

## ABSTRACTS

### SOCIETY OF NEMATOLOGISTS 46<sup>th</sup> ANNUAL MEETING SAN DIEGO, CALIFORNIA July 28–August 1, 2007

IMPACT OF CERTAIN OIL-SEED CAKES AND POWDER IN COMPARISON WITH OXAMYL AND UREA ON *MELOIDOGYNE INCOGNITA* INFECTING EGGPLANT. **A. M. El-Sherif,<sup>1</sup> A. R. Refaei,<sup>1</sup> M. E. El-Nagar,<sup>2</sup> H. M. Salem.<sup>2</sup>** <sup>1</sup>Nematology Research unit, Zoology Department, Mansoura University, EL-Mansoura, Dakhlia, Egypt, <sup>2</sup>Plant Protection Institute, Agricultural Research Center, Giza, Egypt.

Eggplant, *Solanum melongena* L., is one of the economic solanaceae crops in Egyptian agriculture. Root-knot nematodes, *Meloidogyne incognita*, were recently recorded to be associated with eggplant plantations in three soil types surveyed within the cultivated areas of Dakahlia governorate. A pot experiment was conducted to evaluate the influence of certain oil-seed cakes (anise, fennel, sesame and plant dry seed powder of red pepper) as soil amendments in comparison with oxamyl and urea on controlling *M. incognita* infecting eggplant seedlings under greenhouse conditions. Twenty-four plastic pots 10-cm-diam. filled with steam-sterilized sandy-loam soil (v:v, 1:1) were planted with 1 eggplant seedling cv. Black long/pot at 60-days-old. One week after transplanting, each seedling was separately treated with anise, fennel, sesame and dry powder of red pepper seed, oxamyl or urea at the rate of 5 g/pot for each oil cake and powder, or 0.3 ml/pot or 0.01 g/pot for oxamyl or urea, respectively. Oil-cake or dry powder or oxamyl or urea was introduced to eggplant seedlings at the time of adding *M. incognita* eggs. Twenty-one seedlings were inoculated with 1,000 eggs each (N). Treatments were as follows: 1) N + anise cake, 2) N + fennel cake, 3) N + sesame cake, 4) N + red pepper powder, 5) N + oxamyl, 6) N + urea, 7) N alone and 8) plant free of N or any treatment. Each treatment was replicated three times. Pots were arranged in a randomized complete block design on a greenhouse bench maintained at  $27 \pm 5^\circ\text{C}$ . Plants received water as needed. After 45 days from nematode egg inoculation, plants were harvested. Data for lengths and fresh weights of shoot and root, and shoot dry weights as well as number of shoot leaves were recorded. Infected plant roots were examined for number of galls, developmental stages, females and egg masses after staining by lactic acid-fuchsin. All tested materials significantly improved growth of eggplant and suppressed number of galls, females and egg masses of *M. incognita* as compared to nematode alone. Moreover, anise oil-cake gave the best growth of total plant fresh and dry shoot weights percentage increase (160.8 and 64%) followed by sesame oil-cake (143.5 and 64%), respectively. However, the highest values of percentage reduction for number of galls (64.2%), females (58.5%) and egg masses (55.7%) on eggplant roots were achieved by sesame oil-cake application, followed by anise oil-cake treatment with values of 63.0, 59.6 and 51.1% for the same nematode criteria, respectively. Meanwhile, oxamyl or urea treatment showed remarkable reduction percentages for nematode paratemers tested.

EFFECT OF FUMIGATION THROUGH DRIP IRRIGATION ON *ROTYLENCHULUS RENIFORMIS* FOR DRIP TAPE SEPARATED BY TWO METERS. **T. A. Wheeler.** Texas Agricultural Experiment Station.

