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ABSTRACTS

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HOST EFFICIENCY OF SELECTED SOYBEANS TO *MELOIDOGYNE HAPLA* AND *PRATYLENCHUS PENETRANS*. **Abawi, G. S., and J. W. Ludwig**. Department of Plant Pathology, NYSAES, Cornell University, Geneva, NY 14456.

The use of soybeans as a rotation or cover crop in vegetable production systems in New York State is increasing. Greenhouse tests were conducted to assess the host status of 22 varieties and breeding lines to *M. hapla*. Seeds were planted in pasteurized soils infested with 10 eggs of *M. hapla*/cc soil placed in 10-cm clay pots. The pots were maintained in a greenhouse at 20 – 25 C for 6 weeks. Roots were washed, rated for galling severity (RGS) on a scale of 1 (no visible galls) to 9 (>80% of roots galled), and eggs were extracted from roots by the sodium-hypochlorite method. All the soybeans evaluated were found susceptible to *M. hapla*, but varied in their efficiency as hosts to the nematode. The latter was suggested by the differences observed in the average RGS ratings (2.8 to 9.0) and the number of eggs produced/g root (1,406 to 28,910). AG2702, DKB28-51 and AG2302 were most efficient, whereas Stratford, AG2103, and Auburn were among the least efficient hosts in one test. The host status of 8 soybeans (least and highest efficient hosts to *M. hapla*) was also evaluated against *P. penetrans* in soil infested with 4 larvae and adults/cc soil. Results obtained suggested that the 8 materials were good hosts to *P. penetrans*, as the number of nematode recovered after 6 weeks varied between 214 to 302/g root. In addition, the incorporation of the 8 soybean materials as green manures 2 weeks before planting did not reduce the number of *P. penetrans* that penetrated and reproduced in snap bean roots as compared to a green manure of wheat or the non-amended treatment.

EVALUATION OF ONION GERMPLASM FOR RESISTANCE TO *MELOIDOGYNE HAPLA*. **Abawi, G. S., and J. W. Ludwig**. Department of Plant Pathology, NYSAES, Cornell University, Geneva, NY 14456.

The northern root-knot nematode (NRKN) is the most prevalent plant-parasitic nematode on onion grown primarily on organic soils in New York State, often casing considerable yield losses. In 1999, 20 commercial varieties were evaluated for resistance to the NRKN in field microplots infested with 20 eggs of *M. hapla*/cc soil and in a production field heavily infestation with *M. hapla*. Results obtained showed that all the varieties were susceptible to the NRKN. Thus, numerous sources of onion germplasm were evaluated during 2000 – 2002 in pasteurized soil (60 C for 40 min.) infested with 10 eggs of *M. hapla*/cc soil and maintenance in a greenhouse at 20 – 25 C for 8 weeks. Reaction to *M. hapla* was assessed by determining the root-galling severity on a scale of 1 (no visible galling or root thickenings) to 9 (>80% of roots with symptoms) and eggs production/g of root. To-date, 31 promising breeding lines (provided by Dr. Martha Mutschler, Cornell University; and Drs. Irwin Goldman and Mike Havey, University of Wisconsin), 54 Plant Introduction Accessions of *Allium cepa*, 50 accessions of *A. fistulosum*, one accession of *A. roylei*, and one onion cross with *A. roylei* (provided by Dr. M. Muschler) have been tested. Results obtained showed that all materials evaluated were susceptible to the NRKN. However, several accessions of *A. cepa*, *A. fistulosum* and *A. roylei* were found to be of higher tolerance than the commercial varieties included in the tests (Millennium, Stuttgart, and Wolf). The cross of *A. roylei* and onion was the best material among those tested in 2002. Further search for higher level of resistance to NRKN in onions and related species is warranted.

EFFICACY OF ABAMECTIN AS A SEED TREATMENT AGAINST *MELOIDOGYNE HAPLA* AND *PRATYLENCHUS PENETRANS*. **Abawi, G. S.,[1] J. W. Ludwig,[2] H. V. Morton,[3] and D. Hofer[3].** [1]Dept. of Plant Pathology, Cornell University, Geneva, NY 14456; [2]Viva, Inc. 1212 Heathrow Drive, Greensboro, NC 27410; [3]Syngenta Crop Protection, Basel, Switzerland.

Pelleted and non-pelleted seeds of tomato cv. 'Agriset' were treated with Abamectin formulation at 0.03, 0.1 or 0.3 mg a.i./seed provided by Syngenta Crop Protection. Abamectin-treated and non-treated seeds were planted in pasteurized soil (60 C for 40 min) infested with 8 eggs of *M. hapla* or 4 *P. penetrans*/cc soil and maintained in a growth chamber at 25 C for 2 weeks or in a greenhouse at 20 – 25 C for 6 weeks. Seed treatment with Abamectin at all rates was highly effective in reducing the number of larvae of *M. hapla* in roots at 2 weeks after planting. For example, the number of J2 of *M. hapla*/ root system of seedlings derived from non-treated and Abamectin-treated seeds (0.3 mg a.i./seed) averaged 24 and 1, respectively. After 6 weeks in the greenhouse, root-galling severity and the number of eggs produced/g roots were significantly reduced by the Abamectin treatment, especially at the highest rate. For example, root-galling severity and egg production averaged 6.6 and 26,142, respectively for seedlings derived from non-treated seeds and 2.1 and 1,320 for those

derived from Abamectin-treated seeds. Seed treatment with Abamectin also reduced the number of lesion nematode in roots after 2 and 6 weeks incubation in the growth chamber and the greenhouse, respectively, especially at the highest rate. For example, the average number of *P. penetrans*/g root after 6 weeks averaged 304 and 114 for the non-treated and Abamectin-treated seeds (0.3 mg a.i./seed), respectively.

TOXICITY AND ANTAGONISTIC EFFECT OF MARINE ALGAL EXTRACTS ON EMBRYONIC-DEVELOPMENT AND SURVIVAL OF NEMATODES. **Abdel-Rahman, F. H., and R. Salah.** Biology Department, Texas Southern University, Houston, Texas 77004.

The nematicidal activity of methanol extracts of thirteen algal species from the Red Sea (ten brown, two green and one red) was evaluated. Two groups of nematodes were used in the bioassay. The first group consisted of *Acrobeloides* sp., *Dorylaimellus* sp., *Mononchus* sp., and *Plectus* sp. (free-living nematodes). The second group was selected from plant-parasitic nematodes *Helicotylenchus* sp. and *Meloidogyne* sp. The bioassays were conducted on the adult stages of the free-living nematodes, adults and/or the larvae as well as the egg-masses of the plant parasitic nematode. The results revealed that extracts from *Stypopodium zonale*, and *Sargassum latifolium* caused more than 90% mortality of *Meloidogyne* sp. The extract of alga *S. latifolium* showed the most nematicidal activity causing at least 85% mortality in five of the tested nematode species. For nematodes, *Dorylaimellus* sp. was the most susceptible species showing the highest mortality, where 9 out of the 10 tested brown algal extracts caused 90% or higher mortality. The minimal effect was found in the *Aphelenchus* sp., where only *Sargassum latifolium* and *Cystoseira myrica* caused 79 % and 45 % death respectively. Other extracts showed minimum or no effect on *Aphelenchus* sp. The egg-hatching test, was conducted only on the plant-parasitic nematodes, and with the brown algal extracts. The bioassay demonstrated that most brown algal extracts were able to prevent or disrupt the embryonic-development of the eggs in the tested nematode species. The highest hatching of 22 % was recorded when eggs were incubated in *Turbeneria triquetra* extract. The algal extracts of *Cystoseria myrica, Sargassum dentifolium,* and *Dictyota dichotoma* caused low egg hatching of approximately 7 %, 7 %, and 9 % respectively. On the other hand, the rest of the algal extracts caused disruption of the embryonic development and arrested development at the early stages of blastula, gastrula, or tadpole, or at later developmental stages such as the first or second stage-larvae. Images of different embryonic developmental stages were monitored and captured using image analysis and processing software (Optimas 6.2).

HISTOPATHOLOGY OF RENIFORM NEMATODE ON *GOSSYPIUM LONGICALYX* AND INTERSPECIFIC COTTON HYBRIDS. **Agudelo, P. A.,[1] A. F. Robinson,[2] J. McD. Stewart,[3] R. T. Robbins,[1].** [1]Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701, [2]USDA-ARS, College Station, TX 77845; [3]Department of Crop, Soil, and Environmental Sciences, , University of Arkansas, Fayetteville, AR 72701

The histopathology of reniform nematode (*Rotylenchulus reniformis*) on roots of *Gossypium longicalyx*, *G. hirsutum*, and interspecific cotton hybrids was studied by light microscopy. Useful resistance to reniform nematode in *G. hirsutum* appears to be very limited, so recently there has been increased interest in the introgression of resistance to this nematode from *G. longicalyx* and other *Gossypium* species into *G. hirsutum* genotypes. *Gossypium longicalyx* is highly resistant to reniform nematode. The mechanisms for resistance, however, are not known and no observations on the cellular changes induced by the nematode in the plant have been published. In *G. longicalyx* roots, penetration by female nematodes was confirmed, and incipient swelling of the females, indicating initiation of maturation of the reproductive system, was observed. Maturation up to the formation of a single embryo inside the female body was frequent, but no maturation beyond this was observed. In the hybrids, diverse responses were observed among plants. Reactions ranged from highly compatible, with the formation of active syncytia and full development of females, to incompatible and no apparent development of the female. Compatible plants showed characteristic hypertrophied cells, enlarged nuclei, dense cytoplasm, and partial dissolution of cell walls, whereas incompatible plant reactions included lignification of the cells adjacent to the nematode head, or the complete collapse and necrosis of the cells involved. The need to characterize reactions and to carefully select among the hybrid plants during the introgression process, as well as the importance of combining the results of reproduction tests with histological observation of the plant-nematode interactions is discussed.

THE INCIDENCE AND IMPACT OF *MELOIDOGYNE INCOGNITA* IN ILLINOIS SOYBEAN PRODUCTION. **Allen, J. B., J. P. Bond, M. E. Schmidt, and J. S. Russin**. Department of Plant, Soil and General Agriculture. Southern Illinois University Carbondale, IL 62901.

Meloidogyne incognita is an emerging threat to soybean production in Illinois. This pathogen has been identified in nine soybean fields, eight vegetable fields and five peach orchards. The potential impact of *M. incognita* to soybean germplasm in northern latitudes is unknown. In 2001 and 2002, four soybean varieties (Pioneer 9481, Pioneer 9492, Gateway 493 and LS94-3207) were selected and planted in infested fields. Nematode population densities were recorded at planting and every 6 weeks until harvest. Reproduction by *M. incognita* was higher in the plots planted to P 9481. Across the four varieties, the increase in the population density of *M. incognita* was concomitant with a linear decrease in soybean yield.

Yield reduction averaged 40% for the two susceptible lines and 15% for the resistant lines. Fifteen elite germplasm lines developed in southern Illinois environments to possess resistance to *Heterodera glycines* and Soybean Sudden Death Syndrome and eighty commercial varieties (Maturity Groups IV and V) were evaluated for resistance to *M. incognita*. Five of the elite lines had a high level of resistance to root galling and egg production. Approximately, 15% of the commercial varieties tested were resistant.

EFFECT OF CHEMICAL PESTICIDE USE ON ENDEMIC ENTOMOPATHOGENIC NEMATODE POPULATIONS IN TURFGRASS. **Alumai, A., and P. S. Grewal.** Department of Entomology, The Ohio State University, OARDC, 1680 Madison Avenue, Wooster, OH 44691

Entomopathogenic nematodes are effective biocontrol agents that are used commercially to control insect pests of turfgrass. However, little information is available regarding the effects of routine chemical pesticide applications on endemic nematode populations. We investigated the natural occurrence and survival of entomopathogenic nematodes on three golf course surfaces (roughs, fairways, and putting greens) representing an increasing gradient of chemical pesticide use. Of the positive sites, the nematodes were recovered from 67% of the rough areas and 33% of the fairways, with no recovery from the putting greens. *Steinernema carpocapsae* was recovered from 4, *S. glaseri* from 1, and *H. bacteriophora* from 4 of the positive sites. In a replicated field study, we tested the effects of 6 commonly used chemical pesticides on population dynamics of endemic *H. bacteriophora* on a golf course rough area. We found that only fipronil (Chipco Choice^{TM}) significantly reduced the persistence of *H. bacteriophora*. In a related laboratory study, we compared the viability and pathogenicity of *H. bacteriophora* (HP 88 strain) and *S. carpocapsae* (All strain) exposed to 8 selected pesticides at three dilutions. We found no significant variation in viability of *S. carpocapsae* or *H. bacteriophora*. However, the insecticide trichlorfon (Dylox 80) significantly reduced pathogenicity of both *H. bacteriophora* and *S. carpocapsae*, whereas the fungicide, aluminium tris (Aliette) and the insectides, chlorpyrifos (Dursban Pro), and carbaryl (Sevin) had negative effects only on *S. carpocapsae* pathogenicity. The insecticides imidacloprid (Merit 75) and carbaryl (Sevin) had synergistic effects on *H. bacteriophora* pathogenicity at low concentrations. These results suggest that nematodes are more likely to occur in less intensely managed areas that receive fewer to no chemical insecticides than the more intensely managed fairways and putting greens.

MANAGEMENT OF SOYBEAN CYST NEMATODE WITH WINTER WHEAT/LEGUME ROTATIONS. **Anderson, T. A., T. W. Welacky,[1] E. Topp,[2] A. E. Tenuta,[3] E. Riga,[4] and J. Potter.[5]** [1] Agriculture and Agri-Food Canada, Harrow, ON, N0R 1G0; [2] Agriculture and Agri-Food Canada, London, ON, N5V 4T3; [3] Ontario Ministry of Agriculture and Food, Ridgetown College, Ridgetown, ON; [4] Washington State University, Prosser, WA, 99350-8694, [5] Agriculture and Agri-Food Canada, Vineland Station, ON L0R 2E0.

Soybean cyst nematode is the most important disease of soybeans in Ontario. It has been estimated that losses exceed $30 million annually because of this pest. SCN continues to spread into shorter season production araes. Crop rotation with non-hosts is one method that is recommended for managing the disease. In southwestern Ontario, there are limited options available for rotation. One option includes winter wheat which is frequently underseeded with red clover. Although wheat is a non-host, conflicting reports have been published on ability of red clover to support reproduction of SCN. This study was undertaken to determine if wheat/red clover crops result in an increase or decrease in soil inoculum and to determine if soil microflora and non-traditional crops affect hatching and viability of SCN. In micro-plot field trials, wheat/red clover crops decreased soil populations of SCN. Population decreases in soil from all wheat-clover treatments decreased to the same degree as in the fallow plots. This suggests that the wheat-clover treatments were neutral in effect on SCN soil populations. An examination of roots at mid-season and at harvest revealed cysts only on soybean roots. Cysts were not observed on clover or wheat roots. Soybean treatments contained the highest numbers of SCN J_2 juveniles. Clover or wheat did not stimulate hatching compared to fallow control plots. All studies suggested that red clover did not support reproduction of SCN and could be used in rotations to help reduce soil populations of SCN.

MECHANISMS THAT REDUCE NEMATODE DEVELOPMENT IN NEW GRAPE ROOTSTOCKS. **Anwar, S. A., and M. V. McKenry**. Department of Nematology, University of California, Riverside CA 92521.

A decade_long search among *Vitis* spp. for broad and durable nematode resistance culminated in the finding of five new sources from four different parentages. Experiments were initiated to quantify and observe nematode/host interactions during nematode penetration, development, and reproduction. The five sources of resistance were effective against all aggressive *Meloidogyne* populations but also nematodes of other genera. What features were common to the new resistance sources? All the new sources display a plant hypersensitive response (HR) that reduces entry of infective juveniles at the root tip. This HR involves a greater number of plant cells than previously available among commercial *Vitis* rootstocks. Contemporary *Vitis* resistance yields only enough HR to stop the first four or five infective juveniles but aggressive populations enter in greater numbers. The root tip is also a favored feeding site of *Xiphinema index*, another reason to locate adequate HR there. During the period from nematode feeding to reproduction these five new resistance sources also exhibit

additional defense mechanisms unavailable in contemporary grape rootstocks. Several resistance sources induced necrosis of feeding tissues within cortical cells, which restricted female development and overall egg production. One source resulted in a dissolving of the adult female within newly formed galls. Several sources limited the size of the syncytium and gall thus limiting long-term feeding by *Meloidogyne* to young roots. Selection for multiple resistant mechanisms in multiple locations along the root and over time provides resistance to a broader collection of nematode genera including aggressive *Meloidogyne* populations, *X. index, Pratylenchus vulnus*, and *Tylenchulus semipenetrans*. Resistance to this many different nematode species has not previously been available.

SPATIO-TEMPORAL ANALYSIS OF *HETERODERA GLYCINES*, SOYBEAN CYST NEMATODE, AND SOIL FERTILITY. **Avendaño, M. F.,[1] and H. Melakeberhan.[2]** [1]Department of Plant Pathology, Iowa State University, Ames, IA 50011; [2] Department of Entomology, Michigan State University, East Lansing, MI 48824.

This work was conducted to analyze the relationship between the spatial distribution of the soybean cyst nematode (SCN) at planting, mid-season and harvest and soil fertility in two fields in Michigan. SCN and soil fertility were analyzed on soil samples collected at planting following a geostatistical sampling design in 1999 and 2000. Geostatistical analysis was applied in conjunction with correlation analysis. The spatial distribution of SCN, pH, P, K, Ca, and Mg were mapped by kriging. SCN population density was positively correlated with pH ($\alpha = 0.01$) in both fields. Correlation coefficients fluctuated between sampling times without following a clear trend. The spatial distribution of SCN was cross-correlated with soil pH over a range of 70m in Field A, and 130m in Field B consistently over time. SCN population density was correlated with Ca, positively and poorly in Field A ($r = 0.17$ to 0.46), and strongly and negatively in Field B ($r = -0.32$ to -0.66). SCN and Ca were cross-correlated up to a range of 110m in Field B. SCN population density was poorly correlated with Mg in the soil, and uncorrelated with P and K (alpha = 0.05). SCN population density was negatively correlated with clay percentage in the soil in Field B (previous work), and soil pH and clay percentage were negatively correlated, indicating that the spatial distribution of SCN was affected by the combination of soil pH and soil texture. The results presented here in combination with previous work on SCN and soil texture support the notion of delineation of management zones for SCN control based on soil fertility in addition to soil texture.

ABAMECTIN SEED COATING: A NEW NEMATICIDE PLANT PROTECTION TOOL. **Becker, J. O.,[1] H. V. Morton,[2] and D. Hofer.[3]** [1]Department of Nematology, University of California, Riverside, CA 92521; [2]Viva, Inc., 1212 Heathrow Drive, Greensboro, NC 27410; [3]Syngenta Crop Protection, Basel, Switzerland.

All currently registered nematicides are used at relatively high application rates that range from a few to hundreds of kg per hectare. This is in contrast to the recent development of some fungicides, insecticides and herbicides that are effective at rates of a few grams of active ingredient per hectare. Seed coating with the microbially derived abamectin promises to be the first plant protection strategy against plant-parasitic nematodes that would operate at similarly low rates. In greenhouse and field trials the seedling protection by abamectin seed coating against second-stage juveniles of the Southern root-knot nematode *Meloidogyne incognita* lasted for about 3 weeks. This allowed susceptible crops such as cucurbits and tomato to develop an extensive root system during the most sensitive phase of plant development despite the presence of high population levels of infective root-knot nematode juveniles. In short-season crops the protection was sufficient to significantly increase crop yield without additional nematicidal treatments. Abamectin seed coating is an effective and low risk integrated pest management tool that has the potential to significantly reduce pesticide load in the environment.

UTILIZATION OF ABAMECTIN SEED COATING IN VEGETABLE TRANSPLANT PRODUCTION SYSTEMS. **Becker, J. O.,[1] S. Mueller,[1] X. Chen,[1] and D. Hofer.[2]** [1]University of California, Department of Nematology, Riverside, CA 92521; [2]Syngenta Crop Protection, Basel, Switzerland.

Seed coating with the microbially derived nematicide abamectin protects young crop seedlings against various endo-parasitic nematodes from invading and consequently damaging the roots. However, many vegetable seedlings are raised as containerized transplants with a self-enclosed root system. We evaluated whether tomato transplants derived from abamectin coated seeds were protected when planted into root-knot nematode-infested soil. Tomato seed (cv. Tiny Tim) was coated with abamectin at 0.1 or 0.3 mg a.i./seed while untreated seeds served as the control. The seed was placed in seedling trays with UC potting mix and incubated in the greenhouse under ambient light at approximately 24 C. Three weeks later, the seedlings were removed from the trays and transplanted into 600 cc pots containing sandy, root-knot nematode-infested soil (ca. 5800 *Meloidogyne incognita* eggs/100 cc soil). After 8 weeks of incubation at 24 C in the greenhouse, tomato plants from treated seed significantly outperformed the untreated control in respect to plant dry weight and reduction in root galling. Abamectin seed coating was sufficiently effective and long-lasting to provide significant seedling transplant protection against root-knot nematodes.

CHORISMATE MUTASE (HG-CM) OF THE SOYBEAN CYST NEMATODE, *HETERODERA GLYCINES*, CAUSES ALTERED ROOT DEVELOPMENT WHEN EXPRESSED IN SCN RESISTANT SOYBEAN HAIRY ROOTS**. Bekal, S., T. L. Niblack, and K. N. Lambert**. Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

Chorismate mutase (CM) is an esophageal gland protein secreted by plant parasitic nematodes and is thought to play a role in the initial phase of nematode feeding cell formation. Two forms of this gene (Hg-cm-1A and Hg-cm-1B) were recently cloned and characterized in the soybean cyst nematode (SCN). Hg-cm-1B is expressed in SCN that are virulent on some resistant soybeans indicating it may play a role in suppression of host plant resistance. Both forms of the Hg-cm-1 were expressed in SCN resistant soybean hairy roots and cause abnormal root growth. Tests are underway to determine if the roots expressing Hg-cm are more susceptible to SCN and to determine how root cell development is altered.

BACTERIA ASSOCIATED WITH CYSTS OF SOYBEAN CYST NEMATODE (SCN) CAN INFLUENCE SCN EGG HATCHING IN THE PRESENCE OF AN SCN-SUPPRESSIVE SOIL AMENDMENT. **Bent, E.,[1] E. Topp, [1] and T. W. Welacky. [2]** [1]Agriculture & Agri-Food Canada, London, ON;[2] Agriculture & Agri-Food Canada, Harrow ON.

Amendment of agricultural soils with lime-stabilized sewage sludge (LSSS) can reduce infection of soybean (*Glycine max*) by the plant parasitic soybean cyst nematode (*Heterodera glycines,* SCN). The effect of LSSS may be mediated at least in part via soil microorganisms that interact with SCN. Senescent SCN cysts in agricultural soils contain large numbers of bacteria (~10^9 cells/mL cyst volume) including *Lysobacter* sp., a genus that produces proteolytic and chitinolytic enzymes that could affect nematode eggs. Three *Lysobacter* isolates obtained from SCN cysts, as well as an isolate of *Escherichia coli*, were evaluated for their effects on SCN egg hatching, an important stage in the lifecycle of this nematode. Surface-disinfested SCN eggs were suspended in water containing 0–5 g/L LSSS, or 1 g/L calcitic lime, with or without inoculation with 5 ×10^6 CFU test bacteria/mL, and incubated for 12 days. In the absence of LSSS, bacteria had no effect on egg hatching, relative to eggs treated with water. In the presence of LSSS, all 3 *Lysobacter* strains enhanced hatching frequency relative to uninoculated treated eggs, while *E. coli* had no effect. One *Lysobacter* strain promoted hatching relative to uninoculated treated eggs at all concentrations of LSSS as well as in the presence of lime, while the other two promoted hatching at 2.5 or 5 g/L LSSS. Based on these results, cyst-associated bacteria can influence egg hatching of SCN in the presence of LSSS, and could be involved in LSSS-mediated suppression of SCN.

INTERACTIONS OF *MELOIDOGYNE INCOGNITA* AND MONARDA DIDYMA (BEE BALM), A POSSIBLE NEMATODE-ANTAGONISTIC PLANT. **Bernard, E. C., K. D. Gwinn, S. E. Greene, and P. J. Long.** Department of Entomology and Plant Pathology, University of Tennessee, 2431 Center Drive, 205 Plant Sciences, Knoxville, TN 37996-4560.

Bee balm (*Monarda* spp.) is a widely grown, showy genus of perennials in the family Laminaceae. These plants contain a complex mix of essential oils, the composition of which varies among species and cultivars. Incorporation of ground *Monarda* herbage into greenhouse soils can reduce Rhizoctonia seedling disease on tomato. Effects of *Monarda didyma* 'Elsie's Lavender' plants and incorporated herbage on *Meloidogyne incognita* population dynamics were investigated in the greenhouse. Roots of seedlings planted in soil infested with *M. incognita* were readily invaded, but development was slow and most invading juveniles failed to reach the adult stage 60 days after planting. The few females that did develop produced no more than ten eggs each. Giant cell complexes generally were underdeveloped and partially necrotic. Galling was inconspicuous. In another experiment, ground herbage was incorporated into soil (up to 10% by volume) infested with *M. incognita* eggs and planted with a tomato seedling. Egg production was determined 60 days later. Reproduction increased with increasing herbage proportion up to 5%, indicating that incorporated herbage may have stimulated hatch.

FACTORS INFLUENCING FEEDING, ATTRACTION AND AGGREGATION BEHAVIOR OF THE PREDATORS, *LAIMYDORUS BALDUS* AND *DISCOLAIMUS MAJOR* (NEMATODA: DORYLAIMIDA). **Bilgrami, A. L., and R. Gaugler.** Department of Entomology, Rutgers University, New Brunswick, NJ 08816, USA.

The effects of temperature (5–40 °C), prey density (25–250 individuals), duration of predator starvation (0–12 days), and prey incubation (2–12 days) were observed on feeding, attraction, and aggregation behaviors of predatory nematodes, *Laimydorus baldus* and *Discolaimus major* using *Hirschmanniella oryzae* as prey. Time of site formation, number and duration of feeding sites, feeding and post-feeding aggregation and number of predators feeding or showing aggregation at feeding sites were influenced by the above factors. *D. major* was more efficient than *L. baldus.* Minimum time taken by predators to form the feeding site, higher rate of site formation, short durations of feeding site and rates of feeding and aggregation were attributed to the efficiency of 175 prey individuals, which were incubated for less than 12 but more than 16 h either at low (5–10 °C) or higher (40 °C) temperatures. Temperatures, 25–35 °C; prey densities, 200–250 individuals; prey incubation, 12–16 h; and predator starvation, 6–8 days were optimal for various predatory activities at the feeding site. Responses of predators were suppressed when less than 6 or more than 8 days starved individuals were tested against 25–175 prey individuals, which were incubated for less than 12 but more than 16 h either at low (5–10 °C) or higher

(40 °C) temperatures. Present study supports the hypothesis that prey emitted attractants regulated rates of feeding, attraction and aggregation behaviors of predatory nematodes and that changes in prey emitted attractants occurring due to various factors altered rates of feeding, site formation, attraction and aggregation etc.

IMPACT OF AGRICULTURAL SYSTEMS ON NEMATODE COMMUNITY STRUCTURE. **Bird, G. W.,[1] M. F. Berney,[1] P. J. Smeenk,[2] J. E. Sanchez,[2] R. R. Harwood,[2] and F. W. Warner.[1]** [1]Department of Entomology; [2]Department of Crop and Soil Sciences, Michigan State University, East Lansing, MI 48824.

A long-term research site, Living Field Laboratory, was established in 1992, at the MSU, Kellogg Biological Station. The goal was to determine the impacts of four management systems (conventional, integrated fertilizer, integrated compost and transitional organic, associated with continuous corn or a corn-corn-soybean-wheat rotation) on crop yield, soil nutrient mineralization and environmental quality. Each system was replicated four times. Nematode community structure was monitored from 1996–1998 and again in 2001–2002. In general, nematode community structure associated with continuous corn system was more stable than that associated with the rotation system. Following nine years of continuous corn (1993–2001), population densities of bacterial feeding nematodes (June 10, 2002) were significantly ($P = 0.007$) higher (547.5 vs 58.8 per 100 cm^3 soil) in the organic management system with a cover crop, compared to the conventional system without a cover crop. The same was true for fungal feeding nematodes associated with these systems (91.3 vs 20.0 per 100 cm^3 soil, $P = 0.003$). There were no significant differences ($P = 0.05$) between these systems for the plant-feeding, omnivorous or carnivorous nematodes. With continuous corn, initial population densities were higher in the organic system compared to the conventional system in four of the five years. When expressed as a ratio of non-plant parasites/plant parasites, the ratios for the organic system were always higher than those associated with the conventional system. Grain yields of first-year corn were considerably higher than those of second-year corn or yields under continuous corn. Carbon and nitrogen mineralization potentials were significantly greater under the organic, compared to the conventional system.

MOLECULAR BARCODING FOR NEMATODE IDENTIFICATION AND DIVERSITY STUDIES. **Blaxter, M., R. Floyd, and E. Abebe.** Institute of Cell, Animal and Population Biology, University of Edinburgh, Edinburgh EH9 3JT, UK.

We have developed a molecular barcoding system to identify nematode taxa based on the sequence of a segment of the small subunit ribosomal RNA gene (SSU). Sequences are grouped by identity to define Molecular Operational Taxonomic Units (MOTU). This system allows diversity and abundance estimations of nematodes in soils using a standard methodology, without specialist training in nematode taxonomy. Our main study site is the UK Natural Environment Research Council Soil Biodiversity and Ecosystem Function Programme site at Sourhope in the Scottish borders, though we have also sampled other terrestrial and marine sites using the same method. Nematodes extracted using ludox centrifugation and individuals picked at random and digested in sodium hydroxide. A 5′ portion of the SSU is isolated using PCR and sequenced directly. 450–500 bp of sequence was generated per nematode. Sequences were trimmed of low quality bases using PHRED and automatically clustered into MOTU by sequence identity using a custom set of PERL scripts. We have also investigated the use of other markers, such as the internal transcribed spacer, but find that they are too variable to be used reliably. A total of 2039 individual nematode sequences have been generated from across the Sourhope site, from which ~140 distinct MOTU (2 bp variation or less) were defined. The most common MOTU is similar to *Helicotylenchus*, with 835 sequences – around 40% of the total. Other common MOTU show similarity to *Aporcelaimellus* and *Tripyla*. The majority of MOTU found were represented by only a single sequence each. We have investigated the statistical properties of our barcoding dataset, and show that it is a robust, sensitive and efficient way of discovering and describing nematode diversity. In one set of taxa, we have shown that the MOTU correspond more closely to biological species than does analysis using morphological systematics. The rapidly accumulating dataset of SSU sequences from described nematode specimens, and the ever-reducing cost of sequencing makes this approach very attractive.

NEMATODE AND HOST EXPRESSED SEQUENCES IN COMPATIBLE AND INCOMPATIBLE INTERACTIONS WITH POTATO. **Blok, V., M. Armstrong, B. Dixon, J. Wishart, A. Paterson and M. Phillips.** Plant Pathogen Interactions Program, Scottish Crop Research Institute, Invergowrie, Dundee, UK, DD2 5DA.

Nematode sequences isolated from infected potato roots by the suppressive subtractive hybridisation procedure were categorized following database searches according to their EST, BLASTX and BLASTN matches. As well as matches to existing plant parasitic nematode ESTs, either with or without BLASTX matches, other matches were to potential novel nematode sequences. In a compatible interaction, sequences predominantly had matches to ribosomal proteins consistent with increased protein synthesis associated with nematode development. Host sequences with strong matches to Solanum tuberosum or Lycopersicum esculentum ESTs included those implicated in wounding by pathogens and stress responses such as peroxidases, ubiquitin, and HRGP proteins and trancription and signalling factors such as WRKY and receptor like protein kinases.

THE LEVEL OF RESISTANCE IN PROPRIETARY CULTIVARS RESISTANT TO *HETERODERA GLYCINES* IS NOT THE SAME. **Bond, J. P.,[1] T. L. Niblack,[2] and G. R. Noel.[3]** [1]Department of Plant, Soil and General Agriculture, Southern Illinois University, Carbondale, IL 62901; [2]Department of Crop Sciences, University of Illinois, Urbana, IL 61801; [3]USDA, ARS, Urbana, IL 61801.

Approximately 350 cultivars described as resistant to *Heterodera glycines* are sold in Illinois and other Midwestern states. Greater than 90% of these cultivars utilize resistance obtained from PI88.788. In the past few growing seasons, increasing numbers of soybean producers have complained that their resistance to *H. glycines* is not durable. Shifts in the parasitic abilities of nematode populations represent one possibility for the lack of durable resistance. Another possibility is that recent cultivars do not carry the full complement of resistance genes as originally provided by Fayette, a highly utilized resistance source. We determined the level of resistance of 367 cultivars to five populations of *H. glycines* identified as HG Types 3, 7, and 2.5.7 (races 1, 3, and 5), which comprise 90% of the populations in Illinois. Regardless of the HG Type, a large percentage of the cultivars did not carry adequate levels of resistance and would be ineffective in current rotation systems to manage *H. glycines*.

ABSENCE OF *WOLBACHIA* IN NONFILARIID NEMATODES. **Bordenstein, S. R.,[1] D. H. A. Fitch,[2] and J. H. Werren.[3]** [1]J. B. Paul Center for Comparative Molecular Biology and Evolution, Marine Biological Laboratory, Woods Hole, MA 02543; [2]Department of Biology, New York University, New York, NY 10003; [3]Department of Biology, The University of Rochester, Rochester, NY 14627.

Intracellular bacteria of the genus *Wolbachia* are among the most abundant endosymbionts on the planet, occurring in at least two major phyla – the Arthropoda and Nematoda. Current surveys of *Wolbachia* distribution have found contrasting patterns within these groups. Whereas *Wolbachia* are widespread and occur in all three major subphyla of arthropods, with estimates placing them in at least several million arthropod species, *Wolbachia* of nematodes are confined to the filariid nematodes, in which they occur at appreciable frequencies. It has been hypothesized that *Wolbachia* entered the ancestor of modern day filariids in a single acquisition event, and subsequently cospeciated with their filariid hosts since then. To examine the broader distribution of *Wolbachia* in nematodes, we used a polymerase chain reaction (PCR) assay to screen for the presence of *Wolbachia* in a diverse set of nonfilariid species. The assay consisted of 3 different types of PCR screens on adults of 20 secernentean nematode species (14 rhabditids, including 2 strongylid parasites of vertebrates, a diplogasterid, 3 cephalobid relatives, a myolaim, and a filariid) and 2 non-secernentean species (plectids). Two PCR screens were specific to the 16S rDNA and *ftsZ* protein coding genes of *Wolbachia;* a third screen controlled for template presence (nematode 18S rDNA). Based upon our results, we conclude that *Wolbachia* are absent in all 21 non-filariid species. The absence of *Wolbachia* in these non-filariids supports the hypothesis that *Wolbachia* entered the nematode phylum just prior to the divergence of the filariids.

SEGREGATION OF RESISTANCE TO ROOT-KNOT NEMATODES IN A *VITIS RUFOTOMENTOSA* HYBRID POPULATION. **Boyden, L. E.,[1] P. Cousins,[1] and D. W. Ramming.[2]** [1]USDA-ARS, Cornell University, Geneva, NY 14456; [2] USDA-ARS, Postharvest, Quality, and Genetics Research Unit, Parlier, CA 93648.

