and species identification of *Pratylenchus* spp. in Manitoba potato fields using morphometric characters and molecular techniques (PCR). A survey was undertaken sampling 21 potato fields with a history of Early Dying of Potato which is known to be aggravated by *Pratylenchus* spp. Fifteen females were chosen and morphological characters measured for species differentiation. Results of the survey showed that *Pratylenchus* spp. was found in six of the 21 fields with population ranging from 45–631 nematodes/kg soil. Morphometrics of the females indicated the species present in Manitoba is *P. neglectus*. For molecular species diagnostics, approximately one hundred *Pratylenchus* spp. were hand picked from soil from each field sample, their DNA extracted and PCR with species-specific primers used to identify the species of *Pratylenchus*. The molecular diagnostics of the *Pratylenchus* spp. species will be reported. The presence of *P. neglectus* rather than *P. penetrans* is encouraging to producers in Manitoba. The nematode *P. neglectus* has a higher threshold level of individuals in soil without causing disease. Additionally it also does not interact synergistically with *Verticillium* spp. to the same degree as *P. penetrans* to cause Early Dying Disease of Potato.

BIOLOGICAL SOLUTIONS FOR NEMATODE CONTROL. **Marrone**, **P.** AgraQuest, Inc. Davis, CA 95616.

In high value specialty crops such as grapes, tomatoes and flower bulbs, biological pesticides have potential for controlling nematodes as well as and as cost effectively as synthetic chemicals. This talk will provide an overview of the process of discovery of new biological nematicides and review the biological pesticides that are currently on the market and the products in development for nematode control. Examples include products based on *Myrothecium*, *Paecilomyces lilacinus, Pasteuria penetrans* and the biofumigant, *Muscodor albus*.

DEVELOPMENT OF HYPOTHESES ON THE PHYSIOLOGICAL FUNCTIONS OF A COMPLEX PEPTIDE FAMILY IN NEMATODES FROM A PHYLOGENETIC PERSPECTIVE. **Masler, E. P., and L. K. Carta.** Nematology Laboratory, Agricultural Research Service, United States Department of Agriculture, Beltsville, MD 20705.

Neuropeptides control essentially all physiological processes in animals. In vertebrates, and the higher invertebrates such as molluscs and insects, there exist numerous neuropeptide families servicing an equally large array of physiological

functions. Each family consists of relatively small numbers of related amino acid sequences. In the lower invertebrates, including the nematodes, the situation is strikingly different. Few neuropeptide families have been identified, but the number of different sequences within each family is remarkably high. The most prominent neuropeptide family in nematodes comprises the FMRFamides, whose members are characterized by an amidated Arg-Phe C-terminus and an involvement with neuromuscular control. In *Caenorhabditis elegans*, at least 23 different FMRFamide-like peptide genes (*flp*s) encoding over 60 unique FLP sequences have been identified. Multiple genes encoding numerous FLPs have been reported in parasitic species as well. EST and amino acid sequence evidence suggest that FLPs are highly conserved within nematodes and are of fundamental importance to these animals. Basic physiological questions such as "Why are there so many iterations of the FLPs in nematodes?" and "Do these peptides have roles in addition to muscular control?" may be addressed with hypotheses for candidate phenotypes within phylogenetic frameworks. For example, an analysis of recently published data indicates that *flp* ESTs represent a greater percent of all ESTs in parasitic nematodes than in free-living species. Such a hypothetical comparison, and other such comparisons involving nematode behavior, locomotion, viviparity, etc., can be refined using current phylogenies to identify candidate peptides for experimental testing.

EFFICACY OF AGRI-TERRA AGAINST NEMATODES ASSOCIATED WITH MAJOR CROP SPECIES IN LOUISIANA. **McGawley, E. C., M. J. Pontif, J. M. Bruce, and C. Overstreet.** Department of Plant Pathology and Crop Physiology, Louisiana State University, Baton Rouge, LA 70803.

Agri-Terra has proven to be a safe and efficacious material for the management of many economically important plant parasitic nematode species in Louisiana. Popular cultivars of all major row crops plus a wide variety of vegetable species (tomato, bell pepper, cucumber, cabbage and lettuce) were evaluated in microplot and field trials during the period 2000–2005. In every trial, significant reductions in indigenous or introduced nematode populations were observed following application of this material. Harvest data from microplot trials, averaged over years, showed increases in plant dry weights over those of controls which averaged 59% for cotton (four trials), 44% for sugarcane (three trials), 59% for rice (two trials), and 42% for soybean (two trials). In field trials in which Agri-Terra was employed as an at-planting, in-furrow spray treatment (10 GPA of a 100:1 dilution), significant yield responses, when compared with those of non-treated controls, were observed in five consecutive years with cotton and in two of three years with soybean. In a multi-year field trial with sugarcane the first ratoon crop produced a significant increase in the sugar content per ton of sugarcane.

DEVELOPMENT OF A NEMATODE-ROOTSTOCK PROFILE FOR *PRUNUS* SPP. **McKenry, M., S. Kaku, and T. Buzo.** Department of Nematology, University of California, Riverside, CA 92521.

European and American breeders of Prunus rootstocks have been searching for nematode resistance combined with tolerance to nematodes and the replant problem. A two-year evaluation of 45 contemporary and new Prunus rootstocks is underway. This is occurring in field settings with candidates individually infested with *Pratylenchus vulnus, Meloidogyne incognita*, and *Mesocriconema xenoplax*. Thirty of these rootstocks exhibit resistance to *M. incognita*, usually a result of Nemaguard (*Prunus persica* x o.p.) parentage but resistance also comes from peach cultivars such as Okinawa. Only two rootstocks exhibit resistance to *P. vulnus*. Their parentage includes the plum stocks *Prunus cerasifera, P. tomentosa* or *P. incana*. Eight of the candidates reduced *P. vulnus* reproduction to numerically less than that of Nemaguard with Garnem and Bright's Hybrid-4 exhibiting moderate resistance. Many of the peach x almond crosses offer resistance to *M. incognita* and some provide reduced buildup of *P. vulnus*, but also support higher population levels of *M. xenoplax*. In our two-year evaluations Lovell (*P. persica*) supports half the *M. xenoplax* population of Nemaguard. In several seven-year field trials the buildup of *M. xenoplax* on Viking rootstock (Nemaguard x [almond x plum]) is half that of Lovell. Our goal is to identify rootstocks with parentage different from Nemaguard in order to combat the rejection component of the replant problem while limiting the soil pest and disease component of the replant problem.

NEMATOLOGICAL AND AGRONOMIC APPROACHES TO NUTRIENT USE EFFICIENCY: BRIDGING THE GAPS. **Melakeberhan, H.** Department of Entomology, Michigan State University, East Lansing, MI 48824.

In light of multiple restrictions on many pesticides and nematicide uses, emphasis on developing ecologically- and economically-friendly food and fiber production systems is likely to increase. Ecologically sound production systems are likely to include organic plant and animal residues, with significant implications on soil nutrient dynamics, which, in turn, can affect pest management and agronomic practices. For example, increasing soil nutrition can cause a plant to perform better under nematode (and other pest) stress compared to low nutrient conditions. In order to best use soil nutrients regardless of how they are added to the soil, however, it is important to understand nutrient use efficiency (NUE). Defined as the amount of yield per unit of nutrient element (e.g., N, P, K), NUE is often used to identify agronomic and ecological adaptation traits. Where no nutrient elemental analysis is available, fertilizer use efficiency (FUE) will suffice. The terms NUE or FUE however may have slightly different meaning to an agronomist, who looks at plant productivity (eg. growth

or photosynthesis), and to a nematologist, who includes nematode population dynamics; hence, the need to bridge the gaps. For example, when fertilizer is applied, plant productivity (Y-axis) may or may not increase, and with or without correlation to nematode population density (X-axis). By expressing the data on the same scale (as percent of control) and plotting the relationships of X and Y, it is possible to distinguish FUE categories of efficient for: (A) host and nematode, (B) host, (C) nematode, and (D) neither. If the interactions fall in Category A, best case FUE scenario, fertilizer application can be a stand-alone management strategy. If the interactions result in Categories B and C, complimentary nematode suppressing and host productivity improving tactics, respectively, would have to be added. If the interactions result in Category D, there is no need to apply fertilizer. The agro-ecological and economic implications of identifying FUE will be discussed using proven examples.

HISTOLOGY OF RESISTANCE REACTION IN WHITE CLOVER TO *HETERODERA TRIFOLII*. **Mercer, C. F.** AgResearch, Grasslands Research Centre, Private Bag 11008, Palmerston North, New Zealand.

Heterodera trifolii is a pest of white clover (*Trifolium repens*) in New Zealand pasture and a recurrent selection program has developed resistant germplasm. Greenhouse experiments used recently hatched *H. trifolii* second-stage juveniles (J2) pipetted around the roots of 2-week-old stolon tip clones of white clover genotypes resistant or susceptible to *H. trifolii*. Plants were inoculated in petri dishes of sand at 22°C. After 24 hours, roots were washed free of sand particles and suspended, in aerated tap water, at 22°C. After a further 24 hours, roots were stained in acid-fuschin, the J2 counted and their invasion stage categorized. Syncitia, with or without associated J2, were also counted. In susceptible genotypes, more J2 were seen with heads inserted into the stele near a small syncitium, than in resistant material. In resistant genotypes, more J2 with straight bodies and head still in the cortex but near a syncitium, were seen than in susceptible genotypes. There was a large difference between resistant and susceptible genotypes in the number of syncitia which had started to form but were not associated with a nearby J2. Numbers of these unassociated syncitia were greater in resistant genotypes than in susceptible ones and apparently reflected the greater number of J2 which failed to set up an effective syncitium and quit the root before it was stained. Results to date suggest that resistance acts at the syncitium initiation stage, but improved techniques, such as a shorter inoculation period, will be tested before comparing white clover genotypes for successful entry of J2 into the roots.

PLANT-DERIVED COMPOUNDS ACTIVE AGAINST *MELOIDOGYNE INCOGNITA*. **Meyer, S. L. F.,[1] I. A. Zasada,[1] D. P. Roberts,[2] B. T. Vinyard,[3] D. K. Lakshman,[4] J.-K. Lee,[5] D. J. Chitwood,[1] and L. K. Carta.[1]** USDA, ARS, [1]Nematology Laboratory, [2]Sustainable Agricultural Systems Laboratory, [3]Biometrical Consulting Service, [4]Floral and Nursery Plants Research Unit, US National Arboretum, Henry A. Wallace Beltsville Agricultural Research Center, BARC-West, Beltsville; and [5]Nematology Laboratory, Applied Entomology Division, Korean National Institute of Agricultural Science and Technology, Suwon.

Products from two plant genera, *Plantago* and *Eugenia*, were tested for effects on the root-knot nematode *Meloidogyne incognita*. Extracts from *P. lanceolata* and *P. rugelii* were also evaluated for toxicity to the plant-pathogenic fungi *Fusarium oxysporum* f. sp. *gladioli*, *Phytophthora capsici*, *Pythium ultimum*, and *Rhizoctonia solani*, and to the beneficial microbes *Enterobacter cloacae*, *Pseudomonas fluorescens* and *Trichoderma virens*. *Plantago* extracts were prepared in methanol and in water from dried shoots and roots of field-collected plants. Clove oil from *E. caryophyllata* was purchased from Sigma Chemical Company. All *Plantago* and *Eugenia* extracts were lethal to second-stage juveniles (J2) of *M. incognita*, and also inhibited egg hatch. J2 were more sensitive than eggs to lower concentrations of the *Plantago* extracts (25% and 50%), but effects on the two life stages were similar at 75% and 100% concentrations. J2 and egg hatch activity decreased by 65%–72% at the 75% concentration. The *Plantago* extracts were not toxic to the tested bacteria and fungi. Direct exposure to 0.2% clove oil resulted in 100% loss of J2 viability, and inhibited egg hatch 97.5%. Clove oil did not exhibit as pronounced a difference in toxicity to eggs and J2 as was observed with *Plantago* extracts.

NEM-FEM: A DATABASE OF CODING NEMATODE-SPECIFIC MULTI-SPECIES CONSERVED SEQUENCES. **Mitreva, M., Y. Yin, J. C. Martin, S. Abubucker, J. P. McCarter, S. W. Clifton and R. K. Wilson.** Genome Sequencing Center, Washington University School of Medicine, St. Louis, MO 63108.

Proteins with specificity to nematodes may serve as excellent targets for drugs with low toxicity to humans and other vertebrates or for environmentally safe pesticides. Guided by the importance of identifying these proteins and aided by the increase of nematode sequences in the public databases, we applied computational based methods that can detect highly conserved regions in a robust fashion. Our approach identified putative coding sequences conserved across the phylum Nematoda or nematode subgroups; we call these 'Nematode-specific Multi-species Conserved Sequences' (or NMCSs). We identified 274 NMCSs, with on average 8±4 species per group and an average alignment length of 174 ± 123 amino acids (aa). 88 of these (33%) had *Caenorhabditis elegans* gene with known RNAi phenotype following transient gene

knockout. In addition, 307 groups included only parasitic nematode species, therefore assigned as parasitism-specific, with an average of 6 ± 4 species per group and 134 ± 73 aa average alignment length. The strongest NMSC models are the ones spanning across the phylum Nematoda. We will report on the progress of identification of NMCSs including extensive functional classification for those with homology. Projects details available at www.nematode.net; project funded by NIH-NIAID-46593 grant.

DIFFEENTIAL RATE IN SPORE ATTACHMENT OF SEVEN ISOLATES OF *PASTEURIA PENETRANS* ON THE ROOT KNOT NEMATODE *MELOIDOGYNE JAVANICA*. **Mousa, E. M., and Mahdy, M. E.** Department of Agricultural Botany, Faculty of Agriculture, Menofiya University, Shebin El-Kom, Egypt.

Seven isolates of *Pasteuria penetrans* collected from Egypt (Pp EGY); Japan (Pp J); Great Britain (Pp GB); South Africa (Pp3 SA); Malawi (Pp Mal); Papua New Guinea (Pp PNG) and Barbados (Pp Bar) were tested for their difference in the spore attachment on the root-knot nematode, *Meloidogyne javanica*. Spore suspensions of seven *P. penetrans* isolates were adjusted to 5.0 × 105 spores/ml. Attachment studies were carried out in vitro on *M. javanica* juveniles. Results of the study showed that among the seven isolates of *P. penetrans*, three of them originated from Egypt; Japan and Great Britain showed 50% more attachment rate on *M. javanica* juveniles than the other isolates. All other four isolates had low attachment rate while Barbados isolate did not attached on the *M. javanica* juveniles. Greenhouse experiment on tomato plants with the above isolates confirmed the results of the study.