Tests were conducted to determine if control of reniform nematode could be obtained when Telone EC (1,3-D) was applied through the drip tape at 46 liters/ha. Row spacing was 1-m wide, and drip tape was placed every 2 m (every other row). Soil samples were taken before and after fumigation at 15, 30 and 61 cm in the bed and the dry furrow and at 15 and 30 cm over the drip tape. Reniform nematode population density at a depth of 30 and 61 cm increased by 200% in plots not fumigated and decreased by 51% in plots that were fumigated from 26 April to 19 May. Partial control was achieved over the drip tape and bed. At the 15 cm depth and over the drip tape and bed in nonfumigated and fumigated plots, reniform nematode population density increased by 192 and 294% from 26 April to 19 May. No control was achieved at the 15 cm depth. In the dry furrow at all depths, reniform nematode population density increased in the nonfumigated plots by 90% from 26 April to 19 May and decreased by 16% in the fumigated plots. Fumigation did not control the reniform nematode sufficiently, and cotton lint yield was not different between treatments. Yield was reduced by 898 kg lint/ha for LOG10 (reniform nematode on May 19/100 cc soil).

INFLUENCE OF PLANTING DATE AND WATER MANAGEMENT ON RENIFORM NEMATODE POPULATIONS IN COTTON. **S. R. Stetina, W. T. Pettigrew.** USDA ARS Crop Genetics and Production Research Unit, Stoneville, MS, USA.

Planting dates and water management practices were examined for potential impact on reniform nematode (*Rotylenchulus reniformis*) populations in cotton (*Gossypium hirsutum*). A field trial conducted in 2005 and 2006 in Stoneville, MS, examined the influence of early (April 1) or normal (May 1) planting dates and irrigated or dryland production on

early-season root infection and seasonal changes in the reniform nematode population in the soil. Treatment effects were evaluated on six cultivars in a modified split block design with six replications. Soil populations of reniform nematode were measured at planting and harvest each year, and root infection was assessed 3 to 5 weeks after planting. Cultivars did not differ, so data were averaged across them to examine planting date and water management effects. Average reniform nematode population sizes in irrigated and dryland cotton were equivalent, though a significant seasonal interaction was detected. In both years of the study, nematode populations increased from planting to harvest, but significantly more so in irrigated plots than in dryland plots. Planting date did not affect either soil reniform nematode populations or early-season root infection by reniform nematodes. These findings suggest that modifications to either planting dates or water management practices will not suppress reniform nematode populations in cotton.

**OCCURRENCE AND ECONOMIC IMPORTANCE OF PLANT-PARASITIC NEMATODES IN ORGANIC GROWN VEGETABLES IN GERMANY.** **J. Hallmann,<sup>1</sup> A. Frankenberg,<sup>2</sup> A. Paffrath.<sup>3</sup>** <sup>1</sup>Federal Biological Research Centre for Agriculture and Forestry, Toppeheideweg 88, 48161 Muenster, Germany, <sup>2</sup>Bioland Landesverband Nordrhein-Westfalen, Im Hagen 5, 59069 Hamm, Germany, <sup>3</sup>Landwirtschaftskammer Nordrhein-Westfalen, Zentrum für Ökologischen Landbau, Gartenstr., 11, 50765 Köln-Auweiler, Germany.

Plant-parasitic nematodes are an increasing threat to organic vegetable growers in Germany. Since nematode problems are often overlooked or misidentified, the actual damage is expected to be much higher than the reported cases. In an attempt to evaluate the occurrence and economic importance of plant-parasitic nematodes in organic grown vegetables in Germany, a survey was conducted including 55 vegetable producers in seven federal states of the country. The survey included nematode evaluation of 207 soil samples and a questionnaire for growers querying production factors and damage levels. Overall, *Pratylenchus* and *Tylenchorhynchus* were the most prominent nematode genera detected with an incidence of over 90% of the samples, followed by *Paratylenchus* (56%), *Meloidogyne* (51%) and *Globodera/Heterodera* (15%). Yield losses could exceed 50% on carrots and onions and were most pronounced on sandy soils. In many cases, nematode problems started 5–10 years after conversion to organic farming. The survey indicated that plant-parasitic nematodes are wide-spread in organic farming in Germany and can cause severe damage which may result in complete loss of the crop. Awareness of vegetable growers of plant-parasitic nematodes has to be enhanced, and adequate control measures need to be developed to ensure high quality of organic produce in the future.