Development of rootstocks resistant to root-knot nematodes (*Meloidogyne* spp.) is a priority in grape breeding. The nematode resistance of a seedling population derived from a *Vitis rufotomentosa* hybrid was evaluated to investigate the genetic control of resistance in this species. The female parent of the population is the phylloxera resistant rootstock 161-49C, a *V. riparia* × *V. berlandieri* hybrid. 161-49C does not contribute resistance to root-knot nematodes to its progeny. The male parent is 8-10B, a selection of *V. rufotomentosa* x *V. rupestris* from the USDA grape breeding program at the Postharvest, Quality, and Genetics Research Unit. This selection demonstrates resistance to *N*-virulent nematodes, but the genetic nature of its resistance was not known. The source of resistance in 8-10B is its *V. rufotomentosa* parent. *V. rufotomentosa* is native to southeastern North America and is closely related to *V. aestivalis*, accessions of which have been identified as sources of resistance to *N*-virulent root-knot nematode populations. Resistance in the 161-49C × 8-10B cross was assessed in greenhouse pot culture. Seedling roots were stained in an eosin-Y solution six weeks after inoculation with approximately 1500 second-stage *N*-virulent *Meloidogyne arenaria* juveniles. The resistance class of 157 seedlings was determined by assessing the degree of galling and number of egg masses per root system. Segregation in the seedling population was consistent with a 1:1 ratio of resistance to susceptibility, indicating that 8-10B is heterozygous for a dominant allele conferring resistance to *N*-virulent root-knot nematodes. The genetic relationship between this allele and the *N* allele, which confers resistance to *N*-avirulent *Meloidogyne incognita*, has yet to be determined.

MORPHOLOGICAL AND MOLECULAR CHARACTERIZATION OF *MELOIDOGYNE MAYAGUENSIS* FROM FLORIDA. **Brito, J.,[1] T. O. Powers,[2] P. G. Mullin,[2] R. N. Inserra,[1] and D. W. Dickson.[3]** [1]Division of Plant Industry, P. O. Box 147100, Gainesville, FL 32614-7100; [2]Deparment of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722; [3]Department of Entomology and Nematology, University of Florida-IFAS, Gainesville, FL 32611-0620.

The discovery of *Meloidogyne mayaguensis* is confirmed in Florida; this is the first report for the continental USA. *M. mayaguensis* is a virulent species that poses a serious threat to agriculture. Of particular concern is the ability of this

nematode to reproduce on host cultivars specifically bred for nematode resistance. The range of morphometrics of isolates from Florida overlapped with those reported in the literature. Male stylet length values across three Florida isolates were smaller than those of the original description (17.5–21.7 ìm vs. 20.7–24.6 ìm), and similar to those from West Africa. Perineal patterns with high trapezoidal dorsal arches, like those of *M. incognita*, were observed in 30% of female specimens from three isolates. Enzyme analyses of those isolates showed two major bands (VS1-S1 phenotype) of esterase activity, and one strong malate dehydrogenase band (Rm 1.4) plus two additional weak bands that migrated very close together. PCR primers embedded in the COII and 16S genes produced a product size of 702 bp and amplification of the 63 bp repeat region resulted in a single product of 320 bp. Nucleotide sequence comparison of these mitochondrial products together with sequence from 18S rDNA and ITS1 from the nuclear genome were nearly identical with the corresponding regions from a *M. mayaguensis* isolate from Mayaguez, Puerto Rico, the type locality of the species. The nematode reproduced on cotton, pepper, tobacco, watermelon, but not on peanut. Preliminary results indicate these isolates can reproduce on tomato containing the *Mi* gene. Molecular techniques for the identification of this nematode will be particularly useful in cases of mixed species populations and when low numbers of juveniles are recovered from the soil.

BREEDING RESISTANCE TO ROOT-KNOT NEMATODE AND TOBRAVIRUS IN POTATO. **Brown, C. R., and H. Mojtahedi.** USDA Agricultural Research Service, Prosser, WA 99350.

The increased cultivation of potato in warmer and tropical production areas necessitated a search for resistance to southern root-knot nematode (SRN). No useful resistance among cultivated materials was identified. Extensive surveys of wild species identified a number of resistant candidates. The wild species *Solanum sparsipilum* has proven to be a good source that has been introgressed into advanced breeding material. Resistance from *S. sparsipilum* appeared to be highly heritable. *Melodiogyne chitwoodi*, Columbia root-knot nematode (CRN), described first in 1980, has come to dominate root-knot research efforts in certain traditional production areas including the Netherlands and the Western United States. Resistance to CRN was not available in cultivated materials, but was found in *S. bulbocastanum*, *S. hougasii*, and *S. fendleri*. Inheritance studies have determined monogenic dominant resistance in both *S. bulbocastanum* and *S. hougasii*. The genomic location of resistance in both species has been mapped to homologue 11. In contrast, there are numerous varieties and advanced breeding materials presenting resistance to *Tobravirus* tobacco rattle virus (TRV) vectored by Trichodorid nematodes. Although apparently not simply inherited, resistance to TRV is heritable enough to be readily bred into progeny of resistant parents. Transgenic resistance to TRV, utilizing a "pathogen derived resistance" approach has been researched by several groups. Partial levels of resistance have had the undesirable concomitant attributes of being strain-specific or providing resistance to TRV by mechanical inoculation to foliage but not by nematode transmission to tubers. Different strategies of transgenic resistance obviously need to be examined. The introduction of these two types of natural resistance either singly or combined, to new commercial varieties would reduce the burden of soil fumigation that is a particularly heavy cost in the Pacific Northwest of the US.

NOVEL APPLICATION OF SCANNING ELECTRON MICRSCOPY (SEM) FOR INVESTIGATING POSTEMBRYONIC DEVELOPMENT OF THE LIP REGION OF *ACROBELES COMPLEXUS*. **Bumbarger, D., and J. G. Baldwin.** Department of Nematology, University of California, Riverside, CA 92521.

Nematodes of the subfamily Cephalobinae (Rhabditida) are distinct in the presence of cephalic cuticular appendages including modified lips and probolae. There is a gradient from highly complex appendages to a complete lack of elaboration of these structures. Earlier work has documented a wide range of adult head morphologies using SEM, but studies of juvenile morphology are rare and limited to light microscopy. Here we have examined the adult and each of the four juvenile stages of *Acrobeles complexus* with SEM. A new mounting method allows for increased manipulation of specimen orientation and more complete observations of single individuals than was previously possible. Observations demonstrate that the first juvenile stage has a unique morphology compared to other stages with respect to both cuticular and sensory structures. First stage labial probolae are not deeply bifurcate and have a unique tine morphology, primary and secondary lip axils are of markedly different depths, guarding processes and some papillae are apparently missing. Second through fourth stage juveniles, while very similar to adults, differ in the number of tines and in the morphology of tines on the cephalic probolae. Amphidial openings change gradually from ovate in the first juvenile stage to circular in the adult, while increasing in size. Due to dramatic differences between early juvenile and adult cephalic morphologies, characters obtained from juvenile morphologies could prove useful in diagnosing closely related taxa, constructing phylogenies and understanding patterns in the evolution of novel structures.

EFFECT OF BANANA NUTRITIONAL STATUS ON HOST TOLERANCE TO THE BURROWING NEMATODE *RADOPHOLUS SIMILIS*. **Bwamiki, D. P., J. M. Duxbury, and J. Esnard.** Department of Soil and Crop Sciences, Cornell University Ithaca NY 14850.

Banana (Musa spp) is an important source of carbohydrates for many people. *Radopholus similis* causes serious economic losses in Musa ssp. Banana response to *R. similis* attack was studied under different nutritional regimes to assess

whether manipulation of soil nutrition could be used in nematode control. Grande Naine banana plantlets were grown in 11-liter pots under different nitrogen (N), phosphorus (P), and potassium (K) nutritional regimes and nematode populations in a controlled-temperature greenhouse. Nutrient treatment combinations containing N at: 210, 420 and 840; P at: 21, 42 and 84; K at: 610, 1220 and 2440 mg/L were added to soil over 20 weeks. Plantlets were allowed to grow for 8 weeks and then inoculated with 0, 1000, 2000 and 4000 nematodes per plant. Plants were harvested 12 weeks after inoculation and assessed for root necrosis, nematode numbers, and biomass production. NPK at 420:42:610 resulted in the lowest necrotic damage, lowest number of nematodes in the roots and the largest biomass production. These results suggest that the impact of burrowing nematodes on banana is reduced at a lower level of potassium than the high levels often recommended for good crop production. The relationship between nematode populations and plant nutrient uptake will be discussed.

USE OF HSP90 FOR NEMATODE PHYLOGENY. **Carta, L. K., and A. M. Skantar.** USDA-ARS Nematology Laboratory, Beltsville, MD 20705.

We have been examining the nuclear Hsp90 chaperone gene for usefulness in phylogenetic prediction. Only one copy of this gene is known from *Caenorhabditis elegans*. Using degenerate primers for partial Hsp90 sequences, we detected only one copy of Hsp90 in *Heterodera glycines* and *Meloidogyne javanica*. These partial Hsp90 sequences produced well-supported nine-taxon trees with generally expected topology using MrBayes, a powerful new phylogenetic program. However, trees were poorly resolved with the same nematode taxa and similar-size or longer sequences from LSU (28S) and SSU (18S) ribosomal DNA genes. Both LSU and Hsp90 genes supported the monophyly of *Meloidogyne* with *Pratylenchus* instead of *Meloidogyne* with *Heterodera*, a grouping expected from a traditional taxonomic interpretation. On a broader phylogenetic scale, using entire Hsp90 amino acid sequences published for other metazoans, nematodes appeared paraphyletic to insects. This relative position is consistent with most other nuclear protein-coding genes.

PERSISTENCE OF *PASTEURIA PENETRANS*. **Cetintas, R., and D. W. Dickson.** Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620.

Pasteuria penetrans was reported to suppress *Meloidogyne arenaria* race 1 in a peanut field in Levy County, FL. Our objective was to determine the persistence of *P. penetrans* in this site by determining the density of the bacterium following 9 years of continuous cultivation of bahiagrass, rhizomal peanut, and weed fallow. The treatments were chosen to include root-knot nematode nonhosts (bahiagrass and rhizomal peanut) and weed hosts (weed fallow). The plots were established in a randomized complete block with ten replicates in the summer of 1991. In 1999, the bahiagrass and weed fallow plots were plowed, disked, and peanut planted. Glyphosate was sprayed over the rhizomal peanut in the summer of 1999 and the plots were plowed and disked in the spring of 2000. All plots were planted to peanut in 2000–2002. In 1999, the initial density of *M. arenaria* second-stage juveniles (J2) was low in all plots and no J2 with endospores attached were recovered. Approximately 2.5% of root-knot nematode females recovered from peanut grown in weed fallow plots were filled with endospores of *P. penetrans*. No endospore-filled females were recovered from peanut grown in bahiagrass and rhizomal peanut plots. The percentage of J2 with endospores attached reached the highest level of 65.3% between June and August of 2001 in weed fallow, and 34% and 24% in August of 2002 in bahiagrass and rhizomal peanut, respectively. The percentage of endospore-filled females recovered from peanut grown in weed fallow plots increased to 55% in 2002, whereas the percentages in bahiagrass and rhizomal peanut plots were 40% and 15%, respectively.

MELOIDOGYNE JAVANICA ON PEANUT IN FLORIDA. **Cetintas, R., and D.W. Dickson.** Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620.

Analysis was made by PAGE of esterase phenotypes of *Meloidogyne* spp. females extracted from roots of peanut collected from an experimental research site, in Alachua County, Florida. Following 9 years of continuous cultivation of bahiagrass, rhizomal peanut and weed fallow, the site was planted to peanut from 1999 to 2002. In 2002, of 290 females characterized, an average of 71% showed a typical esterase pattern for *M. arenaria* phenotypes A2, whereas 30% of females collected from the site showed typical esterase patterns for *M. javanica* J3 phenotype. A greater percentage of *M. javanica* (42%) was found in weed fallow plots then in the bahiagrass and rhizomal peanut plots, which were 23% and 25%, respectively. These results also were supported by malate dehydrogenase phenotypes and observation of perineal patterns taken from single egg masses of the population from the site. This confirms the diagnosis of *M. javanica* on peanut in Florida.

THE ECOLOGY OF *C. ELEGANS:* PHENOTYPIC PLASTICITY, SURVIVAL, AND FACULTATIVE VIVIPARY. **Chen, J., and E. P. Caswell-Chen.** Department of Nematology, University of California, Davis, CA 95616.

The nematode *C. elegans* is used as a model system in many areas of research, including aging and development. The worm is considered cosmopolitan, although relatively few studies have been conducted on its biology and ecology in nature. The nematode life history includes a well-documented survival and dispersal stage, the dauer. A component of *C. elegans* life history seldom considered in detail is the phenomenon of *endotokia matricida*, or internal hatching of larvae

(referred to as "bagging"). Many nematode species exhibit intra-uterine birth with resulting death of the parent, including *Heterorhabditis spp., Mehdinema alii*, and *Metacrobeles amblyurus*. We have observed internal retention of larvae in *C. elegans* as a response to stress and starvation, and that such retention allows the larvae to feed on the parent body contents and obtain sufficient nutrition to reach the resistant, long-lived dauer stage. We suggest that intra-uterine hatch is a life-history trait in *C. elegans* and is a form of facultative vivipary. This phenotypic plasticity complements androdioecy and the dauer stage to enhance progeny survival and dispersal under stress. Understanding facultative vivipary may also provide insights into the evolution of reproduction and longevity.

GENETIC ANALYSIS OF (A)VIRULENCE IN *MELOIDOGYNE HAPLA* MATCHING RESISTANCE IN COMMON BEAN (*PHASEOLUS VULGARIS*). **Chen, P.,[1] and P. A. Roberts.** [2] [1]Department of Plant Pathology, Cornell University, Geneva, NY14456; [2]Department of Nematology, UC Riverside, Riverside, CA92521.

A collection of seven *Meloidogyne hapla* isolates were differentiated into three groups according to their reactions on susceptible and resistant common bean. Four isolates were virulent to resistant cv. NemaSnap, two isolates were avirulent and one isolate reproduced significantly less on susceptible cv. Yolano than resistant NemaSnap. Crosses were made between an avirulent maternal and two virulent paternal *M. hapla* isolates in modified plant growth pouches. Starting at 14 days after inoculation with the avirulent female line, 3–5 virulent males were hand-picked and added to the females every 3 days over a period of 17 days. Putative F1 egg masses were transferred individually to a new growth pouch to increase the population. Three F1 lines at the F3 stage had significant reproduction on both susceptible and resistant plants, indicating virulence transfer from the male line. The F3 cohort from these three F1 lines and the F4 heterozygous families had a virulence percentage of 25% (AA+Aa+aa: aa = 4: 1), indicating (a)virulence was controlled by a single gene. In the established F4 families, homozygous avirulent, homozygous virulent and heterozygous segregating progenies were observed. Segregation patterns fit a 3 (avirulence) to 1 (virulence) ratio, indicating that virulence was recessive to avirulence.

CHEMICAL INDUCTION OF SYSTEMIC ACQUIRED RESISTANCE (SAR) AGAINST PLANT-PARASITIC NEMATODES IN PINEAPPLE. **Chinnasri, B., B. S. Sipes, and D. P. Schmitt.** Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa.

The potency of acibenzolar-S-methyl, DL-á-amino-n-butyric acid (AABA), DL-â-amino-n-butyric acid (BABA), ã-amino-n-butyric acid (GABA), p-aminobenzoic acid (PABA), salicylic acid (SA), and riboflavin in activating systemic acquired resistance (SAR) against *Rotylenchulus reniformis* and *Meloidogyne javanica* in pineapple was investigated in a greenhouse experiment. All the inducers were applied as foliar sprays on 1-month-old pineapple plants (20 ml/plant) grown in 22-cm-diam pots. Two days after application, 10,000 eggs of *R. reniformis* or *M. javanica* were inoculated onto the plants. The results, obtained 6 months after inoculation, showed that acibenzolar-S-methyl was the only potent inducer. Acibenzolar-S-methyl decreased *R. reniformis* egg production by 58% as compared to the untreated control. With *M. javanica*, however, acibenzolar-S-methyl, BABA, and riboflavin were among the effective compounds, reducing egg production by 60–64%. The point in the SAR pathway that each compound affects may explain the different results. Acibenzolar-S-methyl may act upstream in SAR signaling; thereby controlling the formation of both nematode feeding sites, syncytia formed by *R. reniformis* whereas giant cells formed by *M. javanica*. BABA and riboflavin may act downstream by activating components in the SAR pathway that affects only the formation of giant cells.

RESISTANCE TO *MELOIDOGYNE* IN *ARACHIS HYPOGAEA:* HISTORY AND FUTURE. **Church, G.,[1] J. L. Starr,[2]** and **C. E. Simpson**.[3] [1]USDA, ARS, USHRL, Fort Pierce, FL 34945; [2] Department of Plant Pathology & Microbiology, Texas A&M University, College Station, TX 77843; [3] Texas Agricultural Experiment Station, Stephenville, TX 76401, USA.

Peanut, *Arachis hypogaea* L., is grown worldwide in tropical, subtropical and warm temperate climates as a major oil seed crop. *Meloidogyne* species cause substantial economic losses in most peanut producing countries. The management of nematodes in peanut production is primarily through the application of nematicides. A high level of resistance to *Meloidogyne* spp. was not known among *A. hypogaea* genotypes prior to 1989. Resistance to *M. arenaria, M. hapla,* and *M. javanica* has been identified among the wild *Arachis* species and an interspecific hybrid. After more than a decade of backcross breeding, resistance to *M. arenaria* and *M. javanica* was introgressed into *A. hypogaea* cv. Florunner, resulting in the release of the cultivar 'COAN'. Resistance in COAN is inherited as a single dominant gene. However, resistance in the interspecific hybrid is inherited as one dominant gene and one recessive gene. RFLP markers, linked to the dominant resistance gene, were identified from a genetic map of peanut. These markers were used in marker-assisted selection of homozygous resistant individuals in the development of the cultivar 'NemaTAM'. Many peanut cultivars are susceptible to *Cercospora arachidicola*, *Cercosporidium personatum*, *Sclerotina minor*, *Puccinia arachidis* and tomato spotted wilt virus. Efforts have been made to combine the two nematode resistance genes with resistance to these plant pathogens.

DIFFERENTIAL MORTALITY OF *HETERODERA GLYCINES* MALES AND FEMALES AFFECTS SEX RATIOS ON RESISTANT SOYBEAN. **Colgrove, A. L.,[1] and T. L. Niblack.[2]** [1]Department of Plant Microbiology and Pathology, University of Missouri, Columbia, MO 65211; [2]Dept. of Crop Sciences, University of Illinois, Urbana, IL 61801.

The variability of sex ratios of *Heterodera glycines* on resistant hosts is not fully understood. Males and females are both required for reproduction and typically occur in an approximate1:1 ratio. However, the adult sex ratio is altered on resistant hosts. This could be attributed to either environmental factors that affect sex determination or differential mortality of males and females. Lines of *H. glycines* characterized by zero and high female numbers on resistant soybean were used to observe male numbers. All *H. glycines* lines were inbred isolates that had specific compatibilities with a range of soybean genotypes. Infection rates at five days after inoculation and relative male/female numbers after one generation in hydroponic culture were determined. Resistance that suppressed adult females also reduced numbers of adult males. On Peking, PI 90763, and PI 437654, male numbers were low and close to zero, but on PI 88788, male numbers were much higher. In a separate test, an HG Type 0 was used to investigate male and female numbers on Peking, PI 88788, PI 90763, PI 437654, PI 209332, PI 89772, and PI 548316. Male numbers were similar to female numbers on Peking, PI 90763, PI 437654, and PI 89772 while male numbers on PI 88788, PI 209332, and PI 548316 were higher than that of females. In all tests, total adult numbers on resistant soybean were significantly less than that on susceptible soybean, indicating that resistance influences survival and not sexual development of *H. glycines.*

MULTIPLE EGG HARVESTS FROM *MELOIDOGYNE*-INFESTED TOMATO ROOT SYSTEMS. **Cotter, H. V. T., C. B. Hicks, and J. A. Simmons.** BASF Global Insecticide and Nematicide Biology Research, Research Triangle Park, NC 27709.

Maximizing egg production from greenhouse root-knot nematode (*Meloidogyne*) cultures is an important component of improving the efficiency of root-knot screening programs for both chemical and non-chemical approaches to discovery of new technology to control root knot. We have found that incubation of the naked root systems in a moist box after the initial sodium hyprochlorite extraction of *Meloidogyne* eggs allows production of a second set of eggs that can then be harvested 4 days following the initial harvest. Our *Meloidogyne* cultures are maintained on tomato plants (cultivar Bonny Best) in a 1:1 mixture of sandy loam and coarse sand in 3′ plastic pots. Eggs are harvested typically 2–3 months after the plants are inoculated with second-stage juveniles (J2s). Root systems, devoid of the shoots, are washed free of soil then shaken in three washes of 0.6% sodium hypochorite water to free the eggs; each wash is shaken for 30 seconds. After each wash the released eggs are collected on a 500-mesh sieve and immediately rinsed with water. After the third extraction (wash), the sodium hypochorite is rinsed from the root systems, which are then placed on moist paper toweling in boxes at ambient lab conditions to allow further egg production by the *Meloidogyne* females on the roots. After 4 days incubation these eggs are harvested using the same method. Percent egg hatch from a given second batch of eggs was higher than that from the corresponding first batch. J2s produced from the second egg batch were infective. The second egg harvest increased egg yield by 70% on average, but the increase was highly variable.

MANAGEMENT OF *BELONOLAIMUS LONGICAUDATUS* ON ESTABLISHED BERMUDAGRASS FAIRWAYS USING 1,3-DICHLOROPROPENE. **Crow, W. T.,[1] R. M. Giblin-Davis, [1] and D. W. Lickfeldt. [2]** [1] Dept Entomology & Nematology, University of Florida, Gainesville; [2] Dow AgroSciences, Fayetteville, GA.

Belonolaimus longicaudatus is considered the most damaging of all plant-parasitic nematodes to bermudagrass (*Cynodon dactylon*). Nematicide options for managing *B. longicaudatus* on established turfgrasses have been drastically reduced with the cancellation of several organophosphate nematicides over the past several years. 1,3-Dichloropropene (1,3-D) has been proven an effective nematicide for management of *B. longicaudatus* as a preplant treatment on many crops. The objective of this research was to determine if 1,3-D can be used as a post-plant nematicide to manage *B. longicaudatus* on established bermudagrass fairways without causing unacceptable phytotoxicity. In a series of experiments, 1,3-D applied at 55 kg a.i./ha by slit-injection was effective at reducing population densities of *B. longicaudatus* on established bermudagrass fairways ($P < 0.05$), improving turf health, and enhancing root development. Phytotoxicity was minimal and was within the tolerance of most turf managers.

FROM BOTH SIDES NOW? LOOKING AT THE NEMATODE'S ROLE IN SYMBIOSIS WITH *C. ELEGANS* GENETIC TECHNIQUES. **Darby, C.** Department of Microbiology, University of Alabama at Birmingham, Birmingham, AL 3529.

The interactions between nematodes and bacteria in the entomopathogenic symbioses are not understood. Highly specific associations occur between *Steinernema* sp. nematodes and *Xenorhabdus nematophila* bacteria, and between *Heterorhabditis* sp. and *Photorhabdus luminescens*, but the underlying mechanisms (e.g. signaling, recognition, adherence) are unknown. Complete understanding of a symbiosis requires tools to analyze both participants. Encouraging progress has been made in identifying bacterial genes, but the nematode side remains *terra incognita*. Genetic techniques developed for use with the model nematode *Caenorhabditis elegans* may be adaptable to *Steinernema* sp. or *Heterorhabditis* sp., despite the absence of genome sequences or even genetic maps. RNA interference (RNAi) is a technique in which administration

of double-stranded RNA corresponding to a coding region of a gene mimics the phenotype of a mutation in the gene. Merely feeding *C. elegans* an *E. coli* strain that synthesizes dsRNA is frequently sufficient to produce a phenotype, and the speed and simplicity of this procedure has made possible genome-wide screens for informative genes. RNAi is being developed for *Steinernema carpocapsae*. Following demonstration experiments, a library of *S. carpocapsae* clones will be made in a vector that expresses dsRNA. The clones will be administered singly by feeding, and the nematodes will be screened for symbiosis defects. Clones producing a phenotype will be sequenced to identify immediately the relevant gene. Another potential genetic tool is a transposon mutagenesis procedure developed for *C. elegans*. This technique creates mutations by random insertion of heterologous DNA into the genome, and the polymerase chain reaction is used to amplify nematode DNA flanking the insertion site. The mutated gene therefore can be identified without genetic mapping.

COMPARATIVE GENOMICS OF GENE EXPRESSION IN PARASITIC AND FREE-LIVING NEMATODES. **Dautova Mitreva, M.,[1] J. P. McCarter,[1] J. Martin,[1] C. Murphy,[1] S. Clifton,[1] and R. H. Waterston.[2]** [1] Genome Sequencing Center, Department of Genetics, Washington University School of Medicine, St. Louis, MO 63108; and [2] Department of Genome Sciences, University of Washington, Seattle, Washington 98195.

Molecular characterization of parasites, as well as the development of new techniques for control, can benefit from genomic approaches. As an entrée to characterizing parasitic nematode genomes, we have generated 196,500 expressed sequence tags (ESTs) from 26 nematode species as of April 2003. A key goal of our bioinformatic analysis is to organize this large dataset uniformly across multiple species in a manner that allows simple access by the community of nematologists. We will report on the progress of parasitic EST analysis including: creation of NemaGene clusters to reduce sequence redundancy, identification of common and rare represented genes, identification of stage-specific expression, functional classification based on Gene Ontology assignments, biochemical pathway identification using KEGG, and identification of genes orthologues in *C. elegans* and other nematodes. All sequences are publicly available at www.ncbi.nlm.nih.gov/dbEST . Projects details, NemaGene sequences and a list of collaborators are available at www.nematode.net. The project is funded by grants NIH-NIAID-46593 and NSF-0077503.

CONTROL OF COLUMBIA ROOT-KNOT NEMATODE (*MELOIDOGYNE CHITWOODI*) IN POTATO CV RUSSET NORKOTAH WITH OXAMYL. **David, N. L.,[1] R. E. Ingham,[1] N. D. McKinley,[2] and B. A. Charlton.[3]** [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 Experiment Station, 6941 Washburn Way, Klamath Falls, OR 97603.

Columbia root-knot nematode (*Meloidogyne chitwoodi*, CRKN) is a serious pathogen of potato *(Solanum tuberosum)* in many areas of the western United States. Tuber infection by CRKN reduces tuber quality by inducing surface galling and/or internal brown spots. Trials at Hermiston, OR in 2001 demonstrated that oxamyl (Vydate C-LV®) applications of 1 lb. a.i./a beginning at planting (in-furrow) and continuing through the season at two-week intervals after 950 soil degree-days base 5C (DD_{5C}) had significantly less infection than the non-treated control. However, waiting until crop emergence or 950 DD_{5C} to make the initial application was not significantly different from the non-treated control. Trials in Klamath Falls, OR during 2002 further substantiated findings from Hermiston in 2001. All treatment schedules with 1 lb. a.i./a per application in Klamath Falls had significantly lower infection than the non-treated control. However, treatment schedules with an in-furrow application of oxamyl at planting had significantly less infection than schedules beginning at 950 DD_{5C}. Moreover, the schedule of oxamyl in-furrow, at crop emergence, and at 950 DD_{5C} had significantly less infection than the schedule of oxamyl at 950 DD_{5C}, two-weeks later, and four-weeks later. In-furrow applications clearly enhance oxamyl efficacy against CRKN in potato. Furthermore, the Klamath Falls study suggests that early season oxamyl applications may be adequate to control CRKN in potato in short, cool growing regions.

THE EFFECT OF OXAMYL ON COLUMBIA ROOT-KNOT NEMATODE *(MELOIDOGYNE CHITWOODI)* POPULATION DYNAMICS ON POTATO. **David, N. L., and R. E. Ingham.** Department of Botany and Plant Pathology 2082 Cordley Hall, Oregon State University, Corvallis, OR 97331.

Columbia root-knot nematode (*Meloidogyne chitwoodi,* CRKN) is a serious pathogen of potato *(Solanum tuberosum)* in many areas of the western United States. The first generation of CRKN is able to infect potato roots as soon as they are present; however, tuber infection does not occur until the second generation of CRKN hatches, 950 soil degree-days base 5 C (DD_{5C}) after planting. Applications of oxamyl to control CRKN have generally been initiated at 950 DD_{5C} and continuing on a two-week interval until harvest. Trials at Hermiston, Oregon, were established to study the effects of oxamyl on population dynamics of CRKN in potato. Soil samples were taken every two weeks from planting until 800 DD_{5C} and then once a week until harvest. Trials in 2001 indicated that waiting until 950 DD_{5C} to begin oxamyl applications of 1 lb. a.i./a had little effect on the fecundity of the first generation of CRKN compared to the control, while oxamyl applications beginning at planting or emergence resulted in lower reproduction of the first generation of CRKN. It appears that beginning applications of oxamyl at planting delayed the hatching of the second generation of CRKN, whereas waiting until crop emergence or 950 DD_{5C} did not. Oxamyl applications of 1 lb. a.i./a made after 950 DD_{5C} on a two-week schedule

were sufficient to reduce the hatching of the third generation, which occurs around 1500 DD_{5C}, to undetectable levels. These data indicate that waiting until 950 DD_{5C} for the initial oxamyl application may be too late to reduce second generation hatch of CRKN, increasing the chance for tuber infection.

NEMATODE COMMUNITY STRUCTURE OF A DECLINING NORTHERN HARDWOOD FOREST. **Davidson, W. L., K. B. Nguyen, R. McSorley, and B. J. Adams.** Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620.

A forest survey conducted in the northern hardwoods of the Blue Ridge Mountains revealed that forest decline is occurring in large patches from Mt. Rogers, VA to The Smoky Mountains, TN/NC. Differences between nematode community composition between areas of light and severe decline may help to determine the causal factor(s) of the decline. Nematode communities were assayed from four different sites in the Great Balsam Mountains, NC, with two objectives: 1) to produce the first catalogue of the nematode communities from these mountains, and 2) to ascertain whether or not nematode community data can aid in determining the cause(s) of declining forest health. A stand of yellow birch, *Betula alleghaniensis,* with greater than 50% mortality, a stand of yellow birch with 0% mortality, a stand of American beech, *Fagus grandifolia*, with 100% mortality, and a stand of northern red oak, *Quercus rubra,* with 25% mortality were selected for study. At each site, soil under five trees was sampled. Using a standard sieving and centrifugation procedure, nematodes were extracted from 100-cc subsamples. Each nematode was identified to the lowest practical taxon. The data were then categorized into five trophic groups: bacterivores, fungivores, herbivores, omnivores, and predators. Thus far, 69 nematode genera have been recovered from these sites. *Filenchus, Aphelenchoides, Plectus, Acrobeloides, Prismatolaimus, Cervidellus, Teratocephalus, Alaimus, Criconemoides, Eudorylaimus,* and *Aporcelaimellus* were common at all sites. The healthy birch site had significantly more predacious nematodes (Mononchida) than the birch decline site. Specifically, *Mylonchulus* and *Iotonchus* were found only at the healthy birch site. These results suggest some initial differences among sites, which will be verified with repeated sampling and soil chemistry analyses.

ROOT-KNOT NEMATODES: A POTENTIAL PROBLEM ON VIDALIA ONIONS. **Davis, R. F.,[1] and D. B. Langston.[2]** [1]Crop Protection and Management Research Unit, USDA ARS, Tifton, GA 31793; [2]Department of Plant Pathology, University of Georgia, Tifton, GA 31793.

Vidalia onions may be one of many cultivars of Yellow Granex onions produced in a region, defined by law, surrounding Vidalia, GA. Unidentified *Meloidogyne* species are sometimes found at high densities in onion fields. Our objective was to evaluate the suitability of onion (*Allium cepa* cv. Sweet Vidalia) as a host for *Meloidogyne incognita*, *M. arenaria*, and *M. javanica*, which are commonly found in Georgia. Nematode reproduction was evaluated in two greenhouse trials with six replications each. Single onion transplants were put into individual 15-cm-diam pots and allowed to grow for 5 or 7 weeks prior to inoculation of the first and second trials, respectively. Each pot was inoculated with 8,000 eggs of one nematode species. Reproduction was measured by extracting and counting eggs from onion roots 54 days after inoculation in both the first and second trials. Relative reproduction differed between trials ($P \leq 0.05$), so data were analyzed separately. All three nematode species increased with final egg counts of 19,300 for *M. incognita*, 32,100 for *M. arenaria*, and 40,350 for *M. javanica* in the first trial and 167,200 for *M. incognita*, 71,600 for *M. arenaria*, and 101,950 for *M. javanica* in the second trial. Final egg counts were similar ($P \geq 0.05$) among the three species in the first trial, but *M. incognita* produced more eggs ($^*P \leq 0.05$) than *M. arenaria* in the second trial. Onion can be a good host for all three *Meloidogyne* species tested, but reproduction and damage in the field may be limited by soil temperatures.

MORPHOLOGICAL AND MOLECULAR COMPARISONS OF TWO *BREVIBUCCA* SPECIES . **De Ley, I. Tandingan,[1] W. Sudhaus,[2] and P. De Ley.[1]** [1]Department of Nematology, University of California, Riverside, CA 92521; [2]Freie Universität Berlin, AG Evolutionsbiologie, Institut für Zoologie, Königin-Luise-Straße 1-3, 14195 Berlin-Dahlem, Germany.

The genus *Brevibucca* is traditionally classified in Panagrolaimoidea, but a recent molecular and developmental analysis does not exclude alternative placements, nearer to e.g. Rhabditoidea or Cephaloboidea. No detailed descriptions of *Brevibucca* species were published in the last forty years. We studied two german isolates of *Brevibucca* (SB 117 and SB 261) and report on their morphology and morphometry as observed with light microscopy and Scanning Electron Microscopy. We also obtained sequence data from the Internal Transcribed Spacer region, the D2D3 expansion segment, and small subunit rDNA. SB 117 is identified as *Brevibucca punctata*, while SB261 is identified as *B. saprophaga*. Both species are redescribed here and their relationships with other Rhabditida are analyzed from the perspectives of morphological cladistics and molecular phylogenetics. Taxonomic implications are discussed, particularly with reference to the recently proposed placement of the genus in a separate family.

PHYLOGENY AND SEQUENCE VARIATION IN THE RIBOSOMAL DNA OF SIX SPECIES OF *LONGIDORUS* (NEMATODA). **De Luca, F.,[1] A. Reyes,[2] J. Grunder, [3] P. Kunz,[3] A. Agostinelli,[1] C. De Giorgi,[4] and F. Lamberti.[1]** [1]Istituto per la Protezione delle Piante, Sezione di Bari, CNR, Via Amendola 165/A, 70125 Bari, Italy; [2]Istituto di Tecnologie Biomediche, Sezione di Bioinformatica e Genomica Comparata, CNR, via Amendola 165/A, 70125 Bari, Italy; [3]Nematology Section, Swiss Federal Research Station, Wàdenswill, Switzerland; [4]Dipartimento di Biochimica e Biologia Molecolare, Università di Bari, via Orabona 4/A, 70125 Bari, Italy.

A comparative analysis of the nucleotide sequences of the ITS containing region and the D1-D2 region of the 28S rDNA were used to assess for their potential utility in reconstructing phylogenetic relationships in six species of *Longidorus* from Switzerland. The D1-D2 sequences resulted more conserved than the ITS sequences that varied widely in primary structure and length, and no consensus was observed. Extensive interspecies sequence variation and minor intraspecie sequence variation were observed. The phylogenetic analyses were carried out by using the minimum evolution method with three different sequence data sets: ITS1-ITS2, 5.8S-D1-D2 and combining ITS1-ITS2+5.8S-D1-D2. The trees obtained were almost the same and two main groups were always evident: the first group includes *L. macrosoma* and *L. helveticus*, whereas the second group includes *L. profundorum*, *L. arthensis, L. elongates,* and *L. raskii.*

DEVELOPMENTS WITH *PASTEURIA PENETRANS* AS A BIOLOGICAL CONTROL AGENT. **Dickson, D. W., and R. Cetintas.** Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620.

Pasteuria penetrans is an important parasite of some species of root-knot nematodes. The bacterium, which appears to be density dependent, attaches to the preparasitic second-stage juveniles of root-knot nematodes in soil. These juveniles that are encumbered by spores may infect a host plant, grow and develop to females, thereby allowing the bacterial parasite to develop. An infected female may contain up to 2 to 2.5 million spores, however females lose their ability to lay eggs. With the loss of fecundity there is a proportional loss in healthy juveniles to continue the cycle for *P. penetrans*. At this point the bacterium has developed to a suppressive level thereby abating root-knot disease for some unknown period of time. This raises the question about long-term persistence of *P. penetrans* in an identified suppressive soil. In a site previously known to be suppressive to *Meloidogyne arenaria* the bacterium did not again reach suppressive levels after 9 years of continuous bahiagrass, rhizomal peanut, and weed fallow, and 4 years of continuous peanut.