APPLYING A RIBOSOMAL DNA SEQUENCE "BARCODE" TO ASSESS NEMATODE BIODIVERSITY IN A COSTA RICAN RAINFOREST PRESERVE. **Mullin, P.,[1] T. Powers,[1] T. Harris,[1] A. Esquivel,[2] R. Giblin-Davis,[3] D. Neher,[4] and S. P. Stock.[5]** [1]Department of Plant Pathology, University of Nebraska, Lincoln, USA, [2]de Ciencias Agrarias, Universidad Nacional, Heredia, CR, [3]Institute of Food and Agricultural Sciences, University of Florida, Fort Lauderdale, USA, [4]Plant and Soil Science Department, University of Vermont, Burlington, USA, [5]Department of Entomology, University of Arizona, Tucson, USA.

In March, 2005 we sampled soil, litter, and understory habitats at four discrete locations within the La Selva Biological Station rainforest preserve in Costa Rica. Nematodes were extracted from composite samples and either preserved in formalin or tentatively identified to genus while alive and then prepared for PCR by crushing individual specimens in sterile water. Amplifications were performed on a subset of these specimens using a primer set targeting a portion of the 18S ribosomal repeat. A total of 360 nematode 18S sequences were generated via direct sequencing of the PCR products, and sorted into molecular operational taxonomic units (MOTUs) on the basis of primary sequence. MOTU determination was strict, with a single substitution considered sufficient to designate a unique "taxon". 167 unique nematode MOTUs have been identified and compared with small subunit sequences archived in GenBank to assess putative identifications and likely relationships. Most (109) MOTUs are represented by a single sequence, although multiple representatives of several have also been recovered. Total species richness estimates range from 464 to 502 for the sample area. Most major terrestrial nematode lineages are represented, with *Cephalobinae, Plectinae, Tylenchidae*, and *Dorylaimoidea* among the most frequently encountered. Conversely, relatively few sequences attributable to *Enoplida, Triplonchida, Chromadorida* or *Monhysterida* have been recovered. Phylogenetic analysis of MOTU sequences assigned to *Tylenchidae* along with GenBank sequences representing that family indicate that there are lineages present in the Costa Rican fauna that have not to date been sampled in SSU-based nematode phylogenies.

DNA ADDUCTS: QUANTIFICATION OF NEMATODE RESPONSE TO ENVIRONMENTAL STRESS. **Neher, D. A.,[1] and S. R. Stürzenbaum.[2]** [1]Plant and Soil Science Department, University of Vermont, Burlington, VT 05405; [2]School of Biomedical & Health Sciences, Pharmaceutical Sciences Research Division, King's College London, 150 Stamford Street, London SE1 7NH, UK

Bonger's (1990) nematode maturity index is based on inferences of life history characteristics from morphology. An independent empirical test of nematode response to environmental stress will improve our interpretation and application of these indices. We tested a proof of concept for DNA adducts, a molecular biomarker of DNA damage, in *Caenorhabditis elegans*. The approach is based on the concept that adducts in genomic DNA can block DNA polymerases, thus interfering with replication of the template DNA and decreasing the yield of product following PCR. We quantified bulky DNA adducts in a XL-PCR (16144 bp) target amplicon that was amplified from genomic lysate extracted from single nematodes. Amplification efficiency was assessed by means of a second, fully quantitative PCR. Following the normalization with an invariant control gene, adduct formation was evaluated by the identification of XL-PCR amplifications that were, relative to the control gene, reduced or inhibited. Adduct formation was demonstrated in individual nematodes exposed to 5 μg/g agar benzo[a]pyrene, 3 μg/g fluoranthene, and a mixture containing 0.5 μg/g benzo[a]pyrene and 1 μg/g fluoranthene, but not 20 μg/g copper. We conclude that adducts reflect DNA damage caused by chemical contamination of the environment. To our knowledge, this is among the first applications of DNA adducts as a molecular biomarker to ecological analysis.

NOVEL APPLICATION OF T-RFLP TECHNIQUES TO CHARACTERISE THE DIET OF THREE SPECIES OF PREDATORY *MONONCHIDAE*. **Neilson, R., J. Hesketh, and T. Daniell.** Scottish Crop Research Institute, Dundee, Scotland, DD2 5DA, UK.

Although predatory nematodes have a putative key role within soil foodwebs, little is actually known about the composition of their diet. Due to their small size, traditional gut content analysis prevalent in other soil dwelling organisms is not a consistent and feasible option to investigate diet. However, a few light microscopy studies have utilized sclerotized fragments of nematode prey residing in the gut of predatory nematodes to attempt to characterize the dietary intake of *Mononchidae*. Here we report the novel application of T-RFLP techniques to characterize the diet of specimens representing three species of predatory *Mononchidae, Anatonchus tridentatus, Mylonchulus striatus* and *Prionchulus puncatus*. An initial in silico screen of a database comprising of 18S rDNA nematode sequences with 20 restriction enzymes indicated that ECO-RI, RSA-I and DDE-I had discriminatory cut-sites that had the possibility of separating predatory and potential prey species. PCR products produced by fluorescent tagged primers were digested with the three identified enzymes. Downstream analyses revealed potential prey species for each of the predatory specimens examined. For some specimens investigated, four potential prey species were identified for other specimens a larger potential prey pool was identified. A better resolution could be achieved by a more extensive initial in silico screen of restriction enzymes to achieve a greater discriminatory power.

SOYBEAN CYST NEMATODE IN ILLINOIS FROM 1990 TO 2006: SHIFT IN VIRULENCE PHENOTYPE OF FIELD POPULATIONS. **Niblack, T. L., K. B. Colgrove, and A. C. Colgrove.** Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

Soybean cyst nematode (SCN) infestations in Illinois were surveyed in 1990 for their virulence phenotypes by Sikora and Noel (1991, Journal of Nematology 23:624–628) with the race differential soybean lines Pickett 71, Peking, Plant Introduction (PI) 88788, and PI 90763. They observed race 3 (64%), race 1 (27%), and low percentages of races 2, 4, and 5. Translated into more recent terminology for SCN virulence phenotypes, race 3 would be HG Type 0- (avirulent on Peking, PI 88788, and 90763) and race 1 would be HG Type 2-(virulent on PI 88788 only), the "-" indicating that the complete HG Type Test was not done. In practical terms, their results suggested that few of the SCN populations were virulent on the soybean cultivars available to growers at the time. In 2004 and 2005, samples of 212 SCN infestations in Illinois were collected from fields arbitrarily chosen by GPS coordinates along transects delineated by highways from which soybean fields were accessible. Of these populations, 175 were tested according to the Illinois SCN Type Test, a modification of the HG Type Test that includes only three of the sources of resistance (those currently available in Illinois): 1, PI 548402 (Peking); 2, PI 88788; and 4, PI 437654. We observed that 70% of the SCN populations were virulent on PI 88788, i.e., SCN Type 2 or 1.2. In addition, 27% were able to develop on PI 437654, although not at high enough levels to be designated SCN Type 4. These results suggest that identification and deployment of novel sources of resistance to SCN should be pursued vigorously.

CAL-AGRI PRODUCTS FOR THE MANAGEMENT OF IMPORTANT AGRICULTURAL PESTS. **Nielsen, D. I., R. B. Kemper, R. M. Steckler, and E. C. McGawley.** Cal Agri Products, LLC, Los Angeles, CA 90034.

Over the past six years, entomologists, plant pathologists and nematologists in California, Florida, North Carolina, New York, Minnesota, Louisiana, Idaho and in Spain, China, United Kingdom, Mexico, Saudi Arabia, Italy, Belgium, Australia, India, Indonesia, Korea, Pakistan, Vietnam and Morocco have evaluated the efficacy of the Agri 50 family of foliar pesticides and Agri-Terra against insects, phytoparasitic fungi and nematodes. This pesticide family has demonstrated efficacy against whiteflies, scale insects, selected aphid species and powdery mildew. This material is currently labeled in China, Morocco and in the European Union.

EFFICACY OF AGRI-TERRA AGAINST PHYTOPARASITIC NEMATODES. **Nielsen, D. I., R. B. Kemper, R. M. Steckler, and E. C. McGawley.** Cal Agri Products, LLC, Los Angeles, CA 90034.

Nematologists in California, Florida, North Carolina, Minnesota, Louisiana, Idaho and in Spain, China and Morocco have evaluated the efficacy of Agri-Terra, a reduced risk pesticide, against phytoparasitic nematodes over the last six years. Agri-Terra has been tested on burmuda grass, lantana, pepper, tomato, cotton, soybean, rice, sugarcane, cabbage, endive, lettuce, mustard, tobacco, cucumber, and potato against a wide spectrum of nematodes including reniform, root-knot, soybean cyst, lesion, lance, stunt, ring, stubby-root, dagger and spiral. In all cases the material has been shown to be nematicidal.

OBSERVATION OF SOIL COMMUNITIES IN PEANUT ROTATIONS USING AUTOMATED RIBOSOMAL INTERGENIC SPACER ANALYSIS (ARISA). **Olivaras-Fuster, O.,[1] C. Arias,[1] R. N. Huettel,[2] and K. L. Bowen.[2]** Department of [1]Fisheries and Allied Aquacultures and [2]Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849.

Root-knot nematodes and *Aspergillus* spp. are factors in peanut production causing decreased yield and peanut quality through mycotoxin contamination. Other factors that may be involved in this disease complex include varying soil-borne organisms and other plant parasitic nematodes. Since crop rotation is used to reduce both nematode and fungal populations, the rotation systems may influence the soil community complex. We are evaluating the use of automated ribosomal intergenic space analysis for fingerprinting the bacterial and fungal communities in peanut rotations. ARISA profiles were compared between field plots of continuous peanut, peanut-cotton, peanut-fallow, peanut-bahia and continuous bahia. Complexity of the profiles based on the number of bands observed as well as the relative intensity of the bands in the different systems were analyzed. Overall, ARISA profiles displayed between 40 to 60 bands, depending on the rotation system, ranging from 100 bp to 1000 bp. ARISA profile repeatability using multiple extraction from the same samples was excellent indicating that there is potential for using this approach to observe changes in soil communities.

THE ISOLATION AND FUNCTIONAL ANALYSIS OF PARASITISM GENES OF THE BEET CYST NEMATODE, *HETERODERA SCHACHTII*. **Patel, N.,[1] H. Diab,[1] B. Gao,[2] X. Wang,[4] R. S. Hussey,[2] T. J. Baum,[3] and E. L. Davis.[1]** [1]NC State University, Raleigh, NC; [2]University of Georgia, Athens, GA; [3]Iowa State University, Ames, IA; [4]Cornell University, Ithaca, NY 14853.

Homologs of parasitism genes that encode peptides secreted from the stylet of the soybean cyst nematode, *Heterodera glycines* are being investigated in *H. schachtii* to use the tractable plant model, *Arabidopsis thaliana*, for studies of parasitism gene function. Full-length cDNA clones encoding products with 92% identity to *H. glycines* CLAVATA3 (CLV3)-like protein (SYV46), 97% identity to annexin (4FO1), 88%, identity to a potential elicitor (4EO2), and 95% identity to a nuclear-localized protein (5DO8) have been isolated from *H. schachtii*. Polyclonal antibodies to the *H. glycines* parasitism gene proteins also bind within *H. schachtii* esophageal gland cells, confirming appropriate parasitism gene expression and translation. *H. schactii* infection assays using clv-1 receptor Arabidopsis mutants are being analyzed for their effect on nematode CLV3 (SYV46). Similarly, infection assays using Arabidopsis defective in annexin-1 are being conducted to decipher the role of plant annexin during nematode infections. RNA interference is being investigated as a parallel functional approach based on the potential of ingested dsRNA to knock-out target nematode parasitism genes.

THE EFFECT OF ROOTING AND OUTGROUP CHOICE IN RECOVERING SIGNIFICANT COSPECIATION BETWEEN *HETERORHABDITIS* AND *PHOTORHABDUS*. **Peat, S.,[1] R. French-Constant,[2] N. Waterfield,[2] and B. J. Adams.[1]** [1]Department of Microbiology & Molecular Biology, BrighamYoung University, Provo, UT 84602; [2]University of Bath, Claverton Down, Bath BA27AY, UK.

The entomopathogenic nematode *Heterorhabditis* forms a unique obligate symbiotic relationship with the gram negative bacteria *Photorhabdus*. Together they infect and kill larval insects, using the cadavers as a source of nutrients to grow and reproduce. One would expect that such a close association of host and symbiont would lead to the presence of strict cospeciation. To test this assertion, phylogenies of *Heterorhabditis* and *Photorhabdus* were constructed. Phylogenies were compared using TREEMAP 1.0, TREEMAP 2.0, and PAUP*4.0b10. The number of cospeciation events occurring in this system was not significantly different from chance alone when the *Photorhabdus* tree was rooted with *P. temperata*. Upon re-rooting the *Photorhabdus* tree with *P. luminescens akhurstii* and re-associating the host and symbiont trees, the eight cospeciation events observed in the most parsimonious reconstruction were significant with a p-value of 0.018. The discordant results obtained from the two analyses indicate that tree rooting choice has a significant influence on the number of cospeciation events recovered within the *Photorhabdus/Heterorhabditis* association. Further investigation into the effects of different rooting methods, as well as outgroup choice, were conducted and are discussed.

A PHYLOGENETIC ANALYSIS OF *PHOTORHABDUS* AND THE EVOUTION OF BIOLUMINESCENT INTENSITY. **Peat, S.,[1] R. French-Constant,[2] N. Waterfield,[2] and B. J. Adams.[1]** [1]Department of Microbiology & Molecular Biology, Brigham Young University, Provo, UT 84602; [2]University of Bath, Claverton Down, Bath BA27AY, UK.

Photorhabdus spp., the only known terrestrial bacteria to express bioluminescence, are well known for their symbiotic association with heterorhabditid nematodes. This association, along with their ability to kill insects, has increased the interest in the evolutionary relationships within the genus. Currently, three species are recognized within the genus *Photorhabdus; P. temperata* and *P. luminescens*, which are endosymbionts of the nematode *Heterorhabditis*, and *P. asymbiotica*, which have been isolated from human wounds. Furthermore, *P. luminescens* is divided into three subspecies. This work tests the monophyly of these groups using simultaneous phylogenetic analysis of both molecular and morphological data, and uses the resulting topology to track the evolution of bioluminescent activity within the genus. This analysis revealed the absence of monophyly between clinical isolates and symbionts, while confirming the monophyly of the specific and sub-specific designations. The present study also indicates that bioluminescent intensity shows a pattern of decline through the evolution of *Photorhabdus*.

PLANT PARASITIC NEMATODES ASSOCIATED WITH SEMI-TEMPERATE VEGETABLES IN BENGUET, PHILIPPINES. **Pedroche, N. B.,[1] D. De waele[2] and L. M. Villanueva.[1]** [1]Semi-Temperate Vegetable Research and Development Center, Benguet State University, La Trinidad, Benguet 2601, Philippines; and [2]Laboratory of Tropical Crop Improvement, Faculty of Bioscience.