**CONTROL STRATEGIES FOR THE ROOT-KNOT NEMATODE *MELOIDOGYNE HAPLA* IN ORGANIC FARMING.** **J. Hallmann,<sup>1</sup> F. Rau,<sup>2</sup> H. Buck,<sup>2</sup> M. Puffert.<sup>3</sup>** <sup>1</sup>Federal Biological Research Centre for Agriculture and Forestry, Toppeheideweg 88, 48161 Muenster, Germany, <sup>2</sup>Ökoring Niedersachsen, Bahnhofstr. 15, 27374 Visselhövede, Germany, <sup>3</sup>Landwirtschaftskammer Nordrhein-Westfalen, Gartenbauzentrum Münster-Wolbeck, Münsterstr. 62–68, 48167 Münster, Germany.

The root-knot nematode *Meloidogyne hapla* is a major pest in organic farming causing severe damage, especially on vegetables. Common practices such as high cropping frequencies of legumes and low frequencies of cereals in association with unsatisfactory weed control are assumed to be major factors for nematode build-up in vegetable dominating rotations. Due to the broad host spectrum of *M. hapla*, strategies solely based on crop rotation are often not sufficient in controlling the nematode. A series of field experiments was conducted to develop more efficient control strategies. Based on the results, a recommendation for reducing high nematode densities was developed which is built on black fallow throughout the main vegetation period, buffered by additional measures such as previous growth of a overwintering legume and its incorporation early in spring before *M. hapla* has multiplied, followed by a overwintering cereal to conserve soil nutrients and avoid erosion. In the long-term, build-up of damaging levels of *M. hapla* should be avoided by a higher cropping frequency of non host crops (e.g., cereals, Tagetes), growth of catch crops (e.g., fodder radish), satisfactory weed control, short periods of black fallow between crops and avoidance of clover immediately before the growth of susceptible vegetables.

**DEVELOPMENT, REPRODUCTION, AND ROOT GALLING OF *MELOIDOGYNE INCOGNITA* POPULATIONS ON SEVERAL COTTON CULTIVARS.** **S. A. Anwar,<sup>1</sup> M. V. McKenry,<sup>2</sup> N. Javed.<sup>3</sup>** <sup>1</sup>HEC-Foreign Professor, University of Agriculture, Faisalabad, Pakistan, <sup>2</sup>University of California, Riverside, CA, <sup>3</sup>Department of Plant Pathology, University of Agriculture, Faisalabad, Pakistan.

Roots of irradiated NIAB-78 cotton cultivar were compared with roots of five other cotton cultivars including CIM-534, CIM-506, CIM-496, CIM-499 and CIM-707 in a glasshouse. Each pot or growth tube was inoculated with 500 freshly hatched J2 derived from a single egg mass that had been increased on tomato. Nematode reproduction and “in root” developmental progress were assessed 63 days after inoculation (DAI) utilizing Phloxine B and Acid Fuschin stains, respectively. Egg mass production by three *M. incognita* populations, CIM-534, CIM-496 and CIM-506, was significantly higher on roots of CIM-534, CIM-496, CIM-506, CIM-499 and CIM-707 cotton cultivars compared to that on NIAB-78 cultivar. Developmental stages of *M. incognita* populations originally from CIM-534, CIM-496, CIM-506 and NIAB-78

on the NIAB-78 cotton cultivar were quantified. Equal numbers of J2 penetrated roots, regardless of the population. However, roots of NIAB-78 infected by the NIAB-78 population had double the number of J4 and adult females as compared to three other populations. This translated to greater numbers of egg masses and egg-producing adult females by the NIAB-78 population compared to other populations. Increased virulence was associated with larger gall sizes, larger giant cell formation and improved success of juveniles transitioning into reproducing adults.

**COVER CROPS AND ORGANIC MULCHES FOR NEMATODE, WEED AND PLANT HEALTH MANAGEMENT. K. Wang,<sup>1</sup> R. McSorley,<sup>1</sup> R. N. Gallaher.<sup>2</sup>** <sup>1</sup>Departments of Entomology and Nematology and <sup>2</sup>Agronomy, University of Florida, Gainesville, FL.