TARGETING SOIL SAMPLE COLLECTION OF *MELOIDOGYNE INCOGNITA* IN SOUTHERN MISSOURI SOYBEAN FIELDS FOR MANAGEMENT OF YIELD LOSSES. **Donald, P. A.,[1] J. A. Wrather,[2] and G. Shannon.[2]** [1]USDA ARS Crop Genetics and Production Research Unit, Jackson, TN 38301; [2] Plant Science Unit, Delta Center, Portageville, MO 63873.

Meloidogyne incognita, root-knot nematode, suppresses yield in some soybean production fields in southeastern Missouri. Producers can minimize this yield suppression through wise selection of root-knot nematode management strategies based on the nematode population density in the field during the fall. Data on the best time during the fall for producers to collect soil samples has not existed. Our objective was to measure the M. incognita population density biweekly from beginning bloom to early-December at 10 locations in each of 2 soybean fields during 2001 and 2002. These data were used to predict the optimal time of soil sample collection for predictive management decisions for the next growing season. The number of juvenile plus male root-knot nematodes present in the soil peaked the first part of October in both years and the ratio of plant-parasitic nematodes to total nematodes was also highest at this time. Males were first observed in soil samples in August in both years. There was a secondary peak in numbers of vermiform root-knot nematodes in late November 2001 and early December 2002. Based on these results, southeast Missouri producers should sample fields in early-October to increase the likelihood of detecting root-knot nematodes if present in a soybean production field and to best measure risk of yield loss due to these nematodes.

ECOLOGICAL STUDY OF A MARINE NEMATODE COMMUNITY IN SOUTHERN SAN FRANCISCO BAY. **Duarte, E. O.,[1] J. J. Chitambar,[2] S. B. Opp,[1] and E. B. Lyke.[1]** [1]Department of Biological Sciences, California State University, Hayward, Hayward, CA 94542; [2]Plant Pest Diagnostics Branch, California Department of Food and Agriculture, Sacramento, CA 95832.

A one-year study was conducted of a marine nematode community in southern San Francisco Bay between August 1999 and July 2000. Monthly samples were collected at-two mudflat sites to assess spatial and temporal variations in nematode abundance and community composition. Physical field factors (sediment temperature, salinity, RPD depth, & sediment granulometry) were also monitored for correlation with the nematode community. The nematode community was similar at the two study sites, and was composed of at least six nematode species. The community was dominated by *Sabatieria pulchra*, and included to a lesser degree *Cobbia* sp., *Paramonhystera* sp., *Terschellingia* sp., and *Pelagonema* sp. Significant differences in nematode abundance were observed within and between the study sites, and also over time. Nematode abundance was minimal in winter (February 2000), and maximal during late spring and summer (May, June, July 2000). Mean sediment temperature, salinity, and RPD depth did not vary significantly within or between the study sites,

but differed significantly over time in characteristic regional patterns. Sediment granulometry (particle size distributions) differed significantly at the two study sites, and also varied significantly over time. Seasonal changes in nematode abundance correlated positively with seasonal changes in mean sediment temperature. No other significant correlations were found between seasonal changes in nematode abundance and the remaining physical field factors. Current findings support previous reports that nematode communities of estuarine habitats contain relatively few species, and exhibit marked seasonal differences, which correlate with seasonal temperature changes.

FOOD WEB INVOLVEMENT IN THE REGULATION OF CITRUS PESTS AND DISEASES BY ENTOMOPATHOGENIC NEMATODES. **Duncan, L. W.** University of Florida, Citrus Research and Education Center, Lake Alfred, FL 33850.

The foremost biotic threat to citrus in Florida is a pest-disease complex caused by the weevil *Diaprepes abbreviatus* and fungi in the genus *Phytophthora*. Due to deregulation of effective insecticides, many growers rely on commercial entomopathogenic nematode (epn) products to help manage soilborne stages of the insect. Augmentation of epn to reduce population densities of both the weevil and *Phytophthora* spp. can be profitable in groves located on Florida's central ridge. However, efficacy has been inconsistent and often poor elsewhere. Endemic epn species are a key factor regulating population density of *D. abbreviatus* on the central ridge, in contrast to some orchards examined in other regions. Coincidentally, *D. abbreviatus* is less abundant and damaging in orchards on the central ridge than elsewhere in the state. Regional variation in soil texture may be a factor in the variable response to epn use, the prevalence of endemic epn, and consequently, the distribution of *D. abbreviatus* in Florida. Due to significant natural control of *D. abbreviatus*, a better understanding of key food web relationships involving this insect on the central ridge could help to maximize profitabilily of biological control efforts. For example, evidence of greater persistence of endemic compared to exotic epn raises the possibility that competition from exotic epn, which can reduce the prevalence of endemic epn, may mitigate the value of nematode augmentation. Density dependent factors may govern the observed seasonality of natural control by epn. These factors include competition with free living bactivorous nematodes in the weevil cadaver, antagonism by nematophagous fungi, and parasitism by an epn-phoretic *Paenibacillus* species that reproduces in *D. abbreviatus* and impairs epn motility in soil.

ENTOMOPATHOGENIC NEMATODES IN THE EUROPEAN BIOCONTROL MARKET. **Ehlers, R-U**. Institute for Phytopathology, Department for Biotechnology and Biological Control, Christian-Albrechts-University Kiel, Klausdorfer Str. 28-36, 24223 Raisdorf, Germany,

In Europe total revenues in the biocontrol market have surpassed 200 million . The sector with the highest turn-over is the market for beneficial invertebrates with a 55% share, followed by microbials with approximately 25%. Annual growth rates of up to 20% have been estimated. Entomopathogenic nematodes (EPN) are exceptionally safe biocontrol agents. Until today, EPN are exempted from registration in most European countries, the reason why SMEs were able to offer economically reasonable nematode-based products. The development of technology for mass production in liquid media significantly reduced the product costs and accelerated the introduction of nematode products. Progress in storage and formulation technology has resulted in high quality products which are more resistant to environmental extremes occurring during transport to the user. Today three companies produce EPN in liquid culture. Major markets in the EU are the control of sciarid flies in ornamentals and mushrooms followed by the application against weevils in ornamentals and strawberries. Grubs and other scarabaeid species in orchards and turf are also controlled with EPN. Factors influencing success or failure in these applications will be discussed. Since the introduction of the Western Corn Rootworm *Diabrotica virgifera virgifera* into Serbia in 1992, this pests as spread all over the Balkan Region and has reached Switzerland, Italy, France and Austria in 2002. As larvae are highly susceptible to *H. bacteriophora*, the estabishment of this nematode in agriculture environments is a potential measure to provide sustainable control. Novel adjuvants used to improve formulation of EPN have enabled the foliar application against Western Flower Thrips and lepidoptera. The use of EPN in these markets requires another reduction in product costs.

SYMBIOTIC INTERACTION OF *HETERORHABDITIS BACTERIOPHORA* AND ITS SYMBOTIC BACTERIUM *PHOTORHABDUS LUMINESCENS* DURING NEMATODE DEVELOPMENT AND REPRODUCTION. **Ehlers, R-U.** Institute for Phytopathology, Department for Biotechnology and Biological Control, Christian-Albrechts-University Kiel, Klausdorfer Str. 28-36, 24223 Raisdorf, Germany.

Mass production of nematodes for biocontrol purposes is done in large scale bioreactors in liquid media. The development of the nematodes under artificial conditions differ from the life cycle in vivo. Exit from the dauer stage (DJ) of *H. bacteriophora* (recovery) is induced by a chemical signal excreted by the symbiotic bacterium *P. luminescens*. This signal is composed of at least 2 compounds, one of less than 20 kDa and are negatively charged and another of 5 kDa. Bacteria also secrete an antagonistic signal which inhibits nematode recovery. Since DJ recovery depends on the presence of the bacterial food signals, the percentage of recovering DJ in liquid culture is influenced by the bacterial density

and the bacterial growth phase. The response to the food signal differs from batch to batch. Culture conditions during inoculum production have an impact. At low population density and enough bacteria supply the hermaphrodites lay many eggs into the medium. DJ developing from eggs laid into the medium better respond to the food signal than DJ developing inside the uterus by endotokia matricida. The pH (between 4–12) and the CO2-concentration are positively correlated with DJ recovery. High temperature can inhibit DJ recovery. During the pre-dauer stage the nematodes harbour their symbiont cells in the intestine. Secondary form cells are not retained by the dauer juveniles of H. bacteriophora.

IMPROVEMENT OF THE DESSICATION AND TEMPERATURE TOLERANCE OF HETERORHABDITIS BACTERIOPHORA. **Ehlers, R-U, and O. Strauch.** Institute for Phytopathology, Department for Biotechnology and Biological Control, Christian-Albrechts-University Kiel, Klausdorfer Str. 28-36, 24223 Raisdorf, Germany.

Foliar application of EPN against lepidopteran pests, leaf miners, thrips and white flies is currently tested under greenhouse conditions to investigate the potential for commercial use. After spraying EPN are exposed to low humidity and high temperature. An enhancement of the desiccation and heat tolerance can increase the performance of commercial EPN products. Nematodes are able to adapt to desiccation stress by the production of several protective substances like glycerol, trehalose and proteins. Prior to the selection process, the optimal adaptation conditions were determined. Nematodes were dehydrated in polyethylenglycol (PEG) with defined water activities (Aw-values). Decreasing water activity causes increasing dehydration. The highest desiccation tolerance was observed in populations adapted at an Aw-value of 0.96 for 72 h. The variability of the desiccation tolerance within a population increased during adaptation. The proportion of the genetic variability on the phenotypic variability (heritability–h^2) for adapted was $h^2 = 0.46$ and for not adapted $h^2 = 0.48$. By selection the mean tolerated Aw-value could be reduced from 0.89 to 0.81 including the adaption to low humidity. No reduction of the mean tolerated water activity could be obtained for non-adapted populations. The heritability for the heat tolerance was $h^2 = 0.68$. After 4 selection steps the mean tolerated heat tolerance was increased from 38.5 up to 39.2 °C. By selection for cold active infective juveniles the mean tolerated temperature was continuously reduced from 7.3 down to 5.2 °C during the first 5 steps. Afterwards, however, the mean tolerated temperatures increased to 6.7 °C. A screening among isolates of *Photorhabdus luminescens* resulted in the identification of strains which were growing at lower temperature.

HERITABILITY OF THE LIQUID CULTURE MASS PRODUCTION POTENTIAL OF THE ENTOMOPATHOGENIC NEMATODE *HETERORHABDITIS BACTERIOPHORA*. **Ehlers, R-U., and O. Strauch.** Institute for Phytopathology, Department for Biotechnology and Biological Control, Christian-Albrechts-University Kiel, Klausdorfer Str. 28-36, 24223 Raisdorf, Germany, E-Mail: ehlers@biotec.uni-kiel.de.

The infective stage of entomopathogenic nematodes (*Heterorhabditis* spp.) is the mobile, but developmentally arrested dauer juvenile (DJ). For commercial application, nematodes are produced in liquid culture. Prior to the inoculation of the DJ, their symbiotic bacterium *Photorhabdus luminescens* is cultured. The DJ exit from the arrested stage (recovery) and develop to reproductive adults. Recovery is a response to bacterial food signals. In liquid culture the percentage of DJs recovering from the DJ stage is highly variable, which significantly influences the number of reproducing hermaphrodites and the final DJ yields. The liquid culture yield is defined by the number of DJ/ml harvested from the medium. The heritability of the disposition to recover from the DJ stage and of the final DJ yield in liquid culture has been evaluated. From a hybrid strain of *H. bacteriophora* 30 homozygous inbred lines were established by inbreeding over 7 generations. These inbred lines were propagated in liquid culture several times and DJ recovery and yields were recorded. The calculated heritability for the DJ recovery was low ($h^2 = 0.38$). No significant genetic variability could be detected for this trait. In contrast, a high heritability ($h^2 = 0.90$) was found for the total number of DJs produced in the liquid medium.

A PHORETIC ASSOCIATION BETWEEN A PUTATIVE *PAENIBACILLUS* SP. AND THE ENTOMOPATHOGENIC NEMATODE *STEINERNEMA DIAPREPESI*. **El-Borai , F. E.,[1] L. W. Duncan,[1] J. F. Preston,[2] and D. Dunn.[1]** [1]University of Florida, Citrus Research and Education Center, Lake Alfred, FL 33850; [2]Microbiology and Cell Science Dept., University of Florida, Gainesville, Fl 32611.

Spores of an unidentified bacterium were discovered adhering in large numbers to the cuticle of third-stage infective juvenile *Steinernema diaprepesi* (IJ3) endemic in a central Florida citrus orchard. Based on FAME profiles and 16S rRNA gene sequencing, the bacterium is closely related to the insect pathogens *Paenibacillus popilliae* and *P. lentimorbus*. Unlike the fastidious *P. popilliae*, the putative *Paenibacillus* sp. is readily cultured on standard media. The bacterium did not attach to cuticles of several entomopathogenic or plant parasitic nematodes tested, suggesting host specificity to *S. diaprepesi*. Apparently the relationship between the nematode and the bacterium is phoretic, because no spores have been observed in the body of *S. diaprepesi*. The effect of the bacterium on the nematode population biology was studied using 20-cm-long vertical sand columns contained in 5-cm dia. PVC tubes. A single *D. abbreviatus* larva was confined below 15 cm depth and the soil surface was inoculated with either spore-free or spore-encumbered IJ3 nematodes (20 IJ3 cm-2 soil surface).

After seven days, spore-free *S. diaprepesi* were 100-fold more numerous below 15 cm, 17-fold more numerous at depths between 5 and 15 cm, and only 35% as numerous at 0–5 cm depth, compared to spore-encumbered nematodes. Mortality of *D. abbreviatus* larvae was 72% greater ($P < 0.01$) for spore-free compared to spore-encumbered *S diaprepesi*. Spore-free nematodes produced more than 5 times as many progeny IJ3 ($P < 0.002$), than did spore-encumbered nematodes. Inoculation of the insect *D. abbreviatus* with 108 spores resulted in death of the insect and reproduction of the bacterium. These data suggest that the bacterium is an ectoparasite of *S. diaprepesi* and an antagonist of the nematode in its application for the biocontrol of *Diaprepes*.

THE MORPHOLOGY OF THE EXCRETORY SYSTEM OF *OTOSTRONGYLUS CIRCUMLITUS*. **Elson-Riggins, J. G.,[1,2] E. G. Platzer,[2] and J. G. Baldwin.[2]** [1]Department of Biological and Physical Sciences, Montana State University Billings, Billings, MT 59101; [2]Department of Nematology, University of California, Riverside, CA 92521.

The morphology of the excretory-secretory (ES) system of *Otostrongylus circumlitus*, a strongylid parasite of seals, was examined using light and transmission electron microscopy. This was compared to the ES system of other nematodes, with particular emphasis on the free-living species *Caenorhabditis elegans* (Rhabditida). *Caenorhabditis elegans* has been proposed as a model to study the vertebrate parasites of the order Strongylida, since there is increasing evidence that Rhabditida and Strongylida share a unique evolutionary history. The aim of this work was to identify adaptations of the ES system present in parasitic *O. circumlitus*, but not in free-living *C. elegans*. These are most likely to be specializations for parasitism, while general features of the system are probably shared between the two species. The ES system of *O. circumlitus* is of the rhabditoid type and has two metabolically active associated gland cells. The most prominent difference between *O. circumlitus* and *C. elegans* is in the ES glands. In the former there are 2 unicellular glands, which are very long, extending 1/7 to 1/10 of the body length from below the excretory duct, a distance of 0.5 to 1 cm. In the present observations, these glands were packed with secretory vesicles, which were most concentrated at the anterior end. One gland terminates anterior to the other and specialized structures within and associated with this gland are all displaced anteriorly relative to the same structures in the other gland. We speculate on the nature of a double membrane bound structure 190 mm in diameter that occurs near the posterior terminus of-each gland.

NEW TOOLS FOR HIGH-THROUGHPUT PHENOTYPIC GREENHOUSE SCREENING. **Faghihi, J.,[1] H. Quesada,[2] R. Gazo,[2] R. A. Vierling,[3] and V. R. Ferris.[1]** [1]Department of Entomology, Purdue University, West Lafayette, IN 47907; [2]Department of Forestry and Natural resources, Purdue University, West Lafayette, IN 47907; [3]Department of Agronomy, Purdue University, West Lafayette, IN 47907.

Large-scale breeding efforts are underway by seed companies for new CystX soybean lines, developed from our PUSCN14 germplasm resistant to soybean cyst nematode (SCN). Efficient and cost effective testing for verification of the integrity of the seeds with respect to their resistance to SCN is critical to the development process. We developed new tools and protocols to meet this challenge for large-scale, accurate greenhouse phenotypic screening. A particular need was an efficient method for loading our seedling trays (8 × 16 cells) with equal soil amounts in each cell. This was accomplished by devising three separate pieces of equipment, each comprised of two parts as follows: The first device was constructed by boring 128 holes, 21 mm in diameter in a 210 × 580 mm piece of plywood, 34 mm thick. A sliding piece of sheet metal is placed underneath the holes and the assembly placed over the seedling tray. After the holes are filled with soil, excess soil is brushed off the plywood, and the metal sheet is pulled out allowing the soil to drop into the cells. This insures an equal seedbed depth in each planting cell. A similar device is used to place the seeds on the seedbed. A third such device, 9 mm thick, is used in a manner similar to the first device to place an equal amount of soil over the seeds. Use of these new devices has greatly improved our accuracy, throughput, and overall screening efficiency.

A TRANSPOSON ISOLATED AND CHARACTERIZED FROM THE PLANT PARASITIC NEMATODE *MELOIDOGYNE ARTIELLIA*. **Fanelli, E.,[1] A.Kumar,[2] J. T. Jones,[2] F. De Luca,[3] M. Di Vito[3] and C. De Giorgi.[1]** [1]Dipartimento di Biochimica e Biologia Molecolare Università degli Studi di Bari, via Orabona 4, 70126 Bari, [2]Plant-Pathogen Interactions Programme, Scottish Crop Research Institute, Invergowrie, Dundee, DD2 5DA, UK, [3]Istituto per la protezione delle piante sezione di Bari, CNR via Amendola 165 Bari.

In order to investigate genes expressed by the plant parasitic nematode *Meloidogyne artiellia*, we have performed a small scale expressed sequence tag (EST) project on a cDNA libary generated from this nematode. To date over 500 sequences have been obtained. Housekeeping genes, nematode specific genes and homologues of candidate pathogenicty factors identified from *M. incognita* have been identified in the dataset. One cDNA sequence identified was similar to a putative transposase derived from an Ac-like DNA transposon. Transposable elements are DNA fragments capable of moving within a genome from one locus to another or, more rarely, by horizontal transfer between two genomes. Such elements have rarely been studied in plant parasitic nematodes but are of interest due to their utility in population genetic studies as well as their capacity to influence the genome architecture and biology of the organisms in which they are located.

Further characterisation of the *M. artiellia* element and analysis of the prevalence of the element within other *Meloidgyne* species is currently underway.

QUANTITATIVE REAL TIME RT-PCR ASSAY OF GRAPE FANLEAF VIRUS FROM SINGLE SPECIMENS OF *XIPHINEMA INDEX*. **Finetti-Sialer, M.,[1] and A. Ciancio.[2]** [1] Dipartimento di Protezione delle Piante e Microbiologia Applicata, Università dagli Studi, Bari; [2] CNR, Istituto per la Protezione delle Piante, Sezione di Bari, Italy.

A real-time reverse transcription (RT) PCR assay was developed to assess presence of Grapevine fanleaf virus (GFLV) in single specimens of its vector *Xiphinema index*. The test used fluorescent DNA-based probes and a 660 bp cloned fragment of the GFLV coat protein (CP) gene. The *X. index* population proceeded from a grapevine orchard at Palagiano, Italy. A diluted series of *in vitro* transcripts from the fragment served as standard to quantify the virus load in nematodes. The test allowed recognition of individual specimens with adsorbed GFLV particles. Positive specimens were distinguished from controls or negative samples by the fluorescence emissions observed during amplification. Fluorescence from positive nematodes rised at cycles 30–32, and corresponded to a minimum amount of template RNA of about 800 fg. Considering that GFLV particles have a single CP copy, the minimum amount of virus detectable was approx. $1.06 \cdot 10^{6}$ particles per nematode. The CP fragments amplified and sequenced from other specimens of the Palagiano population showed a low rate of nucleotide variability, as only 0.2 % of positions displayed a nucleotide substitution. They did not involve, however, the region used for detection. This region appeared highly conserved in GFLV, as other strains amplified and sequenced from dry grapevine leaves proceeding from Malta were similar to the italian strain used. The assay allows reliable quantitative detection of GFLV from single specimens of *X. index*, as well as the monitoring of the nematodes capability to acquire, retain and/or transmit the virus.

LABORATORY TESTS WITH *STEINERNEMA SCARABAEI*, A NEW BIOLOGICAL CONTROL AGENT AGAINST WHITE GRUBS. **Fischer, R.,[1] K. Schlueter,[1] O. Strauch,[2] A. Koppenhoefer,[3] and R.-U. Ehlers.[2]** [1]University of Applied Sciences Kiel, D-24 783 Osterroenfeld; [2]Dept. Biotechnology and Biological Control, Inst. for Phytopathology, Christian-Albrechts-University Kiel; D-24 223 Raisdorf, [3]Department of Entomology, Rutgers University, New Brunswick, NJ 08901, USA

White grubs (Coleoptera: Scarabaeidae) are susceptible to entomopathogenic nematodes of the genera *Steinernema* spp. (Rhabditida: Steinernematidae) and *Heterorhabditis bacteriophora* (Nematoda: Heterorhabditidae). In Germany, primary damages to turfgrass are caused by the herbivorous root-chewing larvae of *Amphimallon solstitialis* and *Phyllopertha horticola* and by *Melolontha melolontha* to many highly valuable crops (e.g. asparagus, strawberry, apple trees). Tremendous secondary damages to turfgrass is caused by grub feeding predators such as crows, foxes and wild boars. Although reliable biological control of populations of *P. horticola* can be achieved with *H. bacteriophora*, beetles with perennial life-cycles such as *A. solstitialis* and *M. melolontha* were less or non susceptible. Recently several North American white grubs species were found highly susceptible to *S. scarabaei*, isolated in central New Jersey. Field collected third-instar larvae of *P. horticola*, *A. solstitialis* and *M. melolontha* were tested individually for their susceptibility against *S. scarabaei*. Five different infective juvenile (IJ) concentrations (5 – 400 IJ) of *S. scarabaei* (J3) were applied. Twenty-five grubs per IJ concentration were kept individually in 50 ml cups with 30cc soil and grass seedlings. The assay cups were incubated at 20°C in growth chambers and mortality of the grubs was determined after 14 days. All tested white grubs species were highly susceptible to *S. scarabaei* (*M. melolontha:* LD50/90 = 39/173IJs/grub; *A. solstitialis:* LD50/90 = 9/64IJs/grub). In contrast, at 400 *H. bacteriophora* IJs/grub the mortality remained below 20%. Also the grubs of *P. horticola* are more susceptible to *S. scarabaei*.

STICKING AND SWARMING IN *XENORHABHUS NEMATOPHILA*. **Forst, S., H. He, and D. Kim**. Department of Biological Sciences, University of Wisconsin, Milwaukee, WI 53201.

Xenorhabdus nematophila, a gram negative bacterium belonging to the *Proteus* clade of the *Enterobacteriaceae* family, forms a mutualistic association with the soil nematode, *Steinernema carpocapsae*. The nematode invades insects and releases *Xenorhabdus* into the hemolymph where it participates in insect killing. To better understand the interaction between the bacterium and nematode, the *mrx* operon of *X. nematophila*, which encodes fimbrial appendages that facilitate adhesion to biotic surfaces, was studied. The *mrx* operon contained 5 structural genes (*mrx*ACDGH) but unlike the *mrp* operon of *Proteus mirabilis*, lacked a site-specific recombinase and a *mrpB*-like gene. MrxA fimbriae were produced at high levels in cells grown on agar while the Mrp fimbriae in *Proteus* are not produced on agar surfaces. Thus, the regulation and genetic organization of the *mrx* operon was found to be distinctive in several respects. Competition experiments showed that a strain lacking the MrxA fimbriae could colonize but was not efficiently released from the nematode. *Xenorhabdus* also displays swarming behavior on agar surfaces. Since the regulatory protein, OmpR, controls flagella production and swarm cell differentiation in several enteric bacteria, an *ompR*-minus strain of *Xenorhabdus nematophila* was created. Swarming behavior in the *ompR strain* began 4 hours sooner than in wild type cells. Precocious swarming in

the *ompR* strain was correlated with early flagellation and cell elongation indicating that OmpR was involved in the temporal regulation of these processes in *X. nematophila*. The *ompR* strain also showed a competitive defect in the release of the bacteria from the nematode. Taken together, these results suggest that MrxA fimbriae and OmpR play a role in the interaction between the bacteria and the nematode.

EVOLUTION OF FILTER FEEDING AND CHEWING IN NEMATODES. **Fürst von Lieven, A.** AG Evolutionsbiologie, Institut für Biologie/Zoologie, Königin-Luise-Str. 1-3, 14195 Berlin, Germany.

The distinct morphological regions of the tripartite pharynx in Secernentea reflect several functions of this organ. Besides the basic functions of sucking and pumping food against the body pressure, the pharynx of rhabditids serves two additional functions restricted to two pharyngeal subunits: The corpus traps bacteria behind the stoma and at its posterior end. I recently discovered a pharyngeal pocket valve that helps to trap particles behind the corpus in *Poikilolaimus oxycercus* and *Acrobeles ciliatus*. Trapped bacteria are chewed by the grinder of the terminal bulb. The separated sites of trapping and chewing are connected by the isthmus that transports bacteria towards the grinder. This complex feeding structure originated step by step from a two part pharynx comprising a propharynx and terminal bulb as in "Plectidae" (a paraphyletic taxon from within which the Secernentea are derived). Analysis of video sequences of feeding rhabditids and plectids provided new data to reconstruct this transformation. Within the "Plectidae" two types of grinders occur: (1) The triangular chewing plates of the "*parietinus*-type" can bulge medially when chewing bacteria and can be retracted, drawing some particles into the grinder. (2) The more solid chewing plates of the grinder in *Ceratoplectus*, *Plectus parvus*, and *Wilsonema* are homologous with those of Secernentea. Because these plates can not be retracted, an alternative mechanism to transport bacteria towards the grinder must have been the prerequisite for their origin. The differentiated pattern of lumen closure in *Ceratoplectus*, *Plectus parvus*, and *Wilsonema* can be interpreted as the first step in the origin of a functional separation of trapping bacteria and transporting bacteria towards the grinder which led to the morphologically discernible units corpus and isthmus found in the Secernentea.

IMPACT OF BIOCIDES ON SOIL SUPPRESSION AGAINST *HETERODERA SCHACHTII*. **Gao, X., and J. O. Becker.** Department of Nematology, University of California, Riverside, CA 92521.

The effects of several biocides on a beet cyst nematode-suppressive soil were evaluated in greenhouse trials. The soil was obtained from a location at the University of California Riverside Agricultural Operations that previously had been shown to be inhospitable to *H. schachtii*. The potted soil (2000 g) was drenched with 240 ml of water containing PCNB (100 ppm), benomyl (200 ppm), fludioxonil (100 ppm), mefenoxam (10 ppm), streptomycin sulfate plus penicillin (each1000 ppm), formaldehyde (5.5 ml of 37% solution) or fumigated with methyl iodide (5 ml a.i./m3). The soil was seeded with Swiss Chard and 6 weeks later was infested with 5000 second-stage juveniles of *H. schachtii*. After two nematode generations, the number of cysts in soil treated with PCNB, fludioxonil, mefenoxam, or streptomycin sulfate plus penicillin was not significantly different from the untreated control, while there were significantly more cysts in the benomyl, formaldehyde, and methyl iodide treatments. The numbers of eggs per g soil were significantly higher in the methyl iodide, formaldehyde and to a lesser extent, in the benomyl and fludioxonil treatments than in the non-treated control soil. At 1130 degree-days, 66% of cysts were colonized by fungi in the untreated, suppressive soil. Significantly fewer cysts were colonized in the treatments with methyl iodide, formaldehyde, benomyl, and fludioxonil. The percentages of cysts colonized by fungi were positively correlated with egg parasitism. These results support earlier research that suggested fungi as the primary cause of this beet cyst nematode suppression.

A PHYLOGENETIC ANALYSIS OF THE ORDER TYLENCHIDA (NEMATODA) BASED ON 18S RNA RIBOSOMAL DNA SEQUENCES: CHARACTERS, SUPPORT, AND TESTS OF SYSTEMATIC HYPOTHESES. **Garcia-Varela, M.,[1] P. G. Mullin,[2] T. S. Harris,[2] A. L. Szalanski,[3] T. O. Powers,[2] and B. J. Adams.[1]** [1]Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620; [2]Department of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722; [3]Department of Entomology, University of Arkansas, Fayetteville, AR 72701.

The majority of Tylenchid nematodes are plant parasites, but fungivorous, algal-feeding, and insect parasitic species have also evolved within this group, making it of particular interest to researchers wishing to tease out the origin and maintenance of these trophic habits. Family level phylogenetic analyses of the Tylenchida have not been published to date, yet numerous classification schemes intentionally based on phylogeny have been published that are amenable to testing. The Order Tylenchida encompasses more than 2800 species. Reconstructing the phylogeny of such a large group is a daunting task that requires thoughtful consideration for the number of taxa and characters that can be reasonably sampled and analyzed. Given that not all taxa can be sampled, nor can an unlimited number of characters be analyzed, we sampled a few characters (approximately 650 bases of 18S rDNA) for a large number of taxa (58), and then strategically selected taxa from which to draw complete 18S sequences. We show that contrary to systematic rules of thumb, the addition of more taxa does not always improve phylogenetic resolution by breaking up poorly supported branches, and the addition of more

of the same type of data does not necessarily increase support for nodes where they were previously tenuous. However, the relationships among many Tylenchid families were sufficiently robust to be tested for monophyly and their rational classification into hierarchical categories.

ULTRASTRUCTURE AND DEVELOPMENT OF TWO *PASTEURIA* SPECIES ON *HOPLOLAIMUS GALEATUS*. **Giblin-Davis, R. M.,[1] D. S. Williams,[2] J. A. Brito,[3] D. W. Dickson,[4] and J. F. Preston.[2]** [1]University of Florida-IFAS, Fort Lauderdale Research and Education Center, 3205 College Ave., Davie, Florida, 33314-7799; [2]Microbiology and Cell Science, University of Florida-IFAS, PO Box 110700, Gainesville, FL 32611-0700; [3]Division of Plant Industry, P. O. Box 147100, Gainesville, FL 32614-7100; [4]Department of Entomology and Nematology, University of Florida-IFAS, PO Box 110620, Gainesville, FL 32611-0620.

Two putative *Pasteuria* species, strain L-1 and LS-1, which are obligately endoparasitic Gram-positive prokaryotes that parasitize the phytoparasitic lance nematode, *Hoplolaimus galeatus,* were studied. Attachment and development of the two morphometrically divergent strains were observed in field populations of *H. galeatus* in bermudagrass (*Cynodon* sp.) turf plots in Fort Lauderdale, Florida. These bacteria were distinguished from other described species of *Pasteuria* using ultrastructure of the mature endospores. LS-1 possesses unique laterally digitate projections of the dorsal outer spore coat. L-1 is closest in ultrastructure to '*Candidatus* Pasteuria usgae', but can be distinguished from that species based upon; 1) the more rounded shape of the central body of the spore, 2) fibrous micro-projections of the outer spore coat which project uniformly from the sides and top of the central body versus laterally in '*Candidatus* Pasteuria usgae', and 3) the epicortical wall remnant which appears as a mostly lateral band in L-1 versus a sublateral band in '*Candidatus* Pasteuria usgae'. Development and sporogenesis for both strains were elucidated with TEM and are similar to other nematode-specific ***Pasteuria*.**

MOLECULAR MECHANISMS OF *XENORHABDUS*-NEMATODE INTERACTIONS. **Goodrich-Blair, H., E. C. Martens, K. Heungens, C. E. Cowles, and E. I. Vivas.** Department of Bacteriology, University of Wisconsin-Madison, Madison, WI 53706.

The bacterium *Xenorhabdus nematophila* colonizes the intestine of a non-feeding stage of *Steinernema carpocapsae* nematodes. The nematode is the vector that carries *X. nematophila* into insect hosts, which are killed to obtain nutrients for development and reproduction. In adapting to this specialized life style, *X. nematophila* has evolved functions necessary to be both a symbiont, providing beneficial functions for one animal (the nematode) and a pathogen, causing death of another (the insect). This combination makes it an excellent model to understand both types of relationships. To study mutualism we have identified ten genes affecting *X. nematophila* colonization of *Steinernema carpocapsae* nematodes. Six encode proteins with predicted functions in regulation or metabolism. Another encodes a putative regulatory RNA, NilD RNA (nematode intestine localization), which is required for colonization and which functions, in part, to regulate the translational efficiency a colonization transcription factor. Current experiments are aimed at understanding the role of NilD RNA in colonization and the stimuli affecting its expression. We have identified an additional three genes required for colonization, *nilA, nilB,* and *nilC,* that encode a ~10-kDa protein of unknown function, a b-barrel outer membrane protein, and an outer membrane lipoprotein. Membrane localization suggests that NilA, NilB and NilC function to link an aspect of the external environment to the inner cell. Such a function could be nutrient acquisition, adhesion, signal sensing, or some combination of these. Experiments are underway to examine sub-cellular localization of each protein and their possible association with each other or with nematode factors. Furthermore, we are assessing whether NilA, NilB and/or NilC affect the expression of other genes, and how they themselves are regulated.

MORPHOLOGICAL AND MOLECULAR VARIABILITY AMONG *BELONOLAIMUS* (NEMATA: BELONOLAIMIDAE) POPULATIONS IN FLORIDA. **Gozel,[1] U., K. B. Nguyen,[2] L. W. Duncan,[1] J. Hamil,[2] and B. J. Adams.[2]** [1]University of Florida, Citrus Research and Education Center, 700 Experiment Station Road, Lake Alfred, FL 33850; [2] Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620.

Nominal species of *Belonlaimus* (Nemata: Belonolaimidae) have been shown to exhibit substantial intraspecific morphological, molecular, and bionomic variation. Such variation may be due to differences distributed among different populations or species, suggesting that some nominal species of *Belonolaimu*s may actually represent two or more cryptic species. In order to investigate how these sources of variation are partitioned within an evolutionary context, nematodes were extracted from soil samples collected from five different crops located in different sites in Florida. The five crops included citrus (5 populations, CREC, IF, ALB, PRAT, LA), corn (1 population TR), bermudagrass (1 population PL), and sugarcane (1 population IT). Phylogenetic relationships were reconstructed using ribosomal DNA sequences (ITS and D2/D3 regions of the nuclear ribosomal gene array). DNA sequence length of D2/D3 extension of the 28S region for all of the sampled nematodes was 743 bp and the ITS region was approximately 1087 bp (including partial 18S and complete ITS1, complete 5.8S and complete ITS2 regions). Morphological and morphometric characters were also mapped onto the phylogeny. Two of the populations have numerous molecular and morphological autapomorphies, consistent with the

notion that they represent populations of a species that is distinct from the other sampled taxa. The study suggests that ribosomal DNA sequences complement morphological and morphometric data, and are useful for examining and characterizing phylogenetic relationships and species boundaries of *Belonolaimus*.

MORPHOLOGICAL AND MOLECULAR ANALYSIS OF FOUR *XIPHINEMA* SPECIES BELONGING TO THE *XIPHINEMA AMERICANUM* (NEMATA:LONGIDORIDAE) GROUP IN FLORIDA. **Gozel, U.,[1] K. B. Nguyen,[2] F. Lamberti,[3] L. W. Duncan,[1] and B. J. Adams.** [2] [1]University of Florida, Citrus Research and Education Center, 700 Experiment Station Rd., Lake Alfred, 33850, USA; [2]Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620, USA; [3]Istituto Di Nematologia Agraria, Via G Amendola 165/A, Bari, 70126, Italy.