Benguet Province is known as the "Salad Basket of the Philippines" owing to the verdant vegetable farms cradled on its valleys and the terraced gardens carved out of mountain sides. However, the local vegetable industry is threatened by pests and diseases, one of which is plant parasitic nematodes. A survey was conducted in vegetable growing areas in Benguet to identify the major plant parasitic nematode genera associated with the crops, determine their frequency of occurrence and abundance, and assess their potential as parasites of vegetables. Thirteen genera of plant parasitic nematodes were found associated with semi-temperate vegetables: *Aphelenchoides, Criconemella, Ditylenchus, Globodera, Helicotylenchus, Hoplolaimus, Meloidogyne, Longidorus, Paratrichodorus, Pratylenchus, Trichodorus, Tylenchus,* and *Xiphinema*. Among these nematodes, *Helicotylenchus, Meloidogyne, Longidorus* and *Pratylenchus* were the most prevalent. *Helicotylenchus, Meloidogyne*, and *Longidorus* were found in all the fields surveyed while *Pratylenchus* was detected in 26 out of the 27 fields surveyed. The nematodes were recovered with varying population densities in different crops . The abundance index (AI) for *Helicotylenchus, Meloidogyne, Longidorus* and *Pratylenchus* range from 1.54–4.0, 1.74–3.04, 1.0–1.87 and 1.41–3.34, respectively. In root samples, *Meloidogyne* and *Pratylenchus* were the most prevalent occurring at 87.5% and 81.25% of the samples collected with abundance index of 0.13–1.84 and 0.48–1.82 , respectively. *Globodera* was detected in 2 out of 3 areas planted with potato both in root (AI-2.46) and soil (AI = 1–136) samples with frequency of 36.6%. Based on the criteria proposed by Fortuner and Merny (1973) and Prot and Herman (1992), *Meloidogyne* and *Pratylenchus* were omnipresent and can be considered active parasites of semi-temperate vegetables in Benguet Province, Philippines.

EPIDEMIOLOGY OF *XIPHINEMA AMERICANUM* AND TOMATO RINGSPOT VIRUS ON RED RASPBERRY, *RUBUS IDAEUS*. **Pinkerton, J. N., J. Kraus, R. R. Martin, and R. P. Schreiner.** USDA-ARS, Horticultural Crops Research Laboratory, Corvallis, OR 97330.

Soil samples were collected monthly from 1999 through 2002 from 12 plots in a ToRSV infected "Willamette" red raspberry field in Clark Co. WA. Population densities of *X. americanum* were highest in the winter and lowest in the summer and correlated with monthly precipitation and mean soil temperature. All nematode stages were present in soil at all times of the year, but the proportion of juvenile stages was greatest in the winter. Cucumber seedlings were planted in soil collected each month and evaluated for nematode transmission of ToRSV with ELISA. The proportion of assay plants and plots in which virus was detected showed only weak seasonal trends and were not correlated with nematode densities. Soil samples were collected over the season from 2003 to 2004 in the same field. Transmission of ToRSV to cucumbers was monitored alongside an RT/PCR assay for ToRSV in hand-picked nematodes. ToRSV was detected in nematodes throughout the season with a higher frequency than in the transmission bioassay. Infection and systemic spread of virus was studied in "Meeker" raspberry plants planted in root bags filled with field soil infested with viruliferous nematodes or fumigated soil. Four plants were sampled destructively at each sampling date from May 2001 to May 2003. ToRSV was detected by ELISA in fine roots of one plant 5 months after planting and in all subterranean portions of plants after 12 months. Virus was detected in all aerial and subterranean tissues collected the second year. In 2002, plant growth was reduced significantly in nematode-ToRSV infested plants compared to those planted in fumigated soil.

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, Louisiana State University, Baton Rouge, LA 70803.

Full-season microplot studies were conducted to determine the effects of cotton (LA. 887), soybean (Pioneer 96B21), and three endemic weed species, [morning glory-MG (*Ipomoea purpurea*), hemp sesbania-HS (*Sesbania exaltata*) and johnson grass-JG (*Sorghum halepense*)], on reproduction of the reniform nematode, *Rotylenchulus reniformis*. Treatments were arranged as a RCB design with seven replications of seven treatments: 1) cotton or soybean alone; 2) MG alone; 3) JG alone; 4) HS alone; 5) cotton or soybean co-cultured with MG; 6) cotton or soybean co-cultured with JG; and 7) cotton or soybean co-cultured with HS. Over three years, co-culture of cotton with any of the three weed species suppressed reproduction of reniform nematode significantly. Suppression of reniform nematode could result from physical crowding, and/or from allelopathic compounds produced by weeds. Two-month duration greenhouse studies were conducted to test the allelopathy hypothesis. Five replicates of 11 treatments were established. 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 8–10 were the same as 5–7, but were watered daily with leachates from weed species. Treatments 11 and 12 were controls, which received leachates from cotton or growth medium. Data from these tests strongly supports the allelopathy hypothesis. That is, exudates collected from each of the 3 weed species and added to pots were as effective at reducing reniform nematode

populations as were the intact, co-cultured weed species. Leachates from controls had no significant inhibitory effect on reniform reproduction.

EFFECTS OF AZETIDINE-2-CARBOXYLIC ACID ON EGG HATCHING IN THE SOYBEAN CYST NEMATODE. **Pujara, R., M. Brunner, M. K. Klarich, and P. M. Tefft.** Biology Department, Saint Joseph's University, Philadelphia, PA 19131.

Factors that influence egg hatching and development in the soybean cyst nematode remain largely unknown. Presumably one of the first steps in hatching is changes in the permeability of the eggshell leading to hydration and then metabolic activity in the nematode. How this change in permeability occurs is speculative. Many plants produce defensive compounds to prevent herbivory and parasitism. We investigated the effects of a putative defense compound, azetidine-2-carboxylic acid (A-2-C), found in lily of the valley (*Convallaria majalis*), on nematode egg hatching and egg shell permeability. Azetidine 2 carboxylic acid is a proline analog and results in protein translational defects. Egg hatching in zinc chloride (3.0 mM, a known hatching stimulant) was inhibited in a dosage related manner ($r = 0.94$) with A-2-C (0–32mM). Hatching in 32 mM A-2-C was reduced 43% compared to hatching in zinc chloride alone. In order to determine if A-2-C was inhibiting egg hatching by preventing increases in eggshell permeability we calculated the percentage of eggs staining with Nile blue as an indicator of permeability. The percentage of eggs taking up stain following treatment for 3, 5 and 10 days in 32 mM A-2-C plus zinc chloride was reduced significantly in the two longer treatments compared to the eggs in the zinc chloride controls. Aliquots of these eggs were allowed to hatch and counted after 14 days. The A-2-C also caused a reduction in hatching in these eggs similar to the first study. These changes in hatching and permeability suggest that translation of a protein may be required in the early stages of hatching.

CHALLENGES IN NEMATODE DNA EXTRACTIONS FROM SOIL. **Quintero, T., and B. Sipes,** Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822.

Nematodes have a tremendous potential to serve as biomarkers indicating soil health. There is a need for molecular techniques that can similarly determine diversity and ultimately "soil health". To date, molecular methods involving the extraction of nematode DNA, amplification of the 18S ribosomal DNA gene and separation by denaturing gradient gel electrophoresis have been used to provide information about the health of soil ecosystems, but these methodologies have limitations. Representative sampling for nematodes usually requires 200 to 250 cm^3 (~200 grams) of soil. Alternatively, molecular analysis of soil samples utilizes significantly smaller quantities of soil ranging from 0.5 grams to 1 gram. The potential to omit taxa of low prevalence such as predatory nematodes is increased. The goal of this study was to determine a workable sample size for molecular analysis in nematode communities.

Nematodes representing various trophic groups were used and DNA from all trophic groups was extracted. However, the quality of this DNA was highly variable and this subsequently affected the amplification of the 18S ribosomal DNA gene. *Pratylenchus penetrans*, a small plant parasitic nematode, rendered DNA that was often below the threshold level to be visualized on an agarose gel. Further experiments determined the detection level for molecular extractions. Steinernema glaseri, an insect parasitic nematode, was added to a 0.1 gram soil sample ranging from 1000 nematodes to 1 nematode. Only those soil samples containing 1000 nematodes delivered sufficient quantities of DNA to amplify the 18S ribosomal DNA gene. As a result, direct extraction of nematode DNA from soil will require further development to improve sensitivity.

CONTROL OF THE ORIENTAL FRUIT MOTH, *GRAPHOLITA MOLESTA*, USING ENTOMOPATHOGENIC NEMATODES IN LABORATORY AND FRUIT BIN ASSAYS. **Riga, E.,[1] L. A. Lacey,[2] N. Guerra,[1] and H. L. Headrick.[2]** [1]Washington State University, IAREC, Prosser, WA 99350, USA. [2]Yakima Agricultural Research Laboratory, USDA-ARS, Wapato, WA 98951.

The oriental fruit moth (OFM), *Grapholita molesta* (Busck), is amongst the most important insect pests of peaches and nectarines and it has developed resistance to a wide range of insecticides. We investigated the ability of the entomopathogenic nematodes (EPN) *Steinernema carpocapsae, S. feltiae, S. riobrave*, and *Heterorhabditis marelatus* to control OFM under laboratory and fruit bin conditions. At a dosage of 10 infective juveniles (IJ)/cm2 in the laboratory, *S. carpocapsae* caused 63% OFM mortality; *S. feltiae* 87.8%; *S. riobrave* 75.6%; and *H. marelatus* 67.1% mortality. All four nematode species caused significantly higher OFM larval mortality in comparison to the untreated controls. *Steinernema feltiae* was used for the bin assays due to its ability to search for OFM under cryptic environments. Diapausing cocooned OFM larvae in miniature fruit bins were susceptible to IJ of *S. feltiae* in infested corner supports and cardboard strips. Treatment of bins with *S. feltiae* 10 or 25 IJ/ml suspensions of water with wetting agent (Silwet L77) resulted in 33.3–59% and 77.7–81.6% OFM mortality in corner supports and cardboard strips, respectively. This paper presents new information on the use of EPN, specifically *S. feltiae*, as non-chemical means of OFM control.

NEW NEMATODE MANAGEMENT ISSUES AND OPTIONS IN WASHINGTON STATE. **Riga, E., and J. Wilson.** Washington State University, Prosser, WA 99350.

The efficacy of Metham Sodium and the use of green manures in combination with half the recommended rate of synthetic nematicides against plant parasitic nematodes have been studied. Biotic and abiotic factors affecting Metham Sodium efficacy in controlling *Meloidogyne chitwoodi* have been examined. In laboratory experiments, we determined that low concentrations of Metham Sodium may increase nematode populations. However, at higher concentrations this effect is cancelled out and the majority of the nematodes are eliminated at normal application rates. In the field, application of Metham Sodium at normal application rates may decrease as it moves and diffuses into the soil. This can be a problem, since nematodes can travel great distances vertically. Considering the economic threshold for *M. chitwoodi* is 1 nematode per 250 cc soil, stimulating an increase in any portion of the nematode population may be a source for nematode re-infestation following Metham Sodium applications. Green manures in combination with synthetic nematicides to manage plant parasitic nematodes have been employed to manage plant parasitic nematodes. Arugula, *Eruca sativa,* green manure in combination with half the recommended rate of Telone eliminated *M. chitwoodi* from a field trial. In addition, arugula green manure in combination with Telone, Temik and Mocap eliminated *M. chitwoodi* from a field, reduced lesion nematode, and did not affect beneficial free-living nematodes.

A PRELIMINARY EXAMINATION OF TWO UNDESCRIBED LONGIDORIDS FROM THE USA. **Robbins, R. T.** Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701.

Host associations and general morphology of two *Longidoridae* being described are given herein. The first is a species of *Xiphinema* from the Great Smoky Mountains National Park in Tennessee. Four female specimens and a single juvenile specimen were found in soil about the roots of yellow poplar and in mixed forest, two females from each site, in an October 1985 survey by Ernest Bernard. They belong to Group 4 as defined by Loof and Luc in 1990 in their Polytomous key (two equal genital tracts with Z-organs. They are over 5 mm in length with a total stylet length of over 250 μm and have a rounded posterior terminus. Efforts have been made repeatedly to obtain additional specimens without success. The second species is a *Longidorus* obtained from about the roots of citrus in California in 2000 by Howard Ferris. Numerous specimens of females, males, and juveniles were collected. Males are about as numerous as females. Most of the juveniles are fourth stage. Preliminary hierarchical clustering showed the closest relationships to *L. crassus* and *L. breviannulatus*. The males and females are in the range of 4 to 6 mm in length with a total stylet length of almost 175 μm. The tail is conically rounded. The small subunit ribosomal RNA gene from this species was given the GenBank accession number of AY283163 in a 2004 paper by Neilson et al.

INTERACTION OF ROOT-KNOT NEMATODE WITH FUSARIUM WILT RACE 4 ON COTTON. **Roberts, P. A.,[1] R. M. Davis,[2] and T. R. Mullens.[1]** [1]Department of Nematology, University of California, Riverside, CA 92521; [2]Department of Plant Pathology, University of California, Davis, CA 95616.

Root-knot nematode (*Meloidogyne incognita*) interacts with Fusarium wilt (*Fusarium oxysporum* f. sp. *vasinfectum*) as a disease complex on cotton. In California, mild races of Fusaruim wilt (races 1 and 2) have predominated and result in wilt symptoms only in cotton fields infested with *M. incognita*. Recently, a virulent strain of wilt (race 4) was identified in fields without apparent nematode infestation. In greenhouse pathogenicity tests, three cotton cultivars (*Gossypium hirsutum* cvs. Acala SJ-2 and Acala NemX, and *G. barbadense* cv. Pima S-7) were challenged with Fusarium wilt race 4, with or without co-inoculation of *M. incognita.* The race 4 isolate caused severe wilt symptoms and plant stunting on Pima S-7 with or without nematodes. In contrast, race 4 caused milder wilt symptoms on Acala SJ-2, but the symptoms were greater in the presence of *M. incognita*. On Acala NemX, that has resistance to *M. incognita*, race 4 caused moderate wilt symptoms in the absence of nematodes, and wilt symptoms were less in the presence of *M. incognita*. The results indicated that *M. incognita* can increase wilt induced by race 4 on cotton, but the extent of the effect is cultivar-specific and may be reduced by activation of nematode resistance.

INHERITANCE OF RESISTANCE TO *ROTYLENCHULUS RENIFORMIS* IN AN INTERSPECIFIC CROSS BETWEEN AGRONOMIC COTTON (*GOSSYPIUM HIRSUTUM* CV. NEM-X) AND THE PRIMITIVE ACCESSION GB-713 (*G. BARBADENSE*). **Robinson, A. F.,[1] and O. A. Gutierrez.[2]** USDA-ARS, [1]College Station, TX 77845; [2]Mississippi State, MS 39762.