Field experiments were conducted in 2003 and 2004 to evaluate sunn hemp, *Crotalaria juncea*, and cowpea, *Vigna unguiculata*, as summer cover crops and as organic mulches for root-knot nematodes, *Meloidogyne* spp., and weed suppression, while enhancing beneficial nematodes. Experiments were split-plot designs where the main plots were summer planting of sunn hemp, cowpea or fallow, and the subplots were organic mulch of sunn hemp, cowpea or no mulch. Turnip *Brassica rapa* and lima bean *Phaseolus lunatus* were planted in the fall. Cowpea did not suppress root-knot nematodes, but sunn hemp mulch suppressed the nematodes at the end the turnip crop. Sunn hemp mulch enhanced free-living nematode numbers in 2004 but not in 2003 and suppressed weeds in both years. Both sunn hemp and cowpea mulches increased the N and K content and yield of both crops ( $P < 0.05$ ). Inconsistent performance of treatments on nematode communities in both years was attributed to *Pasteuria penetrans* infestation on root-knot nematodes, which was higher in 2004 than 2003. When *P. penetrans* infestation level was high, root-knot nematode numbers were low, thus relationships between several nematode community indices and lima bean yield were evident ( $P < 0.05$ ).

**EFFECT OF MELOIDOGYNE INCOGNITA INFECTION ON LEACHING OF NITROGEN IN COTTON, P. Timper, T. C. Strickland, R. K. Hubbard.** USDA ARS, P. O. Box 748, Tifton, GA, USA.

Root damage from parasitism by nematodes reduces the ability of the plant to take up water and nutrients. Recently, parasitism of cotton by *Rotylenchus reniformis* was shown to increase mineralization of nitrogen (N). We hypothesized that, because of increased mineralization and reduced uptake, more N would leach from the root zone of cotton parasitized by *Meloidogyne incognita* than from non-parasitized cotton. We constructed lysimeters out of 23-liter plastic buckets to collect leachate from different nematode treatments. The lysimeters were filled with 18 liters of field-collected loamy sand and planted with a single cotton seed. The soil was fertilized with 10–10–10 (NPK) at planting and ammonium nitrate 5 weeks later. The nematode treatments were 0, 8,000 and 20,000 *M. incognita* eggs/plant. The nematodes were inoculated in a split application with ¼ applied at planting and ¾ applied 3 weeks later. When roots were examined 15 weeks after planting, root gall indices (0–10 scale) were 0, 2.6 and 7.6 for the three nematode treatments. Nematodes reduced plant top weight but not root weight. There was an increase in the amount of leachate with increasing nematode inoculum which was probably due to reduced evapotranspiration by nematode-infected plants. However, nematodes did not affect the total amount of N leached from the root zone.

**HISTOLOGICAL CHARACTERIZATION OF ROOT-KNOT NEMATODE RESISTANCE IN COWPEA AND ITS RELATION TO REACTIVE OXYGEN SPECIES MODULATION. S. Das, J. D. Ehlers, T. J. Close, P. A. Roberts.** University of California, Riverside, CA.

Root-knot nematodes (*Meloidogyne* spp.) are sedentary endoparasites with a broad host range. Cowpea (*Vigna unguiculata*) is an important food and fodder legume mainly in sub-Saharan Africa and also in the United States, where root-knot nematodes are a major problem for cowpea growers. Several sources of root-knot resistance have been identified in cowpea, including the widely used *Rk* gene. To elucidate the mechanism of *Rk*-mediated resistance histologically, we compared the response of a resistant cowpea cultivar CB46 with a susceptible near-isogenic line (in CB46 background) against avirulent *M. incognita* feeding. There was no typical Hypersensitive Response (HR) in resistant roots, and nematodes were able to develop feeding sites similar to those in susceptible roots up to 9–14 days post inoculation (dpi), after which giant cell deterioration was observed. These results confirmed that induction of resistance is relatively late in this system. In pathogen resistance, HR is closely associated with an oxidative burst (OB) in infected tissue. We also quantified the level of reactive oxygen species release in both compatible and incompatible reactions. Following a basal OB at the start of infection, no significant OB was detected up to 14 dpi. These results will be useful to design gene expression experiments to dissect *Rk*-mediated resistance at the molecular level.

**EFFICACY OF MIDAS FOR CONTROL OF NEMATODES, PATHOGENS AND WEEDS IN ORNAMENTAL COCKSCOMB (CELOSIA ARGENTEA) PRODUCTION IN FLORIDA. N. Kokalis-burelle, E. N. Roskopf.** USDA, ARS, U.S. Horticultural Research Lab, Ft. Pierce, FL, USA.