Members of Longidoridae in the genus *Xiphinema* are important plant parasites as well as vectors for plant nepoviruses. *Xiphinema* species diversity and taxonomic validity has been somewhat controversial, with the number of recognized species in the *X. americanum* group ranging from 34 to 51 depending on the taxonomic authority and methodological operations used to delimit species. In the *X. americanum* group, morphometric variation is abundant, yet identification is made difficult by overlapping measurements and suites of combinations of characters as opposed to unambiguous autapomorphies. In order to investigate morphological variation as a component of species diversity in *Xiphinema*, we examined molecular (ITS and D2/D3 ribosomal DNA sequences) and morphometric characters from closely related species of the *X. americanum* group in Florida: *X. citricolum, X. floridae, X. georgianum* and *X. tarjanense*. Six different D2/D3 nucleotide substitutions were identified among the 4 taxa, but only one of them was autapomorphic (unique, derived). The ITS DNA sequences were much more variable, yielding numerous apparent autapomorphies that were consistent with several morphological observations. Although further investigation of intraspecific variation is needed prior to making robust taxonomic statements at the species level, molecular and morphological characters presently support the taxonomic validity of *X. citricolum, X. floridae, X. georgianum* and *X. tarnjanense*.

OPPORTUNTIES FOR EXPANDING THE USE OF ENTOMOPATHOGENIC NEMATODES FOR TURFGRASS PEST MANAGEMENT. **Grewal, P. S.** Department of Entomology, Ohio State University, Ohio Agricultural Research and Development Center, Wooster, OH 44691.

Turfgrass is a perennial plant community highly valued for aesthetic, environmental, and recreational uses including lawns, golf courses, parks, athletic fields, and cemeteries. Unfortunately, turf is attacked by a variety of insect pests and chemical pesticides are undesirable due to the high human exposure. However, many pests of turfgrass are highly susceptible to entomopathogenic nematodes (Rhabditida: Steinernematidae and Heterorhabditidae), the efficacy of which has been well demonstrated in the field. Entomopathogenic nematodes are now routinely used on golf course fairways in Japan for the control of hunting billbug and on home lawns in the USA and Canada for the control of white grubs, flea larvae, and molecrickets. Turf is an ideal system for the use of entomopathogenic nematodes due to its perennial nature, dense ground cover, and availability of irrigation, yet nematodes are underutilized in North America. The lack of availability of large quantities of good quality products, their high cost, and inconsistent control, have restricted the use of nematodes on golf course fairways, lawns, athletic fields, sod farms, and public parks, and the availability of inexpensive broad-spectrum chemical pesticides and extremely low damage thresholds has excluded their use on golf course greens. The ban on the use of many broad-spectrum chemical insecticides due to the implementation of the Food Quality Protection Act (FQPA) coupled with recent discoveries of new and more effective nematode species and strains provides an opportunity to expand the use of nematodes in turfgrass. Recent research strongly suggests that predictable control of turf pests can be achieved with new strains of entomopathogenic nematodes that provide equal or better control than the curative chemical insecticides.

INOCULUM PRODUCTION OF NEMATOPHAGOUS FUNGI IN SPAWN BAGS. **Han, P., and J. O. Becker.** Department of Nematology, University of California, Riverside, CA 92521.

Production of consistent quality inoculum is a prerequisite for research investigations on potential biocontrol agents as well as for the development of a reliable and effective commercial product. Various nematode-destroying fungi were cultured on cereal or other suitable substrates in autoclavable Unicorn mushroom spawn bags. The polypropylene bags featured microfilters that allowed gas exchange without compromising the aseptic growing conditions of the biocontrol fungus. The production capacity was varied by the size of the spawn bag that may hold from a few 100 grams to several pounds. Strains of *Verticillium chlamydosporium* or *Fusarium oxysporum* that were parasitic on females and eggs of root-knot nematodes were cultured on sterile millet for two to four weeks at room temperature during which the substrate was rapidly colonized by either fungus. Chlamydospore and/or conida production was as prolific or better than in standard Erlenmeyer flasks with cotton plugs. The flexibility of the bags allows for easy mixing and removal of the inoculum. The final product was aseptically sealed and stored in the spawn bags without further transfer and minimum space requirements. Gas exchange during storage via the microfilter helped to maintain the quality of the product.

A STUDY ON PATHOGENICITY OF BACTERIA CARRIED BY PINE WOOD NEMATODES. **Han, Z., H. Yingdi, and Z. Boguang.** Department of Forest Entomology and Pathology, Nanjing Forestry University, Nanjing 210037, P. R. China.

Three bacterium strains with a high frequency, viz. Njh, Njt and Njw, have been isolated from the xylem of a wilted black pine (*Pinus thuntergii*) tree caused by pine wood nematodes (*Bursaphelenchus xylophilus*) and from the surface of the nematodes. These bacteria were *Pseudomonas fluorescens* biotype I, *Pseudomonas fluorescens* biotype II and a species of the genus *Pantoea* respectively. Both callus and bacterium-free seedlings of black pine were inoculated with only bacteria, a mixture of bacteria and the aseptic nematodes of *Bursaphelenchus xylophilus*, a mixture of bacteria and the aseptic nematodes of *B. mucronatus*, *etc*. to determine their pathogenicity. It is revealed that the inoculation only with either aseptic *B. xylophilus* or *B. mucronatus* wouldn't lead to browning of the callus and wilt of aseptic black pine seedlings. But those inoculated with a mixture of the aseptic nematodes and one of the three bacteria mentioned above shown some wilting or browning symptoms. The combination of nematodes with either Njh or Njt caused severe symptom but the combination of nematodes and Njw weak in symptom. Inoculation with a mixture of *B. mucronatus* which is weak in pathogenicity and the bacteria also caused serious wilt of pine seedlings and browning of the callus. It indicated that seedling symptoms under artificial inoculation in lab conditions were in close correlation with the bacteria carried by pine wood nematodes and poorly correlated with species of nematodes. The bacteria carried by pine wood nematodes played an important role in pathogenicity. In addition, it has been proved by the treatment of callus of black pine with bacterium culture fluid that there were wilt-related toxins existing in bacterium culture fluid. Therefore, it's suggested that the diseases were caused by co-infection of both pine wood nematodes and bacteria.

FIRST RECORD OF RICE-ROOT NEMATODE (*MELOIDOGYNE GRAMINICOLA*) IN FLORIDA. **Handoo, Z. A.,[1] W. Klassen,[2] A. Abdul-Baki,[3] H. H. Bryan,[2] and Q. Wang.[2]** [1]USDA, ARS, Nematology Laboratory, Beltsville, MD 20705; [2]Tropical Research and Education Center, University of Florida, Homestead, FL 33031; [3]USDA, ARS, Beltsville, MD 20705.

A root-knot nematode was discovered on roots and soil of a prominent weed, sandbur (*Cenchrus* spp.) collected from a farm adjacent to Tamiami Airport, south Miami, Florida and was identified as *Meloidogyne graminicola* Golden and Birchfield, based on morphological observations. This is the first report of *Meloidogyne graminicola* from Florida and a new host record for this species. It is a major pest of rice in several countries. The infested farm, dominated by *Cenchrus* spp., was sampled as a pre-planting measure for plant-parasitic nematodes, and *Meloidogyne* juveniles were recovered. During past 14 years this was constantly planted to tomato and sorghum sudangrass (*Sorghum bicolor* x *S. bicolor* var. sudanense (Piper) Stapf) in rotation. The field has heavy infestations of sandbur (*Cenchrus* spp.) and nut-grass, both purple and yellow nutsedge, *Cyprus rotundus* L., and *Cyprus esculentus* L., respectively. The roots from the field did not exhibit any symptoms of galls typical of root-knot nematodes. However, heavily infected roots were dark brown to black-colored, and from each infected root area we recovered clusters of 5-10 root-knot nematode females with egg masses attached. This species' close relationship with two other closely related species, *M. hapla,* and *M. naasi,* are discussed. Additional information regarding distribution of this nematode within the region is needed, especially in rice fields throughout Florida.

AN UNDESCRIBED NEW SPECIES OF NEEDLE NEMATODE (*LONGIDORUS* SP.) FOUND ON LOBLOLLY PINE SEEDLINGS AT A SOUTH GEORGIA NURSERY. **Handoo, Z. A.,[1] S. W. Fraedrich,[2] and M. M. Cram[2].** [1]USDA, ARS, Nematology Laboratory, Beltsville, MD 20705; [2]USDA Forest Service, Athens, GA 30602.

A new *Longidorus* species was found associated with severely stunted and chlorotic loblolly pine (*Pinus taeda* L.) seedlings at a south Georgia nursery. It is characterized in having females with a body length of 7-9 mm, lip region slightly swollen, anteriorly flattened, giving the anterior end a rather truncate appearance, long odontostyle that measures 142-160 μm, odontophore length 43–45 μm, total stylet 185–205 μm long, vulva located at 46–52% of body length, and tail bluntly rounded to almost hemispherical. SEM observations revealed additional details of the head region. Males are rare, but present. The new species is closely related to *L. macrosoma*, *L. saginus*, *L. tarjani* and *L. longicaudatus*, but differs from these species either by the body and stylet length or by the shape of the tail; additional morphological characters further distinguish it from these individual species. Host range studies showed that this nematode prefers pines including loblolly, slash (*Pinus elliottii* Engelm. var. *elliottii*) and longleaf pine (*P. palustris* Mill.), and did not reproduce on nutsedge or grasses used as cover crops by the nursery. Additional information regarding the distribution of this species within the region is needed.

EFFECTS OF HIGH SALINITY IRRIGATION ON *BELONOLAIMUS LONGICAUDATUS* AND *HOPLOLAIMUS GALEATUS*. **Hixson, A. H., W. T. Crow., and R. McSorley.** Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611.

In many coastal areas in the Southeastern United States, water restrictions are limiting the amount of fresh water available for irrigation of golf courses, athletic fields, and lawns. Seashore paspalum (*Paspalum vaginatum*) has great

potential for use in these areas. Because of its tolerance to high salinity irrigation, use of this grass on golf courses, athletic fields, and lawns in coastal areas may aid in conservation of freshwater resources. Plant-parasitic nematodes are damaging pests of turfgrasses in Florida, with *Belonolaimus longicaudatus* and *Hoplolaimus galeatus* being considered among the most damaging. It is unknown how these plant parasitic nematodes may be impacted by high-salinity irrigation used on seashore paspalum. Experiments were performed to examine the effects of increasing irrigation salinity levels on *B. longicaudatus* and *H. galeatus* using seashore paspalum as a host. These experiments were conducted in small pots in an environmentally controlled glasshouse. Irrigation treatments consisting of six rates of salinity were formulated by concentrating deionized water to five salinity levels, (5, 10, 25, 40, and 55 dS/M) and deionized water to serve as a control. Population densities of *H. galeatus* demonstrated a negative linear regression ($P < 0.001$) with increasing salinity. Population densities of *B. longicaudatus* were quadratically ($P < 0.001$) related to increasing salinity from 0 dS/M to 55 dS/M. An increase in population densities of *B. longicaudatus* was observed at 10 dS/M compared to 0 dS/M ($P\pard * = 0.0232$). Reproduction and feeding of *B. longicaudatus* and *H. galeatus* decreased at salinity levels of 25 dS/M and above.

PHYLOGENY OF THE PHYLUM NEMATODA BASED ON SMALL SUBUNIT RIBOSOMAL DNA SEQUENCES. **Holterman, M. H. M., S. J. J. van den Elsen, H. H. B. van Megen, A. W. G. van der Wurff, and J. Helder.** Laboratory of Nematology, Wageningen University, Binnenhaven 5, 6709 PD Wageningen, Netherlands.

On the basis of full-length SSU rDNA sequences (ë 1700 bp), the phylogeny of the phylum Nematoda was reconstructed. The majority of nematode sequences were collected at the Laboratory of Nematology (Wageningen University). Sequences obtained from GenBank were added to these. In order to obtain a SSU rDNA sequence, we isolated DNA from a single individual nematode, which had been identified and photographed beforehand. The SSU rDNA was PCR amplified, cloned into vectors and sequenced. A number of nematode species were sequenced multiple times to assess intraspecific variation and sequence errors. For these species a consensus sequence was constructed. This resulted in an alignment, which included the SSU sequences of 355 nematode species. Ten members of the phyla Arthropoda, Priapulida, Tardigrada, Nematomorpha and Kinorhyncha represented the outgroup. The alignment was constructed with the ClustalW algorithm and checked manually. Two regions of the SSU (99 and 76 bp), which were difficult to align, were omitted from the analysis. Trees were constructed using Neighbour-Joining, Maximum Parsimony, Maximum Likelihood and Bayesian inference of phylogeny. All methods showed consensus in that they revealed five major clades. Our results suggest that the classic division of the nematodes into the Adenophorea and Secernentea is invalid: the Adenophorea are paraphyletic with respect to the Secernentea. This is in correspondence with earlier molecular phylogenetic analyses.

COMPARATIVE MITOCHONDRIAL GENOMICS: A MOLECULAR FRAMEWORK FOR THE FAMILY MERMITHIDAE. **Hyman, B. C.,[1,2,3] E. Platzer,[1] S. Tang,[2] A. Tran,[3] Z. Wu,[3] and R. P. Pacheco.[4]** [1]Departments of Biology and Nematology, Graduate Programs in [2]Genetics, and [3]Cell, Molecular and Developmental Biology, University of California, Riverside, CA 92521, [4]CIIDIR Unidad Oaxaca del Instituto Nacional, Oaxaca, Mexico.

To better understand the evolution of the unusual mitochondrial DNA (mtDNA) within the mermithid nematode *Romanomermis culicivorax*, a molecular phylogeny is being developed for the family Mermithidae. Our goal is to map unusual architectural features of the *R. culicivorax* mtDNA molecule within a phylogenetic framework of the mermithid nematodes using a comparative mitochondrial genomics approach. Nucleotide sequence analysis of the mtDNA encoded cytochrome oxidase subunit 1 (COI) and the nuclear rDNA D3 regions has enabled resolution of intergeneric and interspecific affinities among the mermithids. Among seven genera analyzed, grouping into clades appears to follow host-preference. Species-level associations within *Romanomermis* and *Strelkovomermis* include representatives of a lengthy longitudinal transect ranging from Northern Canada to Argentina. Sequence data from these isolates, and from *R. iyengari* (India), are being interpreted in a biogeographic context. Mitochondrial gene order analysis has revealed profound differences between Secernentean and Adenophorean mtDNAs with respect to transcriptional organization, which is near constant in the Secernentea, and with the presence of extensive mtDNA sequence amplification, which appears relegated to the Adenophorea. Mermithid mitochondrial gene orders differ from each other and from all other nematode mtDNAs. Computer-assisted modeling enables a deduction of the most parsimonious steps leading to variation in mitochondrial gene order and the molecular evolution of the complex *R. culicivorax* mitochondrial genome.

EFFECT OF COVER CROPS, METAM SODIUM AND OXAMYL ON COLUMBIA ROOT-KNOT NEMATODE. **Ingham, R. E., [1] N. L. David,[1] and D. A. Horneck,[2] G. H. Clough,[2] and P. B. Hamm.[2]** [1]Department of Botany and Plant Pathology, 2082 Cordley Hall, Oregon State University, Corvallis, OR 93771, [2]Hermiston Agricultural Research and Extension Center, Oregon State University, P.O. Box 105, Hermiston, OR 97838.

Fall cover crops of clover, mustard cv Martigena, and oil seed radish cv Colonel were compared to 38 gpa of metam sodium (MS) for control of Columbia root-knot nematode (*Meloidogyne chitwoodi*). Cover crops were planted 17 August 2001 and incorporated on 25 October. MS was applied on 8 November. Potato cv Russet Norkotah was planted on 19 April 2002 and harvested on 5 September. Half of each plot received oxamyl (as Vydate C-LV®) at 1 lb. a.i./a. applied in furrow

at planting, banded at hilling (29 May) and by chemigation in ½ in. water at 950 soil degree-days base 5 C after planting (5 July). Nematode populations across the plot area averaged 128/250 g soil when cover crops were planted, 9/250 g soil at incorporation and 6/250 g soil when potato was planted. There was no difference between treatments as populations declined dramatically in all plots, even wet fallow. At potato harvest, populations were lower than in control plots in MS only, MS + oxamyl, mustard + oxamyl, and radish + oxamyl treatments. Tuber damage from nematode infection was less than controls with MS alone, but not with cover crops alone. However, damage in mustard + oxamyl plots was less than in the control or the oxamyl only treatment, and equal to that in the MS + oxamyl treatment. Damage in MS + oxamyl or mustard + oxamyl treatments was reduced close to acceptable quality standards, and one more application of oxamyl may have been sufficient.

THE HEAT-STABLE ROOT-KNOT NEMATODE RESISTANCE GENE *MI-9* IS LOCATED IN THE SAME GENETIC INTERVAL AS *MI-1*. **Jablonska, B., D. Noyes, P. A. Roberts, and I. Kaloshian**. Department of Nematology, University of California, Riverside, CA 92521.

A heat-stable root-knot nematode resistance gene, *Mi-9*, was identified in *Lycopersicon peruvianum*, LA 2157. LA 2157 is the only self-compatible accession of *L. peruvianum*. Previous work has localized *Mi-9* to the short arm of chromosome 6 of tomato where *Mi-1* is also located. We used an intraspecific cross to generate an F2 segregating population. Eight hundred and seventeen individuals of this population were used to fine map *Mi-9*. Five PCR-based markers and two RFLP markers, spanning the *Mi-1* region, were used to screen the population. In addition, we designed primers that enable us to distinguish between *Mi-1* and its homologues. These primers were used to identify *Mi-1* homologues cosegregating with the heat-stable resistance. We have identified four *Mi-1* homologues on the short arm of chromosome 6 in LA 2157 and two of these homologues are required for the heat-stable resistance. Recombinants indicated that these homologues are in very close proximity to the location of *Mi-1*. In addition, using these primers we have determined the absence of *Mi-1* in LA2157. We are also investigating the mechanism of resistance mediated by *Mi-9* in LA 2157.

FIELD INTERACTION OF SOYBEAN CYST NEMATODE, *HETERODERA GLYCINES*, AND *FUSARIUM SOLANI* F. SP. *GLYCINES*, CAUSAL AGENT OF SUDDEN DEATH SYNDROME OF SOYBEAN. **Jackson, T. A.,[1] T. L. Niblack,[1] and G. S. Smith[2].** [1]Dept. of Crop Sciences, Univ. of Illinois, Urbana, IL 61801; [2]Missouri Dept. of Agriculture, Jefferson City, MO 65102.

Soybean cyst nematode (SCN), *H. glycines*, is the greatest disease threat to soybean production nationwide and is common in Illinois fields. Sudden death syndrome (SDS), caused by *Fusarium solani* f. sp. *glycines* (Fsg), causes economic losses throughout the Midwest and is frequently observed in fields infested with SCN. Previous research showed increased SDS severity in the greenhouse when plants were also infected with SCN, but the relationship between SCN and Fsg is poorly understood and there is little data on their interaction in the field. During 2000 – 2002, four soybean near-isogenic lines varying in resistance to each pathogen were planted in a total of six naturally-infested locations, two in Missouri and four in Illinois, to determine the effects of SCN population levels on SDS development and Fsg infection. Four treatments were assigned according to the relative SCN population density at planting and replicated six times. Nematode densities, fungal colonization of taproots (cfu/g), and SDS development were each evaluated. In 2000 and 2001, nematode reproduction and fungal colonization was highest on the SDS-resistant SCN-susceptible line, but there was no significant nematode treatment effect on SDS severity or fungal colonization. SDS foliar symptoms were mild or absent at all locations due to late planting. In 2002, SDS disease indexes at growth stage R5 were significantly higher and mean seed weights were significantly lower in the two SDS-susceptible lines than in the resistant lines. 2002 nematode reproduction and Fsg colonization data will also be presented.

THE PHYLOGENETIC POSITION OF *DELADENUS SIRICIDICOLA*. **Jackson, P. A.,[1] R. W. Poon,[2] T. D. Phan,[3] M. Garcia-Varela, [4] K. B. Nguyen,[4] S. Pearlman,[5] U. Gozel,[4]and B. J. Adams.[4]** Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620.

The Neotylenchid *Deladenus siricidicola* is currently used as a biocontrol agent in the pine forests of Australia against the woodwasp *Sirex noctilio*. The wasp causes significant damage to pine trees (*Pinus radiata)* through the use of phytotoxic mucus and a symbiotic fungus, *Amylostereum areolatum*. The nematode is both mycetophagous and entomoparasitic. It relies on the wasp to vector it into the pine tree along with its symbiotic fungus. The woodwasp bores into the pine several times, leaving an egg to develop in each cavity. One of the cavities bears phytotoxic mucus and fungal spores. The spores develop and the plant-parasitic fungus destroys the tree but also provides a nutritional source for the nematode during the early stages of its life cycle. The woodwasp larvae also feed on the fungus. Upon completion of the free-living stage, the nematode parasitizes the woodwasp. Taxonomically, *D. siricidicola* has been placed in the Order Tylenchida, possibly as sister taxon to the Anguinoidea. In order to determine the phylogenetic position of *D. siricidicola*, we sequenced the entire 18S ribosomal RNA gene and compared it to other published nematode sequences. We discuss the phylogenetic

position of *D. siricidicola* in context of its morphological and bionomic affinities, particularly with regard to the origin of insect parasitism.

OVERWINTERING SURVIVAL OF *APHELENCHOIDES FRAGARIAE* AND THE EFFECTIVENESS OF A HOT WATER DRENCH. **Jagdale, G. B., and P. S. Grewal.** Department of Entomology, Ohio State University, OARDC, Wooster, OH 44691.

Foliar nematode, *Aphelenchoides fragariae* is a serious pest of many ornamentals. In the Midwest, symptoms of nematode infection on hosta (*Hosta* sp) leaves are first noticed in July but their overwintering strategy is unknown. We compared the survival of overwintering nematodes in potted hosta plants maintained either in a polyhouse or covered under plastic with those planted in a home garden. Nematodes survived as juveniles and adults in the soil, dry leaves, and dormant buds (crown) but not in the roots. Buds from garden plants and in containers under plastic cover harbored 46–50% juveniles, 29–37% females, and 13–25% males, while plants overwintered in a polyhouse harbored 32% juveniles, 45% females, and 23% males. Plants in the polyhouse had the highest number of buds that harbored the nematodes, whereas the bare home garden had the highest number of plants supporting the nematodes in the soil. Effectiveness of a hot water (100oC) soil drench was studied against *A. fragariae* on hosta and fern (*Matteuccia pensylvanica*) in the greenhouse for two consecutive years to target overwintering nematodes. Hot water drench consistently reduced the number of nematode infected leaves and the size of chlorotic lesions relative to the control 150 days after treatment for both years in hosta but not in the fern. Hot water caused 34–67% reduction of *A. fragariae* in hosta leaves, 50% in fern fronds and 61–98% in the soil over the control. We conclude that the hot water soil drench early in the spring could prove effective in managing foliar nematodes in nurseries and landscapes.

EVALUATION OF WINTER COVER CROPS FOR MANAGEMENT OF *ROTYLENCHULUS RENIFORMIS* IN COTTON. **Jones, J. R., and K. S. McLean**. Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849.

The reniform nematode (*Rotylenchulus reniformis*) has become one of the most dominant pests on cotton (*Gossypium hirsutum*) in Alabama. Winter cover crops that serve as non-hosts could suppress *R. reniformis* populations for the next growing season. In the greenhouse, thirty winter cover crops were evaluated for host suitability to *R. reniformis*. Each cover crop was planted in autoclaved soil and inoculated with 2000 *R. reniformis* juveniles and vermiform adults per 500 cm3 of soil. Greenhouse pots were arranged in a RCBD with 6 replications and allowed to grow for 60 days. Eight cover crops were selected from the greenhouse trials and planted in a field naturally infested with *R. reniformis* immediately following the cotton harvest. Fallow treatments were added as controls and nematodes were sampled monthly. In the greenhouse, oats (*Avena strigosa*), wheat (*Triticum aestivum*), rye (*Secale cereale*), ryegrass (*Lolium multiflorum*), radish (*Raphanus sativus*), lupin (*Lupinus albus*), white mustard (*Sinapis alba*), black mustard (*Brassica nigra*), turnip (*Brassica rapa*), and canola (*Brassica campestris*) produced Rf values less than 1, indicating *R. reniformis* did not increase on those hosts. However, *Rotylenchulus reniformis* did increase on crimson clover *(Trifolium incaratum)* and vetch *(Vicia sativa)* with Rf values of 1.4 and 1.26, respectively. In the field trial, AU Robin crimson clover and subterranean clover (*Trifolium subterraneum*) supported the highest *R. reniformis* populations at 30 days after planting. However, by 90 days after planting, no differences in *R. reniformis* populations were observed between the winter cover crops.

BIOLOGICAL CONTROL IN NEMATOLOGY. **Kaya, H. K.** University of California, Department of Nematology, Davis, CA 95616

Biological control in nematology has two major aspects. One aspect is the use of nematodes as biocontrol agents against various soil invertebrate pests, and the other deals with biocontrol agents against pestiferous plant-parasitic nematodes. (Unlike herbivorous insects of weeds, plant-parasitic nematodes have not been exploited against weeds.) In the former case, major advances have been made, in particular, with entomopathogenic nematodes along with their associated bacteria where a number of species are commercially available for insect suppression in many countries throughout the world. In the latter case, a few biological agents are available commercially in several parts of the world, but in general, research is still ongoing to optimize the use of these agents against plant-parasitic nematodes. One of the goals in biological control is to introduce the biological agent to provide permanent reduction of a pest organism. In this situation, the biological agent operates in a density-dependent manner. With entomopathogenic nematodes, there is an example of classical biological control, but these nematodes are primarily used as augmentative agents (i.e., in a density-independent manner). In biological control of plant-parasitic nematodes, the approach is augmentative as well, though *Pasteuria penetrans* has provided long-term control (density-dependent parasitism) under some circumstances. Once entomopathogenic nematodes or the natural enemies of plant-parasitic nematodes are applied to soil, the lack of persistence has often affected their efficacy. Although entomopathogenic nematodes and natural enemies of plant-parasitic nematodes occur naturally in soil, these biocontrol agents have enemies of their own. For example, the natural enemies of plant-parasitic nematodes,

especially nematophagous fungi, infect entomopathogenic nematodes. However, the interrelationships between nematodes used in biological control of invertebrates and natural enemies of plant-parasitic nematodes have not been studied in detail.

SPECTRAL CLASSIFICATION OF RENIFORM NEMATODE INFESTED COTTON PLANTS USING SELF-ORGANIZED MAPS. **Kelley, A. T.,[1] G. W. Lawrence,[1] R. L. King,[2] J. Vickery, [2] and H. K. Lee.[1]** [1]Department of Entomology and Plant Pathology, Mississippi State University, MS State, MS 39762, [2]Department of Electrical and Computer Engineering, Mississippi State University, MS State, MS 39762.

Accurate and timely detection of the reniform nematode (*Rotylenchulus reniformis*) on infested cotton plants is essential for improving cotton crop yields. Through proper implementation of a *R. reniformis* management program, yield losses can be minimized. Presently, the *R. reniformis* detection technique is dependent upon soil analysis. Soil analysis is time consuming and oftentimes expensive especially for farmers with large fields. This study was conducted to determine if spectral characteristics of *R. reniformis* infested cotton plants can be used to identify *R. reniformis* and estimate population levels of *R. reniformis* infesting the plant. Microplots and production fields were used to collect hyperspectral and nematode data. Hyperspectral (HS) data were collected using three cotton plant targets: single leaf, plant canopy, and canopy plus soil. HS data were analyzed using a Matlab based hyperspectral toolkit (MHTK) featuring Kohonen's self-organizing map (SOM). MHTK classification accuracies were as high as 100% for selected cotton targets. Sixteen wavelengths pertinent to *R. reniformis* population estimation on cotton were identified by the toolkit and analyzed using linear discriminate analysis (LDA). LDA results identified single leaf as the best target over multiple locations and varying environmental conditions for *R. reniformis* population estimation. Plant canopy was shown to be the best target to evaluate in-season *R. reniformis* population levels. Future research should evaluate the 16 wavelengths characteristic of *R. reniformis* infested cotton plants using aerial imagery in order to develop an easily implemented detection software package for farm use.

CAENORHABDITIS ELEGANS: A NEMATODES CONTRIBUTIONS TO SCIENCE. **Kemphues, Kenneth.** Department of Molecular Biology and Genetics, Cornell University, Ithaca New York 14853.

The germ line is a specialized cell type that holds the heritage of the species. Germ cells are often set aside early in development and are protected from differentiation until late stages of development. Theodore Boveri used nematode embryos for a series of key experiments providing some of the first evidence for the existence of germ plasm, a region of egg cytoplasm that contains determinants of the germ line. More recent work in *Caenorhabditis elegans* has revealed molecular details of the germ plasm and how it comes to reside in a particular region in the egg and a single cell in the early embryo. Two key features of the specification of the germ line are the establishment of embryonic polarity that guides the accumulation of germ line determinants in the posterior of the embryo and a system for transcriptional repression that protects the germ line from inappropriate differentiation in the early embryo.

HOW DOES *POCHONIA CHLAMYDOSPORIA* LIMIT POPULATIONS OF ROOT-KNOT NEMATODES? **Kerry, B. R., [1] S. D. Atkins, [1] P. Gray ,[1] L. Hidalgo-Diaz, [2] P. R. Hirsch, [1] T. M. Mauchline, [1] and C. O. Morton. [1]** [1]Nematode Interactions Unit, Rothamsted Research, Harpenden, Herts., AL5 2JQ, UK, [2] Centro Nacional de Sanidad Agropecuaria, Havana, Cuba.

Pochonia chlamydosporia (synonym *Verticillium chlamydosporium*) is a facultative, generalist parasite in soil, which infects fungal spores, invertebrate eggs and colonises the rhizosphere of a range of plants. However, in molecular and population studies, individual isolates of the fungus demonstrate specific interactions with their nematode hosts and during their saprophytic phase in the rhizosphere. Differences in host preference between isolates are difficult to detect in standard bioassays on agar but may be crucial for the selection of isolates for exploitation as biological control agents. Infection of root-knot nematode eggs involves the secretion of a basic serine protease, which degrades the outer vitelline membrane of the eggshell and exposes the chitin layers. Differential production of this enzyme and variation in its amino acid sequence between isolates suggest it may be a virulence and host range determinant. In culture, some isolates produced a nematicidal metabolite, phomalactone. An isolate from Cuba has been selected and a simple bi-phasic fermentation method developed for mass-production of inoculum in that country. Single applications of chlamydospores of the fungus introduced into soil when poor hosts for root-knot nematodes are planted, have significantly reduced the numbers of healthy root-knot nematodes. Soil factors influence the establishment of the fungus but once established it survives between crops. The final stages of toxicology testing of the fungus are in progress and a product for use by local farmers should be available within a year.

ROOT-KNOT NEMATODES CONTAINED IN ROOT GALL OF ORIENTAL MELON. **Kim, Don G.,[1] Sung K. Choi,[1] Jae T. Yoon,[1] and Ye H. Choi.[2]** [1]Department of Agricultural Environment, Gyeongbuk Agriculture Technology Administration, Daegu, Korea; [2]Korea Biochemical, Dongdong-Ri, Euiryong-Gun, Gyeongnam, Korea.

Oriental melon, *Cucumis melo* L. *cv*. Geumssaragi-euncheon, grafted on Shintozoa (*Cucurbit maxima* ×*Cu. moschata*) was planted on February 04 in a greenhouse infested with *Meloidogyne arenaria* and root gall were examined five months

after planting. A gram of root gall was volumed ca. $10cm^3$ and contained in an average of 363 females (170 developing and 193 matured females), 2,120 second-stage juveniles (J2), and 13,074 eggs. In addition, there was 56 J2 per cm^3 soil around the infested plant. An oriental melon had an average of 134.6 g of root gall (70% of total root weight) per 0.72 m^2 area. In a conservative estimation, an oriental melon plant could accommodate ca. $1.2x10^7$ eggs and J2 per 0.72 m^2. The eggs contained in root tissues could be an important inoculum source to the next crop and the fate of these eggs are well worth of further investigation.

ASSESSMENT OF NEMATICIDAL PROPERTIES OF PROPYLENE OXIDE AND ALLYL ISOTHIOCYANANTE. **King, P. S., and R. Rodríguez-Kábana.** Department of Entomology and Plant Pathology, Auburn University, AL 36849.

The nematicidal properties of three aqueous solutions containing 2.0% (v/v) propylene oxide [PO], 0.2% (v/v) allyl isothiocyanate [AI], and [PO + AI], were studied in a greenhouse experiment using soil infested with *Meloidogyne incognita*. Rates (mg ai/kg soil) were 40, 80,120, 160 and 200 for [PO], 4, 8, 12, 16 and 20 for [AI], and 44, 88, 132, 176 and 220 for [PO + AI]. Pots were covered with plastic immediately after application of the materials. After 2 weeks, nematological analysis of soil was conducted which showed *M. incognita* J_2 were eliminated in response to all applications of [AI] and [PO +AI], whereas [PO] had no effect on the nematode. Rutgers tomatoes were planted in each pot. After 8 weeks, nematological analyses of soil and roots were conducted. Data were also collected for shoot height, shoot weight, root weight and root condition. *M. incognita* J_2 in both soil and roots were eliminated in response to [AI] at rates $8 mg a.i./kg soil and to [PO + AI] at rates $88 mg a.i./kg soil, and again [PO] showed no effect on the nematode at any rate. Shoot height, shoot weight and root weight increased directly in response to all [PO] rates, but the effects on these variables were further enhanced in response to all rates of [PO + AI] and the two lowest rates of [AI]. Root condition did not improve in response to [PO], but did improve directly to all rates of [PO + AI]. Root condition also improved in response to [AI] when compared to the control, but the effect leveled off at higher rates. Results indicate that there is a degree of synergism between [PO] and [AI].

MOLECULAR PHYLOGENGY AND MORPHOLOGICAL EVOLUTION OF RHABDITID NEMATODES. **Kiontke, K., S. Chiou, N. G. Gavin, Y. Raynes, and D. H. A. Fitch.** Department of Biology, New York University, New York, NY 10003.

To reconstruct a phylogenetic tree of "Rhabditidae" we have sequenced small subunit ribosomal RNA genes (SSU rDNA) for more than 100 species of "Rhabditidae" and Diplogastrina representing almost every major group within these taxa. Using SSU rDNA sequences, we can show that the parasitic Strongylida and *Heterorhabditis* are part of "Rhabditidae". Our analysis supports several monophyletic groups within "Rhabditidae" (e.g. *Caenorhabditis; Oscheius;* a clade consisting of *Pelodera*, *Mesorhabditis* and *Teratorhabditis;* Diplogastrina). *"Pellioditis"*, *"Rhabditis"* and *"Rhabditoides"* are shown to be para- or polyphyletic. Some relationships at the base of our phylogeny are not well resolved as are some relationships within otherwise well resolved groups (e.g. the species most closely related to *Caenorhabditis*). Therefore we are sequencing the 3′ half of the largest subunit of RNA polymerase II and about 3000 bases of large subunit rDNA. Our phylogeny can be used to trace the evolution of molecular, morphological, developmental, and even behavioral characters. For example, many introns in the RNA polymerase II gene were lost in the lineage leading to *Caenorhabditis*, whereas only one intron was clearly gained. The position of the vulva changed at least 4 times from median to posterior which correlates with the loss of the posterior gonad arm. Within Rhabditidae these 2 characters are never reversed. Hermaphroditism evolved from gonochorism at least 8 times independently. In only one case, our data suggest evolution of gonochorism from hermaphroditism. Heterogony evolved twice within the group. The position of phasmids relative to genital papillae in the male tail has changed several times in a saltational manner. Phylogenetic correlation may also be made for size of the male bursa and mating type, suggesting that natural selection may have a role in diversifying male morphology.