Rotylenchulus reniformis is a major nematode problem in the central U.S. cotton belt. Resistant cultivars would offer an economic and environmentally sound solution but are not available, and information regarding the inheritance of resistance is limited. This study's objectives were: 1) employ generation means analysis to study inheritance of resistance to *R. reniformis* in an interspecific cross between a highly susceptible cultivar of upland cotton (*Gossypium hirsutum*) and the most highly resistant primitive genotype known within the most closely related species (*G. barbadense*); 2) collect seed from selected resistant and susceptible progeny for subsequent resistance inheritance analyses. A hybrid (F_1 generation) was created, with GB-713 as the resistant female parent (P_1) and *Meloidogyne incognita* resistant Acala NemX as the

susceptible male parent (P_2), and also with opposite sex assignments. Assays for nematode resistance were conducted under growth chamber conditions during 2004–2005 to assign resistance scores to 720 plants representing F_1 and F_2 generations as well as backcross generations, BC_1P_1 and BC_1P_2, that resulted from crossing parents with F_1 plants. Reproduction was evaluated by extracting and counting vermiform nematode stages from soil. Results were consistent with inheritance of resistance by a single, partially dominant gene. Resistance was not sex-linked. Seed for subsequent study were collected after self-pollinating the following plants: 24 resistant BC_1P_2 plants; 12 resistant $BC_1P_2 \times P_2$ plants; 12 susceptible $BC_1P_2 \times P_2$ plants. The frequencies of highly resistant, moderately resistant, and susceptible progeny are being examined to confirm that inheritance of resistance in these progeny sets is consistent with that predicted from the generation means analysis study.

ANALYSIS OF POPULATIONS OF *STEINERNEMA FELTIAE* AND H*ETERORHABDITIS DOWNESI* FROM BULL ISLAND, IRELAND, USING AFLP. **Rolston, A. N., C. Meade, and M. J. Downes.** Department of Biology, NUI Maynooth, Ireland.

Three species of entomopathogenic nematodes occur in Ireland, two of which, *Steinernema feltiae* and *Heterorhabditis downesi* have been found to co-exist in the sand dune system of Bull Island, Dublin. Earlier work identified differing distributions of the two species: *S. feltiae* prevalence increases with distance from the beach into the sand dunes; *H. downesi* is most common 20–30 m into the dunes. Two phenotypes of *H. downesi* also occur on Bull Island: Yellow and Purple, with the Purple phenotype being more common in the front of the dunes. Although morphologically identical and sharing the same RFLP pattern, it has been suggested that each phenotype is unable to utilize the other's symbiotic bacteria. If this is true then both phenotypes should represent distinct populations of *H. downesi* with no gene flow between these populations. Little is known about intraspecific EPN ecology, particularly with regards to gene flow and gene diversity within and between populations. This has significant implications with regards to inundative application of alien nematodes and their effect on local EPN populations. Previous work studying the molecular phylogeny of Bull Island S. feltiae β-tubulin introns using EPIC-PCR highlighted surprising variability given the small geographical distances between the isolates, suggesting multiple colonization events of the island. Here we use Amplified Fragment Length Polymorphism (AFLP) to analyze the EPN populations of Bull Island. Are Purple and Yellow *H. downesi* distinct populations, and if so, what levels of gene flow occur between the two? Are *S. feltiae* populations restricted in their gene diversity and gene flow? What are the implications for inundative application of alien EPN?

SUPPRESSIVE SERVICE OF THE SOIL FOOD WEB. **Sánchez-Moreno, S., and Ferris, H.** Department of Nematology, University of California Davis, Davis, CA 95616.

Soil food webs perform the important ecosystem services necessary to maintain both agricultural productivity and ecosystem health. Higher trophic levels in soil food webs can play a role suppressing plant parasites and affecting nutrient dynamics by modifying abundance of intermediate consumers. Natural and agricultural landscapes were sampled to compare soil faunal structure and function. Top-down soil suppressiveness of *Meloidogyne incognita* was determined in laboratory assay. Five treatments, including two nitrogen fertilizers, two herbicides, and simulated tillage, were established in experimental microcosms to evaluate the effects of simulated agricultural practices on top-down suppressiveness. Soil food web indices were calculated to infer soil food web condition. Structured soil food webs in natural areas, with abundant predatory nematodes, effectively suppressed plant-parasite populations while disturbed communities in agricultural soils did not. Soil suppressiveness was related to the ratio of predators to prey and with the prevalence of omnivore and predator species. Agricultural management led to a reduction in the suppressive capacity of the soil food web. Abundance of predatory nematodes was correlated with soil ammonium, probably due to its association with nematodes and other organisms grazing on microbes. Trophic cascades were detected in the low-fertility natural soil. Soil suppressiveness is strongly affected by soil food web dynamics and agricultural management.

WHAT BROUGHT US TO WHERE WE ARE IN NEMATODE CONTROL? **Schmitt, D. P.** Society of Nematologists, Marceline, MO 64658.

How well we endure in sustaining crop production depends, in part, upon how well the crops are protected from pests such as nematodes. Soon after humans began cultivating crops some 15,000 years ago, management to maintain crop vigor became necessary. Even at that early time, plant-parasitic nematodes surely were one of the key obstacles in maintaining crop vigor. Today, common nematode management tools in annual crops include rotation, resistance and nematicides. Management on perennial crops depends heavily on chemical preplant treatments and chemical postplant treatments. Understanding the history of agriculture helps us to understand how we arrived at the present choice of management tools and programs. History also reveals strategies that have not been successful to date. The successful methods evolved over hundreds or even thousands of years of trial and error. For example, fallow, a type of rotation, has been utilized for several millennia. The earliest cultivated fields were small with short periods of cultivation followed by long periods of fallow. Although many factors could account for the rationale to use this practice, one of the major biological factors was very

likely plant-parasitic nematodes. Likewise, humans have been selecting animals and plants for a wide variety of purposes. The first corn on the cob was developed some 7,500 years ago by Stone Age farmers who made genetic selections from a Mexican wild grass (an illustration of early genetic engineering). We can speculate with some confidence, that centuries ago, improved crop vigor and yield was accomplished with genetic manipulations that resulted in accidental, but effective resistance to nematodes. Chemical control of nematodes seems to be a relatively recent practice. Julius Kuhn tested carbon disulfide as a soil fumigant for control of the sugar-beet cyst nematode in the 1870's-1880's. These examples illustrate how, by trial and error, humans could have selected plant production practices and modified these practices in ways that would purposely and serendipitously control nematode populations.

SHANK-INJECTED AND DRIP-APPLIED FUMIGANTS AS ALTERNATIVES TO METHYL BROMIDE FOR PROPAGATIVE MATERIAL NURSERIES. **Schneider, S.,[1] J. Gerik,[1] H. Ajwa,[2] T. Trout,[3] and A. Shrestha.[4]** [1]USDA ARS San Joaquin Valley Agricultural Sciences Center, Parlier, CA 93648; [2]Department of Plant Sciences, University of California, Davis 95616; [3]USDA ARS Water Management Research, Ft. Collins, CO 80526; [4]UC Kearney Agricultural Center, Parlier, CA 93648.

Nematode-free propagative material is critical to the establishment of healthy orchards and vineyards. In a series of field trials, performance of shank-injected and drip-applied fumigants were compared to the industry standard methyl bromide under nursery conditions. Nematode control at the time of planting and presence of root knot nematode (*Meloidogyne spp.*) galls on roots at harvest were documented. In a sandy loam soil, shank-injected and drip-applied iodomethane (IM) + chloropicrin (Pic) (50:50, 270–448 kg ha^{-1}) and 1,3-dichloropropene (1,3-D) + Pic (65:35, 468 L ha^{-1}) controlled plant parasitic nematodes to a depth of 150 cm and resulted in gall-free plants at harvest. Drip-applied Pic (448 kg ha^{-1}) and sodium azide (336 kg ha^{-1}) controlled plant parasitic nematodes at planting, but 20–80%, depending on treatment and variety, of the plants had galled roots at harvest. In a commercial nursery trial, bags of soil infested with citrus nematode (*Tylenchulus semipenetrans*) were buried at 15, 30, 60, and 90 cm depths in a deep alluvial loam soil prior to fumigation, because the plant parasitic nematode populations in the field were not uniformly high enough for a valid trial. In this soil, shank-injected IM+Pic (67:33, 336 kg ha^{-1}), 1,3-D (327 L ha^{-1}), and 1,3-D + Pic (65:35, 460 L ha^{-1}) resulted in nematode mortality comparable to methyl bromide at all depth, but drip-applied formulations, at the same rates, did not perform as well at the 60 and 90 cm depths. Nematode control by drip-applied fumigants was comparable to industry standard methyl bromide in sandy loam soils, but was not as effective in a deep alluvial loam.

MARKET AND FINANCIAL FACTORS USED TO MAKE NEW PRODUCT DEVELOPMENT DECISIONS. **Schroeder, M. E.** DuPont Crop Protection, Newark, DE 19714.

Growth in the conventional agrochemical market has slowed in recent years. Industry sources forecast modest compound annual growth in the global crop protection market of about 0.7% overall, to a total value of about $28B in 2009. Market factors affecting the decision to invest in new product discovery research include the shifting focus of agricultural production, the impact of transgenic crops in key market segments, competition from generic manufacturers and the increasing costs associated with meeting regulatory requirements for registration. Although some industry consolidation will continue, the major crop protection companies will continue to invest in basic discovery research. New technology will continue to drive new product discovery. High throughput screening on agricultural pests will provide the search engine necessary to identify new active ingredients, now estimated to be 1 new product for every 150,000 compounds tested. New products will provide not only outstanding biological performance, but will also be safe and deliver value-added qualities such as yield enhancement or quality improvement. A stage gate process employing Six Sigma principles of process analysis is used to assess the potential of a new product as a viable business opportunity.

UPTAKE OF FITC BY *HETERODERA GLYCINES* IS INHIBITED BY STOMAL OCCLUSION AND QUIESCENCE. **Schroeder N. E., and A. E. MacGuidwin.** Department of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706.

Previous research showed that fluorescent compounds are taken up by non-feeding plant-parasitic nematodes. *Fluorescein isothiocyanate* (FITC) was used to study chemical uptake in the second-stage (J2) soybean cyst nematode, *Heterodera glycines*. The percent of body area that fluoresced was evaluated for individual nematodes using image analysis. Treatments were compared using ANOVA. In order to examine the mode of entry, a veterinary cyanoacrylate adhesive was applied to either the anterior or posterior end of 6 nematodes. An additional control group was included that did not receive any adhesive. After the adhesive had solidified, nematodes were incubated overnight in 0.1 mg/ml FITC. Nematodes were washed repeatedly in water. The adhesive was removed and nematodes not killed by the adhesive were examined for fluorescence. Nematodes glued at the anterior end showed a significant reduction in fluorescence compared with the other treatments. This suggests that a major route of chemical entry is through the stoma. Eggs were mechanically broken to

obtain quiescent J2. Nematodes were incubated in 0.1 mg/ml FITC for 5 hours and compared to active and heat-killed J2 for fluorescence. Quiescent nematodes showed significantly less fluorescence than actively moving nematodes while heat-killed nematodes showed a much higher intensity of fluorescence than actively moving nematodes with a greater distribution within the nematode. In a parallel experiment similar results were obtained from J2 rendered inactive by exposure to cold temperature when compared with actively moving nematodes.

CONTROL OF PESTS IN PECAN AND PEACH USING ENTOMOPATHOGENIC NEMATODES: CHALLENGES AND SUCCESSES. **Shapiro-Ilan, D. I., and T. E. Cottrell.** USDA-ARS, Southeastern Fruit and Nut Research Lab, Byron, GA 31008.

Pecan and peach orchards may offer a suitable environment for using entomopathogenic nematodes (Steinernematidae and Heterorhabditidae) to control key insect pests such as pecan weevil (*Curculio caryae*), plum curculio (*Conotrachelus nenuphar*), peachtree borer (*Synanthedon exitiosa*), and lesser peachtree borer (*Synanthedon pictipes*). For example, high levels of field suppression have been observed for larvae of plum curculio (>90% control), and peachtree borer (87% control), whereas field suppression of adult pecan weevil has been variable (50–80% control). Depending on the target insect and commodity, key barriers for adopting entomopathogenic nematode technology within grower systems include economics and competitiveness with other control strategies, efficacy, and environmental persistence. Nematode strain improvement and improved application or formulation methods may enhance efficacy. Hybridized strains have shown improved efficacy for pecan weevil in the laboratory and are being incorporated into field studies. Application of nematode infected-hosts for peachtree borer, and formulations that prevent rapid desiccation for lesser peachtree borer may improve control of these pests. Some of these strategies for improving pest suppression should be applicable to other systems.

ENTOMOPATHOGENIC NEMATODES AND INSECTS: DIRECT AND ACTIVE INFECTION. **Shapiro-Ilan , D.,[1] J. Campbell,[2] and E. Lewis.[3]** [1]USDA-ARS, Byron, GA 31008; [2]USDA-ARS, Manhattan, KS 66502; [3]Department of Nematology and Department of Entomology, University of California, Davis, CA 95616.

In nature, entomopathogenic nematodes (*Steinernematidae* and *Heterorhabditidae*) must infect an insect host in order to complete their life cycle. The decision to infect or not is critical because once inside the host there is no turning back. In this presentation, we review and analyze infection behavior of entomopathogenic nematodes. Specifically, what are the factors that affect the nematode's decision to infect or not? At what point do the nematodes determine that an already infected host is unsuitable? Differential foraging strategies, ranging from a sit and wait ambusher approach to an active host-seeking or cruiser approach, clearly affect transmission and infectivity. Yet, further studies are required to elucidate aspects of host attraction and infection dynamics that vary among and within ambusher and cruiser species. Certain chemical or physical cues appear to attract or repel entomopathogenic nematodes to their host. However, the relative importance and interaction among these cues remain unclear. We believe that the next steps in understanding entomopathogenic nematode infection dynamics will include characterization of specific cues, assessing the nutritive status of host, and defining interactions between population dynamics and infectivity.

DISTRIBUTION OF A MARINE NEMATODE FAMILY, *COMESOMATIDAE*, IN THE GULF OF MEXICO. **Sharma, J.,[1] R. Huettel,[2] and J. Baguley.[3]** [1]University of Texas at San Antonio, San Antonio, TX 78249, [2]Auburn University, Auburn, AL 36849, [3]University of South Carolina, Columbia, SC 29208.