Midas (iodomethane:chloropicrin, 50:50) was evaluated for control of root-knot nematodes (*Meloidogyne incognita*), pathogens and weeds in *Celosia argentea* in a cut-flower production field in Florida. Midas (224 kg/ha) was compared to

methyl bromide:chloropicrin (MeBr, 98:2, 224 kg/ha) and an untreated control. Nematodes, pathogens and weeds were evaluated throughout the season. At 97 DAP, nematodes were extracted from roots, which were evaluated for galling and disease. *Pythium* and *Fusarium* cfu were enumerated on selective media. Midas reduced populations of nematodes in soil compared to the control post-fumigation. At 40 DAP, soil nematodes rebounded and were evenly distributed among treatments. At 97 DAP, both Midas and MeBr reduced root-knot nematodes in roots and galling and increased root weight compared to the control, with Midas having higher root weights than MeBr. Stem diameter and height were lowest in the control, while incidence of wilted and stunted plants and number of *Fusarium* and *Pythium* cfu were highest in the control with no differences between fumigants. Number of marketable stems was higher in fumigant treatments than the control. Weed counts and weights were highest in the untreated check throughout the season with no difference between MeBr and Midas. Midas provided nematode, weed and disease control and *Celosia* yields comparable to MeBr.

**PINE WILT DISEASE: THE FIRST RECOGNIZED CASE OF A PLANT DISEASE INDUCED BY A MUTUALISTIC NEMATODE-BACTERIAL SYMBIOSIS. B. Zhao, X. Liu.** Nanjing Forestry University, Nanjing, P. R. China

Pine wilt disease (PWD) has become a worldwide threat to pine forests. It has been generally believed that *Bursaphelenchus xylophilus*, the pine wood nematode (PWN), was the only pathogen involved. Our recent work, however, indicates that the bacteria symbiotically associated with the nematode play a significant role in the disease. We conducted a series of inoculation experiments with a new inoculation device, a cold water jacket, by which the nematode and its associated bacteria were confined within the inoculated zone of a treated pine branch. The results showed that PWD was induced without movement of the two pathogens from the inoculation site to other parts of the treated pine. Also, in the absence of the nematode, the pathogenic bacterium, *Pseudomonas fluorescens* (GcM5-1A), mixed with a crude extract of the surface coat proteins from axenic PWN was able to cause the disease. The bacterium was re-isolated from the dead host in accordance with Koch's postulate. Finally, inoculation of a cell-free supernatant from a culture of *P. fluorescens* without PWN resulted in PWD. We conclude that systemic toxins produced by the pathogenic bacteria associated with PWN cause PWD and tree death. This is the first experimentally confirmed plant disease induced by a mutualistic nematode-bacterial symbiosis.

**EFFECT OF PLANT GROWTH-PROMOTING RHIZOBACTERIUM *PAENIBACILLUS POLYMYXA* GBR-1 ON PLANT-PARASITIC NEMATODES AND ROOT-KNOT DISEASE. Z. Khan,<sup>1</sup> S. Son,<sup>1</sup> S. Kim,<sup>1</sup> Y. Jeon,<sup>2</sup> H. U. Khan,<sup>3</sup> Y. Kim,<sup>1</sup> H. Moon.<sup>1</sup>** <sup>1</sup>Department of Agricultural Biotechnology, Seoul National University, Seoul 151-921, Korea, <sup>2</sup>Bio-resource Research Group, KT&G Central Research Institute, Suwon 440-746, Korea, <sup>3</sup>Pakistan Science Foundation, Islamabad, Pakistan.

*Paenibacillus polymyxa* is a plant growth-promoting rhizobacterium and is used for the biological control of plant diseases. *Paenibacillus polymyxa* strain GBR-1 was isolated from rotten ginseng roots and was grown in brain heart infusion broth (Difco) at 28°C for 48 hours. After culturing, the bacterial cells were precipitated through centrifugation (6,000 rpm), and either the culture filtrate or aqueous bacterial cell suspension (dissolved in distilled water) was used for treatment. *In vitro* and pot experiments were conducted to evaluate nematocidal potential of *P. polymyxa* GBR-1. Exposure of plant-parasitic nematodes, *Bursaphelenchus xylophilus*, *Pratylenchus pratensis*, second-stage juveniles of *Heterodera glycines* and *Meloidogyne incognita* to various concentrations of culture filtrate of *P. polymyxa* GBR-1 under *in vitro* conditions caused substantial mortality of these nematodes and significantly reduced egg hatch of *M. incognita*. The exposure duration and increasing concentration of culture filtrate had direct bearing on nematode mortality and inhibition of egg hatching. Culture filtrate as well as aqueous bacterial cell suspension (dissolved in distilled water, 10<sup>7</sup> cfu/ml) of *P. polymyxa* caused significant reduction in root galling and *M. incognita* population when applied at the rate of 5 ml/pot and increased plant growth 2-3 fold compared to untreated control.