USE OF GRAM-POSITIVE BACTERIA AS BIOLOGICAL CONTROL AGENTS FOR PLANT PARASITIC NEMATODES. **Kokalis-Burelle, N.,[1] and D. A. Samac.[2]** [1]USDA, ARS, U. S. Horticultural Research Laboratory, Fort Pierce, FL 34945, [2]USDA, ARS Plant Science Research Unit, St. Paul, MN 55108.

Gram-positive bacteria are well-suited for development as biological control agents for plant parasitic nematodes because many are easy to culture, produce a wide array of chemical compounds, and form spores that enable drying and storage of formulations. Two types of gram-positive bacteria with potentially different modes of action will be discussed. The first type is plant growth-promoting rhizobacteria (PGPR). Numerous PGPR are gram-positive bacteria, and of these, many are *Bacillus* spp. Mechanisms for disease reduction with PGPR are primarily considered to be indirect. PGPR often induce systemic resistance or increase tolerance to pathogens in the host plant, resulting in increased plant growth and yield. In Florida field trials, two gram-positive PGPR isolates (*Bacillus subtilis* strain GBO3 and *B. amyloliquifaciens* strain IN937a) in a formulation containing chitin, reduced root-knot nematode (*Meloidogyne incognita*) galling and improved

root condition of pepper (*Capsicum anuum*) and muskmelon (*Cucumis melo*) when added to transplant media at seeding. A second type of gram-positive bacteria with biological control activity against nematodes is *Streptomyces*. *Streptomyces* spp. have been isolated from many disease suppressive soils and some exhibit direct antagonism of plant parasitic nematodes through the production of antihelminthic compounds such as macrocyclic lactones. Several isolates of *Streptomyces* previously shown to suppress potato scab disease (*Streptomyces scabies*) in the field were shown to also reduce populations of root lesion nematodes (*Pratylenchus penetrans*) in both susceptible and resistant alfalfa (*Medicago sativa*) varieties. Studies are underway to evaluate the effects of these *Streptomyces* isolates on root-knot nematode populations and to determine if they also induce systemic resistance responses in the host.

PANAGRELLUS REDIVIVUS AS A MOLECULAR MODEL FOR CYST NEMATODES. **Kovaleva, E. S.,[1] K. A. Kromina,[2] N. V. Girsova,[2] E. P. Masler,[1] V. G. Dzhavakhiya,[2] and D. J. Chitwood.[1]** [1]Nematology Laboratory, USDA/ARS, Beltsville, MD 20705, [2]Molecular Biology Laboratory, All-Russian Research Institute of Phytopathology, Golitsino, Moscow Region 143050, Russia.

The use of molecular biology techniques opens up a number of possibilities for discovery of cyst nematode control strategies. Because of the paucity of information on genes from cyst nematodes, it is logical to exploit sequences from well-studied species such as *Caenorhabditis elegans* and a few animal-parasitic nematodes. Nonetheless, in our initial studies with more than 10 genes from *Heterodera glycines* and *Globodera rostochiensis*, this approach was inefficient for primer and probe design. In some cases the usage of sequences from non-nematode species, such as insects or vertebrates, was more successful. The primary reason for this lack of utility of other nematodes is the dramatic difference in GC composition and codon usage between cyst nematodes versus *C. elegans*, the animal parasites, and *Meloidogyne species*. Therefore, we examined another free-living nematode, *Panagrellus redivivus*, as a candidate for a molecular model. This species is conveniently handled in the laboratory but is very poorly studied on the molecular level. We first selected for investigation the actin/heat shock protein-70 superfamily, which contains ubiquitous, highly expressed and strongly conserved proteins. Several genes from this superfamily were identified from *P. redivivus*. Nucleotide and amino acid sequences were analyzed and compared with those for corresponding genes from other nematodes. Comparative analysis of the base composition and codon usage indicated that *P. redivivus* is a much better molecular model for cyst nematodes than *C. elegans* or any animal-parasitic nematode.

HEAT SHOCK PROTEINS 70 IN *HETERODERA GLYCINES*. **Kovaleva, E. S.,[1] E. P. Masler,[1] S. S. Sardanelli, [2] and D. J. Chitwood.[1]** [1]Nematology Laboratory, USDA/ARS, Beltsville, MD 20705 and [2]Plant Nematology Laboratory, University of Maryland, College Park, MD 20742.

Heat shock proteins 70 (Hsp70) are a ubiquitous and highly conserved group of molecular chaperones which bind nascent and denatured proteins, through an ATP-dependent mechanism, preventing aggregation and improper folding. Some members of this family are expressed at increased levels in response to heat shock. The other, constitutive, members are not heat-inducible and may vary during development. The copious functions of Hsp70 are especially important for endoparasitic nematodes, such as *Heterodera glycines*, that are exposed to a variety of environmental challenges during their life cycle, including temperature extremes and dietary restriction. Using Western blots we detected several Hsp70 proteins in homogenates of *H. glycines*, as well as *Caenorhabditis elegans*. The Hsp70 antiserum, known to react with both vertebrate and invertebrate Hsp70s, revealed at least two distinct proteins in *H. glycines*, which were membrane-associated. In contrast, at least three proteins were detected on the *C. elegans* blots, one of which was soluble. To investigate the multi-gene family of *H. glycines* Hsp70 proteins further, two Hsp70 genes were isolated and characterized. The deduced proteins have molecular weights of 71 and 73kDa, and share high homology (80–88 percent amino acid identity) to Hsp70s from a number of nematodes and other invertebrates. The amino acid sequence of the 73kDa Hsp70 from *H. glycines* is highly homologous to the glucose regulated protein type of Hsp70s. Experiments are being conducted to determine the effect of heat shock and diet on members of the *H. glycines* Hsp70 protein family.

A MATRIX METALLOPROTEINASE GENE FROM THE SOYBEAN CYST NEMATODE, *HETERODERA GLYCINES*. **Kovaleva, E. S., A. M. Skantar, and D. J. Chitwood.** Nematology Laboratory, USDA/ARS, Beltsville, MD 20705.

Despite considerable attention given to proteases as good targets for nematode control strategies, genes encoding metalloproteinases (MP) from plant-parasitic nematodes have not yet been characterized. The available information about MP sequences is restricted by several fragments beyond the catalytic domain of the enzyme. Because the majority of hatching enzymes from non-nematode species are MPs, this class is particularly attractive for study in plant-parasitic nematodes. Here we report the isolation and characterization of a full-length mRNA clone from the soybean cyst nematode *Heterodera glycines* encoding a putative matrix metalloproteinase (Hg-MMP). The deduced preproenzyme with molecular mass of 65.9 kilodalton shares the major structural characteristics of interstitial collagenase M10A subfamily. The catalytic domain includes the three histidine residues involved in coordination of the catalytic zinc atom, a catalytic glutamic acid

residue, and a methionine turn common for MPs. The catalytic domain of Hg-MMP exhibits limited homology to corresponding proteases from plants, including host plant *Glycine max*, and to several hatching enzymes from non-nematode species. The nucleotide homology of Hg-MMP to any known genes is so low that the BLAST search of more than 1.6 million sequences against the 1880-bp long Hg-MMP sequence did not return any homologous genes with an E-value below 0.1. The singularity of the newly discovered MP from *H. glycines* and its involvement in hatching will be studied further.

DEVELOPMENTAL AND BIOCHEMICAL ANALYSIS OF NEMATODE CHORISMATE MUTASES FROM PLANT PARASITIC NEMATODES. **Lambert, K. N., S. Bekal, E. A. Doyle, T. V. Akraiko, and J. A. Painter**. Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

Nematode chorismate mutases (CMs) are esophageal gland proteins secreted by plant parasitic nematodes. These CMs are thought to play an important role in the initial phase of root-knot and cyst nematode feeding cell formation by the direct manipulation of the plant's shikimate pathway. Nematode CMs could alter many plant processes such as plant cell development, cell wall structure, or suppress host plant resistance mechanisms. Nematode CMs are developmentally regulated and several CM forms are expressed. We have expressed a variety of nematode CMs with differing enzymatic activity in soybean hairy roots and Arabidopsis thaliana, and have noticed a number of alterations of plant growth and development such as alterations in flowering time, leaf color and altered leaf and root morphologies. Analytical analysis of chorismate-derived compounds is underway to determine which metabolites are altered that cause the plant growth defects.

DIFFERENTIAL HOST STATUS OF ROTATION CROPS TO DAGGER, LESION AND ROOT-KNOT NEMATODES. **LaMondia, J. A.,[1] and J. M. Halbrendt.[2]** [1]The Connecticut Agricultural Experiment Station, Windsor CT 06095 and [2]The Fruit Research and Extension Center, Biglerville PA 17307.

The host status of *Avena strigosa* (Saia oat); *Brassica napus* ('Dwarf Essex' rapeseed); *Pennisetum glaucum* ('101' forage pearl millet); *Rudbeckia hirta* (small black-eyed-Susan); *Sorghum sudanense* ('Trudan 8' sudangrass); and *Tagetes minuta* ('Polynema' marigold) to the dagger nematode, *Xiphinema americanum*, the lesion nematode, *Pratylenchus penetrans*, and the root-knot nematode, *Meloidogyne hapla* were determined in greenhouse, microplot and field plot experiments in Connecticut and Pennsylvania. A rye control was the best host of *P. penetrans* under greenhouse conditions and all other rotation crops were either nonhosts, poor hosts, or maintenance hosts. *Meloidogyne hapla* did not reproduce on Saia oat, sudangrass or pearl millet after 10 weeks under greenhouse conditions. Low populations were maintained on marigold, *R. hirta* and rapeseed (0.1; 1.3 and 2.3% of numbers extracted from a susceptible tomato control, respectively). In greenhouse pots, *X. americanum* numbers decreased after 8 months on *R. hirta* and rapeseed, increased slightly on Saia oat, millet and marigold, and increased nearly three times on sudangrass. Shoot incorporation or removal influenced *P. penetrans* densities in field microplots. *Rudbeckia hirta* and marigold resulted in very low or undetectable densities regardless of shoot incorporation. Incorporation of millet, sudangrass, Saia and rapeseed shoots reduced densities by 96 to 100% compared to shoot removal, indicating that plant breakdown products may be nematicidal. *Meloiodgyne hapla* and *X. americanum* population changes in microplots and field plots were similar to results obtained from greenhouse pots. Our results show that none of these nematode-antagonistic rotation crops control all three genera of plant parasitic nematodes.

SITE SPECIFIC MANAGEMENT OF THE RENIFORM NEMATODE. **Lawrence, G. W.,[1] S. Samson [2], A. T. Kelley[1], H. K. Lee [1], W. A. Giverns [2] and K. S. McLean.** [3] [1] Department of Entomology & Plant Pathology, and [2] Department of GeoResources Institute, Mississippi State University, Mississippi State, MS 39762. [3] Department of Entomology & Plant Pathology, Auburn University, Auburn, AL 36849,.

Variable rate applications of the nematicide metam-sodium were examined for the management of the reniform nematode. The test was established in a field that was naturally infested with the reniform nematode and applied in the spring 23 days prior to planting the crop. The test location was sampled on one-half acre grids to determine nematode numbers and to georeference their locations in the field. Nematode numbers ranged from 0 to 19,900 reniform per 500 cm^3 soil. Nematode numbers were placed into four classes, no nematodes (0), low (250–5,000), medium (5,001–10,000), and high (greater than 10,000). A metam-sodium prescription map was developed based on the four classes. Treatments included metam-sodium applied at the single conventional rates of 28, 47, and 76 l/ha and a variable rate application of 28–76 l/ha. The experiment was a completely random design with four replications. Each treatment consisted of 12 rows 30.5 m in length with a 96.5cm row spacing. Metam-sodium was injected 41cm deep in the row using a RowTill Ripper and the rows were immediately hipped with attached middle buster plows. Cotton lint yields averaged 1938 kg/ha in the variable rate (28–76 l/ha) metham-sodium treatment. This compared to the conventional single rate lint yields of 1784, 1934 and 1551 kg/ha where metham-sodium was applied at 76, 47 and 28 l/ha, respectively. The variable rate application treatment averaged the lowest amount of metam-sodium applied per hectare (8.6 liters) and provided the highest economical returns.

CULTURAL PRACTICES AND THE DISPERSION OF THE RENIFORM NEMATODE IN MISSISSIPPI. **Lee, H. K., [1] G. W. Lawrence, [1] J. L. DuBien, [2] and A. T. Kelley. [1]** [1] Department of Entomology & Plant Pathology, [2] Department of Mathematics & Statistics, Mississippi State University, Mississippi State, MS 39762.

Cultural practices and in field dispersion of the reniform nematode, *Rotylenchulus reniformis* ,were examined in cotton and in soybean fields during the growing season. The fields were naturally infested with the reniform nematode. During each phase of cotton and soybean production, the agricultural producer will use various types of equipment. Soil samples were collected from this equipment at planting, cultivation, and at harvest. At planting, soil samples were collected from various parts of a Row Till Ripper and the tractor. Nematode numbers from the guage wheels, ripper shanks, middle buster plows and tool bars averaged 726, 963, 896, and 659 nematodes / 100 cm3 soil, respectively. A total of 633 reniform / 100 cm3 were recovered from the tractor tires. During cultivation, soil samples were collected from the tractor tires, cultivator wheels and rolling coulters. The average nematode numbers for each site were 600, 180, and 1,210 nematodes / 100 cm3 soil, respectively. At harvest, soil samples were collected from the tires of the harvestor. An average of 26 reniform / 100 cm3 were recovered. Nematode numbers varied from the specific types of equipment and from the different collection sites. The differences in the average number of nematodes recovered was directly related to the amount of soil adhering to the equipment.

LIFE HISTORY RESPONSE OF *ACROBELOIDES* AND *APHELENCHUS* TO COPPER AND BENZO(A)PYRENE. **Li, F., and D. A. Neher.** Department of Earth, Ecological and Environmental Sciences, University of Toledo, Toledo, OH 43606.

Maturity index values represent life history characteristics often inferred by morphology. We tested the hypothesis that *Acrobeloides* and *Aphelenchus* are sensitive to chemical pollutants, opposite of what their maturity index value of 2 suggests. Controlled experiments were conducted to quantify survival, development and reproduction when exposed to various concentrations of copper and benzo(a)pyrene (BaP). *Acrobeloides* and *Aphelenchus* were reared at 19 °C and provided diets of *Escherichia coli* and *Rhizoctonia solani*, respectively. LC50 values for *Aphelenchus* exposed to copper or BaP are greater than *Acrobeloides*. Copper impedes growth of *Acrobeloides* at 10 mg/kg, resulting in no survival at 20 mg/kg. In contrast, *Aphelenchus* is more tolerant, with no visible impact at concentrations of 20 mg/kg. In contrast to copper, a threshold in concentration of BaP occurred, below which there is little to no impact on growth and above which was fatal. Threshold values were approximately 0.5 mg/kg for *Acrobeloides* and 2 mg/kg for *Aphelenchus*. Early results suggest that copper reduces numbers but not size of offspring; BaP has the opposite effect. Apparently, *Acrobeloides* is more sensitive than *Aphelenchus* to copper and BaP, implying that a maturity index value of 2 is inaccurate. Refinement of index values based on empirical evidence can be used to improve sensitivity and interpretation of nematode community indices.

MOLECULAR RESPONSES DURING SOYBEAN CYST NEMATODES/SOYBEAN ROOTS INTERACTION. **Lu, G, X. Hu, R. Ruff, and W. Schuh**. Pioneer Hi-Bred International Inc., A DuPont Company, P. O. Box 1004, Johnston, IA 50131.

The soybean cyst nematode (SCN) (*Heterodera glycines*) is one of the most devastating pathogens of soybean on a worldwide basis with an approximately $2.5 billion in annual crop loss. After penetrating into soybean roots, the SCN second-stage juveniles (J2) inject secretions that modify root cells and transform them into a specialized feeding structure called syncytium. The syncytium provides nourishment essential for nematode growth and reproduction. Our research is focusing on the molecular responses to the secretions in the syncytium in a comparable interaction. A cDNA library was made from syncytium-rich root tissues. A number of SCN-inducible and SCN-repressive genes were identified from the cDNA library. We also studied the nematode interaction using promoter::GUS-transgenic soybean hairy roots. Under this study, we found that SCP1 promoter activity was repressed, whereas UCP3 promoter activity was significantly induced in the syncytia. These results indicate that there are active molecular changes that are associated with syncytium formation during the nematode/root interaction. Some of the changes may be relevant to the syncytium formation, and other responses may be the results of the syncytium establishment. The information can facilitate us to understand the regulation of the syncytium formation and control nematode reproduction in plants.

EFFECT OF AUXIN ON EGG HATCHING IN THE SOYBEAN CYST NEMATODE. **Lucchi, J., D. O'Gurek, P. Merella, and P. M. Tefft.** Biology Department, Saint Joseph's University, Philadelphia, PA 19131.

Egg hatching and development of the soybean cyst nematode (SCN) is influenced by many factors including the physiological status of the host plant. We investigated the effects of a plant growth regulator on nematode egg hatching. SCN eggs were incubated in different concentrations of auxin (indole-3-acetic acid) ranging from 10^{-7} – 10^{-3} M. Control eggs were incubated in zinc chloride (a known hatching stimulant) and water. Percentage of hatched worms was determined for each of the concentrations. Low concentrations of auxin (10^{-5}–10^{-7} M) yielded hatch rates similar to the controls. At high concentrations (10^{-3}, 10^{-4} M) hatching percentage was decreased to about 50% of the controls. Eggs pre-treated for

as little as one hour in auxin prior to incubation in zinc chloride hatched at rates significantly less than controls. In another study to test the efficacy of host-plant root diffusates on egg hatching in nematode eggs, soybeans were treated with auxin (10^{-5} and 10^{-7} M). SCN eggs were placed in root diffusate collected from treated plants and hatching percentages were determined. The root diffusate from auxin treated plants hatched fewer eggs than the controls.

GENETIC ANALYSIS OF ROOT-KNOT NEMATODE, *MELOIDOGYNE HAPLA*. **Lui, Qingli, [1,2] and V. Williamson.** [1] Departments of [1]Nematology and [2]Plant Pathology, University of California, Davis, CA 95616.

Root-knot nematodes (*Meloidogyne spp.)* are obligate endo-parasites, for which the genetics is poorly understood due to their complex reproduction modes. Among the agriculturally important root-knot nematodes, some isolates of M.hapla undergoes meiosis and sexual reproduction making genetic crosses possible. We have inbred five strains of M. hapla, UCR, VC1R, HSL, HRO and HSMB for 16 generations by single eggmass transfer on tomato. Cytology analysis demonstrated that these five strains are sexual forms; reproduce by a facultative meiotic pathogenesis (Triantaphyllou, 1988). These strains show different interactions with potato host Solanum bulbocastanum. DNA molecular fingerprinting revealed that DNA polymorphisms are very common among these strains. We crossed VC1R (male) to HSL (femaile). Currently we are harvesting "F1" progeny. PCR-based markers will be used to detect the crossed progenies. Then we are going to use more AFLP analysis to follow the progenies and segregations of the markers to develop a genetic map for this nematode.

EMBRYONIC DEVELOPMENT AND THERMAL TIME REQUIREMENTS FOR THE LIFE CYCLE OF *BELONOLAIMUS LONGICAUDATUS*. **Luo, D., J. Smith-Becker, and J. O. Becker.** Department of Nematology, University of California, Riverside, CA 92521.

The sting nematode, *Belonolaimus longicaudatus,* was detected in California's Coachella Valley at the beginning of 1990. Despite its currently limited distribution, it must be considered as an established invasive species for the state. The objective of this project was to gather information concerning biological and ecological aspects of the nematode that are essential to devise rational strategies for its management and containment. The effect of temperature on development of *B. longicaudatus* was monitored on excised root culture plates. In addition, growth chamber pot cultures with *B. longicaudatus* and ryegrass were used to verify the in vitro results. The minimum time for one generation (J2 to J2) under optimum conditions, expressed as the rate of development, was linearly related to temperature between 20?C and 28?C. The threshold temperature below which there is no development was estimated at 12.8?C by back projection of the linear regression line to where it intercepted the temperature axis. This result is similar to the previously reported base temperature for sting nematode embryogenesis. The heat sum requirement was determined as the reciprocal of the slope at 365 degree days. Growth chamber trials with sting nematode-infested ryegrass cultures confirmed the thermal time requirements obtained with the in vitro cultures.

ASSOCIATION OF SOIL FACTORS AND NEMATODE COMMUNITY INDICES WITH THE POTATO EARLY DYING DISEASE IN COMMERCIAL POTATO FIELDS IN WISCONSIN. **MacGuidwin**, **A. E.** Department of Plant Pathology, University of Wisconsin-Madison, 53706.

The potato early dying disease (PED) is caused by the interaction of *Verticillium dahliae* and *Pratylenchus penetrans*. Management decisions are based on initial inoculum densities of both organisms. This simple model is adequate for some, but not all fields. We conducted demographic studies in commercial potato fields during 2002 to identify other factors associated with PED. Twenty-three potato fields representing four enterprises were studied. Based on inoculum densities of pathogens and disease thresholds, the fields represented a range of risk potential for PED. Soil samples from five geo-referenced locations in each field, collected in May and August, were assayed for nematodes, *Verticillium dahliae*, soil texture, soil pH, carbon, nitrogen, and phosphorus in May, and soil fauna and plant parasitic nematodes in August. Soil collected in May was used in a bioassay to detect infection of 'Russet Burbank' potato by *V. dahliae* and other soil-borne fungi and nematodes. All fields were infested with plant pathogenic nematodes and all but two fields were infested with *P. penetrans*. A PED risk score assigned to the samples on the basis of initial inoculum recovered was significantly correlated with PED symptomology in the bioassay only when fields were grouped by enterprise, and even then only for fields from two enterprises. Some soil factors were significantly correlated with PED, but the structure index, based on nematode community analysis, was most highly correlated with initial levels of *V. dahliae* in the soil and disease scored in the bioassay. As the representation of taxa with high C-P values increased in the nematode community, population densities of *V. dahliae* decreased. The study is being repeated in 2003.

EVOLUTION OF NEMATODE EMBRYOGENESIS. **Malakhov, V. V.** Department of Invertebrate Zoology, Moscow State University, Moscow, 119992, Russia.

Investigations of nematode ontogeny clearly reveal that there are three principal types of embryonic development: 1) nematodes of the order Enoplida have regulative development with a variable arrangement of blastomeres; 2) nematodes of all orders of the subclassis Enoplia (except Enoplida) have mosaic development with an anterior origin of endoderm

precursor; 3) nematodes from two other subclassis (Chromadoria and Secernentea) have mosaic development with a posterior origin of endoderm precursor. The regulative enoplid development seems to be the most primitive type of nematode embryogenesis, being ancestral to the two other developmental types. Unlike other nematodes, bilateral symmetry appears late in the enoplid embryogenesis. The mouth and anus of the enoplid embryo develop from the anterior and posterior openings of the closing blastopore (amphistomy). The primitive features of enoplid embryogenesis correlate with other unusual characters. Enoplid spermatozoa have a normal nuclear envelope, which is a unique character among nematodes. Eutely is not a characteristic of marine enoplids and, furthermore, this group of nematodes can regenerate internal tissue to repair damaged organs. Molecular phylogeny also supports the idea of a unique position of the Enoplida among nematodes. There is no obvious way to derive the nematode cleavage from the classical spiral or radial cleavage. Nematode embryogenesis is more likely to be a derivation from the arthropod mode of embryonic development, which is to say that the idea of ecdysozoan kinship of Nematodes cannot be rejected.

COMPARISON OF NEMATODE COMMUNITIES IN THREE LONG TERM POLLUTED SITES IN THE BOURGOYEN-OSSEMEERSEN (GHENT, BELGUIM). **Manhout, J.,[1] M. Spildooren, [2] R. Van Gansbeke, [2] R. Vandriessche, [2] W. Bert, [2] G. Borgonie, [2] and W. Decraemer.[1,2]** [1] Department of Recent Invertebrates, Royal Belgian Institute of Natural Sciences, Vautierstraat 29, B-1000 Brussels; [2] Section Nematology, Ghent University, K.L. Ledeganckstraat 35, B-9000 Ghent.

Nematodes have a large potential as biological indicators. They are the most abundant multicellular organisms on the planet and are numerous in a large number of different habitats including heavily polluted sites. Their lifecycles varies from a few days to years which implies that a nematode community will be sensitive to short as well as long-term changes in their habitat. The nature reserve Bourgoyen-Ossemeersen used to be partially made up of long-term polluted (especially heavy metal pollution) sites, such as an old municipal waste site, a tar waste site and a sludge disposal site. These sites differ mostly in pollution degree and soil structure. From each waste site and their nearby control sites 300 individuals were identified to genus level. The concentrations of the most common heavy metals (Pb, Zn, Cu and Cd) were measured for these three sites. Especially high concentrations were found for Cd and Pb. A comparison between these different sites was made using different ecological indices (the Maturity Index, Trophical Index and others) in correlation with the pollution degree and vegetation.

AN ALDOLASE GENE FROM THE CYST NEMATODES *HETERODERA GLYCINES* AND *GLOBODERA ROSTOCHIENSIS*. **Masler, E. P., D. J. Chitwood, and E. S. Kovaleva.** Nematology Laboratory, USDA/ARS, Beltsville, MD 20705.

Fructose-bisphosphate aldolase (E.C. 4.1.2.13) is the key enzyme of the sixth step of glycolysis. Aldolase isozymes take part in developmental stage-specific and tissue-specific sugar phosphate metabolism. The expression levels often correlate with diet or other environmental conditions, therefore the enzyme may regulate important aspects of pathogenesis in plant-parasitic nematodes. Here we report the isolation and characterization of full-length coding sequences for aldolase from the cyst nematodes *Heterodera glycines* and *Globodera rostochiensis*. The respective sequences encode 40-kilodalton secretory proteins of 366 and 365 amino acids. The homology of the two deduced proteins is very high: 89% amino acid identity. Comparison with available amino acid sequences from other nematodes revealed that both proteins from cyst nematodes have higher homology to aldolase 2 (Ce-2) rather than aldolase 1 (Ce-1) from *Caenorhabditis elegans* or aldolase from *Onchocerca volvulus* (72, 60 and 61% amino acid identity, respectively). Total aldolase-like activity in tissue extracts from *H. glycines* and *C. elegans* was quantified by spectrophotometric assay using fructose-1,6-biphospate as the substrate. The measured affinity was similar for both species (Km in micromolar range) whereas the amount was much greater for *C. elegans* than for any developmental stage of *H. glycines*. Inhibition analysis also revealed metabolic differences between the species. Experiments are underway to examine the effects of diet on the activity of aldolase. For *H. glycines*, this is achieved by exposure of soybean plants to conditions that retard root growth such as reduced light or leaf removal.

ALTERATIONS IN GENE EXPRESSION AND METABOLITE LEVELS IN SOYBEAN ROOTS DURING INVASION BY THE SOYBEAN CYST NEMATODE. **Matthews, B.,[1] R. Khan,[1] N. Alkharouf,[1] and L. W. Sumner.[2]** [1]USDA, ARS, Plant Science Institute, Soybean Genomics & Improvement Laboratory, BARC-West, Beltsville, MD 20705, USA; [2]Noble Foundation, Ardmore, OK, USA.

The soybean cyst nematode (SCN), *Heterodera glycines* is the most devastating pest of soybean in the US, causing an estimated one billion dollars in damage each year. The defense response of soybean to SCN is a multigenic trait and varies depending upon the genotypes of soybean and SCN. More than 6,000 cDNA inserts from several soybean cDNA libraries were printed and monitored using microarrays to identify genes involved in the response of soybean to SCN. RNA was harvested from roots of soybean cv. Peking resistant to SCN strain NL1-RHp (reflecting a race 3 phenotype) and susceptible cv. Kent, either not infected or at 0, 6, 12 hr, 1, 2, 4, 6, and 8 days after infection by SCN strain NL1-RHp.

Two independent biological samples were used for microarrays and-metabolic profiles. The RNA was fluorescently labeled as cDNA for hybridization to the microarrays. A number of defense-related genes were identified statistically as being altered due to SCN invasion, as were genes encoding potential regulatory factors, such as kinases and transcription factors, genes involved in sugar metabolism and cell wall formation, and a number of genes encoding proteins of unknown function. Alterations in metabolic profiles combined with gene expression analysis allowed a better interpretation of the events that occur in the root during nematode attack. These results are being used to identify pathways, genes, and metabolites important to the defense response of soybean against SCN attack. See our web site at http://bldg6.arsusda.gov/benlab/ for further information.

SELECTION OF HOST-PLANT RESISTANCE TO ROOT-KNOT NEMATODES IN CARROT USING FIELD SCREENING AND MOLECULAR MARKERS. **Matthews, W. C.,** [1] **P. W. Simon,** [2] **and P. A. Roberts.**[1] [1]Department of Nematology, University of California, Riverside, CA 92521. [2]USDA-ARS,Department of Horticulture, University of Wisconsin, Madison, WI 53706.

Host plant resistance in carrot (*Daucus carota*), conferred by the *Mj-1* locus derived from breeding line Brasilia 1252, is highly effective against the root-knot nematode, *Meloidogyne javanica*. In multiple field evaluations of Brasilia 1252-derived entries, including hybrid combinations, this resistance was variable against *M. incognita*. Screening of derivatives for reaction to *M. incognita* and *M. javanica* suggested this variability is due to a lack of homozygosity of the *Mj-1* locus. The derivatives most uniformly resistant to *M. incognita* lacked evidence of segregation to *M. javanica*. Some derivatives that were segregating to *M. javanica* were also more susceptible to *M. incognita*, while other derivatives, though showing no evidence of segregation to *M. javanica*, were also less resistant to *M. incognita*. These latter derivatives may still have individuals that are heterozygous for *Mj-1*, but were not observed in the *M. javanica* tests. The field evidence confirms previous findings of a phenotypic separation of homozygosity and heterozygosity of the *Mj-1* locus, and that this gene dosage effect is more apparent with *M. incognita*. In addition, many hybrid entries were more severely damaged by *M. incognita* in field tests conducted at high temperatures, indicating that the gene dosage effect to *M. incognita* is greater at high temperature. These results illustrate the necessity that *Mj-1* needs to be homozygous to be fully effective, especially against *M. incognita*. The application of *Mj-1* linked STS codominant flanking markers to Brasilia 1252 derivatives is being made to confirm the phenotypic responses in field screenings and to select advanced lines homozygous for *Mj-1* resistance.

NEMATICIDAL ACTIVITY OF WALNUT EXTRACTS AGAINST ROOT-KNOT NEMATODES. **McKenry, M. V., and S. A. Anwar**. Department of Nematology, University of California, Riverside, CA 92521.

Reducing branches, limbs, trunks, and roots of *Juglans* spp. to a powder of less than 100-mesh can provide a water extractable tea. The powder exhibits a specific gravity greater than 1.0, enabling easy separation of particulates from the tea. *Juglans* spp. contain antioxidants, phenolic compounds, tannins, and other soluble ingredients that, in combination, produce a nematicidal effect. A bioassay was performed to investigate the nematicidal activity of walnut extract against second stage juveniles (J2) of *Meloidogyne incognita* at 24, 48, and 144 hours after exposure to various extract concentrations. Freshly hatched J2 were placed into sealed plastic vials at four concentrations of *Juglans* tea, including 50, 25, 10, and 2.5 g/l. The three higher concentrations consistently caused greater mortality, compared to the 2.5 g/l concentration and a water control. Forty-eight hours later, the two higher concentrations resulted in 100% nematode mortality with vacuoles observed in the intestinal region. The 10 g/l concentration took 96 additional hours to–produce 100% nematode mortality. By comparison, synthetic juglone provides 100% kill after 48 hr exposure to 1 g/l. In a microplot setting, newly planted grapevines inoculated with *M. incognita* J2 received 50 g/l *Juglans* tea six times over a two-year period. Nematode control approximated 75%, equivalent to the standard phenamiphos comparison. Phytotoxicity was not observed with repeated applications of the tea. This tea product provides the opportunity for delivery of botanically derived nematicidal agents deep into the soil profile around roots of perennial crops.

BIODEGRADATION OF ALDICARB IN *ROTYLENCHULUS RENIFORMIS* INFESTED COTTON FIELD SOILS. **McLean, K. S.,**[1] **and G. W. Lawrence.**[2] [1]Department of Entomology & Plant Pathology, Auburn University, Auburn, AL 36849, [2]Department of Entomology & Plant Pathology, Mississippi State University, Mississippi State, MS 39759.

The microbial degradation of aldicarb was examined in soils from four cotton fields naturally infested with *Rotylenchulus reniformis* with a history of aldicarb use. In test 1, treatments included soils that were sterilized by autoclaving or natural (not autoclaved) and in which aldicarb was incorporated at 0.59 kg a.i./ha or no aldicarb was added. In test 2, the aldicarb rates were expanded to 0.29, 0.59, 0.85, and 1.19 kg a.i./ha. In both tests, the autoclaved soil was infested with *R. reniformis* to equal the original natural soil population level. In test 1, the addition of aldicarb at 0.59 kg a.i./ha to natural soil decreased R. reniformis numbers only 36% compared to 95% in the same soils which were autoclaved. The use of increasing rates of aldicarb in test 2 did not increase the efficacy of aldicarb in the natural soils. Nematode numbers were reduced 40, 23, and 7 % where aldicarb was applied at 0.29, 0.59, and 0.85 kg a.i./ha and increased 6 % when aldicarb was applied at 1.19 kg a.i./ha in one of the natural soils. Autoclaving restored aldicarb toxicity in all soils tested. *Rotylenchulus reniformis*

numbers were reduced 97% as compared to the autoclaved soil without aldicarb. Bacterial populations isolated from these soils increased in the natural soils with the addition of aldicarb. However, no bacterial species was consistency associated with aldicarb degradation.

DEVELOPMENT OF RESISTANCE IN CLOVERS FOR NEW ZEALAND PASTURE. **Mercer, C. F.,[1] R. N. Watson,[2] B. A. Barrett,[1] G. Spangenburg, [3] J. van den Bosch,[4] and K. K. Moore.[1]** [1]AgResearch Grasslands, PB 11-008, Palmerston North, New Zealand, [2]AgResearch Ruakura, PB 3123, Hamilton, New Zealand, [3]Agriculture Victoria, La Trobe University, Bundoora, Victoria, Australia, [4]Wrightson Research, PO Box 939, Christchurch, New Zealand.

White clover (*Trifolium repens*) is the key legume in New Zealand pasture but does not reach its potential in part because of root-infecting nematodes. Recurrent selection programs in white clover improved resistance to *Meloidogyne trifoliophila* (clover root-knot nematode) (CRKN) and *Heterodera trifolii* (clover cyst nematode) over seven and five generations respectively. A concurrent four-generation selection program improved tolerance to nematodes. Two-year field trials testing germplasm from these programs under grazing, showed that resistant material yielded better than susceptible and less than the tolerant material and cultivar controls. Further selection of resistant germplasm for agronomic performance is underway. Twenty New Zealand pasture populations each of CRKN and CCN were found to be the same pathotype for each nematode. In comparisons of foliage dry weight yields from pots of nematode-infected resistant and susceptible lines, all lines were reduced by nematode infection but the resistant ones outyielded the susceptible ones. Studies of stained roots revealed that invasive juveniles of CRKN entered roots of resistant and susceptible genotypes in similar numbers and moved towards the root tip in both plant types. However, at 3 days after inoculation, the reaction of the juveniles differed. Screening of a mapping population of *T. semipilosum* using SSRs developed for white clover confirmed that the CRKN resistance is conferred by a single locus. White clover mapping populations for reaction to CRKN and CCN are being evaluated with the aim of developing markers.