Nematodes are generally the dominant component of meiobenthic communities. Their abundance ranges from 1.6 × 105 to 4 × 104 individuals m-2 in the shallow estuarine and deep abyssal habitats, respectively. The *Comesomatidae* is a cosmopolitan family of marine nematodes with a global distribution and is a major representative of the deposit feeders. Their abundance ranges from 107–104 individuals m-2 in the shallow estuarine and deep abyssal habitats, respectively. A study of nematode diversity is underway in the northern Gulf of Mexico deep sea ranging from the Texas to Florida slopes. All samples thus far examined are well represented by this family. However, of the four genera examined, *Cervonema* sp. and *Sabatieria* sp. appear to have broad distributions from 200–3000 m. depth; whereas, *Paracomesoma* and *Dorylaimopsis* sp. have thus far only been observed in samples at depths ranging from 200–1650 m. Studies of the distribution range of such cosmopolitan groups are crucial to understanding the biodiversity in the deep sea.

SPATIAL ZONATION OF NEMATODES IN A WEST TEXAS SALT MARSH. **Sharma, J.,[1] R. N. Huettel,[2] and J. K. Bush.[3]** [1]Department of Biology, [3]Department of Environmental Science, University of Texas, San Antonio, TX 78249; [2]Department of Entomology and Plant Pathology, Auburn University, AL 36849.

The Diamond-Y Preserve is a salt marsh in West Texas on the eastern edge of the Chichuahuan Desert. It overlies the Permian Basin, an area covered by a shallow sea during the Mesozoic era and now of interest for its vast petroleum deposits. It is home to six endangered or threatened species of plants and animals and thus now under management by the

Nature Conservancy of Texas. Soil samples were taken along a horizontal transect at varying levels of submergence that are characterized by specific vegetation. Nematode species abundance was highest in the *Distichlis spicata* (saltgrass) zone with *Helicotylenchus* spp. being most predominant. *Hoplolaimus* sp. and *Helicotylenchus* spp. were associated with an endangered plant species *Helianthus paradoxus* (puzzle sunflower). An unknown root-knot nematode species was found in association with *Scirpus americanus* (three-squared bulrush) in brackish water. Bacterivores, fungivores and predators were found across the transect. Aquatic nematode species of the *Monhysteridae* occur in the brackish water zone and stream. The importance of recognizing the soil microfauna in such endangered habitats as an indicator of environmental changes is discussed.

CAN *STEINERNEMA CARPOCAPSAE* PLAY A ROLE IN CONTROLLING NAVEL ORANGEWORM (*AMYELOIS TRANSITELLA*) IN PISTACHIOS: THE CHALLENGE OF SCALEUP. **Siegel, Joel P.,[1] L. A. Lacey,[2] B. Higbee,[3] and R. Fritts Jr.[4]** [1]USDA/ARS, SJVASC, Parlier, CA 93648; [2]USDA/ARS, YARL, Wapato, WA 98951; [3]Paramount Farming Company, Bakersfield, CA 93308; [4]Advan LLC, Clovis, CA 93619.

Previous small plot studies with commercially produced *Steinernema carpocapsae* demonstrated that this species could infect overwintering larvae of the Navel Orangeworm (*Amyelois transitella*) infesting pistachios on the ground. Applications were made from October through April and the nematodes were effective when soil temperatures did not exceed 32°C during the day or fall below 5°C at night. Although these small-scale applications were successful this nematode needed to be evaluated in large scale trials. Applications were made to the berm as well as the entire orchard floor in some trials as well as by herbicide applicator and chemigation. Nematodes were applied at a concentration of 1.24 billion per ha in a volume of 1,869 l per ha by herbicide applicator and through microsprinklers and double drip irrigation. Applications failed when the nematodes were applied using double drip irrigation but succeeded when microsprinklers were used (output of 22 liters per hour, run for 6 hours). Applications made by herbicide applicator were successful when post application irrigation was supplied by microsprinklers.

PLANT-PARASITIC NEMATODE SURVEY OF CITRUS IN HAWAII. **Sipes, B.** Department of Plant and Environmental Protection Sciences. University of Hawaii. Honolulu, HI 96822.

A survey was conducted to identify plant-parasitic nematodes found in citrus orchards in on the islands of Oahu and the Big Island of Hawaii. Samples were collected from grapefruit, lemon, lime, orange, pomelo, tangelo, and tangerine trees. Carrizo citrange and C-35 were common rootstocks but in most cases the rootstocks associated with the different scions were unknown. A soil sample of up to 250 cm^3 was processed by elutriation and centrifugation to collect nematodes. A citrus root sample of up to 20 g was processed by mist extraction. *Xiphinema* sp. was the most common plant-parasitic nematode recovered in the soil and root samples, 23% of the total. *Tylenchulus semipenetrans* was recovered from 11% of the soil samples and 2% of the root samples. *T. semipenetrans* was found predominately on Oahu. *Rotylenchulus reniformis* was found in 19% of all samples and also predominately on Oahu (37% of the Oahu soil samples). *R. reniformis* was never found in the root samples. *Meloidogyne* sp. were recovered in 7% of soil samples, all from the Big Island, and in only 5% of root samples. *Meloidogyne* sp. population densities were less than 100/per root sample in all but one sample in which the root population exceeded the soil population density by 100X. This population of *Meloidogyne* will be identified. *Helicotylenchus* sp. and several species of ring nematodes were also commonly recovered from the soil samples.

MOLECULAR CHARACTERIZATION OF 17 β-HYDROXYSTEROID DEHYDROGENASE GENES FROM *HETERODERA GLYCINES*. **Skantar, A. M.,[1,2] and D. J. Chitwood.[1]** [1]Nematology Laboratory and [2]Molecular Plant Pathology Laboratory, USDA-ARS Plant Sciences Institute, Beltsville, MD 20705.

Nematodes are nutritionally dependent upon their hosts for sterols, making steroid metabolism an attractive area of investigation for the development of new methods for managing plant-parasitic nematodes. We recently identified two genes in *Heterodera glycines Hg-hsd-1* and *Hg-hsd-2*) with similarity to 17β-hydroxysteroid dehydrogenases involved in the synthesis of steroid hormones in mammals. Sequence comparisons revealed similar gene sequences in other plant parasites, including *Meloidogyne* spp., *Pratylenchus penetrans*, and *Globodera pallida*. Quantitative real-time PCR of *Hg-hsd-1* and *Hg-hsd-2* showed the highest expression in developing females, with relatively low levels in second-stage juveniles, eggs, or males. Decreases in expression of both genes in aging females corresponded to increased cuticle pigmentation of the cyst. Transcription was localized to the cyst contents and absent from developing embryos, suggesting that *Hg-hsd-1* and *Hg-hsd-2* are most likely involved in gonad development or oogenesis. These 17 β-hydroxysteroid dehydrogenases genes may play a role in the conversion of ingested sterols into steroid hormones in nematodes. Additionally, using the free-living nematode *Caenorhabditis elegans* we have demonstrated the inhibitory effects of azacoprostane and other inhibitors on sterol metabolism and revealed the impact of such perturbations on nematode development and responses to UV and temperature stress. Together, these studies provide new insights into the role of sterols and how they are processed in nematodes.

EFFECT OF VARYING RATES OF ABAMECTIN AS A SEED TREATMENT OF SUGAR BEET ON SEED GERMINATION, EMERGENCE, AND CONTROL OF HETERODERA SCHACHTII. **Smith, H. J.,[1] F. A. Gray,[1] and S. Malone.[2]** [1]Department of Plant Sciences, University of Wyoming, Laramie WY 82071; [2]Syngenta-NAFTA, Stanton, MN 55018.

The sugar beet nematode, *Heterodera schachtii* parasitizes roots of the sugar beet plant and can severely reduce yield. Control measures consist of crop rotation and application of soil-applied nematicides or fumigants. Experiments were conducted under environmentally controlled conditions to evaluate the insecticide/nematicide abamectin on seed germination, plant emergence from soil, and suppression of *H. schachtii*. Abamectin was applied to sugar beet seed at 0, 0.05, 0.1, 0.2 and 0.3 mg. Experiments were conducted twice with six replicate pots/treatment, and presented results are an average of the two experiments. Percent seed germination on filter paper in petri dishes after six days at 21°C with increasing rates of abamectin ranged from 72.5% in the untreated check to 8.75% with the high rate of 0.3 mg. Seedling emergence from soil after 15 days at 21°C ranged from 69.5% in the untreated check to 61.1% with the high rate of 0.3 mg. Although abamectin appeared to be phytotoxic in the petri dish germination tests, it did not appear to be phytotoxic in the soil emergence studies. Final populations of *H. schachtii* and the reproductive factor (Rf) for the five rates of abamectin was; 6,684 eggs/juveniles with a Rf of 3.34 for the untreated check to 3,786 eggs/juveniles with a Rf of 1.89. Parasitism by *H. schachtii* on sugar beet roots was reduced with increasing rates of abamectin. Abamectin should provide early protection of sugar beet seedlings against invasion and parasitism by the sugar beet nematode.

COTTON CULTIVARS TOLERANT TO RENIFORM NEMATODE. **Stetina, S. R.,[1] G. L. Sciumbato,[2] J. A. Blessitt,[2] and L. D. Young.[1]** [1]USDA ARS Crop Genetics and Production Research Unit, Stoneville, MS 38776; [2]Delta Research and Extension Center, Mississippi State University, Stoneville, MS 38776.

Thirty-six commercial cotton (*Gossypium hirsutum*) cultivars were evaluated to identify those that are most tolerant to reniform nematode (*Rotylenchulus reniformis*) and therefore do not exhibit reduced yield due to this pest. Field trials were conducted for three years at two locations in the Mississippi Delta that were naturally infested with reniform nematode at population levels exceeding the action threshold for Mississippi. Main plots were cultivars and subplots were either not treated or treated with nematicide (1,3-dichloropropene or aldicarb) at recommended rates. Reniform nematode population densities were assessed at planting, midseason, late season, and harvest. Plots were mechanically harvested to determine seed cotton yields. Data from all years and locations were pooled, and tolerant cultivars were identified using one-tailed t-tests ($P = 0.05$). Cultivars for which nematicide significantly reduced the nematode population at either the midseason or late season sampling intervals, but for which yields were not significantly higher due to nematicide application, were judged to be the most tolerant. Only Deltapine 449 BR, Paymaster 1218 BR, and SureGrow 215 BR met these criteria. In addition, tolerance indices [index = (yield of nontreated plot/yield of nematicide-treated plot)*100] were calculated and compared for nine cultivars that showed suppressed nematode populations in nematicide-treated plots at any point during the growing season. Differences of least squares means showed that SureGrow 215 BR's tolerance index of 106.4 was significantly higher than Deltapine 20 B's index of 85.8, though neither cultivar differed from the others evaluated. The three cultivars identified as being tolerant based on the t-tests ranked first (SureGrow 215 BR), second (Paymaster 1218 BR), and fifth (Deltapine 449 BR) with respect to tolerance index. These cultivars may be beneficial to growers because they do not appear to suffer significant yield loss in the presence of reniform nematode.

INFLUENCE OF HOST DESICCATION ON THE EMERGENCE AND INFECTIVITY OF INFECTIVE JUVENILE ENTOMOPATHOGENIC NEMATODES. **Stevens, G. N.,[1] H. Arimoto,[1] J. Ruiz-Vega,[2] H. K. Kaya,[1] and E. E. Lewis.[1]** [1]Department of Nematology, University of California, Davis, CA 95616; [2]Centro Interdisciplinario de Investigacion para el Desarrollo Integral, Instituto Politecnico Nacional, Oaxaca, Mexico.

Application of entomopathogenic nematodes in host cadavers appears to confer many advantages compared to application of infective juveniles (IJ) in water suspensions. For example, IJ emerging directly from cadavers may be more infective and less vulnerable to desiccation in field settings, improving their utility as biological control agents. In addition, formulation of cadavers may have a reduced capital requirement compared to production and formulation of aqueous-phase emerged IJ. Producing a formulation based on desiccated cadavers requires infection of the host insect, desiccation of the infected host, and subsequent cadaver storage, transport, and application. However, we have little knowledge of how the age of the infection at which desiccation begins or the degree of desiccation may influence eventual IJ emergence and quality. This experiment was designed to examine the influence of desiccation on emergence and infectivity of *Heterorhabditis bacteriophora* IJ. *Galleria mellonella* cadavers were placed in a standard food dehydrator either four or six days after initial exposure to *H. bacteriophora* IJ, and dried at 29°C. Cadavers were removed two, four, or six days after they were added to the dehydrator, at which time they retained on average 76, 54, and 39% of their original mass, respectively. We assessed the influence of infection stage (four vs. six days) and desiccation period (two, four, or six days) on the total number of IJ emerging from each cadaver. Cadavers began producing IJ two weeks after they were placed on emergence traps. While emergence rates did not differ significantly between the two infection stages, increasing the desiccation period

appears to reduce the emergence rate. This desiccation technique appears promising for field applications, as some desiccated cadavers produced significant numbers of IJ. Further investigations into the effects of this desiccation regime are underway.

ECOLOGY AND OVER-WINTERING ABILITY OF *ROTYLENCHULUS RENIFORMIS* ON COTTON IN ARKANSAS. **Still, J. A.,[1] and T. L. Kirkpatrick.[2]** [1]Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701 and [2]University of Arkansas, SWREC, Hope, AR 71801.

The reniform nematode, *Rotylenchulus reniformis*, was first reported in Arkansas in 1988. Incidence of this nematode in the state is greatest in cotton fields in east-central and southern Arkansas. In 1998, reniform nematodes were first detected in three fields in extreme northeastern Arkansas (Mississippi Co.). Soil samples submitted to the Arkansas Nematode Diagnostic Laboratory from production fields in this area of the state indicate an increase in incidence of the reniform nematode in this region. Preliminary investigations indicate that *R. reniformis* population densities in the spring in northeastern Arkansas are much lower than population densities in the southern part of the state at the same time of year. An experiment to evaluate the impact of soil texture and temperature on the temporal population dynamics and over-wintering survival of the reniform nematode on cotton in these two regions was initiated in 2005. Ten individual sampling points were established in each of the three soil types in a single production field in Mississippi Co., AR (MS) and in one soil type in a production field in Monroe Co., AR (MR) in June, 2005 with a Global Positioning System. The soil types in the MS field were: loamy sand (79% sand, 16% silt, 5% clay), sandy loam (63% sand, 27% silt, 10% clay), and sandy clay loam (63% sand, 17% silt, 20% clay). The soil type in the MR field was relatively uniform across the entire field and was a silt loam (19% sand, 70% silt, 11% clay). Monthly soil samples were collected from all sites from June, 2005–March, 2006. In addition, samples were collected vertically to a depth of 120 cm from each site in October, 2005 and in February and April, 2006. Nematode population densities in the MS field during the winter months stayed low in the soils where there was a higher percentage of sand and increased as clay content increased and had little variation over time. Nematode population densities in the MR field increased during the winter months and stayed high through March, 2006.