**RECESSIVE RESISTANCE TO THE ROOT-KNOT NEMATODE *MELOIDOGYNE INCOGNITA* DERIVED FROM THE GRAPEVINE ROOTSTOCK 3309 C. P. Cousins,<sup>1</sup> D. Johnston,<sup>1</sup> S. Switras-Meyer,<sup>1</sup> C. Meyer.<sup>2</sup>** <sup>1</sup>USDA ARS, Grape Genetics Research Unit, Geneva, NY, USA, <sup>2</sup>Cornell University, Department of Horticultural Sciences, Geneva, NY, USA.

Grapevine genetic resistance to root-knot nematodes (*Meloidogyne* species) is known to be conferred by dominant alleles found in principal nematode resistant rootstock varieties. 3309 C is a root-knot nematode-susceptible grapevine rootstock. Although typically staminate flowered, 3309 C occasionally produces perfect flowers; rare perfect flowers were self-pollinated to produce a 3309 C self population. Seedlings growing in individual pots in a greenhouse were inoculated with 1,500 *M. incognita* second-stage juveniles once the seedlings had two true leaves, at age about 1 month. Seven weeks after inoculation, nematode reproduction was assessed by washing the roots free of potting medium, staining the roots for 1 hour in an aqueous solution of eosin-Y (0.25 g/liter) and counting the egg masses present on each root system. Five seedlings demonstrated very low levels of nematode reproduction (fewer than 0.60 egg masses/g fresh weight of roots); 28 suscep-

tible seedlings showed more than 1.0 egg mass/g fresh weight of roots. This segregation pattern is consistent with 3309 C being a heterozygous carrier for a recessive allele that provides resistance against *M. incognita*.

**THE TOBACCO CYST NEMATODE AFFECTS PHOTOSYNTHESIS OF SHADE-GROWN CIGAR WRAPPER TOBACCO. J. A. Lamondia.** The Connecticut Agricultural Experiment Station Valley Laboratory, Windsor, CT 06095, USA.

The tobacco cyst nematode (TCN), *Globodera tabacum tabacum*, reduces growth and yield of shade-grown cigar wrapper tobacco. The effects of nematode infection on photosynthetic functions were investigated in 65 field microplots in 2005 and 2006. Nematode-susceptible shade tobacco cv. '8212' was planted in 2005, and either 8212 or TCN-resistant '125B' was planted in 2006. Photosynthetic and transpiration rates per 6.25 cm<sup>2</sup> were recorded for two leaves per plant on 21 Sep 2005 and on 14, 19 and 20 Jul and 9 Aug 2006. Initial nematode densities (Pi) were placed into four classes: 0–50, 50–75, 75–100 and > 100 J2/cm<sup>3</sup> soil. Previous microplot studies determined that leaf yield losses were less than 5% below 100 J2/cm<sup>3</sup> soil and increased exponentially above that density. TCN Pi class affected photosynthetic and transpiration rates in both years. Rates were higher for the 50–75 and 75–100 classes than for higher and lower TCN Pi. Shoot weights averaged 2.3, 2.1, 2.1 and 1.8 kg/plot for the low to high Pi classes, respectively, in 2006. Resistant plants had higher weights and higher rates of photosynthesis and transpiration. The combination of reduced photosynthetic and transpiration rates per unit leaf area and reduced leaf area for plants grown in plots with > 100 J2/cm<sup>3</sup> soil of *G. t. tabacum* represents a significant effect of the nematode on total plant photosynthesis.