ON THE ORIGINS OF DORYLAIMIDA: A TALE WITH TEETH. **Mullin, P. G.** Department of Plant Pathology, University of Nebraska, Lincoln, NE USA

In any given terrestrial soil sample, a significant proportion of the nematodes present will be “free living” forms. The majority of these are often representatives of the subclass Dorylaimia, which includes the orders Dorylaimida and Mononchida. Dorylaimida are considered by many workers to be the most diverse order of soil nematodes, and can be found in all habitats worldwide. The origins and deep phylogenetic relationships of this order, however, have not been clearly determined. Some have proposed the evolution of both Dorylaimida (*via* the Nygolaimina) and Mononchida from a common ancestor resembling modern Bathyodontidae, while others have envisioned a more basal position for the Mononchida, which then gave rise to the bathydontids and subsequently the Dorylaimida (again *via* the nygolaims). Phylogenetic analyses using 18S SSU rDNA suggest that bathyodontids share common ancestry with a combined clade comprising Mononchida and Mermithida, and that the origins of Dorylaimida predate the divergence of Bathydontidae and the mermithid-mononchid lineage. Since many of the nematodes classified in Dorylaimia are soil predators, discussion of key adaptations will center on modifications of feeding apparatus that facilitate this mode of existence. Subsequent adaptation of these structures in the Dorylaimida has allowed this group to diversify and exploit a wide range of terrestrial ecological niches.

EFFECT OF ORGANIC AMENDMENTS ON SOIL NEMATODE COMMUNITY, MICROBIAL BIOMASS AND NUTRIENT CYCLING. **Nahar, M. S.,[1] P. S. Grewal,[2] S. A. Miller,[1] and D. Stinner.[2]** [1]Dept. of Plant Pathology & [2]Dept. of Entomology, The Ohio State University, OARDC, Wooster, OH 44691.

The impact of two organic soil amendments on nematode abundance, community structure and soil characteristics was studied in tomatoes in a transitional organic system. The field experiment was conducted during 2001–2002 in Wooster, OH on silt loam soil previously cropped in a maize-clover rotation. Treatments (raw or composted dairy cow manure amendments and an untreated control) were arranged in a randomized complete block design. Amendments were applied in the spring at a rate estimated to deliver 101 kg N ha-1, and incorporated prior to planting. Soil samples were collected in the spring (before adding amendments) and fall (after harvesting) to determine nematode abundance, community structure and soil characteristics. Shannon-Weiner (*H f*), Simpson (*f*Ē), Pielou (*J f*), and maturity indices were used to compare nematode community structure in amended and non-amended plots. Spring incorporation of organic amendments increased the abundance of bacterial feeding, fungal feeding, omnivorous and predatory nematodes, but decreased plant parasitic nematode populations. Shannon diversity (*H f*) and combined maturity (‡”MI) indices of soil nematodes were reduced in manure amended plots. Organic amendments increased total organic matter, particulate organic matter, microbial biomass-N, potentially mineralizable-N and C but decreased bulk density. Particulate organic matter, potentially mineralizable- N and C were negatively correlated with Shannon diversity (*H f*) and combined maturity index (‡”MI). Particulate organic matter showed a positive correlation with abundance of bacterial feeding ($r = 0.49$ $p<0.0004$) and total

free-living ($r = 0.41$ $p<0.004$) nematodes across all sampling times. We also found a strong negative relationship ($r = -82$, $p<0.0001$) between the abundance of free-living and plant parasitic nematodes, which may be used as a quality indicator.

INTEGRATED MANAGEMENT OF ROOT-KNOT AND PURPLE BLOTCH DISEASES IN GREEN ONION. **Nahar, M. S.,[1] M. H. Rahman,[1] H. S. Jasmine,[1] and S. A. Miller.[2]** [1]Department of Plant Pathology, Bangladesh Agricultural Research Institute, Bangladesh & [2]The Ohio State University, Wooster, OH 44691.

Green onion, *Allium fistulosum* L., is a high value crop cultivated throughout the year in peri-urban farming communities surrounding Dhaka, Bangladesh. Due to intensive cultivation of green onion in rotation with rice, root-knot disease caused by *Meloidogyne graminicola*, and purple blotch caused by *Alternaria porri*, are common and devastating. An on-farm experiment was conducted in summer 1998 and 1999 in naturally infested fields at Kashimpur to evaluate the effects of soil organic amendments, nematicide, and fungicides in managing these diseases. Plots incorporated with poultry litter (3t/ha) plus standard fertilization (composted cow manure 10 t, nitrogen 69 kg, phosphate 90 kg, potash 96.6 kg, sulfur 19.8 kg, zinc 4.29 kg, and boron 0.45 kg per hectare) alone, and in combination with a foliar application of iprodione at a rate of 1000 ppm produced taller, heavier and healthier green onion plants with minimum diseases compared with those of the traditional farmer practice (composted cow manure 20 t, nitrogen 92 kg, phosphate 299.7 kg, and potash 199.8 kg per hectare) and the control treatment (only standard fertilizers). Both treatments also resulted in about twice the yield of the plots managed using the traditional farmer practice. Plant growth and yield were negatively correlated with purple blotch and root-knot diseases, while purple blotch disease incidence and root-knot nematode gall indexing values were positively correlated. Different stages of *M. graminicola* (J1, J2, J3, J4 and adult) were also negatively correlated with plant growth and yield. Root gall production was positively correlated with egg, adult and total *M. graminicol* populations in the roots.

THE ILLINOIS SCN TYPE TEST: PRACTICAL APPLICATION OF THE HG TYPE CLASSIFICATION SYSTEM. **Niblack, T. L.,[1] G. R. Noel, [2] and K. N. Lambert.[1]** [1] Department of Crop Sciences, University of Illinois, Urbana, IL 61801, [2]USDA-ARS, University of Illinois, Urbana, IL 61801.

The HG Type classification system for soybean cyst nematode (SCN) populations is based on the female index, a measure of SCN virulence, on seven soybean plant introductions (PI) and the susceptible cultivar Lee 74. Of the seven indicator lines, only three have been used to develop SCN-resistant soybean cultivars that are currently available to soybean producers in Illinois: PI 548402 (Peking), PI 88788, and PI 437654 (indicator lines 1, 2, and 4 in the HG Type test). An incomplete HG Type test named the Illinois SCN Type test includes only these three indicator lines and Lee 74, and may include one or more additional cultivars for comparison. Conditions of the test are as specified for HG Type tests, except that it is conducted in a hydroponic system so that labs without greenhouse facilities are able to perform it after training. The results can be used in conjunction with data from yield trials, SCN-resistance screening, and lists of sources of resistance to aid cultivar selection decisions. Results suggest that this test will be useful for growers whose SCN-resistant cultivars are suffering damage due to SCN. SCN Type tests can be adapted easily to other states or regions.

THE PHYLOGENETIC POSITION OF THE NEMATODA. SYNAPOMORPHIES OF THE ECDYSOZOA? **Nielsen, C**. Zoological Museum (University of Copenhagen), Universitetsparken 15, DK-2100 Copenhagen, DENMARK.

Nematoda has traditionally been placed in the group Aschelminthes or Cycloneuralia, in a position quite distant from the arthropods. However, almost all molecular studies place them together with the arthropods, in a clade called Ecdysoza, which also comprises Kinorhyncha, Priapula, Loricifera, and Nematomorpha. Several studies have searched for morphological characters which could be interpreted as ecysozoan synapomorphies, but the only character complex which apparently qualifies is that of the cuticle and its molting. The ultrastructure of the cuticle with an outer trilaminar epicuticle shows considerable similarities, but the arthropod cuticle consists of chitin and that of the nematodes of collagen. Molting appears to be governed by hormones called ecdysones, especially 20-hydroxy-ecdysone, but there are considerable differences between the mechanisms in crustaceans and insects, and the mechanism in nematodes is almost totally unknown. It seems uncertain whether the nematodes are able to synthesize the hormones, but with the present knowledge of the whole genome of Caenorhabditis it should be possible to investigate the whole process in detail. The results may even turn out to have economic importance, with possibilities for biological control.

POTENTIAL YIELD LOSS WHEN PLANTING *HETERODERA GLYCINES*-RESISTANT CULTIVARS. **Noel, G. R.,[1] and S. J. Bauer.[2]** [1]USDA, ARS, Urbana, IL 61801, [2]Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

Soybean producers are concerned that planting *Heterodera glycines*-resistant soybean cultivars either in non-infested fields or fields with low numbers of *H. glycines* sacrifices yield. Seven fields located in central Illinois and western Indiana, ranging from no infestation to moderate levels of infestation with *H.glycines,* were planted with a susceptible cultivar, a cultivar with a low level of resistance from PI88.788 resistance, a cultivar with a high level of resistance from PI88.788, and a cultivar carrying resistance from Peking. Plots ranged from 100 to 800 m long and 40 to 150 m wide. Each cultivar

was arranged in a randomized complete block design and replicated three times. All farming operations were done with the producers' equipment. Numbers of *H. glycines* eggs were determined at planting and at harvest. Yield was determined with a weigh wagon, and moisture was corrected to 13%. Nematode reproduction differed among cultivars and locations. Yield of the susceptible cultivar was greater in fields with no *H.glycines* or low levels of infestation. When the level of infestation reached 200 eggs/150 cm^3 soil, there was a yield advantage when the cultivar with the high level of resistance from PI88.788 and the cultivar with Peking resistance were planted. Higher levels of infestation were required before planting the cultivar with the low level of PI88.788 resistance became advantageous. Although progress has been made in equalizing the yield potential of *H glycines*-susceptible and resistant cultivars, developing equivalent yield potential in fields either with no *H glycines* or low levels of infestation remains elusive.

ROLE OF WEED HOSTS IN POPULATION ENHANCEMENT OF ROOT-KNOT NEMATODE. **Noling, J. W.,[1] and J. P. Gilreath.[2]** [1]University of Florida, IFAS, Citrus Research & Education Center, Lake Alfred, FL 33850;[2] University of Florida, IFAS, Gulf Coast Research & Education Center, Bradenton, FL 33314.

Seven commercial vegetable fields in Florida were surveyed in 2002 to evaluate the host status of various weeds to root-knot nematode *(Meloidogyne* spp.*)*. Weed roots collected from each field were indexed for root gall severity and relative density of egg masses per gram root. In five of the seven fields surveyed, *M. incognita* was the exclusive root-knot nematode species recovered from weed roots. An apparently new root-knot nematode species to Florida, *M. mayagensis,* was recovered from one of the seven field sites. In general, nematode galling and egg production were observed on the roots of fifteen weed species. With some weed species, nematode galling and egg production were variable between survey site locations but were not correlated with differences in nematode species. The results of three field trials demonstrated the importance of weed density and management to nematode population suppression. In general these results suggest that nematodes cannot be effectively managed without effective weed management. Weeds which were allowed to grow and increase in numbers, particularly between mulch covered rows, served to increase soil population densities of nematodes and perpetuate the nematode problem from one cropping season to the next. The lack of weed control appears to mandate the continued need for broad spectrum soil fumigants for nematode control.

REPRODUCTION RATES OF FUNGAL-FEEDING *FILENCHUS* SPP. (TYLENCHIDAE) AS AFFECTED BY FOOD SOURCE AND CULTURE MEDIA. **Okada, H., and I. Kadota** Laboratory of Crop Protection, National Agricultural Research Center for Tohoku Region, Arai, Fukushima-city, 960-2156, Japan.

Studies of their fungal-feeding habits will allow better understanding of the ecology of nematodes in the genus *Filenchus* and in other genera of the Tylenchidae. In previous studies we measured reproduction rates of *Filenchus misellus*, collected from decomposing rice straw, when feeding on fungi on PDA plates. Studies on the other species of *Filenchus* are necessary to determine how commonly fungal-feeding occurs in the genus. Additionally, to evaluate the importance of the fungal-feeding habit under natural conditions, it is necessary to determine whether *Filenchus* spp. feed on fungi in soil. We found that *Filenchus discrepans*, collected in a crop field in Japan, also feeds on fungal hyphae. In this study we report reproduction rates of the nematode over 46 days of incubation at 25 ?C on six fungal species as food sources on PDA plates and in soil amended with chopped soybean stems and pods. On PDA plates, reproduction rates were high on *Rhizoctonia, Coprinus* and *Chaetomium*, while low to moderate on *Fusarium, Pythium* and *Pleurotus*. This host status of the fungi for *F. discrepans* is similar to that we found for *F. misellus* in previous studies. In soil, however, high reproduction rates of *F. discrepans* occurred only on *Rhizoctonia* and *Coprinus*. We will report also the results of feeding habit experiments for the other species of *Filenchus*.

NEMATODES ASSOCIATED WITH COTTON IN LOUISIANA DURING 1990-2001. **Overstreet, C**., **and M. Wolcott**. Department of Plant Pathology and Crop Physiology, Louisiana State University AgCenter, Baton Rouge, LA 70803.

During 1990-2001, 12,936 soil samples were processed by the Nematode Advisory Service from cotton fields in Louisiana. Fifteen genera of plant-parasitic nematodes were found occurring in these fields. *Rotylenchulus* was the most common genus found (45.4% of the samples). *Meloidogyne* sp. was found in only 9% of the fields. Nematodes from these two genera include the most important plant-parasites on cotton in Louisiana. Other nematode types that were fairly common included *Helicotylenchus* sp. (43.9%), *Tylenchorhynchus* sp. or *Melinius* sp. (18.2%), *Pratylenchus* sp. (9.6%), and *Hoplolaimus* sp.(9.5%). Fewer occurrences were found with *Criconemella* sp. (6%), *Paratrichodorus* sp. (3.1%), *Paratylenchus* sp. (2.2%), *Heterodera* sp. (1.3%), *Xiphinema* sp. (0.5%), and *Scutellonema* (0.1%). Three genera, *Criconema*, *Gracilacus*, and *Hirschmanniella,* were found in only a single field for each type. The highest incidence of *Rotylenchulus* sp. was found in Franklin, Morehouse, Rapides, and Richland parishes located in Central and Northeast Louisiana (primarily Southern Mississippi Alluvium or Silty Uplands and Ouachita Valley Alluvium soils). *Meloidogye* sp. was found to occur in the largest number of fields in Morehouse, Ouachita, Red River, and Tensas parishes (Red River, Ouachita and Mississippi Alluvium soil types). *Meloidogye* sp. and *Rotylenchulus* sp. normally are not found together in

Louisiana as evident from only a 3% concomitant occurrence. There was also not a significant correlation between populations of these two nematodes.

HOST SUITABILITY OF WHEAT TO *MELOIDOGYNE GRAMINICOLA* IN THE RICE-WHEAT SYSTEM OF BANGLADESH. **Padgham, J. L.** Department of Crop and Soil Sciences, Cornell University, Ithaca, NY 14853.

The potential of wheat as an alternate host to *M. graminicola* in Bangladesh's rice-wheat cropping system was investigated. A greenhouse experiment and a soil bioassay test of rice-wheat production fields were undertaken to determine the host suitability of wheat to *M. graminicola* and to assess root-galling severity on wheat in production fields, respectively. The four cultivars of wheat (Gowrab, Kanchan, Satabdhi, and Sourav) tested for host suitability were determined to be excellent hosts of *M. graminicola*, and had mean root-galling severity ratings between 6.3 to 7.7 (on a 1 to 9 scale). Reproduction of *M. graminicola* on rice (cv. BR11), which was included for comparison, was twice as great as that of wheat and had a mean root-galling severity rating of 7.0. Root-galling severity ratings on wheat in the soil bioassay test ranged between 2 and 6, with the highest root-galling severity observed on plants grown in soil not treated with nematicide in the previous rice crop. The results of these two experiments illustrate the potential of wheat to maintain high soil populations of this nematode between monsoon rice seasons, and suggest that wheat may be susceptible to significant damage from *M. graminicola* in a rice-wheat rotation.

INTERACTION OF *ROTYLENCHULUS RENIFORMIS* AND SPECIES OF *FUSARIUM* ON SEEDLING DISEASE OF COTTON. **Palmateer, A. J. ,[1] K. S. McLean,[1] E. van Santen,[2] and G. Morgan-Jones.[1]** [1]Department of Entomology & Plant Pathology, Auburn University, AL 36849, Department of Agronomy and Soils, Auburn University, AL 36849.

A greenhouse study examined the interaction of *Rotylenchulus reniformis* and ten *Fusarium* species on cotton seedling disease. Treatments consisted of *Fusarium chlamydosporum, F. equiseti, F. lateritium, F. moniliforme, F. oxysporum, F. oxysporum* f.sp. *vasinfectum*, *F. proliferatum, F. semitectum, F. solani,* and *F. sporotrichioides. Rhizoctonia solani* and *Thielaviopsis basicola* were included for disease comparisons. Pots containing 500 cm^3 of natural and autoclaved soil were infested with an inoculum concentration of 1% (v/v) of fungal treatments alone and in combination with 2000 *R. reniformis* juvenile and vermiform adult nematodes in a factorial arrangement. *Rotylenchulus reniformis* increased cotton seedling root hypocotyl disease index (RHDI) when combined with *F. chlamydosporum, F. oxysporum* f. sp. *vasinfectum, F. solani, R. solani* or *T. basicola*. *Fusarium lateritium, F. moniliforme*, *F. oxysporum* f.sp. *vasinfectum, F. solani, R. solani, T. basicola*, and both controls displayed shorter seedlings in *R. reniformis* infested soil. *Fusarium lateritium, F. moniliforme*, *R. solani* and *T. basicola* caused damping-off in natural and autoclaved soil. *Rhizoctonia solani* had a greater impact on seedling dry weight in *R. reniformis* infested soil. *Fusarium chlamydosporum, F. lateritium,* and *F. oxysporum* did not impact *R. reniformis* reproduction, whereas seedlings growing in soil infested with *F. moniliforme, R. solani* and *T. basicola* supported fewer numbers of *R. reniformis* than the control with millet. *Rotylenchulus reniformis* reproduction on cotton seedlings was greater in natural soil compared to autoclaved soil. This study suggests the importance of *Fusarium* species and *R. reniformis* on cotton seedling disease.

IN VITRO, MORPHOLOGICAL AND MOLECULAR EVALUATION OF MALAYSIAN ISOLATES OF *PAECILOMYCES LILACINUS* AGAINST ROOT KNOT NEMATODES. **Pande, S., [1] S. D. Atkins,[2] D. Crump,[3] A. Jama, [1] and S. Gowen. [1]** [1]Crop Protection Group, University of Reading, UK, RG6 6AR; [2] Rothamsted Research, Harpenden, UK, AL5 2JQ.;[3] BioNem, Harpenden, UK, LU1 4AS.

Paecilomyces lilacinus is a facultative parasite of cyst and root-knot nematode eggs. Isolates of *P. lilacinus* were collected from various regions of Malaysia. Morphological observations, culture on semi-selective media and PCR detection using specific primers confirmed them as *P. lilacinus*. They were evaluated *in vitro* for egg parasitism by placing sterilised *Meloidogyne* eggs on sporulating cultures. Fifteen isolates infected the eggs to varying degrees (ie from 2-88 %). These isolates displayed a variety of cultural phenotypes. The pigment of the fungal colonies varied from light yellow to dark pink. The pinkness of the colony was positively correlated with sporulation. Egg pathogenicity of the isolate was positively correlated to sporulation. Genetic profiles were generated for the isolates using the arbitrary RAPD (random amplified polymorphic DNA) primer V17. The RAPD profiles demonstrated the existence of six genetic groups. The isolates from the same groups showed similar growth rates and optimum growth temperature values, but sporulation and pathogenicity varied. All three isolates in the same genetic group as Pl 251 (a commercially available product) had optimum growth at 30 °C but only two were as highly sporulating and as pathogenic as Pl 251. Growth temperature reaction of each group might be related to their geographical distribution in Malaysia. Selective primers based on ITS sequence detected all of the isolates tested as well as *P. lilacinus* isolates from other countries and the commercial strain Pl 251. The PCR reaction was optimised to detect the presence of the fungus from environmental samples including soil, roots, and eggs.

MORPHOBIOMETRIC STUDIES OF *BURSAPHELENCHUS XYLOPHILUS* DEVELOPMENT STAGES, FROM PORTUGAL. **Penas, A. C.,[1] M. A. Bravo,[1] and M. Mota.[2]** [1]Dept. Protecção de Plantas, Estação Agronómica Nacional, Qtª Marquês, 2780 Oeiras, Portugal, [2]NemaLab-ICAM/Departamento de Biologia, Universidade de évora, 7000 évora, Portugal.

Morphobiometric studies of developmental stages of *Bursaphelenchus xylophilus* were conducted using a Portuguese population collected from *Pinus pinaster* and reared on *Botrytis cinerea* at room temperature. Nematodes were extracted by the Baermann funnel technique and measured after staining in 1% acetic orcein for 24 hours for the propagative forms and approximately 48 hours for the dispersal stages. Adults were measured from temporary mounts without staining. Measurements of total body length and reproductive system length of molting specimens allowed separation into four distinct groups: 2nd molt, 3rd molt, female 4th molt and male 4th molt. The same measurements were made for individuals that were not molting; as a result, four groups, J2, J3, J4 and adults, were established which fit in between molts. Cell multiplication of the reproductive tract occurred throughout nematode development, even though this multiplication did not display the same intensity. Genital primordium developed until the end of the 2nd molt very slowly, followed by a much faster developmental rate. Genital primordium length of resistance juveniles found in *P. pinaster* and from *B. cinerea* was closer to that of the 3rd propagative juvenile stage although body length was identical to that of 4th propagative juvenile stages. However, the resistance juveniles from pine were slightly smaller than those from *B. cinerea*. Adult females showed a longer reproductive tract than pre-adult females mainly due to the growth of post-uterine sac.

IDENTIFICATION OF *MELOIDOGYNE* SP. ASSOCIATED WITH THE WATER WILLOW, *JUSTICIA AMERICANA*. **Phan, T. D., [1] P. A. Jackson,[1] R. W. Poon,[1] K. B. Nguyen,[1] K. M. Fritz,[2] and B. J. Adams.[1]** [1]Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620, [2]Department of Biological Sciences, Auburn University, AL 36849.

Ecological studies, both terrestrial and aquatic, are becoming increasingly complex in their attempts to account for the contributions of multitudinous numbers of species and trophic relationships to ecosystem processes and function. The movement towards more inclusive, multidisciplinary ecological research stems from the acknowledgement that many large-scale patterns of above ground plant and animal community structure cannot be completely explained without considering the contribution of the belowground members of the ecosystem. As part of a study to investigate the contribution of riparian vegetation to the substrate stability of an aquatic-terrestrial interface in a southeastern stream ecosystem, a population of *Meloidogyne* sp. was isolated from water willow, *Justicia americana*. The nematode was present at levels high enough to cause significant damage to its host, and perhaps play a role in the water willow community structure, which, in turn, could impact the stability of the riparian ecosystem. Because of difficulties obtaining and culturing the sampled nematodes, morphological identification was intractable. However, we were able to extract DNA from ethanol preserved specimens and sequence a portion of ribosomal DNA (a 5′ portion of the 18S rRNA gene). We aligned the sequence to other published 18S *Meloidogyne* sequences and performed phylogenetic analyses. Relative to the other taxa, the nematode found on water willow is closely related to *M. arenaria, M. javanica,* and *M. incognita*. However, polymorphisms at several nucleotide positions and at least one autapomorphy suggest the initiation of lineage independence. This seems to fit the trend that as ecosystems are studied in greater detail, the known diversity of nematode taxa increases accordingly.

USE OF GROWER FIELD DATA TO PARAMETERISE POTATO CYST NEMATODE MANAGEMENT MODEL AND TO ASSESS NEMATICIDE EFFICIENCY. **Phillips, M. S., M. J. Elliot, and D. L. Trudgill.** Scottish Crop Research Institute, Invergowrie, Dundee, DD2 5DA, UK.

A computer programme has been produced to model the yield losses caused by the potato cyst nematodes (PCN) *Globodera pallida* and *G. rostochiensis* and to indicate the population dynamics following various management strategies. The models were originally developed using data from detailed experimental trials. The inputs for the programme include the tolerance and resistance of the cultivars, rotation length and nematicide use for a particular environment. In order to make the model more applicable to particular farms or fields within farms a series of grower trials were conducted in potato crops treated with nematicide. Small plots were then left untreated and the initial and final population densities together with yield were assessed. The data resulting from these trials was assessed in terms of their utility in parameterising the model. The results showed that if the range of initial population densities was sufficiently broad customising the model to that environment was possible. More interesting however was the information comparing the results from nematicide treated and untreated plots that suggested that the nematicide efficiency varied greatly and that the estimated efficacy of nematicide treatment was greater in respect of yield then with respect to nematode population control.

EFFECTS OF *MESOCRICONEMA XENOPLAX* ON *VITIS VINIFERA* AND ASSOCIATED MYCORRHIZAL FUNGI. **Pinkerton, J. N., and R. P. Schreiner.** USDA ARS Horticultural Crops Research Laboratory, Corvallis, OR 97330,

Surveys reported that *Mesocriconema xenoplax* was present in 85% of vineyards in western Oregon, but yields were not depressed in established vines. Microplots studies were initiated in 1997 in a Willamette Valley vineyard to determine the

impact of *M. xenoplax* on vine establishment. Plots were infested with 0.03, 0.6, and 3.0 nematodes g^{-1} soil and planted with self-rooted Chardonnay and Pinot Noir vines. Control plots were not infested. In 1998 with ca 2000 degree-day base 9C accumulation, population densities increased 32× and 44× on one-year-old Chardonnay and Pinot Noir vines. Nematode population dynamics and pruning data suggest that the carrying capacity of vines in microplots was 5-8 *M. xenoplax* g^{-1} soil. In November 2000, 40 months after planting, pruning weights, fine root weights and fruit yield were reduced greater than 58%, 75%, and 33%, respectively, for all vines planted in infested soil compared to control vines. In November, greater than 80% of the fine root length was colonized by arbuscular mycorrhizal fungi in all treatments. The frequency of fine roots containing arbuscules (the site of nutrient transfer between plant and fungus), however, was significantly depressed in plants from *M. xenoplax* infested treatments as compared to controls. Competition for photosynthate within the roots system is proposed as the mechanism by which nematodes suppressed arbuscule frequency.

A NEMATODE APPROACH TO MANAGEMENT AND CONTROL OF *CACTOBLASTIS*? **Poon, R. W.,[1] P. A. Jackson,[1] T. D. Phan,[1] K. B. Nguyen,[1] K. A. Bloem,[2] and B. J. Adams.[1]** [1]Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620, [2]USDA-APHIS-NBCI, Center for Biological Control, Florida A&M University, Tallahassee, FL.

The cactus moth, *Cactoblastis cactorum*, is an eminent menace to prickly pear cacti in Florida, pushing "threatened" *Opuntia. spinosissima*. and *O. tricantha* near to extinction. Since its first detection in Florida in 1989, the invasive moth has extended its range north to Charleston SC and west to the extent of the Florida panhandle. Because the current range of the cactus moth is sympatric with rare or endangered butterflies, such as the Schaus swallowtail (*Papilio aristodemus ponceanus*), Florida leaf-wing (*Anaea floridalis*) and Bartram's hairstreak (*Strymon acis*), chemical control or inundative release of lepidopteran parasites are not considered viable control strategies. Therefore, we set out to explore the possibility that entomopathogenic nematodes could be used as part of a management program aimed at slowing the spread of the cactus moth. Accordingly, field collected cactus moth larvae were exposed to several different entomopathogenic nematode species (*Heterorhabditis* spp. and *Steinernema* spp.) at various densities under controlled laboratory conditions. These included nematodes indigenous to Florida as well as commercially available species. In all treatments, the cactus moth larvae were extremely susceptible (100% mortality). Since approximately 40% of cactus moth larvae drop from their feeding sites (inside the cactus pads) to the ground prior to pupation, we theorize that a modicum of control could be achieved using entomopathogenic nematodes. Following a survey of soil invertebrates associated with *Opuntia* communities (to explore the possibility of non-target effects), we will perform inundative field releases of entomopathogenic nematodes to gauge their impact on field populations of the cactus moth.

THE BIODIVERSITY AND BIOCHEMISTRY OF CRYOCONITE HOLES FROM ANTARCTIC GLACIERS. **Porazinska, D. L., [1] A. G. Fountain,[2] T. H. Nylen,[2] and D. H. Wall.[3]** [1]Fort Lauderdale Research and Education Center, University of Florida-IFAS, Fort Lauderdale, FL 33314, [2]Department of Geography and Geology, Portland State University, Portland, OR 97207, [3]Natural Resource Ecology Laboratory, Colorado State University, Fort Collins, CO 80523.

The landscape of the McMurdo Dry Valleys (MCM) of Antarctica is dominated by glaciers and glacier melt is the primary water source for life in soils, streams, and lakes. The glaciers, despite their cold and lifeless appearance, offer functioning habitats for life. The major objective of this study was to examine biogeochemical characteristics of mini-ecosystems called cryoconite holes and to determine linkages to other components (soils, streams, and lakes) of the dry valley landscape. We examined cryoconite holes from five glaciers spanning the length of Taylor Valley, one of many valleys in the MCM. Cryoconite biotic communities were composed of the same species observed in streams and lakes namely cyanobacteria (*Chlorococcus*, *Chroococcus*, *Crinalium*, *Oscillatoria*, *Nostoc*, and *Sprirulina*), rotifers (*Philodina gregaria* and *Cephalodella catellina*), tardigrades (*Acutuncus antarcticus* and *Hypsibius* spp.) and ciliates. Nematodes, the most common invertebrates in the Dry Valleys, were not found in any of the cryoconite holes. Biotic communities did not reflect the composition of the immediately surrounding environments suggesting the effects of aeolian mixing and transport of sediments and biota across the valley. Gradients of chemistry and biotic abundance in cryoconite holes reflected the position of each glacier in the valley. Nitrogen and organic carbon concentration patterns across glaciers potentially resulted from biological activities in cryoconite holes.

EFFECT OF EXTRINSIC FACTORS ON THE BEHAVIOUR OF STEINERNEMATID AND HETERORHABDITID INFECTIVE JUVENILES. **Powell, J. R.,[1] M. Lin,[2] and J. M. Webster.[1]** [1]Department of Biological Sciences, Simon Fraser University, Vancouver, BC V5A 1S6, CANADA, [2]Department of Plant Protection, Nanjing Agricultural University, Nanjing 210095, CHINA

The infective juvenile (IJ) stage of *Steinernema* and *Heterorhabditis* is subjected to a variety of extrinsic factors that affect its dispersal and infectivity. In this talk, we present evidence from two different studies that IJ behaviour is affected by factors associated with the respective bacterial symbiont, the insect host, and crop plants. Two different plant species (*Lycopersicon esculentum* and *Triticum aestivum*) differed in their attractiveness to IJs of *Heterorhabditis bacteriophora*.

Movement of IJs on agar plates increased when both a *Galleria mellonella* larva and *T. aestivum* were present, but *T. aestivum* presence resulted in reduced IJ-induced mortality of *G. mellonella*. Activity of *Steinernema carpocapsae* IJs (defined as the percentage of IJs actively moving) was greater following incubation in a cell-free (CF) filtrate of the symbiotic bacterium, *Xenorhabdus nematophila*, than following incubation in bacterial growth medium or distilled water. However, incubation in CF filtrate negatively affected *S. carpocapsae* dispersal in sand. The link between increased activity and reduced dispersal is not known but it is apparent that interacting factors play a significant role in the overall search for a host insect.

INCORPORATING MOLECULAR DIAGNOSTICS IN A REGIONAL POTATO PEST SURVEY. **Powers, T. O., and P. G. Mullin.** Department of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722.

The detection of a single specimen of *Meloidogyne chitwoodi* in a potato can result in the quarantine of a shipment of potatoes for export. Several countries require phytosanitary certification indicating that field soils are free of regulated nematodes before accepting foreign potatoes. In 2002, we conducted a USDA/APHIS sponsored survey of potato field soils in the central plains of the U.S., extending from North Dakota to Texas. Over 1,600 samples were analyzed. Approximately 5% of the samples contained *Meloidogyne* species, often found in barely detectable levels. A single *Meloidogyne* juvenile was found in twenty-five samples. Representatives of all *Meloidogyne* species recovered were analyzed by PCR/RFLP and in many cases, DNA sequencing. There were no findings of *M. chitwoodi* or any other nematode of regulatory concern. *M. incognita* was the most commonly encountered root-knot species; others include *M. arenaria*, *M. graminis*, and *M. hapla* (two distinct genotypes). We also observed juveniles of *Cactodera*, a cyst nematode of no known economic significance, in high frequency in the southern production region. The occurrence of *M. graminis* and *Cactodera* juveniles in these soils illustrates the need for the ability to distinguish among agronomic pests and those existing on native vegetation.

DEVELOPMENT OF A RAPID DIAGNOSTIC TEST FOR DETECTION OF THE STUBBY ROOT NEMATODE, *PARATRICHODORUS ALLIUS*. **Riga, E.,[1] K. Eastwell, [1] R. Larsen, [2] and G. Vandemark. [2]** [1]Washington State University, IAREC, Prosser, WA; [2]USDA-ARS, Vegetable and Forage Research Unit, Prosser, WA 99350.

Paratrichodorus allius, the stubby root nematode, which transmits Tobacco rattle virus (TRV), is becoming of increasing importance to the potato industry in the Pacific Northwest. The current non-qualitative method of evaluating a field for the presence of *P. allius* requires several days while the detection of viruliferous *P. allius* requires at least 2 months. Technology for rapidly identifying *P. allius* and viruliferous *P. allius* has not previously been utilized. Using sequence data obtained from the internal transcribed spacer (ITS) region *of Trichodorus primitivus*, a series of DNA primers were designed in attempt to specifically detect *P. allius* by PCR. A 600 bp DNA amplification product was obtained using the following primer pair: 5′- CCCGTCGCTACTACCGAT- 3′ , and 5′- TTCACTCGCCGTTACTAAG- 3′ from all total nucleic acid preparations of *P. allius.* Sequence data obtained from the 600 bp fragment is being used in the development of a sequence-characterized amplified region (SCAR) to increase specificity to *P. allius.* Work is currently in progress to refine an RT-PCR system that will detect the presence of TRV in viruliferous *P. allius.* Results from these diagnostic tools will be a great benefit to growers because population numbers of both *P. allius* and viruliferous *P. allius* per field can be obtained within 48 hours. With this information, growers will be able to make decisions by early fall with respect to necessary field preparations.

EVALUATION OF NEMATICIDES AGAINST ROOT-KNOT NEMATODES AND STUBBY ROOT NEMATODES. **Riga, E., and J. Wilson.** Washington State University, IAREC, Prosser, WA.

Nematicides were evaluated for control of the Columbia Root-Knot nematode, *Meloidogyne chitwoodi,* and the stubby root nematode, *Paratrichodorus allius* on "Russet Burbank" potatoes, *Solanum tuberosum,* in a loamy sand field. Field plots were 3 rows wide (13.4 cm row spacing), 7.6 m long, and arranged in a randomized complete block design with 5 replications. Treatments included fosthiazate 900 EC, at 4.86 and 6.48 Kg a.i./ha and 1,3-Dichloropene (1,3-D) at 187 l/ha. Aldicarb 15G at 3.35 Kg a.i./ha served as the standard nematicide check and untreated plots served as controls. 1.3-D was applied as a broadcast by tractor-drawn chisels 45.7 cm deep, spaced 45.7 cm wide apart and packed immediately with a cultipacker 3-4 weeks before potato planting. Fosthiazate was applied as a broadcast spray with a CO_2 pressurized backpack sprayer, rototilled 15 cm deep ands sealed with a cultipacker just before planting. Results showed that both 1,3-D at 187 l/ha, and fosthiazate at 4.86 and 6.48 Kg a.i./ha provided good control by protecting the potato tubers from the nematodes. In addition, both fosthiazate and 1.3-D increased potato yield in comparison to the untreated control. However, aldicarb at 3.35 kg a.i./ha did not protect the potatoes tubers from the nematodes.

NEMATODE SURVIVAL MASKS EXPRESSION OF RESISTANCE OF COTTON TO *ROTYLENCHULUS RENIFORMIS* IN FIELD EXPERIMENTS IN FOUR STATES. **Robinson, A. F.,[1] R. Akridge,[2] J. M. Bradford,[3] W. S. Gazaway,[2] E. C. McGawley,[4] and L. D. Young.[5]** [1]USDA-ARS, College Station, TX 77845, [2]Auburn University, Auburn, AL 36849, [3]USDA-ARS, Weslaco, TX 78596, [4]Louisiana State University, Baton Rouge, LA 70803, [5]USDA-ARS, Stoneville, MS 38776.