SOIL HEALTH BENEFITS FROM A SUGARANE FARMING SYSTEM INVOLVING CROP ROTATION, MINIMUM TILLAGE, CONTROLLED TRAFFIC AND TRASH RETENTION. **Stirling, G. R., A. L. Garside, M. J. Bell and B. G. Robotham.** Sugar Yield Decline Joint Venture, C/- Sugar Research and Development Corporation, Brisbane, QLD, Australia.

In 1993, the Australian sugar industry established a multi-disciplinary research team to identify reasons for a decline in the productive capacity of sugarcane soils. At that time, sugarcane was grown on beds 1.5 m apart, the wheel spacing of harvest machinery did not match crop row spacing and crop residues were often burnt rather than retained. After a plant and 2–4 ratoon crops, fields were ripped and cultivated to alleviate compaction and sugarcane was replanted. Initial studies showed that soils farmed in this manner were degraded from a physical and chemical perspective, while yield increases of 20–40% in fumigated soil indicated that biological constraints were also limiting productivity. Since yield responses of almost the same magnitude were obtained by breaking the sugarcane monoculture with legumes, a new cropping system was devised that included a legume crop in a short fallow. Soils were cultivated to remove existing compaction and permanent wide beds were established with separate cropping and traffic zones. Sugarcane and soybean were grown using minimum tillage techniques and crop residues were retained as mulch on the soil surface. This new farming system has been adopted by growers because productivity has been maintained while profitability and sustainability have improved. From a nematological perspective, losses from *Pratylenchus zeae* have been reduced because nematode populations are lower, damage thresholds have increased and suppressive biological control mechanisms are operating more effectively. Current studies aim to match nutrient availability from crop residues with the nutrient requirements of the crop and the free-living nematode community is being used as a biological indicator.

ENTOMOPATHOGENIC NEMATODES AND BIOLOGICAL CONTROL OF THE ROOT WEEVIL, *DIAPREPES ABBREVIATUS*, IN FLORIDA CITRUS: OPTIMIZING CONTROL ACROSS SITES AND SOILS. **Stuart, R. J.** University of Florida, Institute of Food and Agricultural Sciences, Citrus Research and Education Center, 700 Experiment Station Road, Lake Alfred, FL 33850.

Entomopathogenic nematodes (EPN) are being recognized as important elements for integrated pest management (IPM) in many fruit and nut crops. One of the best examples is the use of EPN in Florida citrus to control root weevils, especially *Diaprepes abbreviatus* (L.) (*Coleoptera: Curculionidae*). In citrus, larval feeding damages roots, reduces yield and kills trees by girdling or by facilitating infection by plant pathogenic *Phytophthora* spp. At present, the only recommended control for larvae is the application of EPN twice per year. Commercial nematode products have been successfully marketed in Florida citrus for weevil control for over 15 years, and many factors have contributed to this success. Biologically, the most important factor was the discovery of an EPN that could be applied to effectively control this weevil. Ecologically, Florida citrus groves have several important characteristics that facilitate EPN effectiveness including sandy

soils, irrigation, and a limited shaded area under the canopy for application. Economically, *D. abbreviatus* is capable of killing trees and destroying groves, and so grower demand for effective control was high. Moreover, many chemical insecticides were eliminated due to regulatory pressures and the newer chemicals have not been able to compete with nematodes in price or efficacy. The relatively low cost of nematode application compared to the high value of the citrus crop was undoubtedly a key factor. However, certain citrus growing areas in Florida have a rich fauna of endemic EPN, and these populations can be responsible for considerable natural biological control of *D. abbreviatus*. Moreover, applications of exotic EPN can have a suppressive effect on endemic EPN. Thus, determining the best approach for using EPN in conservation biological control, and when, where, and how often to apply EPN for inundative biological control are current research issues being faced in an effort to optimize control of *D. abbreviatus.*

MOLECULAR PHYLOGENY OF THE ORDER TYLENCHIDA: ANALYSIS OF NUCLEAR RIBOSOMAL RNA GENES. **Subbotin, S. A.,[1] D. Sturhan,[2] B. J. Adams,[3] T. O. Powers,[4] P. G. Mullin,[4] V. N. Chizhov,[5] N. Vovlas,[6] and J. G. Baldwin.[1]** [1]Department of Nematology, University of California, Riverside; [2]Institut für Nematologie und Wirbeltierkunde, Münster, Germany; [3]Microbiology and Molecular Biology, Brigham Young University, Provo; [4]Department of Plant Pathology, University of Nebraska, Lincoln; [5]Institute of Parasitology, Moscow, Russia; [6]C.N.R. Istituto per la Protezione delle Piante, Bari, Italy.

To study the evolutionary relationships among more than 70 representatives of the order Tylenchida, datasets containing sequences of LSU D2-D3 expansion segments and partial sequences of the SSU were analyzed using maximum parsimony, maximum likelihood and Bayesian inference. Species included were selected to represent the known breadth of taxonomic and morphological diversity of the group. Phylogenetic analyses indicate that Tylenchida contains well-supported lineages that largely correspond to Siddiqi's (2000) Hoplolaimina and Criconematina. Several significant results also derived from our study include: (i) the basal position of groups that include entomoparasitic nematodes within tylenchid trees, (ii) paraphyly of the superfamily Dolichodoroidea *sensu* Siddiqi (2000) with placement of Merliniinae representatives outside Hoplolaimina; (iii) paraphyly of Tylenchoidea; (iv) paraphyly of the genus *Ditylenchus*; (v) evidence for a *Pratylenchus, Hirschmanniella* and *Meloidogyne* clade; (vi) lack of support for traditional placement of *Radopholus* within Pratylenchidae. Congruence and discordance of molecular phylogeny and traditional classifications and morphological-based hypotheses of phylogeny of tylenchids are discussed. It is suggested that in some cases phylogenetic reconstruction of Tylenchida using the SSU alone might be influenced by long branch attraction occurring as a result of unequal rates of evolution among independent tylenchid lineages. The need for a better understanding of informative morphological characters, and the need for additional representatives, particularly of some basal groups, is discussed.

HETERODERA GLYCINES AFFECTS SOYBEAN INFECTION BY *CADOPHORA GREGATA*. **Tabor, G. M., C. R. Bronson and G. L. Tylka.** Department of Plant Pathology, Iowa State University, Ames, Iowa 50011.

Growth-chamber experiments were conducted to assess the effects of *Heterodera glycines* on infection of soybean, *Glycine max*, by *Cadophora gregata* (formerly *Phialophora gregata*), the fungus that causes brown stem rot (BSR) of soybeans. Soybean cultivars with various combinations of resistance and susceptibility to *H. glycines* and *C. gregata* were inoculated with *C. gregata* alone or *C. gregata* plus *H. glycines* (1,200 eggs/100 cm^3 soil). In most *H. glycines*-susceptible soybeans, incidence and severity of internal stem discoloration characteristic of BSR disease was greater with *H. glycines* than without, regardless of the cultivars' susceptibility or resistance to *C. gregata*. The effect of *H. glycine*s on BSR stem symptoms was less in cultivars resistant to both *C. gregata* and *H. glycines* than cultivars only resistant to *C. gregata*. *Cadophora gregata* colonization of *C. gregata*-resistant cultivars was increased by *H. glycines*, and stems of both a *C. gregata*-resistant and a *C. gregata*-susceptible soybean cultivar were colonized earlier by *C. gregata* when the plants also were infected with *H. glycines* than when plants were only infected with the fungus. Additional growth-chamber experiments were conducted to determine the effect of *H. glycines* population densities on incidence and severity of stem colonization by *C. gregata*. Soybean cultivars with three combinations of resistance and susceptibility to *H. glycines* and *C. gregata* were inoculated with *C. gregata* alone or with *C. gregata* plus one of two *H. glycines* population densities (1,500 or 10,000 eggs/100 cm^3 soil). There was earlier colonization of stems of *H. glycines*-susceptible soybeans by *C. gregata* with the higher *H. glycines* population density than the lower density. Severity of *C. gregata* stem colonization did not increase with increasing *H. glycines* population density in a *C. gregata*- and *H. glycines*-resistant soybean cultivar. Microplot experiments are being conducted to study the interaction of these organisms in the field.

SUSTAINABLE MANAGEMENT OF NEMATODES IN EAST AND SOUTHERN AFRICA REQUIRES CAPACITY BUILDING IN THE REGION. **Talwana, H. A. L.,[1] J. W. Kimenju,[2] Z. Sibanda,[3] W. J. Wanjohi,[4] S. R. Gowen,[5] D. J. Hunt,[6] and B. R. Kerry.[7]** [1]Department of Crop Science, Makerere University, P. O. Box 7062, Kampala, Uganda; [2]Faculty of Agriculture, University of Nairobi, Kenya; [3]Sibanda Consultancy, Harare, Zimbabwe; [4]School of Pure and

Applied Sciences, Kenyatta University, Nairobi, Kenya; [5]School of Agriculture, Policy and Development, University of Reading, Earley Gate, PO Box 237, Reading, RG6 6AR, UK; [6]CABI Bioscience, Bakeham Lane, Egham, Surrey, TW20 9TY, UK; [7]Nematode Interactions Unit, Rothamsted Research, Harpenden, Hertfordshire, AL5 2JQ, UK.

Methods for sustainable nematode management need local evaluation and advice provided to growers to ensure successful uptake. In many developing agricultural regions there are too few trained personnel to test strategies and provide advice; most nematologists work alone and often spend only part of their time dealing with nematode problems. The Nematology Initiative for Eastern and Southern Africa (NIESA) aims to develop and sustain a critical mass of expertise within the region to develop locally-adapted management methods and provide much needed diagnostic support. Nematologists in Kenya, Malawi, Uganda, Tanzania and Zimbabwe in organizations committed to retaining a full time nematologist work in partnership with colleagues within the UK. NIESA scientists aim to raise awareness of nematode problems in the region, pool and share information and technical skills, and conduct collaborative research. Six nematology laboratories have been established in participating African countries and an interactive website has been established (http://www.africannematology.info/index.asp.) to provide access to the major nematological journals and to exchange information. Initial collaborative research aims to develop strategies for the management of root-knot nematodes in vegetable and flower production. NIESA scientists have taken part in a 6-week training course in 2005 and will provide further courses in the region to improve nematode diagnosis and the management of pests, and currently three PhD students are being trained. Once established, NIESA will provide a core of expertise linked to a network of nematologists to underpin nematological research inputs in projects to improve crop protection methods in the region.

ROLLING CIRCLE MITOCHONDRIAL DNA AMPLIFICATION UNMASKS NEW MERMITHID PARASITISM OF COMMON TERRESTRIAL ISOPODS. **Tang, S.,[1] G. O. Poinar, Jr.,[3] G. Wang,[2] E. G. Platzer,[2] and B. C. Hyman.[1,2]** [1]Graduate Program in Genetics, Genomics, and Bioinformatics and [2]Departments of Biology and Nematology, University of California, Riverside, CA 92521; [3]Department of Zoology, Oregon State University, Corvallis, OR 97331.

Previous studies have shown that the mermithid nematode *Thaumamermis cosgrovei*, parasitizing local populations of the common terrestrial isopod *Armadillidium vulgare* (Latr.), carried numerous mitochondrial DNA (mtDNA) haplotypes. Recent surveys for additional haplotypes using rolling circle mtDNA amplification revealed the presence an unusual RFLP variant within a mermithid whose macroscopic appearance was strikingly similar to that of *T. cosgrovei*. Exposure of this nematode to distilled water resulted in spontaneous body fragmentation, whereas *T. cosgrovei* remained intact under identical hypotonic conditions. Guided by a morphology-based phylogeny that divides the nematode family Mermithidae into seven subfamilies (Gafurov, 1997), a Maximum Likelihood molecular phylogenetic analysis employing mitochondrial COI and nuclear 18S and 28S rDNA nucleotide sequences consistently yielded a single tree after a series of full heuristic searches. The subfamily Agamermethinae was resolved as monophyletic, and within this clade the tested nematode was a sister taxon to *Agamermis*. Morphological characters from juvenile and adults also support a tentative assignment as *Agamermis* (Cobb, Steiner and Christie, 1923). The complete nucleotide sequence of the *Agamermis* sp. mitochondrial genome has been determined; unlike the hypervariable *T. cosgrovei* mitochondrial genome, only a single *Agamermis* sp. haplotype is found. We infer an ancient, large duplication encompassing the mitochondrial genes for NADH dehydrogenase subunits 3, 4, and 6, (ND3, ND4, ND6) and the small mitochondrial rRNA (rrnS), followed by random loss of duplicate gene copies. These observations provide the first report of any mermithid parasite found in the common pillbug *A. vulgare* beyond that of *T. cosgrovei*.

EXAMINATION OF THE USE OF LIQUID HOG MANURE TO CONTROL *PRATYLENCHUS PENETRANS.* **Tenuta, M., A. Mahran, K. Conn, and G. Lazarovits.** Department of Soil Science, University of Manitoba, Winnipeg, Manitoba, Canada.

Developing new, practical and inexpensive options for the control of plant parasitic nematodes continues to be a challenge. Previously we have reported the use of liquid hog manure to disinfest soil of plant pathogens. Volatile fatty acids in manure reacting in acid soil were shown to be the mechanism that killed pathogens. We report here the observations of two microplot, and one field study extending that work to potential use to control *Pratylenchus penetrans* in potato soil. In May 2004 and 2005 microplot experiments using two slightly acid potato soils (pH 6.5 and 6.1) receiving factorial combinations of hog manure (6,000 US gal/acre) and sulphuric acid (180 US gal/acre) were conducted. Liquid hog manure in combination with acid gave good reduction of P. pentrans in the more acid soil (to <150/kg soil) in both study years. A field experiment was conducted in 2005 on a neutral pH potato soil. Liquid hog manure at 5,800 US gal/acre was added alone or in combination with 300 and 580 US gal/acre of sulphuric acid. By one week after application, the populations of *P. pentrans* decreased to 15% of that prior to application for the liquid hog manure alone and manure with 580 US gal/acre added (to 323 and 83/kg soil, respectively). The results indicate liquid hog manure may be an effective and inexpensive means of killing *P. penetrans* in slightly acid and neutral soil when application is combined with low rates of acid. In acid soils (pH < 5.5), liquid hog manure is inferred alone to be effective.

RESISTANCE OF WATERMELON (*CITRULLUS LANATUS* VAR. *CITROIDES*) GERMPLASM TO ROOT-KNOT NEMATODES. **Thies, J. A., and A. Levi.** U.S. Vegetable Laboratory, USDA, ARS, Charleston, SC 29414.