**LABORATORY AND GREENHOUSE STUDIES WITH QL AGRI AND THE ROOT-KNOT NEMATODE, MELOIDOGYNE INCOGNITA. L. J. Marais,<sup>1</sup> R. Otero,<sup>2</sup> M. A. McClure.<sup>3</sup>** <sup>1</sup>Monterey AgResources, Fresno, CA, USA, <sup>2</sup>Desert King International, <sup>3</sup>Plant Sciences Dept., University of Arizona, Tucson, AZ, USA.

QL Agri is a commercially available extract from the bark of the South American tree, *Quillaja saponaria*. Preliminary work showed that QL Agri can inhibit reproduction of *Meloidogyne incognita* in roots of transplanted tomatoes, but its target is not known. Experiments were conducted to determine if QL Agri is directly toxic to *M. incognita* infective juveniles (J2) and to measure its effect on plant health and nematode reproduction. In vitro assays for J2 motility were conducted in the laboratory in 24-well culture plates with QL Agri concentrations of 1,000 to 100,000 ppm and exposures of 24, 48 and 72 hours. Infectivity after exposure to QL Agri was determined by inoculating individual cucumber seedlings in blotter rolls with J2 treated with QL Agri for 72 hours followed by rinsing and incubation in distilled water for 5 hours. QL Agri reduced the motility of *M. incognita* J2 in vitro, depending upon concentration and length of exposure. At 100,000 ppm, 94% of the J2 were immobilized after 72 hours. However, 95% of immobilized J2 regained motility after rinsing and incubation in distilled water, and their ability to infect cucumber roots was not diminished compared to controls. Greenhouse studies were conducted in 10-cm-diam. pots containing week-old cucumber seedlings with QL Agri applied as a soil drench containing 1,000, 10,000, 50,000 or 100,000 ppm of the commercial concentrate. QL Agri inhibited reproduction of *M. incognita* and enhanced shoot and root growth of infected seedlings.

**EFFECTS OF INOCULUM TYPE, INOCULUM LEVEL, INOCULATION DATE AND ASSESSMENT DATE ON EVALUATING RESISTANCE TO MELOIDOGYNE ARENARIA IN PEANUT. W. Dong,<sup>1</sup> C. C. Holbrook,<sup>2</sup> P. Timper,<sup>2</sup> T. B. Brenneman.<sup>1</sup>** <sup>1</sup>Department of Plant Pathology, University of Georgia, Tifton, GA 31793, USA, <sup>2</sup>USDA-ARS, Coastal Plain Exp. Stn., Tifton, GA 31793, USA.

Use of resistant cultivars is a desirable approach to manage the peanut root-knot nematode (*Meloidogyne arenaria*), and reliable and efficient screening methods are needed. To optimize the resistance screening protocol, a series of greenhouse tests were done using seven genotypes with three levels of resistance to *M. arenaria*. The three resistance levels could be separated based on gall index as early as 2 weeks after inoculation (WAI) using 8,000 eggs/plant, while 4 or more weeks were needed when 1,000–6,000 eggs/plant were used. High inoculum densities (> 8,000 eggs/plant) were needed to separate the three resistance levels based on eggs per gram root within 8 WAI. A gall index based on percentage of galled roots could separate the three resistance levels at lower inoculum levels and earlier harvest dates than other assessment methods. The use of eggs vs. second-stage juveniles (J2) as inoculum provided similar results; however, it took 3–5 more days to collect J2 than to collect eggs from roots. Plant age affected gall index and nematode reproduction on peanut, especially on the susceptible genotypes AT201 and D098. The genotypes were separated into their correct resistance classes when inoculated 10–30 days after planting, but were not separated correctly when inoculated on day 40.

**THE MULTI-YEAR, CUMULATIVE EFFECTS OF MONOCROPPING COTTON RESISTANT TO THE SOUTHERN ROOT-KNOT NEMATODE, MELOIDOGYNE INCOGNITA. R. F. Davis,<sup>1</sup> R. C. Kemerait.<sup>2</sup>** <sup>1</sup>USDA-ARS, CPMRU, Tifton, GA, USA, <sup>2</sup>University of Georgia, Tifton, GA, USA.