Rotylenchulus reniformis is considered the major nematode problem of Upland cotton (*Gossypium hirsutum*) in Alabama, Mississippi, Louisiana, and the Texas Lower Rio Grande Valley. Because no known *G. hirsutum* genotypes are resistant to *R. reniformis*, several breeding projects are underway to introgress resistance into *G. hirsutum* from other species. Until recently, the level of resistance in such accessions has been measured only in pot experiments. Surprisingly, the first field experiment, conducted at a South Texas site in 2001 showed resistance to be almost entirely suppressed and data from supplemental micro plot and growth chamber experiments indicated some soil factor other than nematode origin was responsible. Therefore, in 2002, the hypothesis that resistance suppression was site-independent was tested by planting nine resistant accessions of *G. barbadense*, *G. arboreum*, *G. herbaceum*, and *G. longicalyx* in parallel experiments in infested fields in Alabama, Mississippi, Louisiana, and Texas. Root and nematode population densities in soil were measured at harvest at 15-cm increments from the soil surface to 105 cm deep. Resistant accessions did not suppress nematode population in the soil adequately at any site. However, population densities at harvest in soil samples from fallow areas were 29-44% of those under the susceptible control, and when nematode densities under fallow were subtracted from those under resistant accessions, the relative levels of population suppression were similar to those in pot and micro plot experiments, indicating resistance was expressed in the field but masked by survival.

RELATIONSHIP OF THE NEMATODE HYPERPARASITE *PASTEURIA* SPP. TO *MELOIDOGYNE GRAMINIS* AND *TYLENCHORHYNCHUS* SPP. IN GOLF GREENS. **Rungrassamee, W., and R. L. Wick**. Department of Microbiology, University of Massachusetts, Amherst, MA 01003.

Fenamiphos, the only nematicide registered for nematodes in golf greens, will no longer be available for use after May 2005. Many "softer" alternatives to fenamiphos are available commercially but have not been efficacious in our trials. We have noticed that high populations of nematodes in putting greens often decline after several years without nematicide treatment. *Pasteuria* spp. are known to parasitize many species of nematodes and may play a role in suppression of nematode populations. For these reasons, we began an investigation of *Pasteuria* as a natural suppressant of nematodes in golf greens. Four golf greens with *Pasteuria*-infested root-knot nematode (*Meloidogyne graminis*) and three greens with stunt nematode (*Tylenchorhynchus* spp.) were evaluated for nematode populations and percent of *Pasteuria* infestation. To ensure the estimation of nematode population and percent *Pasteuria* infestation within 95% C.I., the number of soil cores necessary was determined. Root-knot nematodes were found to be highly aggregated, 70 to 100 soil cores were necessary to estimate the population. However, only 16 to 43 soil cores were necessary to estimate the percent of root-knot nematode infested with *Pasteuria*. Distributions of stunt populations were also aggregated but less than root-knot. Nine to 15 soil cores were necessary to estimate the stunt population and 10 to 27 soil cores were necessary to estimate the percent of nematodes infested with *Pasteuria*. The relationship of *Pasteuria* to root-knot and stunt was found to be inversely correlated with R^2 values equal to 0.75 and 0.44 respectively. Further studies to monitor population dynamics of *Pasteuria* and plant parasitic nematodes in golf greens are underway.

COMPUTERIZED KEY TO THE GENUS *BURSAPHELENCHUS* FUCHS. **Ryss, A.,[1] P. Vieira,[2] M. Mota,[2] and O. Kulinich.[3]** [1]Zoological Institute RAS, St. Petersburg, 199034 Russia, [2]Universidade de évora, 7000 évora, Portugal, [3]Institute of Parasitology, Leninsky prospect, 33, Moscow 117071, Russia.

Polytomous multyentry computerised key to the genus *Bursaphelenchus* was constructed using Bikey-Pickey system of Lobanov & Dianov. Key includes 70 species (targets) which are all the valid species of the genus for the world fauna, and 35 characters. Characters were selected from the taxonomic revisions and reviews, as well as from the taxonomic diagnosae of species descriptions. All taxonomic descriptions were analysed and the available collection slides were studied. Identification may be started from any character, but the key system, according the algorithm of Dallwitz, proposes at each identification step the order of characters depending of their diagnostic value (i.e. capability to reach the identification for the minimum number of steps). The most efficient characters at the first step of identification and their calculated weights (the start value is 1) are: male spicular tip (3.3); type of spicule structure (2.8); male spicular condylus shape (2.5); number of pairs of male postanal papillae (2.3); male bursa shape (2.1); ratio of male spicule length to its width (2); capitulum depression index (2); male spicular rostrum shape (2); female tail tip shape (1.9). Index of key perfection–9.56 (for the maximum value of 10); average length of identification path–3.85; minimum and maximum length of identification path–3.49 & 32.62, correspondingly; average of the character states in a character–4.56. Automatically produced by the Pickey system the traditional (monoentry) key has 50 nodes; average of alternative character states in a node–2.76, and 89 final targets of the key.

THE EFFECTS OF A BACTERIAL BIOLOGICAL CONTROL AGENT ON TAXIS OF *CLARKUS PAPILLATUS* IN A LABORTORY *BIOASSAY*. **Salinas, K. A., S. L. Edenborn, and J. B. Kotcon.** Division of Plant and Soil Sciences, West Virginia University, Morgantown, WV 26506.

A bacterial biological control agent was examined for non-target effects on a predatory nematode *in vitro*. The effect of the bacterium *Bacillus subtilis* GBO3 (Kodiak biological fungicide, Gustafson, McKinney, TX) on repulsion of the

predatory nematode *Clarkus papillatus* was assayed in 6 cm petri dishes containing Winogradsky Salts and 1% agar. *B. subtilis* GBO3 and a *Bacillus* sp. isolated from soil (isolate KB-1) were grown to log phase in nutrient broth cultures. Bacterial cells were suspended in Winogradsky Salts solution. Three solutions were used for the treatments: Winogradsky Salts control and each bacterium in these salts. Combinations of each bacterial solution, and the control solution, were developed in pairs alone or together for a total of six treatments. A treatment consisted of one pair of solutions being applied per plate. Each dish was sectioned into thirds and the solutions were applied to the first and third sections, while six *C. papillatus* were applied to the center section of each plate. The number of nematodes counted in each third section was recorded. The predators did not show preference for either bacterial solution or the control solution. Neither bacterial solution exhibited nematicidal properties on the nematodes. This experiment has revealed that *B. subtilis* GBO3 does not repel *C. papillatus*.

SELECTION OF SOYBEAN LINES POSSESSING SOYBEAN CYST NEMATODE RESISTANCE, SUDDEN DEATH SYNDROME RESISTANCE AND YIELD. **Schmidt, M. E., J. P. Bond, J. H. Klein, and R. E. Whelan.** Department of Plant, Soil and General Agriculture, Southern Illinois University, Carbondale, IL 62901.

Soybean cyst nematode (SCN) and soybean sudden death syndrome (SDS) are both major production constraints in the North Central Region. Ninety-six F_6 derived random inbred lines from the cross Asgrow 5403 × Cordell were greenhouse evaluated for SCN race 3 and race 5 resistance, and field evaluated for SDS resistance and yield. Selection of lines possessing SCN resistance did not insure selection for lines possessing SDS resistance. In a field environment with SCN at a level above threshold (500 eggs/100cc soil), and where SDS was severe selection for lines possessing SDS resistance alone proved more beneficial than selection of lines possessing SCN resistance alone when selection of lines possessing both traits is desired. In the same field environment, selection for yield alone or SDS resistance alone was equally effective for extraction of lines possessing both traits. Regression analysis indicated that SDS accounted for 73% of the variation in yield. The yield of SCN or SDS resistant lines did not differ from their susceptible counterparts when tested in a field environment exhibiting a low level of SDS and SCN. Hence, under these conditions selection for yield alone resulted in the culling of lines resistant to both SCN and SDS. Yield was not correlated to SCN resistance in any environment. High yielding lines possessing resistance to SCN and SDS were identified.

PRUNING COFFEE SHOOTS TO MAXIMIZE YIELDS FROM *MELOIDOGYNE KONAENSIS* STRESSED TREES. **Schmitt, D. P., M. Serracin, and B. S. Sipes**. Plant and Environmental Protection Sciences, University of Hawaii, Honolulu, HI 96821.

Shoot removal ("stumping") of coffee trees at regular intervals is a standard coffee production practice in Kona, HI. Shoot regrowth after stumping in *M. konaensis* infested soil is frequently poor. In heavily infested fields, the tree dies. The objective of this research was to determine the optimal vertical shoot management of nematode-infected coffee to maximize tree vigor and survival. Four-year-old coffee trees, half infected with *M. konaensis* and half nematode-free, at the Kona Experiment Station were pruned by removing the shoot above 1.7-m (T), at 0.6-m and allowing one vertical shoot to grow (S1), or at 0.6-m and allowing four vertical shoots to grow (S4). Fruit (cherry) were harvested after 2-years of regrowth. Average cherry yields (kg per tree) by treatment were: *Coffea arabica* 'catuai', S1–4.6, S4– 6.8, T–5.9; *C. arabica* 'typica' scions grafted onto *C. dewevrei* rootstocks, S1–3.2, S4–8.6, T–9.9; typica planted as 6-month-old seedlings, S1–3.5, S4–6.0, T–8.1; and typica planted as 12-month-old seedlings, S1–3.4, S4–7.0, T–9.6. The nematode generally had similar detrimental affects on yields in each pruning treatment within tree treatments, thus only yield responses to tree treatment are presented. Mean cherry yields (kg per tree) as affected by the nematode (indicated by + or − after the abbreviated tree treatment) were: catuai(−)- 5.6, catuai(+)–5.9, grafted(−)–10.6, grafted(+)–7.2, 6-month-old seedlings(−)–6.7, 6-month-old typica seedlings(+)–4.9, 12-month-old typica seedlings(−)–7.3, and 12-month-old typica seedlings(+)–6.0. In *M. konaensis* infested soil, scions growing on resistant rootstocks and pruned to 1.7-m or to 0.6-m retaining four vertical shoots produced trees with acceptable yield potential.

GALLING RESPONSE OF SELECTED COFFEE GERMPLASM TO *MELOIDOGYNE KONAENSIS*. **Schmitt, D. P., M. Serracin, and B. S. Sipes**. Plant and Environmental Protection Sciences, University of Hawaii, Honolulu, HI 968212.

Coffee decline, a disease affecting 85% of the coffee, *Coffea arabica*, L. in Kona, Hawaii, is caused by *Meloidogyne konaensis*. The best control of this nematode is with resistant rootstocks.. *Coffea liberica* W. Bullex Hiern var. *dewevrei* is the primary rootstock now being used in commercial plantings on the island of Hawaii. The objective of this project was to determine the galling reactions of some 60 coffee cultivars and selections in response to infection by *M. konaensis*. Seeds were collected from Hawaii, Brazil, Guatemala, Costa Rica, and Panama. They were germinated and the seedlings transplanted into 15-cm diameter clay pots filled with a heat sterilized Andisol soil. When the plants had 6-8 true leaves and were about 15-20 cm in height, they were inoculated with 10,000 eggs and juveniles of *M. konaensis*. Plants were evaluated for galling 6-8 months later, depending on the run of the experiment. The susceptible standard was a land race 'Guatemalan' of *Coffea arabica* cv. Typica. Tomato *Lycopersicon esculentum* Mill. var. *esculentum* cv. 'Pixie' was used

as an indicator of inoculum viability. Most *C. arabica* accessions evaluated were heavily galled with galling ratings ranging from 4-5 on a 0-5 scale (0 = no galling, 5 = 75-100% of the root system was galled). The *C. arabica* cultivars Columnaris, Caturra, and Eeritrean Moca exhibited moderate galling with ratings between 2 and 3. The reactions of two selections of *C. canephora* Pierre ex A. Froehner evaluated varied; with one selection being heavily galled and the second one having few galls. Galling was light on *C. liberica*. Since the least amount of galling occurred on selections of *C. liberica,* cultivars of this species are the best candidates to test as rootstocks for graft compatibility with scions having the most desirable taste qualities.

RHABDITIDAE: SYSTEMATICS AND IDENTIFICATION. **Scholze, V. S., A. W. G. van der Wurff, and T. Bongers**. Laboratory of Nematology, Wageningen University, Binnenhaven 5, 6709 PD Wageningen, The Netherlands.

Rhabditids occur in almost every niche with a high microbial activity; as a group they are cosmopolitan and vary in life strategy from free-living bacterial feeders to animal parasites. Interest in this family increased especially with the introduction of *Caenorhabditis elegans* (Maupas, 1899) Dougherty, 1953 as a model organism. To assess the functioning of soils, they are considered important indicators of enrichment as they show a short live cycle and form dauerlarvae as the microbial activity decreases. The insect associated rhabditids show potential as biocontrol agents. Since Dujardin (1845) established the first genus Rhabditis, about 250 species have been added. Because of the scattered information in literature and the absence of a handbook, rhabditids are difficult to identify. Presented here is a compendium which includes identification keys to all valid species, descriptions of all known species in terms of morphology as well as ecology, a glossary, a broad introduction on Rhabditidae, a review on nomenclature including synonyms, present status and were applicable: contrasting opinions. Furthermore a data matrix is added containing most important character states. The keys differ from traditional approaches owing to relevant drawings of species in tables which enables easy comparison; emphasis on clear morphological characters in stead of highly variable ratio's; possibility to identify females and/or males separately.

A COMPARISON OF BENEFICIAL TRAITS AMONG STRAINS OF *STEINERNEMA CARPOCAPSAE*. **Shapiro-Ilan, D. I.,[1] R. J. Stuart,[2] and C. W. McCoy[2]**. [1]USDA-ARS, Southeastern Fruit and Tree Nut Research Lab, Byron, GA 31008, [2]University of Florida, Citrus Research and Education Center, Lake Alfred, FL 33850.

Our objective was to compare beneficial traits among strains of *Steinernema carpocapsae* to identify or develop a superior biocontrol candidate for suppression of the pecan weevil, *Curculio caryae*. Virulence, environmental tolerance to heat and desiccation, and reproductive capacity were compared among eight strains. Fitness (*in vitro* growth) of the symbiotic bacteria, *Xenorhabdus nematophila* isolated from six of the nematode strains, was also compared. Significant differences were detected among nematode and bacteria strains for each trait. All nematode strains were more virulent to *C. caryae* adults than larvae. No single *S. carpocapsae* strain was superior for all beneficial traits measured. Based on a novel beneficial trait ranking system, Breton, DD136, Italian, and Kapow strains were graded inferior to other strains. Agriotos, All, and Sal strains were superior when desiccation was a factor. When desiccation tolerance was removed as a factor, the Mexican strain also tended to fall into the superior rankings.

A BIOGEOGRAPHICAL NOTE ON MONONCHIDS (NEMATODA: DORYLAIMIDA) OF JAPAN. **Shishida, T.,[1] and Y. Shishida.[2]** [1]Lab. of Forest Zoology, University of Tokyo, Yayoi 1-1-1, Bunkyo-ku, Tokyo, 113-0032, [2]Section of Phytopathol. And Entomol., Gunma ARC, Egimachi 1251, Gunma 371-0002, Japan.

Soil inhabiting nematodes were recovered mainly from the forest soil in Okinawa Island and predatory nematodes (Mononchoidea) were studied for the first time. Populations of Mononchoidea sampled from 37 different sites, mainly from rhizosphere of woody plants, revealed a total of 19 species belonging to 7 genera: *Clarkus papillatus*, *Mylonchulus muradi*, *M. lacustris*, *M. mulveyi*, *M. brachyuris*, *M. sigmaturus*, *M.* sp.1, *M.* sp.2, *M*. sp.3, *M*. sp.4, *Coomansus parvus*, *Prionchulus punctatus*, *P. muscorum*, *Miconchus thornei*, *M. citri*, *M*. sp., *Mononchus aquaticus, Iotonchus risoceiae* and *Iotonchus* sp. near. *risoceiae*. Details and measurements for 3 known species (*Mylonchulus muradi*, *Miconchus thornei*, *M. citri*), representing the first records from Japan, are given. Four species belonging to the genus *Mylonchulus*, one species of the genus *Miconchus* and one species of the genus *Iotonchus* are considered to be undescribed species and are mentioned without being named or described, until more materials are to be found. Little is known about Mononchid fauna of Japan. Hitherto, only *Clarkus papillatus*, *Mononchus truncates*, *M. tunbrigdensis*, *M. aquaticus*, *Coomansus parvus*, *C. zschokkei*, *Prionchulus punctatus*, *P. muscorum*, *Mylonchulus polonicus*, *M. ubis*, *M. brachyuris* have been repoeted with or without description and/or morphometric data. In these few years, we surveyed nematode fauna, especially of forest solids, of Taiwan and southernmost and central part of Japan, and many Mononchids have been added to a nematode fauna of Japan. Up to now, some forty species (including undescribed species) of suborder Mononchina have been identified: they include genera *Mononchus*(3 spp.), *Clarkus*(3 spp.), *Coomansus*(3 spp.), *Prionchulus*(4 spp.), *Mylonchulus*(14 spp.), *Nullonchus*(1 sp.), *Cobbonchus*(1 sp.), *Anatonchus*(2 spp.), *Miconchus*(4 spp.) and *Iotonchus*(5 spp.).

SURVEY OF PLANT-PARASITIC NEMATODES ON GOLF COURSES IN QUÉBEC. **Simard, L.,[1] G. Bélair,[2] and J. Dionne.[3]** [1]Centre de recherche en horticulture, Université Laval, Québec, Canada G1K 7P4, [2]Agriculture and Agri-Food Canada, St-Jean-sur-Richelieu, Québec, Canada J3B 3E6, [3]Department of plant agriculture, University of Guelph, Guelph, Ontario, Canada N1G 4W1.

A survey of plant-parasitic nematodes associated with turfgrass was done on nineteen golf courses from different regions of Quebec. Soil samples were taken from greens and fairways from August to October 2002. Plant-parasitic nematode numbers were estimated by processing two sub-samples of 50g by the modified Baermann pan method. Soil samples were analyzed: pH, total nitrogen, phosphorus, potassium, sodium, calcium, iron, zinc, organic matter, and soil particles size distribution were determined for each sample. Twelve genera of plant-parasitic nematodes were observed. The most commonly encountered genera were *Pratylenchus, Criconemella, Paratylenchus, Tylenchorhynchus, Helicotylenchus*. These five genera of plant parasitic nematodes were present on 90% and 63% of the golf courses on fairways and greens, respectively.

PROBING THE ROLE OF HSP90 IN NEMATODE DEVELOPMENT. **Skantar, A., K. K. Agama, S. L. F. Meyer, and L. K. Carta.** USDA-ARS, Nematology Laboratory, Beltsville, MD 20705.

We have identified Hsp90 as a potential target for plant-parasitic nematode control. Hsp90 proteins are highly conserved molecular chaperones that ensure proper folding and activation of critical signaling proteins and hormone receptors during conditions of stress. In *Caenorhabditis elegans*, Hsp90 is an essential single copy gene (*daf-21*) and part of a chemosensory pathway that controls nematode development in response to environmental signals. Our objective was to gain insight into Hsp90 function in nematodes through the use of a specific Hsp90 inhibitor called geldanamycin (GA). Varying amounts of GA were incubated with heat shocked or non-heat shocked *C. elegans* eggs and with non-heat shocked eggs from three populations of *Heterodera glycines:* NL1-RH, TN-17 and TN-18. We discovered that GA interferes with egg hatch and juvenile motility in both *C. elegans* and *H. glycines*. These results are the first demonstration of GA inhibition in any species of nematode, validating GA as a molecular probe of Hsp90 function during development.

GENOMIC DIVERSITY OF THE BACTERIAL SYMBIONTS OF ENTOMOPATHOGENIC NEMATODES. **Smith, H. L,[1] J. B. Jones,[2] F. J. Louws,[3] and B. J. Adams.[1]** [1]Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620, [2]Department of Plant Pathology, University of Florida, Gainesville, FL 32611-0680, [3]North Carolina State University, Department of Plant Pathology, Raleigh, NC 27695-7616.

Entomopathogenic nematodes and their bacterial endosymbionts are important biological control agents against a broad range of soil inhabiting insect species. The bacterial endosymbionts have been screened and assayed for their insecticidal toxin properties, yet their evolutionary and taxonomic diversity is only beginning to emerge. For the present study, strains of *Xenorhabdus* and *Photorhabdus* bacteria were retrieved from 94 entomopathogenic nematode isolates and 16 additional bacterial cultures. Identification was based on morphological growth characteristics that were exhibited when cultured on Tergitol-7 agar supplemented with 0.004% triphenyltetrazolium chloride, dividing the bacterial isolates into 52 and 58 strains of *Xenorhabdus* and *Photorhabdus* respectively. Relationships among the strains were analyzed by generating genomic fingerprints based on the amplification of repetitive DNA (BOX element, repetitive extragenic palindromic [REP], and enterobacterial repetitive intergenic consensus [ERIC]) sequences distributed throughout the chromosome (rep-PCR). The rep-PCR products were analyzed by agarose gel electrophoresis, revealing strain-specific patterns. Analysis of the combined BOX, REP, and ERIC fingerprints showed the formation of 6 distinct clusters that are correlated with the species of nematode from which the bacteria were isolated. However, some strains that were isolated from *Steinernema glaseri* are dispersed paraphyletically throughout the dendrogram. Rep-PCR may provide an efficient and sensitive diagnostic tool for identifying and characterizing the bacterial endosymbionts of entomopathogenic nematodes.

IMPACT OF PHYSICAL, CHEMICAL, AND BIOLOGICAL DISTURBANCES ON NEMATODE COMMUNITY STRUCTURE. **Smith, J. J., and G. W. Bird.** Department of Entomology, Michigan State University, East Lansing, MI 48824.

Soil from a carrot ecosystem was used for evaluation of the impact of disturbance on nematode community structure. Three disturbances were tested under greenhouse conditions: 1) physical (simulated tillage), 2) chemical (acetic acid), and 3) biological (germination and growth of a carrot seedling). Soil for the research came from an organic farm, certified by the Organic Growers of Michigan for 18 years. Cores measuring 10.16 cm × 30 cm were removed from the site using a hydraulic soil corer and immediately placed in a non-disturbed manner in 10.16 cm diameter PVC pipes. Six replicate cores were exposed to each of the three disturbances and placed in a greenhouse for 21 days. Replicate undisturbed control (undisturbed core) and a completely disturbed control (core pushed through 0.25 in. screen) were also included. All treatments were hand weeded at emergence. At the end of the trial period, each core was split into 0-15 cm and 15-30 cm soil depths. Nematodes were extracted from each depth using 100 cm^3 of soil, using a centrifugal-flotation technique. Nematodes recovered were classified into seven trophic groups: bacterivores, fungivores, algavores, herbivores, carnivores,

omnivores, and unknowns. Irrespective of treatment, all groups had higher absolute population densities in the 0-15 cm depth, than at the 15-30 cm depth. Population densities of fungivores were significantly lower in the disturbed control and the physically disturbed treatment compared to the non-disturbed control. All trophic groups had similar population densities in the non-disturbed control, biological, and chemical disturbance treatments. Nematode community structure of this site was compared to an adjacent wooded area and a neighboring conventional farm and characterized based on its bacterial, fungal, and protozoan biomass.

NEMATODES IN OAK WOODLANDS FROM SOUTHERN ARIZONA'S SKY ISLANDS. **Stock, S. P., J. Buckner, L. Grossmiller, and P. Coyne**. Department of Plant Pathology, University of Arizona. Tucson, AZ 85721-0036.

Soil samples were collected from two mountain ranges (Santa Catalina and Santa Rita) in southeastern Arizona. Sampling at each mountain range contemplated the upper Sonoran lifezone (4500-7500 ft elevation), where several oak species (Emory oak, *Quercus emoryi;* silverleaf oak, *Q. hypoleucoides;* desert scrub oak, *Q. turbinella* and Arizona white oak, *Q. arizonica*), are the dominant vegetation type. Free-living and phytophagous nematodes were extracted from mineral soil and litter using a modified Seinhorst mist apparatus. Insect-pathogenic nematodes were extracted from soil samples using the modified insect baiting technique. GPS coordinates, soil parameters (pH, texture, and organic matter content), altitude, associated vegetation and soil invertebrates, were recorded for each site sampled. Soils varied from slightly acidic (pH 6.98) to slightly alkaline (pH 7.83) and were rich in organic matter content (mean 3.8%). Four trophic groups of nematodes were identified based on morphological features at the generic level: fungivores, bacterivores, phytophages and omnivore-predators. Bacterivores were the most dominant trophic group, followed by phytophages, fungivores, and predator-omnivores. Most common bacterivore families recovered were: Cephalobidae, Rhabditidae, Diplogasteridae, Plectidae, as well as the entomopathogenic Steinernematidae and Heterorhabditidae. Plant-parasitic nematodes were represented by Tylenchidae and Tylenchorhynchidae, among others. Most numerous fungivores were *Aphelenchus, Ditylenchus* and *Aphelenchoides*. Mylonchulidae, Mononchidae and Dorylaimidae were among the most abundant omnivore-predators. This is the first report of nematode communities in oak woodland soils in Arizona.

IDENTIFICATION AND PHYLOGENETIC RELATIONSHIPS WITHIN THE STEM NEMATODE *DITYLENCHUS DIPSACI* SPECIES COMPLEX (TYLENCHUDA: ANGUINIDA) AS INFERRED FROM ANALYSIS OF THE ITS-rDNA SEQUENCES. **Subbotin, S. A.,[1,2] M. Madani,[2] E. L. Krall,[3] D. Sturhan,[4] and M. Moens.[2]** [1]Institute of Parasitology of RAS, Leninskii prospect 33, Moscow, 117071, Russia, s.subbotin@clo.fgov.be, [2]Crop Protection Department, Agricultural Research Centre, Burg. Van Gansberghelaan 96, 9820 Merelbeke, Belgium, [3]Institute of Plant Protection, Estonian Agricultural University, 51014 Tartu, Estonia, [4]Institut für Nematologie und Wirbeltierkunde, Biologische Bundesanstalt, Toppheideweg 88, 48161 Münster, Germany.

The stem nematode *Ditylenchus dipsaci* has considerable economic importance as a parasite of agricultural and horticultural crops. It is distributed worldwide, infects over 500 plant species, and causes stunting and swelling of various plant organs. Internal transcribed spacer (ITS) fragments of rDNA from 30 populations of the *D. dipsaci* species complex were amplified and sequenced. Most of the populations collected from cultivated plants (red clover, onion, alfalfa, oat, sugar beet, corn, garlic, tobacco, strawberry) had very similar ITS sequences. Different sequences and unique RFLPs were obtained for populations from some wild plants and broad bean. Phylogenetic analysis revealed at least six distinct clades within the *D. dipsaci* species complex. Molecular data support suggestions by several authors that differences in karyotype could indicate that "forms" or biological races presently grouped under the nominal species *D. dipsaci,* such as the giant race from *Vicia faba* and populations from *Cirsium setosum*, *Plantago maritima*, *Falcaria vulgaris, Pilosella officinarum* or *Crepis praemorsa*, probably deserve species status. Using the energy minimization approach and comparative mutation analysis it was shown that the secondary structure of ditylenchid ITS2 was organized in three main domains emerging from a preserved central core.

RECONSTRUCTION OF THE STEM SPECIES PATTERN OF SECERNENTEA. **Sudhaus, W.** Arbeitsgruppe Evolutionsbiologie, Königin-Luise-Str. 1-3, 14195 Berlin, Germany.

The first radiations of nematodes ("Adenophorea") occurred in a marine enviroment. Several lineages became terrestrial. Most important among these was the monophyletic Secernentea with many thousands of species which coevolved with spermatophytes, insects, tetrapods and other organisms. The "Bauplan" of Secernentea represents plesiomorphic and apomorphic features inherited from a stem species that may have existed in the Silurian period. Some of these characters had already evolved in the preceding lineage of aquatic bacteria feeders and were preadaptive for a terrestrial mode of life. Other characters are novel adaptations to terrestrial life and ephemeral habitats that fluctuate in moisture and other factors. Setae, epidermal glands and the three caudal glands were lost. Lateral longitudinal cuticle differentiations assisted locomotion in contact with particles of sediment or soil. Pharyngeal tubes served in filtering bacteria; valve plates in a terminal bulb served to crush bacteria. A new H-shaped secretory-excretory system with lateral canals in the epidermis and a cuticularized median duct helped to cope with changing osmotic pressures. A key novelty was the formation of a special

non-feeding dauerlarva, that enabled resistance to adverse conditions for some time. The anterior sensilla became papilliform and became arranged in two circlets of 6+10 by a shift of the four cephalic sensilla anteriorly. Amphids were reduced to a small roundish pore and were shifted into the lip region. Three pairs of lateral sensilla evolved: deirids and postdeirids with their dendritic processes embedded in the cuticle, and phasmids with pores on the tail. In the male, precloacal unpaired supplementary structures were substituted by paired series of subventral pre- and postcloacal genital papillae. A spiral copula type was retained. The posterior testis was lost; the ovary became reflexed within the growth zone.

DEVELOPMENT AND USE OF RESISTANT PEPPERS FOR MANAGING ROOT-KNOT NEMATODES. **Thies, J.,[1] R. Fery,[1] and J. Mueller.[2]** [1]USDA-ARS, Charleston, SC, 29414; [2]Clemson University, Blackville, SC, 29817.

Root-knot nematodes (RKN) (*Meloidogyne* spp.) are major limiting factors to pepper (*Capsicum* spp.) production globally, and fumigation with methyl bromide (MeBr) is the primary control used by U.S. growers. Pepper production accounts for 12% of MeBr used for pre-plant treatments in the U.S. The proposed 2005 ban of MeBr in the U.S. and loss of other nematicides due to environmental concerns has focused major interest on host resistance to RKN as a viable management alternative to nematicides. Scientists at the U.S. Vegetable Laboratory, USDA, Charleston, S.C. developed two open–pollinated bell pepper cultivars with RKN resistance conferred by the *N* gene. These cultivars (Carolina Wonder and Charleston Belle) are the only RKN-resistant bell cultivars available in the U.S., and are being used extensively by commercial pepper breeders as sources of resistance for bell pepper hybrids. Carolina Cayenne, a well-adapted RKN-resistant cayenne-type pepper co-developed by USDA and Clemson University, has proven useful as a rotation crop; susceptible bell peppers grown after Carolina Cayenne exhibited reduced root-galling and greatly enhanced yields. The resistance in Carolina Cayenne is extremely high (equal to MeBr fumigation) and is controlled by the *N* gene and a recessive gene. Three RKN-resistant Scotch Bonnet-type cultigens (PA-353, PA-398, and PA-426) were recently released by USDA. Resistance of these cultigens is controlled by an allele of the *N* gene. The USDA breeding program is currently using PA-426 as the source of resistance for developing resistant habanero cultivars. Host plant resistance should provide an economical and environmentally compatible alternative to MeBr and other nematicides for managing RKN in commercial pepper plantings.

THE EFFICACY OF ALFALFA AS A ROTATION CROP FOR SIMULTANEOUS SUPPRESSION OF *MELOIDOGYNE INCOGNITA* AND NUTSEDGES. **Thomas, S. H.,[1] J. Schroeder,[1] J. M. Fuchs,[1] C. Fiore,[1] I. Ray,[2] and L. W. Murray.[3]** [1]Department of Entomology, Plant Pathology, and Weed Science, [2]Department of Agronomy and Horticulture, [3]University Statistics Center, New Mexico State University, Las Cruces, NM 88003.

The *Meloidogyne incognita*/yellow nutsedge (*Cyperus esculentus*)/purple nutsedge (*C. rotundus*) complex poses a serious management problem for chile pepper (*Capsicum annuum*) producers in New Mexico. This study compared the effects of *M. incognita*-resistant ('Magna 8') and susceptible ('Dona Ana') alfalfa cultivars at two seeding rates and cotton on pest populations during a three-year rotation and on subsequent chile yield. Nutsedge biomass and *M. incognita* J2 numbers from four 0.25 m quadrats were measured after each of five alfalfa harvests per year, and from cotton plots each September. During the fourth season, plots previously planted with cotton were fumigated with 61 l/ha 1,3-dichloropropene and all plots were later planted with chile pepper. J2 numbers were not consistently detectable from either alfalfa cultivar or seeding rate throughout the three-year rotation, but increased steadily in cotton. Nutsedge biomass decreased nearly 80% in all alfalfa plots over the course of the study, with the greatest biomass persisting in the low seeding rate of the susceptible cultivar 'Dona Ana.' Purple and yellow nutsedge biomass was 60 and 300 times greater, respectively, in cotton plots than in alfalfa plots prior to rotating to chile. Red chile yield from all plots previously cropped to alfalfa was greater in the absence of fumigation than in fumigated plots previously cropped to cotton. These results indicate that rotation to alfalfa may provide a suitable means of *M. incognita*, yellow nutsedge, and purple nutsedge suppression in a chile pepper production system.

PHYLOGENY OF *MELOIDOGYNE* SPP. BASED ON 18S R DNA SEQUENCES. **Tigano, M. S.,[1] R. G. Carneiro,[2] M. Garcia-Varela,[1] I. T. DeLey,[3] and B. J. Adams.[1]** [1]Entomology and Nematology Department, University of Florida, Gainesville, Fl 32611-0620, USA, [2]Embrapa Recursos Geneticos e Biotecnologia, C.P. 02372, 70849-970, Brasilia, DF, Brazil, [3]Department of Nematology, University of California, Riverside, CA 92521.

The identification of *Meloidogyne* spp. is rarely straightforward, and often taxonomists are faced with diagnosing populations that display aberrant isozyme patterns or have morphological features that do not fit published descriptions. To investigate patterns of variation among several unidentified species of *Meloidogyne* within an evolutionary context, we sampled 22 populations of *Meloidogyne* spp. from various plant hosts and different countries and reconstructed their phylogenetic relationships using 18S rDNA sequences. Nominal species sampled that were readily identified were: *M. arenaria* (2 populations), *M. incognita*, *M. javanica*, *M. paranaensis* (2 populations), *M. arabicida*, *M. morociensis*, *M. oryzae*, *M. hapla*, *M. exigua*, *M. mayaguensis* and *M. ethiopica.* In addition, nine unidentified populations that demonstrated atypical morphological characters and/or isozyme patterns were sequenced and included in the analysis. DNA

sequences from the sampled taxa were aligned to previously published sequences bringing the total number of ingroup taxa to 33. Interestingly, despite morphological and isozyme variation, nearly all of the unidentified samples form well-supported clades with identified taxa, and none of them exhibit molecular autapomorphies that would suggest they are evolving independently of their inclusive clade. That this might be an artifact of the conservative nature of 18S rDNA sequences is discussed in light of the taxonomic, morphological and molecular diversity represented by the taxa included in the analyses.

DNA FINGERPRINTING COMPARISONS OF ALABAMA POPULATIONS OF RENIFORM NEMATODE, *ROTYLENCHULUS RENIFORMIS.* **Tilahun, Y.,[1] A. Zipf,[1] D. Deng,[1] K. Soliman,[2] K. McLean,[1] and G. C. Sharma.[1]** [1]Department of Plant and Soil Science, Alabama A&M University, Normal, AL 35762, [2]Entomology and Plant Pathology, Auburn University, Auburn, AL 36049

Cotton, a major crop in the world market, is subject to many biotic stresses, such as nematodes, particularly *Rotylenchulus reniformis*, the reniform nematode. One of the methods toward combating reniform nematodes is the development of genetic resistance in the crop. However, there are no cultivars of Upland cotton available which are resistant to reniform nematodes. Prior to focusing on pest management strategies, it is important to assess molecular and physiological differences which may exist within the pest species, indicating a possible conflict with resistance strategies. This study focuses on the molecular diversity within this rapidly emerging pest species. Three populations of reniform nematodes were collected, one each from 3 distant Alabama counties. Reniform adults were extracted from soil and DNA was isolated, subjected to PCR, cloning, and sequencing. PCR utilized 20-base primers spanning a ribosomal RNA region – the ITS1 spacer region. Amplified product sizes from individual samples were approximately 700 bp. Sequences were initially aligned using ClustalW, a multiple sequence alignment program, and optimized manually, through the use of Jalview, a program at the EMBL website. Comparisons of aligned clonal sequences showed both inter- and intra-population differences. A phylogram of ITS1 sequences revealed 2 superclusters, indicating two distinct lineages. However, each supercluster could not be traced to a single population or region (i.e., isolates from the populations were represented in both clusters), suggesting sequence similarity across populations in Alabama.