Root-knot nematodes (*Meloidogyne incognita, M. arenaria*, and *M. javanica*) cause serious damage to watermelon and resistance to these pests has not been identified in any watermelon cultivar. Root-knot nematodes are currently controlled in watermelon by pre-plant fumigation of soil with methyl bromide or treatment with other nematicides. However, the pending removal of methyl bromide from the U.S. market has increased the urgency of finding alternative management methods, such as host plant resistance, for root-knot nematodes in watermelon. In greenhouse tests, we evaluated 265 accessions from the U.S. Citrullus Plant Introduction Collection for resistance to *M. arenaria* race 1 and identified 22 Plant Introductions (PIs) that were moderately resistant. In further tests, the same 22 PIs exhibited low to moderate resistance to *M. incognita* race 3 and *M. arenaria* race 2. Three watermelon cultivars (*C. lanatus* var. *lanatus*), three *C. colocynthis* PIs, and four *C. lanatus* var. *citroides* PIs, all previously shown to be susceptible to *M. arenaria* race 1, also were susceptible to *M. incognita* race 3 and *M. arenaria* race 2. These results demonstrate that there is significant genetic variability within the U.S.P.I. Citrullus collection for resistance to *M. incognita* and *M. arenaria* races 1 and 2. Several accessions of *C. lanatus* var. *citroides* are potential sources of resistance that may be useful in the development of root-knot nematode resistant watermelon cultivars.

THE INFLUENCE OF CORN ON *MELOIDOGYNE INCOGNITA* POPULATION DYNAMICS. **Thomas, A. C.[1], and T. L. Kirkpatrick.[2]** [1]Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701, [2]Southwest Research and Extension Center, Hope, AR 71801.

Corn is grown in Arkansas on about 100,335 hectares annually, and fits well into cropping systems that include soybean or cotton. In areas of the state where incidence of *Rotylenchus reniformis* is high, rotation to corn has significantly enhanced cotton producers' ability to manage this nematode. However, corn may significantly increase *Meloidogyne incognita* population densities, and can create a problem for either cotton or soybean in the subsequent crop. There is little recent research information available in Arkansas on the influence of corn on *M. incognita* population dynamics, the host status of currently popular hybrids to this nematode species, or the relationship between nematode numbers and plant damage or yield suppression. Seventy-three commercial hybrids and four resistant selections were evaluated in greenhouse trials to assess their suitability as hosts for *M. incognita* and to determine if useful levels of *M. incognita* resistance existed in any of them. Of the hybrids screened, twenty-five percent, including two of the resistant selections with the lowest M. incognita reproduction and three hybrids with high reproduction were evaluated in a second test. In the first test, nematode reproduction was considerably lower on the resistant selections Mp 709 and Mp 712 than on the other hybrids and selections. Reproductive factors (RF) for Mp 709 and Mp712 in the second trial were 21.8 and 12.1, respectively, while RF for the remaining hybrids ranged from 46.0 to 233.3. In microplots, the population dynamics of M. incognita in a susceptible hybrid planted in March (early planting) and in mid-May (late planting) were similar, although there appeared to be a trend toward lower final population densities in the late planted crop. It appears that planting date does not play a significant role in *M. incognita* populations in corn, and the commercial hybrids that are currently popular in Arkansas will not likely help in managing root-knot nematodes in cropping systems that include corn.

SURVEY OF PLANT-PARASITIC NEMATODES IN NEW MEXICO PECAN ORCHARDS USING AN ATV-MOUNTED HYDRAULIC SOIL AUGER. **Thomas, S. H., J. M. Trojan, and J. A. Couch.** Department of Entomology, Plant Pathology and Weed Science, New Mexico State University, Las Cruces, NM 88003.

Pecan is the second-ranking crop commodity in New Mexico, generating $90 million in revenue annually. *Meloidogyne partityla*, a potentially serious nematode pathogen of pecan, was recovered from declining trees in a single orchard in the state in 1996. During 2005–2006 a systematic survey was conducted in which 10% of the pecan hectares in each county were sampled to determine the incidence of plant-parasitic nematodes among orchards. Samples were collected between October and April, which prior studies found to be the period during which most *M. partityla* reproduction occurred. Manual collection of soil cores was impractical due to: the number of orchards to be sampled; the need to collect soil from a 60 cm profile; and dry soil conditions resulting from reduced irrigation during winter. Therefore a hydraulically-driven auger was adapted to mount on an ATV and used to collect 20 sub-samples of 1.9 cm diameter by 60 cm depth per orchard. Nematodes were extracted from soil using elutriation and centrifuge flotation. The most frequently-encountered and abundant plant-parasitic genus was Mesocriconema, which was recovered from 52% of the orchards surveyed, at a mean density of 116 nematodes per 100 cm^3 soil. Species of *Pratylenchus* and *Tylenchorhynchus* were encountered with moderate frequency from 26% and 24% of orchards, respectively. Species of *Meloidogyne*, which were the principal target of the survey, were recovered from 8% of the orchards sampled. Results from this study indicate that species of *Meloidogyne* are infrequent in pecan orchards in contrast to their relatively high frequency as the predominant plant-parasitic nematodes in annual crops in New Mexico.

REPRODUCTION OF *PARATRICHODORUS MINOR* ON CORN HYBRIDS AND INBREDS. **Timper, P., and M. D. Krakowsky.** USDA ARS, Tifton, GA 31793.

In the southeastern United States, stubby-root nematode (*Paratrichodorus minor*) causes more damage to corn (*Zea mays*) than any other plant-parasitic nematode. Currently, there is no known source of resistance in corn to this nematode. Our objective was to evaluate corn hybrids and inbreds for resistance to *P. minor*. In all experiments, Pioneer 3223 served as a susceptible control. Corn seed was planted in sterilized soil (five replicate pots per hybrid or inbred) and inoculated with 500 *P. minor* 2 weeks after planting. Nematodes were extracted from soil 50 days after inoculation. Of the 33 commercial corn hybrids tested, three supported less reproduction of *P. minor* than Pioneer 3223; reproduction on these three hybrids was 18% to 42% that of the susceptible control. None of the 10 public inbreds tested supported less reproduction than Pioneer 3223. In conclusion, moderate resistance to *P. minor* can be found within commercial corn hybrids.

PATHWAYS OF TRANSMISSION OF THE PINEWOOD NEMATODE FROM INSECT VECTOR TO PINE TREE. **Togashi, K.,[1] and Y. Arakawa.[2]** [1]Laboratory of Forest Zoology, University of Tokyo, Tokyo 113-8657, and [2]Mihara, Hiroshima 723-0148.

The pinewood nematode, *Bursaphelenchus xylophilus*, is the causative agent of pine wilt. It enters the tracheal system of cerambycid beetles of the genus *Monochamus* at the beetle emergence. Then the nematode comes out of the tracheae and enters the tree body via feeding and oviposition wounds made by the adult beetles. So far the transmission of nematodes was believed to occur directly from nematode-infected beetle to trees. Using nematode-free and nematode-infected beetles, we found some pathways of nematode transmission from nematode-infected beetles to trees. First, the nematodes moved between two beetles of different sexes during mating behavior of beetles in laboratory. When the nematode-free female beetles obtained the nematodes from the male beetle, one of them transmitted the nematode to the pine bolts via the oviposition wounds. When one or two pairs of beetles with a nematode-infected beetle were placed in each of 64 outdoor cages in the night, the nematodes were isolated from nematode-free beetles of different or both sexes. Heavily nematode-infected adult beetles carried the nematodes in reproductive system at the emergence. A three-week rearing of nematode-infected beetles at a density of one per container showed the presence of the nematodes in the reproductive system. When 42 nematode-free female beetles paired with nematode-infected males for 12 hours, two of them carried the nematodes in the spermathecae, the reservoir of sperms, suggesting the transmission via the oviposition wounds. We will give the effects of environmental factors on various transmission pathways and discuss their significance in the persistence and features of nematode population.

HOST SUITABILITY OF YELLOW NUTSEDGE AND PURPLE NUTSEDGE FOR *MELOIDOGYNE JAVANICA, M. HAPLA* AND *M. INCOGNITA* RACES 1, 2, AND 3. **Trojan[1], J. M., S. H. Thomas[1], J. Schroeder[1], and L. W. Murray.[2]** [1]Department of Entomology, Plant Pathology, and Weed Science and [2]University Statistics Center, New Mexico State University, Las Cruces, NM 88003.

Yellow nutsedge (*Cyperus esculentus*, YNS) and purple nutsedge (*C. rotundus*, PNS) are prolific, perennial weeds. Their competitive nature and worldwide distribution make them among the worst weed pests affecting crop production. The distribution of PNS is primarily restricted to tropical and sub-tropical latitudes while the distribution of YNS reaches beyond that of PNS to include the temperate climates of the world. Greenhouse studies were conducted in 2005 and 2006 to determine the ability of YNS and PNS to host *Meloidogyne javanica, M. hapla*, and races 1, 2 and 3 of *M. incognita* based on nematode egg production per unit of belowground plant biomass. To compensate for differences in egg viability among the five nematode sources used to inoculate the study, a subset of eggs from each inoculum source was hatched and the percent viable eggs determined. Final egg production data was adjusted by this percent viability to reflect egg production per infective juvenile and enable relative host suitability comparisons. All three races of *M. incognita* examined were capable of reproduction on both PNS and YNS. Yellow nutsedge proved to be a good host for both *M. javanica* and *M. hapla*. Purple nutsedge supported no significant egg production for the northern root-knot nematode, *M. hapla* and was a poor host for the tropical root-knot nematode, *M. javanica*. This is the first report of yellow nutsedge as a host for *M. hapla*.

COMPARING SOIL SOLARIZATION TEMPERATURE TO LABORATORY DETERMINED LETHAL TEMPERATURE FOR *MELOIDOGYNE INCOGNITA*. **Wang, K.-H., and R. McSorley.** University of Florida, Gainesville, FL 32611.

Eggs or second stage juveniles (J2) of southern root-knot nematode, *Meloidogyne incognita*, were incubated in sand: peat mix (4:1) with 6% moisture in test tubes immersed in a water bath heated to 38, 40, 41, 42, 44 and 45°C for a series of time intervals. Controls were maintained at 25°C. Surviving or hatching J2 were collected at weekly intervals from Baermann trays until no nematodes were collected from the control. Regression analyses between percent survival or egg hatch compared to the control, and hours of heat treatment were performed for each temperature. Complete suppression of

egg hatch required 15.9 and 13.4 hours at 41 and 42°C, respectively, whereas, 100% mortality of J2 required 30.8, 25.3, and 13.9 hours at 40, 41, and 42°C, respectively. Root-knot J2 were killed within one hour at 44 and 45°C. Nematodes were not completely killed at 38°C within 40 hours of treatment. Unlike nematode development, effect of temperature on nematode mortality is not determined by heat units. Heat units required for 100% mortality of root-knot were 1230, 1038, and 585 degree-hours at 40, 41, and 42°C, respectively. Heat units required to suppress egg hatching completely were 651 and 562 degree-hours at 41 and 42°C, respectively. Assuming a cumulative lethal effect is occurring for M. incognita, a typical soil solarization treatment during summer in Florida for 6 weeks had accumulated 144 and 32 hours above 42°C at soil depth of 5 and 15 cm, respectively. These exposure times had surpassed the lethal exposure time for *M. incognita* at 42°C, and should eliminated *M. incognita* at 0 to15-cm-deep in the soil. The challenge in using solarization for nematode management is not the length of the solarization period but rather the ability to generate heat deeper down the soil layer from which *M. incognita* could resurge.

EFFECTS OF COVER CROPPING ON NEMATOPHAGOUS FUNGI AND NEMATODES. **Wang, K.-H., and R. McSorley.** Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611.

One of the mechanisms by which cover crops can suppress plant-parasitic nematodes is by enhancing natural enemies of plant-parasitic nematodes. This presentation will focus on the effects of cover cropping and incorporation of cover crop residues into the soil on nematophagous fungi and predatory or omnivorous nematodes. Literature reveals that leguminous cover crops enhance the abundance and diversity of nematode-trapping fungi better than non-legumes. However, performance of cover crops in enhancing nematophagous fungi depends on indigenous fungi present in the soil, time after nematicide application, soil organic matter content, and histories of farming practices. Omnivorous nematodes respond more consistently than predatory nematodes to cover cropping. Planting sunn hemp (*Crotalaria juncea*) or cowpea (*Vigna unguiculata*) as cover crops consistently increased omnivorous nematode numbers at the end of cover cropping season. The incorporation of cover crop residues into the soil initially increased the numbers of omnivorous nematodes as well as nematophagous fungi, but these numbers decreased over time. Use of cover crops can also compensate for disruptive impacts from fumigation or solarization on nematopahagous fungi or omnivorous nematodes. New cover cropping approaches are under investigation to enhance these beneficial fungi and nematodes over a longer period of time. Some new approaches include using cover crop residues as organic mulch, practicing strip-till cover cropping, or integrating cover cropping with augmentative biocontrol.

EFFECT OF ROTATION CROPS ON *HETERODERA GLYCINES* HATCH, VIABILITY, AND DEVELOPMENT IN LABORATORY AND GREENHOUSE STUDIES. **Warnke, S. A.,[1] S. Chen,[1,2] D. L. Wyse,[3] P. M. Porter,[3] and G. A. Johnson.[2,3]** [1]Department of Plant Pathology, University of Minnesota, Saint Paul, MN 55108; [2]Southern Research and Outreach Center, University of Minnesota, Waseca, MN 56093; and [3]Department of Agronomy and Plant Genetics, Saint Paul, MN 55108.

Sunn hemp (*Crotalaria juncea*), Illinois bundleflower (*Desmanthus illinoensis*), oilseed rape (*Brassica napus*), perennial ryegrass (*Lolium perenne*), red clover (*Trifolium pratense*), corn (*Zea mays*), and SCN-susceptible soybean (*Glycine max*) were compared for their effects on hatch, viability, and development of the soybean cyst nematode (SCN, *Heterodera glycines*) in laboratory and greenhouse experiments. In the laboratory experiments, root exudates in soil leachates, extracts from fresh plant tissues, and extracts from residues of sunn hemp, red clover, and soybean stimulated SCN hatch, while there was no hatch stimulation from the other crops. All crop extracts appeared to contain hatch inhibitors as well, compared to water and $ZnCl_2$ controls. There was no apparent effect of the leachates and extracts from any of the crops on egg viability *in vitro*. When SCN second-stage juveniles (J2) were exposed to the leachates and extracts for 72 hours, only the extracts from sunn hemp, red clover, and soybean reduced viability of J2. In the greenhouse experiments, residues of all crops except for Illinois bundleflower reduced egg population density, probably due to induced egg hatch, with sunn hemp providing the greatest reduction. The residues of sunn hemp, red clover, and perennial ryegrass in soil reduced the reproduction factor (number of females formed in soybean/initial egg population density), suggesting the residues not only reduced egg population density but also reduced the nematode infectivity. While all crops were penetrated by J2, minimal development (to third/fourth-stage juvenile only) occurred only in sunn hemp, red clover, oilseed rape, and Illinois bundleflower, and full development occurred only in soybean. The results suggest that sunn hemp and red clover were the most effective rotation crops for managing SCN, and stimulating SCN egg hatch to J2 that died in absence of hosts was the main mechanism involved in reducing the SCN population density.

COVER CROPPING FOR NEMATODE SUPPRESSION IN ANNUAL AND PERENNIAL CROPS. **Westerdahl, B. B.** Department of Nematology, University of California, Davis, CA 95616.

The use of cover crops in nematode management is not new, but has seen a resurgence of interest in recent years owing to the loss or restriction of use of nematicides. The literature leads one to believe that previous generations of nematologists were all too happy to abandon or minimize cover crop research in favor of working with the more successful soil fumigants.

The development of molecular techniques for identification of plant-parasitic nematodes to species, online databases to rapidly search out nematode resistant crops, computerized soil temperature monitoring equipment, computer models for calculating nematode degree days, and a greater understanding of nematode biology and population dynamics offer new tools to fine tune the use of cover cropping in nematode management scenarios. The development or selection of cover crop varieties for use against specific nematodes, and the use of green manures, biofumigation, and trap cropping are promising areas of current research in the area of cover cropping. From replicated trials that have been conducted, it is possible to suggest cover crops that will not exacerbate a nematode problem in a perennial planting provided the nematodes present have been identified. For preplant use on annual or perennial crops, it is possible to suggest crops that will reduce damaging nematode populations, although the control obtained is still likely to be less than that provided by traditional fumigant nematicides.

A BRIEF HISTORY OF INTRODUCED AND INVASIVE SPECIES IN HAWAII. **Wilkinson, M.** Hawaii Department of Land and Natural Resources, Honolulu, HI USA.

As on all islands, indigenous species arrived in Hawaii after being carried by wind, under their own power, hitchhiking on other species, or via the ocean. The extreme isolation of the islands has led to astounding levels of endemism. Today about 39% of all species found are endemic. On the other hand, about 20% of Hawaii's species are nonindiginous reflecting the long and complex interaction of introduced species and the Hawaiian biota. Hawaiian ecosystems are so fragmented that general landscape level management is not a matter of individually removing all invasive species but rather mitigating for the most damaging species that threaten rare species and ecosystems as well as working to prevent the introduction of additional invasive species. Polynesian settlers brought a suite of approximately 34 species, 24 were plants with high value as food, medicine or textile fiber. There were also Polynesian rats, pigs, dogs, several small hitchhiking snails, and head lice. Even over the millennia that they have now lived in Hawaii, only about 6 of the plant species persist or naturalize. Captain Cook's arrival was the starting point for the introduction of a familiar suite of species that travel with European settlers. In December, 2003 the Bishop Museum published an occasional paper, Hawaii's Biodiversity: A Detailed Assessment of the Numbers of Species in the Hawaiian Islands. Of the total 25,615 species counted, 5175 are introduced. This reflects the immense propagule pressure created by intentionally imported agricultural, pet species, the hitchhiking species associated with both the vessels and their cargo, smuggled species and associated diseases. Ultimately the goal of preventing the introduction of additional invasive species, controlling what has already arrived and regaining land where possible is to maintain the Hawaiian quality of life for the people, plants and animals that make up this unique island home.

EARLY STAGES IN PARASITISM OF ROOTS BY ROOT-KNOT NEMATODES. **Williamson, V. M., Q. Liu, and S. Lower.** Department of Nematology, University of California, Davis, CA 95616.

Root-knot nematodes are obligate parasites of a wide range of crop species. To complete their life cycle, they must recognize and move to host roots, penetrate the host and migrate intercellularly to the appropriate tissue where they can initiate a feeding site. These parasites must also avoid host defenses and be able to direct development of host feeding cells from which they can obtain sufficient nutrients to complete their life cycle. Microscopic examination and molecular studies have shed light on parasitism mechanisms, and molecular studies have identified numerous candidate parasitism genes. Genetic analyses in *Meloidogyne hapla* provide a complementary approach. Clear differences in attraction to potato roots can be measured between inbred strains of this species using an in vitro assay in which nematodes are suspended in pleuronic gel. These strains also differ in their ability to infect and to develop in roots of the wild potato *Solanum bulbocastanum* and in ability to reproduce on common bean carrying a nematode resistance gene. Genetic crosses have been carried out between these strains, and cultures representing segregating progeny lines have been produced and monitored using DNA markers. Genetic mapping and eventual cloning of the genes corresponding to these traits will contribute to our understanding of the parasitism process.

QUANTITATIVE GROWTH RESPONSE OF WATERMELON TO INFECTION BY MELOIDOGYNE INCOGNITA. **Xing, L.[1], G. Kruger[1], D. S. Egel,[2] and A. Westphal[1].** Department of Botany and Plant Pathology, Purdue University, [1]West Lafayette, IN 47907 and [2]Vincennes, IN 47591.

Root knot nematodes, *Meloidogyne incognita* (RKN) regularly cause damage in watermelon production. In southern Indiana, determining initial population densities of RKN is difficult because low soil temperatures in early spring may hinder detection with standard methods. A microplot trial was conducted to establish how to determine RKN population densities in spring and how these relate to plant damage. In 2004, two sandy soils, typical for southern Indiana watermelon fields, were placed in 45-cm diameter and 55-cm deep polyethylene tubes. Methyl bromide was applied at 390 kg/ha, and after aeration, egg suspensions of greenhouse-raised RKN were added to the upper 15 cm of soil to establish 0, 100, 1,000 or 10,000 eggs/100 cm^3 of soil. On 14 May 2004, 4-week old seedlings of watermelon cv. Royal Sweet were planted, soil samples were collected and used for extraction of second-stage juveniles (J2) by Baerman funnel or a greenhouse (GH) bioassay. In this latter assay, watermelon was seeded into the test soil in small containers and incubated in a heated

greenhouse. After 5 weeks, GH roots were washed free of soil and nematode galls were counted. At harvest of the microplots, plant biomass was determined and entered into the Seinhorst function using soil counts, galls on bioassay roots, or initial infestation levels of RKN as the nematode numbers. Biomass of watermelon decreased with increasing infestation levels of RKN. The bioassay gall counts were a good predictor for watermelon biomass. Soil counts of J2 were not an effective predictor of biomass but the bioassay method for soil collected in early spring has promise to overcome challenges of determining RKN in Indiana and will be useful in pest management systems.

TRANSFERRING SOIL SUPPRESSIVENESS AGAINST HETERODERA GLYCINES *UNDER FIELD CONDITIONS.* **Xing, L., and A. Westphal.** Department of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907.

The soybean cyst nematode (SCN, *Heterodera glycines*) is one of the most damaging soybean pathogens in the U.S. In a survey of >100 soils for suppressiveness against SCN, several soils were identified that allowed for less reproduction of SCN than others. The collection site of one of these soils was chosen for a field trial. On 10 April 2005, three treatments were applied to 9 m × 3 m wide plots: two treatments received 500 kg/ha Dazomet and one treatment remained untreated. Dazomet acts as a biocide against weeds and soil-borne pathogens but was found previously to have limited effects in reducing SCN infestations. On 10 May 2005, equivalents of 5% soil volume of the top 15-cm of soil from non-treated plots were collected and incorporated into one of the biocide treated plots. Numbers of SCN were determined under a SCN-susceptible soybean cultivar. At planting, SCN population densities were similar among the different treatments. At harvest, cyst and egg numbers per 500 g soil and eggs per cyst were similar in the non-treated control (188; 15,860; 85) and the 5% amended treatment (208; 18,920; 92) and were significantly higher in the Dazomet-treated plots (329; 45,967; 143). Therefore, biological suppressiveness was present in this field and was transferable to conducive plots under field conditions.

IDENTIFICATION AND COMPARATIVE ANALYSES OF INTESTINAL GENES IN *ASCARIS SUUM*, *HAEMONCHUS CONTORTUS*, AND *CAENORHABDITIS ELEGANS*. **Yin, Yong,[1] S. Abubucker,[1] J. Martin,[1] J. P. McCarter,[1,2] S. W. Clifton,[1] R. K. Wilson,[1] and M. Mitreva.[1]** [1]Genome Sequencing Center, Department of Genetics, Washington University School of Medicine, St. Louis, MO 63108; [2]Divergence Inc., St. Louis, MO 63141.

Projects are currently underway to analyze over 270,000 expressed sequence tags we have generated during the past 5 years from ~30 parasitic nematode species (www.nematode.net). The nematode intestine is a prominent target for both chemotherapy and immune control against parasitic nematodes. In this report, we treated intestinal ESTs as digital gene expression profiles, analyzed 9,947 EST clusters from *Ascaris suum*, which mainly consumes semi-digested contents of host gut or tissue fluids of the host, and 5,058 EST clusters from the flood-feeding *Haemonchus contortus*. Using our rigorous significance test based on Poisson distribution, we successfully identified 150 and 60 intestine-biased clusters for the two parasitic nematodes, respectively. In addition, we performed the same statistical test on a *Caenorhabditis elegans* SAGE dataset surveying 6,903 genes and identified 254 intestine-biased genes from this free-living nematode. Extensive comparative bioinformatic analyses revealed commonalities and differences in characteristics connecting gene functions, parasitism, adaptation and evolution in Nematoda.

DESS: A VERSATILE SOLUTION FOR PRESERVING MORPHOLOGY AND DNA OF NEMATODES. **Yoder, M.,[1] I. Tandingan De Ley,[1] I. King,[1] M. Mundo-Ocampo,[1] J. Mann,[2] M. Blaxter,[2] L. Poiras,[3] and P. De Ley.[1]** [1]Department of Nematology, Univ. California, Riverside, CA 92521; [2]School of Biological Sciences, University of Edinburgh Institute of Evolutionary Biology Ashworth Laboratories, King's Buildings, Edinburgh EH9 3JT UK; [3]Institute of Zoology, Academy of Sciences, Str. Academie 1, 2028 Chisinau, Moldova.

A solution of 20% dimethyl sulfoxide and 0.25M disodium EDTA, saturated with NaCl, pH 8.0 (first published by Seutin *et al*., 1991 and abbreviated here as DESS) was tested for applications in preservation of nematodes for combined morphological and molecular analyses. DESS can be used to preserve individual nematodes, nematode suspensions or entire soil/sediment samples. Preserved material can be stored for months at room temperature, shipped by mail, or carried in luggage. Morphological features are well preserved; quality is comparable to formalin and much better than ethanol fixation. Specimens can be transferred from DESS to glycerin with traditional protocols. Unlike formalin-preserved material, routine PCR can be performed on individual nematodes after any of these procedures, with success rates and amplification sizes comparable to fresh specimens. Scanning electron microscopy (SEM) of DESS-preserved nematodes followed by critical point drying yields results comparable to specimens similarly processed from formalin. The DESS protocol, examples of multifocal vouchers of nematodes preserved in DESS, and the resulting DNA sequences derived from these specimens are deposited in the NemATOL database (http://nematol.unh.edu/). Data is yet available on preservation quality after multiple years. Nevertheless, DESS clearly simplifies a wide range of applications, due to its suitability for combined morphological and molecular analyses, as well as being less hazardous than most alternatives. DESS is recommended for a wide range of applications and is expected to become a staple method in the nematological repertoire.

FOUR SPECIES OF *PRISMATOLAIMUS* FROM SOUTHERN CALIFORNIA. **Yoder, M., K. Carter, I. Tandingan De Ley, S. Esfahani, and P. De Ley.** Department of Nematology, Univ. California, Riverside, CA 92521.

Four species of *Prismatolaimus* de Man, 1880 (Nematoda: Prismatolaimidae) are described from two locations in southern California. *Prismatolaimus n.sp.* A and B were isolated from two vernal pools within the Santa Rosa Plateau Ecological Reserve, Murrieta. *Prismatolaimus n.sp.* A most closely resembles a population of *P. parvus* Milne, 1963 as described by Brzeski, 1997, but has a smaller stoma and shorter labial setae. Some specimens have a visible dorsal pore and gland, whereas P. parvus was described as lacking these. *Prismatolaimus n.sp.* B resembles *P. leptolaimus* Andrássy, 1969 in orientation of the terminal mucro, but differs in arrangement of the labial and cephalic setae. The other two species were isolated from soil and mulch of avocado trees in Fallbrook. One of these closely resembles the neotype of *P. intermedius* (Butschli, 1873) de Man, 1884 as described by Brzeski, 1997 but differs in being more slender with a longer tail, as well as having a smaller stoma, and longer labial setae. In view of the great variability reported for *P. intermedius* in the literature, we refrain from identifying our material as a new species. A single individual of another species resembles *P. mulcoomus* Brzeski, 1997 with exception of having a shorter tail and longer labial setae. More individuals are needed to properly identify this species and its identity is provisionally designated as *Prismatolaimus sp. cf. mulcoomus*. An individual from each species can be viewed as an online voucher at http://faculty.ucr.edu/~pdeley/vce.html. A tabular key of *Primatolaimus* is also presented.

UNDERSTANDING HOW SOIL CONDITIONS AFFECT THE ABILITY OF ORGANIC AMENDMENTS TO SUPPRESS PLANT-PARASITIC NEMATODES. **Zasada, I. A.** USDA, ARS Nematology Laboratory, Beltsville, MD 20705.

The influence of soil properties on the efficacy of nematicides has always been an important consideration in their use. The role which soil chemical and physical conditions determine the fate, availability and exposure levels to nematodes of compounds derived from organic amendments deserves equal consideration. Hydroxamic acids are the chemically active component of rye cover crops implicated in nematode suppression. They are formed in soil after incorporation of plant material and the enzymatic degradation of parent compounds. The partitioning coefficients of these compounds in three soil types were determined. Availability and exposure times in soil varied with soil type. Similarly, the influence of soil conditions upon the nitrogen cycle is an important consideration when utilizing nitrogenous amendments for nematode management. Ammonia is present at high soil pH, whereas low soil pH encourages the formation of nitrous acid. Both compounds are lethal to nematodes. *Meloidogyne incognita* was exposed to a biomineral soil amendment with or without urea supplementation under varying conditions of temperature. Significant formation of ammonia and nematotoxicity were achieved only when high soil pH was present. Incubation temperature also had a strong influence on ammonia accumulation with the amendment being more effective in producing ammonia and killing the nematode at higher temperatures. The formation of nitrous acid in a diverse set of soil types was evaluated; sand and low organic matter content soils were the best candidates from promoting nitrous acid availability. The use of organic amendments for nematode management is a complex process requiring an understanding of the transformation and generation of active compounds and how soil conditions regulate exposure levels of the compounds. Consistent and reliable nematode suppression with organic amendments will be achieved only through an understanding of these complex processes.