REPRODUCTION OF *MELOIDOGYNE INCOGNITA* ON WINTER COVER CROPS USED IN COTTON PRODUCTION. **Timper, P., R. F. Davis, and P. G. Tillman**. USDA ARS, Crop Protection and Management Research Unit, P.O. Box 748, Tifton, GA 31793.

Traditionally, small grains such as wheat and rye are planted as a winter cover before cotton in Georgia. However, there is growing interest in legumes because they contribute nitrogen to the subsequent crop and because they provide habitat and a food source (nectar) for beneficial insects. The objectives of this study were to determine reproduction of *Meloidogyne incognita* on rye and various legume cover crops in the greenhouse, and determine the effect of these same cover crops on nematode populations in the field and yield of the following cotton crop. In the greenhouse experiment, pots were inoculated with 8,000 eggs and reproduction assessed after 8 weeks. Crimson clover, berseem clover, and hairy vetch were good hosts for *M. incognita*, producing over 90,000 and 125,000 eggs in the first and second trial, respectively. Rye cv. Wrens Abruzzi, vetch cv. Cahaba White, and red clover cv. Cherokee were relatively poor hosts for nematode reproduction. Rye and Cahaba vetch produced less than 10% of the eggs of hairy vetch whereas Cherokee red clover produced 14 to 25% of the eggs of hairy vetch. In the field experiment, the cover crops grew poorly because of cool temperatures after planting in the fall. Cotton was strip planted into the cover crops in the spring. In most cases, cover crop had very little effect on nematode damage to cotton. Only hairy vetch cv. AU Early Cover caused more galling (0-10 index) on cotton than occurred in fallow plots (3.5 vs 1.2). Cotton yields were also lower in AU Early hairy vetch (701 lbs lint/acre) than in fallow plots (933 lbs lint/acre).

MORPHO-ANATOMICAL NOTES ON POPULATIONS OF *PRATYLENCHUS PENETRANS* AND *P. CRENATUS* FROM NEW YORK STATE (USA). **Troccoli, A.,[1] G. S. Abawi,[2] J. Ludwig,[2] and F. Lamberti.[1]** [1]Istituto per la Protezione delle Piante – Sezione di Bari, CNR, 70126 Bari, Italy; [2]Dept. Of Plant Pathology, NYSAES, Cornell Univ., Geneva, NY 14456.

Selected samples examined from a previous nematological survey on several onion producing areas of the N.Y. State revealed the presence of several naturally occurring populations of root lesion nematodes (*Pratylenchus* spp.). Because of the extensive morphological variability exhibited by a few members of some populations, that made identification difficult, a careful morpho-comparative study was done using 35 morphological and morphometric characters obtained by light microscopy with camera lucida attachment. Specimens were previously extracted from roots and from soil, fixed, and processed to pure glycerin to be examined in permanent mounts. All specimens revealed to belong to the widespread species *Pratylenchus penetrans* and *P. crenatus*. *P. penetrans* was most prevalent, being recovered in 47% of root samples analyzed, whereas *P. crenatus* was found in one sample only (6%), in mixture with *P. penetrans.* No root lesion specimens were recovered from soil samples. Members of *P. penetrans* are known to be highly variable with respect to certain

morphological characters, such as the shape and length of tail, the shape of stylet knobs, the shape of spematheca, and the number of lines in the lateral fields, but no deviation from the normal 3-annule lip pattern was ever recorded. Data of the present study largely confirm previous findings, with the exception of a narrow range of specimens of two populations having 2 lip annules, which are reported and illustrated.

COMPARATIVE MORPHOLOGY AND MOLECULAR CHARACTERIZATION OF *PRATYLENCHUS MEDITERRANEUS* AND *P. THORNEI.* **Troccoli, A. and F. De Luca**. Istituto per la Protezione delle Piante – Sezione di Bari, CNR, 70126 Bari, Italy.

Pratylenchus mediterraneus Corbett and *P. thornei* Sher & Allen share evident morphologic similarity. Members of this genus are characterized by a morphological homogeneity that makes proper identification problematic. Taxonomic difficulties are even complicated by intraspecific morphological variability, and often by the occurrence of mixed populations. In order to better define the two species, a comparative study was conducted on three populations of *P. thornei* of different geographic origin and one topotype population of *P. mediterraneus*. The species were characterized both morphologically and molecularly. Specimens for morphometric studies were examined under light microscopy on permanent mounts, whereas for molecular studies the ITS containing region was amplified and compared by nucleotide sequencing and PCR-RFLP analysis. Data obtained by morphometric and molecular investigations are presented and briefly discussed.

RESPONSE OF SELECTED COTTON VARIETIES TO THE RENIFORM NEMATODE IN ALABAMA. **Usery Jr., S. R., [1] K. S. McLean,[1] C. H. Burmester,[2] E. van Santen,[2] and B. A. Meyer.[3]** [1]Department of Entomology and Plant Pathology, Auburn University, Auburn, AL 36849, [2]Department of Agronomy and Soils, Auburn, AL 36849, [3]Delta and Pine Land Co., Hartselle, AL.

Management of the reniform nematode (*Rotylenchulus reniformis*) in cotton production is currently limited to nematicides and crop rotation. However, cotton cultivars with tolerance to *R. reniformis* would be a great asset and management tool for producers. Selected cotton cultivars were screened for tolerance in the field and resistance in the greenhouse to *R. reniformis*. Transgenic cotton cultivars were evaluated with and without aldicarb in two producers fields naturally infested with *R. reniformis*. Field 1 was monocultured cotton, conventionally tilled, under irrigation, while field 2 was an annual corn cotton rotation, no-tilled, without irrigation. In field 1, Rf values varied from 0.83 for PM 1218 BG/RR to 3.17 for FM 989 RR, with no variety exhibiting an Rf value below 1.0 in both the aldicarb and disulfoton plots. Seed cotton yields ranged from 2532 to 3361 lbs/A for DPL 458 B/RR and SG 215 BG/RR, respectively. In field 2, following corn, Rf values varied from 22.4 for DPL 5415 RR to 66.2 ST 4892 BR. Seed cotton yields ranged from 2097 to 2726 lb/A for ST 4793 R and SG 521R, respectively. Greenhouse trials indicated that all of the commercial cultivars tested were susceptible to *R. reniformis*. Rf values ranged from 13.3 on SG 747 to 39.5 on Phytogen GA 161. All varieties tested were susceptible to *R. reniformis*. However, in the field trials variety tolerance may have been observed at the $P < 0.10$ level.

ARE SOME NEMATODES BUILT TO FLOAT? A FUNCTIONAL MORPHOLOGICAL STUDY OF THE EXTRACUTICULAR STRUCTURES OF THE BUNONEMATIDAE (NEMATODA). **Van Gansbeke, R.,[1] A. Govaerts,[1] J. Manhout,[2] M. Claeys,[1] R. Van Driessche,[1] W. Bert,[1] and G. Borgonie.[1]** [1]Section Nematology, Ghent University, K.L. Ledeganckstraat 35, B-9000 Ghent; [2]Department of Recent Invertebrates, Royal Belgian Institute of Natural Sciences, Vautierstraat 29, B-1000 Brussels.

In general, the phylum Nematoda has a very uniform morphology. The Bunonematidae however are a strongly deviant group, which makes them very interesting organisms to study. The Bunonematids are free-living, terrestrial nematodes who were described throughout the world, but never seem to occur dominantly. They have a very unique morphology: they only posses a dorso-ventral symmetry as a result of the present cuticular and extracuticular structures. On the left side, the cuticle has a rather simple structure. When counting the dorsal and ventral wing, six to seven longitudinal ridges are identified. The cuticle on the right side is covered by a complex pattern of cuticle rods, which form a network of meshes. Depending on species and genus, warts or tubercles of varying form are found within this network. Observations using Scanning- and Transmission-Electron Microscopy and observations during specifically motility experiments show that Bunonematids are able to survive for a long period of time on the surface of water. We postulate the hypothesis that the building plan of these nematodes could be an adaptation to floatation.

PARASITISM GENES OF *HETERODERA SCHACHTII*. **Vanholme, B.,[1] T. Tytgat,[2] and G. Gheysen.[1]** [1]Department of Molecular Biotechnology, Faculty of Agricultural and Applied Biological Sciences, Ghent University, Coupure links 653, B-9000 Ghent, Belgium. [2]Institute of Zoology, Faculty of Sciences, Ghent University, K.L. Ledeganckstraat 35, B-9000 Ghent, Belgium.

The remarkable ability of sedentary plant parasitic nematodes to transform differentiated plant cells into complex feeding structures intrigued scientists for decades. Thanks to their interest, a considerable amount of descriptive knowledge is now available. However, the underlying molecular and biochemical mechanisms of this intimate relationship have still not been

elucidated. It is generally accepted that secreted proteins, originating from the nematodes pharyngeal glands should be responsible for this transformation. Different molecular techniques were used to identify putative parasitism genes coding for such proteins, and significant progress has been made in this field over the past few years. In our lab, we combined two-dimensional gelelectrophoresis with micro-sequencing to identify proteins secreted by the sugar beet cyst nematode *Heterodera schachtii*. Although some proteins could be identified (endoglucanases and pectate lyase) most of the spots sequenced likely are pioneer proteins not present in the existing databases. In a second approach we focused on the transcriptome of the nematode. By using cDNA-AFLP we were able to compare gene expression from anterior with posterior dissected parts of different developmental stages. Genes exclusively expressed in anterior parts of the parasitic stages were considered as being candidate parasitism genes and were isolated for further characterization. Again, many of those genes have no homologues in the existing sequence database, confirming the protein results. This extensive set of novel sequences is remarkable and interesting. Unraveling their role in the parasitic process can be seen as a new challenge for the future.

LONG-TERM EFFICACY OF *POCHONIA CHLAMYDOSPORIA* AGAINST *MELOIDOGYNE JAVANICA* IN GREENHOUSE CROPS. **Viaene, N., V. Van Damme, A. Hoedekie, and M. Moens.** Department of Plant Protection, Agricultural Research Center, 9820 Merelbeke, Belgium.

Long-term efficacy of *Pochonia chlamydosporia*, a fungal egg parasite of root-knot nematodes, was tested in two cropping systems simulating commercial greenhouse practices. One consisted of three crop cycles of lettuce, the other of one tomato crop followed by two lettuce crops. In two tests, plants were grown in 10-liter pots in soil inoculated with 5,000 chlamydospores of *P. chlamydosporia* per cm^3 soil at the beginning of both cropping systems. Soil in the control treatments contained no chlamydospores. In the first test, 50 second-stage juveniles (J2) of *Meloidogyne javanica* per 100 cm^3 were applied to all pots, but this density appeared too high to be controlled by *P. chlamydosporia;* many lettuce plants died when replanted in this infested soil. Interestingly, in pots with surviving lettuce plants, significantly fewer J2 were found in soil treated with *P. chlamydosporia* than in untreated soil. In the second test, initial nematode infestation was reduced to 25 J2 per 100 cm^3, while *P. chlamydosporia* infestation was the same as in the first test. After the second crop in the lettuce cropping system, the nematode population was half of that in the control, but there was no significant effect of the fungus after 3 crop cycles. Application of *P. chlamydosporia* in the tomato – lettuce – lettuce cropping system resulted in 46% and 95% fewer egg masses per root system in the second and third crop cycle, respectively, compared to the control treatment. There was also a 60% reduction in number of J2 per 100 ml of soil after the second crop cycle. *P. chlamydosporia* was re-isolated from the soil during all crop cycles.

FIRST REPORT OF *BURSAPHELENCHU*S SP. FROM TURKEY, ASSOCIATED WITH *PINUS NIGRA*. **Vieira, P.,[1] S. Akbulut,[2] M. Mota,[1] and V. Valadas.[3]** [1]NemaLab-ICAM/Departamento de Biologia, Universidade de évora, 7000 évora, Portugal, [2]Abant Izzet Baysal University, Duzce Orman Fakultesi, Beciyorukler Kampusu, 81620, Duzce, Turkey, [3]Dept. Protecção de Plantas, EAN/INIA, Quinta do Marquês, 2780 Oeiras, Portugal.

Forestry plays a very important role in the Turkish economy. Most species are distributed along the shores, in particular the Mediterranean Sea. The total forestry area consists of over 20 million ha, 71.5% conifer trees, 18.8% broad leaved trees and 9.7% mixed forest, comprising 22 major species. Slightly half of this area is productive. Among conifers, 4 species of *Pinus* predominate: *P. brutia* (3,729,866 ha), *P. nigra* (2,527,685 ha), *P. sylvestris* (757,426 ha), and *P. pinea* (46,490 ha). Beside this, Turkey is a country that import larges quantities of pine timber from Asia (special from Russia), via the Black Sea, and also a long coast line facing the Mediterranean Sea. The recent detection of the pinewood nematode, *Bursaphelenchus xylophilus*, in Europe, has raised concerns about the possibility of the presence of this serious A1 pathogen in this areas. In 2003, a survey has been initiated, wood samples have been collected from declining *P. nigra* trees from several sites (Duzce, Istanbul, Ankara). Nematodes were extracted using a standard tray method, observed and identified with a compound microscope. A *Bursaphelenchus* species has been identified for the first time in Turkey. Identification using morphobiometric and DNA analysis (ITS-RFLP) is being undertaken.

PINEWOOD NEMATODE GENERAL INFORMATION DATABASE. **Vieira, P.,[1] J. D. Eisenback,[2] and M. Mota.[1]** [1]NemaLab-ICAM/Departamento de Biologia, Universidade de évora, 7000 évora, Portugal, [2]Department of PPWS, Virginia Tech, Blacksburg, VA 24061, USA.

With the development of computer technology, acess to information from different scientific domains has become very easy to obtain from various sources (web, librarys, etc.). However, it may be frequently difficult to obtain a compilation of relevant information about a specific subject, which may take some time to organize. For the last two years, a project of retrieving, collecting and updating a broad set of data related to the pinewood nematode, *Bursaphelenchus xylophilus*, which has recently been reported from Europe, has been developted. The first cd volume ("PWN-CD: Taxonomy of *Bursaphelenchus* species") contains the original descriptions of all *Bursaphelenchus* species, and is presently available. The cd presented here is the second volume of this project. The main objective was to develop a database with all the

relevant information (mainly papers) concerning *Bursaphelenchus xylophilus*. The cd covers a wide variety of information including text (research articles, legislation, bibliography), images (nematodes, insect vector, host), video clips (historic, field work, techniques). This cd should be able to reach a wide audience from different areas, and is useful for teaching, research, and for political decision-makers, who will have in one cd-rom a practical and updated source of information on this extremely important pest and pathogen.

INFLUENCE OF NEMATODES ON PEACH PRODUCTION IN SOUTHERN ILLINOIS. **Walters, S. A., J. B. Russell, B. H. Taylor, and J. P. Bond**. Department of Plant, Soil and General Agriculture, Southern Illinois University, Carbondale, IL 62901.

The influence of plant-parasitic nematodes on peach production in Illinois is poorly defined. A survey of peach orchards was conducted during 2000-2002 to determine the nematodes associated with mature peach trees (> 10 years old) in southern Illinois. In most orchards, soils are silty clay loams and a standard forage-type fescue is grown between tree rows with a bare strip within the drip-line of trees. Nematode population densities were also monitored throughout the growing season on eight different rootstocks ('Bailey', 'Chui Lum Tao', 'Guardian', 'Higama', 'Ishtara', 'Lovell', 'Rubira', and 'Stark's Redleaf') with yields collected from the 'Redhaven' scions. Eight genera of plant-parasitic nematodes, *Helicotylenchus*, *Meloidogyne*, *Mesocriconema*, *Paratylenchus*, *Pratylenchus*, *Trichodorus*, *Tylenchorhynchus*, and *Xiphinema*, were identified in peach orchards. Over the two growing seasons, populations of *Mesocriconema* and *Xiphinema* were found at the highest densities. Although *Meloidogyne* spp. were found at low densities in a limited number of samples, they have potential to cause excessive damage to peach crops. Rootstocks maintained similar nematode densities and no correlations were observed between nematode densities and 'Redhaven' peach yields.

NEMATODE COMMUNITY DYNAMICS FOLLOWING DECOMPOSITION OF SUNN HEMP (*CROTALARIA JUNCEA*) AMENDMENT IN LITTER BAGS. **Wang, K.-H ,[1] R. McSorley,[1] R. N. Gallaher,[2] and A. Marshall.[2]** 1Department of Entomology and Nematology, 2Department of Agronomy, University of Florida, Gainesville, FL 32611.

Effects of sunn hemp (*Crotalaria juncea*) hay decomposition on the dynamic of free-living nematodes involved in soil nutrient cycling were examined in plots planted with sweet corn (*Zea mays*). Sunn hemp shoot tissues (C:N = 19:1) equivalent to 200 kg N/ha were placed in 0.03 m2 woven glass fabric bags (0.7-mm2 pore size) and buried at 10-cm soil depth. Nematode communities within the sunn hemp bag (SHB), soil directly under the bag (SI), and soil from an adjacent corn plot without the bag (SO) were assayed on 0, 14, 28, 42, 56, and 70 days after burying bags. Population densities of fungivores and bacterivores increased tremendously in SHB at 14 days, followed by an increased in population of omnivores at 42 days. Higher population levels of bacterivores and fungivores were maintained in SHB compared to SI and SO through day 56 ($P \leq 0.05$), but numbers in SHB and SI were similar on day 70. In contrast, the SHB maintained higher population levels of omnivores than SI and SO through the termination of the experiment ($P \leq 0.05$). Numbers of predatory nematodes were low in all treatments, but were highest ($P \leq 0.05$) in SI on most sampling dates. Although nematode community richness, diversity, and maturity index were lower in SHB at the beginning of decomposition, their values gradually increased to levels similar to SI and SO. Decomposition of the sunn hemp hay within the bag was relatively rapid and produced abrupt changes in nematode community structure, but after 70 days, most measures of community structure had returned to values similar to surrounding soil.

GENETIC AND MOLECULAR CHARACTERIZATION OF HOST-PLANT RESISTANCE TO ROOT-KNOT NEMATODES AND FUSARIUM WILT IN COTTON. **Wang, C., and P. A. Roberts.** Department of Nematology, University of California, Riverside, CA 92521.

Root-knot nematode (*Meloidogyne incognita*) is a major pest of cotton (*Gossypium* spp.), both alone and as part of the root-knot-Fusarium wilt (*Fusarium oxyspofum* f.sp. v*asinfectum*, FOV) disease complex. Host plant resistance is the most economical, effective method to control nematode and wilt disease on cotton. Two cultivars differing in resistance to *M. incognita* and FOV were characterized phenotypically for use in genetic analysis; Acala NemX (*G. hirsutum*) has resistance to *M. incognita* and susceptibility to FOV, while Pima S-7 (*G. barbadense*) has resistance to FOV and susceptibility to *M. incognita*. F_1 plants showed resistance to *M. incognita* and an intermediate disease reaction to FOV, suggesting a gene dosage effect in response to FOV. The combined infection of root-knot nematode-Fusarium wilt was also characterized in the two genotypes; Acala NemX showed high resistance to the complex infection, indicating that resistance to root-knot nematode can block the infection of Fusarium wilt, whereas Pima S-7 expressed reduced resistance to Fusarium wilt in the presence of nematode infection. Genetic analysis of resistance to *M. incognita* and FOV is being made by phenotyping F_2, F_3, BC_1F_1 and BC_2F_2 progenies. Amplified fragment length polymorphism (AFLP) analysis yielded many polymorphisms between the two parents and these are being assessed via bulked segregant analysis (BSA) to determine their potential as molecular markers linked to the disease resistance traits. The identification of useful markers will allow wider and earlier selection of resistance in preferred cotton backgrounds based on marker-assisted selection.

PHYLOGENETIC ANALYSIS OF A NEW *PASTEURIA PENETRANS* STRAIN BY 16S rRNA GENE SEQUENCING. **Wang, Y.,[1] X. Gao,[2] and X. Deng.[1]** [1]Department of Plant Pathology, South China Agricultural University, Guangzhou, 510642 P. R.China; [2]Department of Crop Sciences, University of Illinois at Urbana and Champaign, AW-101 Tuner Hall 1102 South Goodwin Ave Urbana,IL 61801-4798.

*Pasteuria penetr*ans is a root-knot nematode-parasitic bacterium. Recently, we isolated a new strain PpTH from *Meloidogyne incognita* in Guangzhou, Guangdong, China. The Pasteuria DNA was extracted from young tomato roots infected by nematodes containing PpTH in its early developing stages, and a partial 16S ribosomal RNA gene was amplified using primers 39F and 654R. The PCR product was digested with *Bam*HI in order to verify the *P. penetrans*. A partial sequence of 560 bp was sequenced and a Blast analysis (NCBI) showed similarity of 97% to *P. penetrans* P100 (AF077672), 98% to *P. penetrans* Pp (AF375881), 92% to *Pasteuria* S-1 (AF254387), 93% to Pasteuria NA (AF134868) and 87% to P. ramosa (U34688). Phylogenetic analysis of these six strains showed that the three *Pasteuria penetrans* strains: PpTH, *P. penetrans* P100 and *P. penetrans* Pp constituted a separate clade with bootstrap support 97%, which excluded the other three *Pasteuria* strains. *Pasteuria* S-1 and *Pasteuria* NA belong to another individual clade with a lower bootstrap support (68%). *P. ramosa* was positioned alone at the exterior branch of the phylogenetic tree. Within the three *Pasteuria penetrans* strains, *P. penetrans* P100 and *P. penetrans* Pp constituted a sister-taxa with bootstrap support 99%, demonstrating they might be closer in genetic relationships than PpTH. The current sequence is still too short to draw a definite conclusion, and further efforts are ongoing to isolate the full length of the sequence of 16S rRNA of PpTH.

EVOLUTION OF A NOVEL GENE EXPRESSION PATTERN IN *CAENORHABDITIS ELEGANS.* **Wang, X., and H. M. Chamberlin**. Department of Molecular Genetics, Ohio State University, Columbus, OH 43210.

We are interested in the molecular, genetic, developmental and morphological changes that underlie characters that differ between species. To investigate these changes, we are studying differences in the excretory system in *Caenorhabditis* species. The *C. elegans* excretory system, which is proposed to mediate osmotic and ionic regulation, comprises four cell types: gland cell, excretory cell, duct cell and pore cell (1). Previous studies have shown that *C. elegans ovo*-related gene *lin-48* expresses in a small number of cells including the excretory duct cell. In the related species *C. briggsae*, the expression is conserved in all cells except the excretory duct. This *lin-48* expression difference affects excretory duct morphogenesis. In *C. briggsae*, as well as in *C. elegans lin-48(sa496)* mutants, the excretory duct is more anterior than in *C. elegans* wild type. This indicates that *C. elegans lin-48 (Ce-lin-48)* is involved in duct morphogenesis and positioning, but this gene function is absent in *C. briggsae* (2). We have made reporter transgenes composed of the *lin-48* regulatory sequences from *C. elegans* or *C. briggsae* driving expression of green fluorescent protein (GFP). Tests of these clones in each species showed that only *Ce-lin-48* is expressed in the excretory duct cell in *C. elegans* animals. These results indicate that there are differences in both *cis*-regulatory sequences and trans-acting proteins between the two species. By creating chimeric reporter transgenes including *C. elegans* and *C. briggsae* regulatory sequences, we have found that one difference between the two species is the presence of regulatory sequences in *Ce-lin-48* that respond to the bZip protein CES-2 (2). The lin-48 gene expression differences between *C. elegans* and *C. briggsae* could result from loss of excretory duct expression in the *C. briggsae* lineage or acquired expression in the *C. elegans* lineage. To distinguish between these possibilities, we have analyzed three additional *Caenorhabditis* species (*C. remanei*, *C. sp.* CB5161 and *C. sp.* PS1010). We found these species have a duct morphology similar to *C. briggsae* indicating the *C. elegans* morphology is unique to this species. For comparison to *C. elegans* and *C. briggsae*, we have isolated the *lin-48* gene from *C. remanei* and *C. sp.* CB5161. Alignment of the lin-48 regulatory sequences reveals that the sequences are more conserved among *C. briggsae*, *C. remanei* and *C. sp.* CB5161. Several conserved domains are absent from *C. elegans*, whereas the previously identified CES-2 binding sites are absent from the other species. Currently, we are creating *lin-48::gfp* reporter transgenes for each species to observe the gene expression patterns. Further experiments with these transgenes will allow us to test whether the differences between *C. elegans* and the other species result from a loss of repressor elements or gain of activator elements in the *C. elegans* gene.

NEMATICIDAL AND HERBICIDAL PROPERTIES OF A FURFURAL-DAZOMET EMULSIFIABLE FORMULATION. **Weaver, C. F., and R. Rodríguez-Kábana.** Department of Entomology and Plant Pathology, Auburn University, AL 36849.

A greenhouse experiment was conducted to assess the nematicidal properties of an emulsifiable concentrate [EC] of furfural containing 12.5% dazomet [furadaz] compared with those of two other EC's, one containing furfural alone, and the other with dazomet only. The EC's were applied to soil as 2% aqueous emulsions to deliver increasing rates of active ingredients ranging from [mg a.i./kg soil] 35-175 for furadaz, 30-150 for furfural, and 5-25 for dazomet. The soil used was infested with *Rotylenchulus reniformis*. Nematological analyses of soil three weeks after application of materials showed that all three formulations resulted in exponential decline in *R. reniformis* in response to increased dosages. Furadaz was the most effective formulation and when applied 100 mg a.i./kg soil, it resulted in total control of *R. reniformis*. Similar patterns were observed for microbivorous nematodes. A second greenhouse experiment was conducted to assess

the herbicidal properties of furadaz 87.5 EC applied in a 20% aqueous emulsion at doses ranging from 175-1,750 mg a.i./kg soil. The soil used was infested with crabgrass, pigweed and other weed species. Half of the pots in the experiment were covered with polyethylene immediately after treatment while the other half remained uncovered. Covers were removed after 12 days and weeds were counted on a weekly basis for one month. Populations of all weeds declined sharply in response to furadaz doses. Covering pots with polyethylene increased herbicidal activity by 40-60%. At the final count, total weed control was achieved 700 mg a.i./kg soil. Results showed that furfural + dazomet is an effective nematicide and that when applied at doses 4-6 times higher than those required for nematode control, the mixture has considerable herbicidal activity.

OCCURRENCE OF *MELOIDOGYNE* SPP. AND *PASTEURIA PENETRANS* IN SWITZERLAND AND THE POTENTIAL FOR INTEGRATED CONTROL. **Weibelzahl-Fulton, E.,[1] and J. Grunder.[2]** [1]Collaborator, OECD Co-operative Research Programme: Biological Research Management for Sustainable Agriculture. [2]Nematology Laboratory, Swiss Federal Research Station, Waedenswil, CH.

In the fall of 2002, a nematological survey of intensively cultivated agricultural soils was conducted in Switzerland. Most of the targeted sites revealed infestations of the northern root-knot nematode (RKN), *Meloidogyne hapla*. *Meloidogyne incognita, M. javanica,* and *M. arenaria* seemed to be limited to greenhouses and tunnels used to produce tomato, cucumber, eggplant and bell pepper. The quarantined species, *M. chitwoodii*, was not detected. At several sites, the RKN populations were managed well by the use of resistant-tolerant tomato and(or) cucumber root stock cultivars. However, *M. hapla* appeared to have resistance breaking capabilities, and built up to high numbers in the soil. Good nematode management also were observed at locations where nematophagous fungi, *Arthobotris superba* and *Hirsutella* sp., were applied, but need to be confirmed in designed experiments. At locations converted to soil-less culture techniques, nematodes were not a problem. In two locations, *Pasteuria penetrans,* a bacterial parasite of RKN, was found, isolated, and tested for its potential as a biocontrol agent. In comparison with a Florida and a Dutch isolate of *P. penetrans,* the Swiss strain was less pathogenic to most *Meloidogyne* sp. tested. The survey was concluded with individual recommendations for integrated nematode control.

TRAP CROPPING FOR MANAGEMENT OF ROOT-KNOT NEMATODE. **Westerdahl, B. B.,[1] J. D. Radewald,[2] J. Nunez, [3] E. P. Caswell-Chen, [1] and C. A. Anderson. [1]** [1]Department of Nematology, University of California, Davis, CA 95616; [2] Department of Nematology, University of California, Riverside, CA 92521; [3] University of California Cooperative Extension, Bakersfield, CA 93307.

Trap cropping is a nematode management technique that has been tested periodically since the late 1800's. A susceptible host is planted and larvae of a sedentary parasitic nematode such as root-knot are induced to enter and establish a feeding site. Once this has occurred, and the female begins to mature, she is unable to leave the root. The plants are then destroyed before egg laying by nematodes is initiated, trapping nematodes within the root. Field trials on carrots have been conducted using six different trap crops destroyed at various times after planting. Trap crop treatments destroyed either with tillage, or with Roundup herbicide, or with a combination of the two were compared to untreated (dry fallow) and 1, 3-dichloropropene treatments. In these trials, several treatments yielded marketable carrots or reductions in root-knot nematode (*Meloidogyne javanica*) populations at harvest that were similar to 1, 3-dichloropropene and significantly greater than the untreated ($P = 0.05$).

MORPHOLOGICAL OBSERVATION ON *LONGIDORUS CRASSUS* THORNE, 1974 (NEMATODA: *LONGIDORIDAE*) AND ITS INTRASPECIES VARIATION. **Ye, W., and R. T. Robbins.** Department of Plant Pathology, Nematology Laboratory, 2601 N. Young Ave., University of Arkansas, Fayetteville, AR, 72704.

Longidorus crassus is widely distributed in Arkansas (36 populations), Alabama, Iowa, Kansas, Nebraska, South Carolina, Wisconsin and Canada. A few males were found. Four juvenile stages were identified. Twenty_three Arkansas populations (3 or more specimens) were studied for variability using standard measurements, mean comparison, coefficent of variation, and hierarchical cluster analysis (HCA). Intraspecies variation was observed among different populations of this species. Most Arkansas populations have a shorter body than the lectotype. Populations Long_63, Long_80 and Long_88 closely resemble the lectotype. Populations Long_10 and Long_80 are different from each other and all other populations. The majority of morphometric characters of this species had a high degree of variability within and between populations. The means of many morphometric characters had significant difference between populations. HCA (Ward method) based on the female morphometric character means of the 23 Arkansas populations and the lectotype were used to examine the morphometric relationships and create dendrograms. Means used were body length, distance from vulva opening to anterior end, head width, odontostyle length, esophagus length, body width, tail length and tail width. Four major groups were distinguished. Long_10 (longest body and shortest odontostyle) was in alone, a second group (Long_63, Long_80, Long_88 and lectotype) had the second longest bodies, widest body widths, most posteriorly located guide ring,

and the longest esophagus. All the remaining populations are in two groups that had some differences compared to the lectotype. *Longidorus crassus* has a high degree of morphometric intraspecific variation, however no morphological differences such as head or tail shape were observed.

SOME OBSERVATIONS ON POPULATIONS OF *LONGIDORUS BREVIANNULATUS* AND *L. FRAGILIS*. **Ye, W., and R. T. Robbins**. Department of Plant Pathology, Nematology Laboratory, 2601 N. Young Ave., University of Arkansas, Fayetteville, AR, 72704.

In a taxonomic study of over 260 populations of *Longidorus*, a total of 27 populations of *L. breviannulatus* were identified, eight populations from Arkansas, two from California, two from Canada, three from Iowa, one from Illinois, one from Kansas, two from Nebraska, one from Maine, two from Michigan, two from New Jersey, one from New York, and two from Wisconsin. *Longidorus breviannulatus* were identified and are reported for the first time from Arkansas, California, Kansas, Maine, Michigan, Nebraska, and New York. A few males were found in one Nebraska (Long-160) and one New York (Long-39) population. One Arkansas population was from Turf grass and seven were from hardwood trees along streams. Twelve of the remaining populations were from corn, four were from turf, one was from sorghum, one from mint, and one host was not known. Seven populations of *L. fragilis* were identified from Arkansas and one from Indiana. Males were not found. Four juvenile stages were identified. This is the first report of finding this species since the original description of two specimens from Minnesota.

AN ONLINE DATABASE OF VIRTUAL NEMATODE VOUCHERS. **Yoder, M., I. Tandingan De Ley, and P. De Ley.** Department of Nematology, University of California, Riverside, CA 92521.

Video Capture and Editing (VCE) allows the construction of "virtual vouchers" of nematode specimens, each voucher consisting of multifocal video clips that are accessible via an overview image map embedded in a HTML document. We have started compiling a collection of these vouchers, including some with annotations labeling diagnostic structures and other morphological features. The resulting "virtual slide collection" is available on theWorld Wide Web at *http://faculty.ucr.edu/~pdeley/lab/labhome.html* and can be accessed using any internet browser with QuickTime (R) plug-in. Genera and species can be searched through an alphabetical index, through nested phylogenetic trees, through a nested classification based on the recently proposed system of De Ley & Blaxter, or via the search page of the university website. At present, the database contains virtual vouchers of 58 permanently preserved specimens belonging to 28 species. This includes vouchers of type material of nine species, vouchers of six cultured strains, as well as fully annotated virtual specimens of six species useful for teaching purposes. Other examples of VCE applications are also accessible online, including virtual vouchers of active or immobilized live nematodes belonging to 5 species of marine nematodes, as well as links to websites for related approaches, such as *http://nematol.unh.edu/* and *http://www.vce.be.tf/*.

THE COMPLEXITY OF IMPLEMENTING AMENDMENT-BASED MANAGEMENT SYSTEMS FOR PLANT-PARASITIC NEMATODE SUPPRESSION. **Zasada, I. A.,[1] and H. Ferris.[2]** [1]USDA, ARS Nematology Laboratory, Beltsville, MD 20705, and [2]Department of Nematology, University of California, Davis, CA 95616.

Organic amendments with potential for nematode management range from nematotoxic plants to manures to industrial by-products. They have been applied to soil as mulches, green manures and extracts. Before an organic amendment can be considered a reliable and consistent nematode management tool, it requires the same rigor of testing and evaluation that has been applied in pesticide development. An essential starting point is the need to understand the mechanisms of nematode suppression associated with the material. Factors that are usually unknown include the chemical composition of the active components; lethal concentration values of the active components and/or the amendment material for specific target nematodes; impact of the material on soil physical and chemical properties; and the influence and impact on soil biotic factors. Experiences with the Brassicaceae, other plant-derived amendments and composted municipal waste demonstrate that organic amendments can be applied for consistent and reliable nematode management when the chemical and biological components of suppressiveness are understood.

DISTRIBUTION AND PATHOGENICITY OF BACTERIA CARRIED BY PINE WOOD NEMATODE IN CHINA. **Zhao, B., H. W., H. Sufen, and H Zhengming.** Department of Forest Entomology and Pathology, Nanjing Forestry University, Nanjing 210037, P. R. China.

Investigation on identification and pathogenicity of bacteria carried by pine wood nematode, *Bursaphelenchus xylophilus,* from both healthy and diseased black pine, *Pinus thunbergii*, and masson pine, *P. mssoniana,* were carried out in five main epidemic provinces in P. R. China. Results showed that no bacterium was found in healthy pines and bacteria were found on pine wood nematodes in all of the samples collected from trees died of pine wilt disease. Twenty-four strains of bacteria were isolated on the nematode from the samples and identified by combination of classical and ATB (automatic testing bacteriology) Expression methods. Bioassay showed that seventeen of the twenty-four identified strains were

phytotoxin producers. Eleven strains of the seventeen phytotoxin producers were in genus *Pseudomonas.* Field tests in which sterilizing measures as strict as possible were taken showed that three of the eight replicate trees inoculated with aseptic nematodes showed no symptom of the disease in an observation more than ten- months later after inoculation and no bacteria and fungi were found in the samples of the tree trees in the recovery. Death was found in seven over eight of the trees inoculated with unsterilized nematodes and in six over eight with both the aseptic nematodes and a strain of bacteria, a strong phytotoxin producer, in the bioassay by using four months old seedlings of black pine. A hypothesis was proposed that pine wilt disease is a complex, which was induced by both pine wood nematode and its carrying toxin-producing bacteria.