This study documents the cumulative effect of moderate resistance on nematode population density and yield loss when a moderately resistant genotype was grown for three years. Cotton genotypes were Acala NemX (highly resistant, but not

adapted to the Southeast), Phytogen PH98–3196 (moderately resistant) and Deltapine DP458 B/R (susceptible). Cotton was grown in fumigated and non-fumigated plots to measure yield loss. Each genotype and nematicide combination was planted in the same place for three years at two sites to document cumulative effects. In 2006, following three years of the different genotypes, all plots at one site were planted with susceptible cotton to document residual effects of planting resistant genotypes. Root galling and nematode population densities in the soil were significantly lower and percentage yield suppression was numerically lower when moderately resistant cotton was grown compared to the susceptible standard in both fields in all three years. Differences between susceptible and moderately resistant genotypes are established quickly (after only one season) and then maintained at similar levels in subsequent years. Moderately resistant cotton genotypes are more beneficial than previously believed and should be pursued for their significant contribution to nematode management.

**NEMATODE-BACTERIAL SYMBIONTS: TO INSECTS AND BEYOND! J. M. Webster,<sup>1</sup> L. Tang,<sup>2</sup>** <sup>1</sup>Simon Fraser University, Vancouver, British Columbia, Canada, <sup>2</sup>Welichem Biotech Inc., Burnaby, British Columbia, Canada.

Entomopathogenic nematodes and their bacterial symbionts live mutualistically in soil and insects. The nematodes and bacteria collaborate to overcome the innate humoral and cellular defences in the insect haemocoel. Some of the bacterial metabolites modulate the insect's defence system and/or function as antimicrobials to minimize competition by fungi and other bacteria. After extraction, synthesis and assaying of selected metabolites show some chemotherapeutic properties. One of these metabolites, a heterocyclic compound (WBI-2100), has demonstrated not only cancer inhibition but also neutrophil stimulation and the prevention of chemotherapy-induced neutropenia when tested in mouse models. In the human-mouse xenograph melanoma model, WBI-2100 achieved 80% tumour growth inhibition when combined with the chemotherapeutic Taxol, in contrast with 40% tumour growth inhibition with Taxol alone. No obvious side effects, such as body weight loss, were observed with WBI-2100 dose rates of up to 20 mg/kg/day. Innate immunity, in contrast to acquired or adaptive immunity, is the immediate defence mechanism of many, perhaps most, organisms, including plants, insects and humans and contributes to the delicate homeostasis of healthy organisms. Treatment with small molecule, non-antigenic substances, such as WBI-2100, may provide the necessary supplement to overcome cancerous growths and maintain a healthier organism.

**IMPORTANCE OF MULTITROPHIC INTERACTIONS FOR THE EFFICACY OF *PAECILOMYCES LILACINUS* STRAIN 251 TO CONTROL ROOT-KNOT NEMATODES. S. Kiewnick.** Agroscope Changins-Waedenswil, Research Station ACW, Plant Protection Ecotoxicology and Soil Zoology, Waedenswil, Switzerland.

The nematode egg pathogenic fungus *Paecilomyces lilacinus* 251 (PL251) is commercially available in several countries. This biocontrol agent has demonstrated efficacy in reducing root-knot, cyst and free-living nematodes on a range of crops. In the US, it is registered as MELON WG for use on a variety of crops. Multitrophic interactions with the host plant, targeted nematode and other soil antagonists were investigated, and it was demonstrated that the biocontrol efficacy was neither linked to the presence of the target nematode nor to the host plant. Some host plants seemed to provide unsuitable conditions in their rhizosphere which resulted in a more rapid decline of PL251 density in soil. In addition, it was found that rhizosphere competence is not the key factor for the efficacy of PL251. Co-application of PL251 with other soil antagonists increased biocontrol efficacy against root-knot nematodes. The efficacy of PL251 depended strongly on the ratio between application rate and inoculum density in soil. As pre-plant treatment, a rate from 1.5 to 7.5 × 10<sup>5</sup> cfu/g soil resulted in significant control at inoculum densities of 100 to 400 root-knot nematode eggs/100 ml soil. However, due to the rapid decline of PL251 in soil, repeated applications are needed to maintain a sufficient density of PL251 for season-long protection.

**LONG- AND SHORT-TERM TILLAGE EFFECTS ON *HETERODERA GLYCINES* REPRODUCTION. P. A. Donald,<sup>1</sup> D. D. Tyler,<sup>2</sup>** <sup>1</sup>ARS USDA, Jackson, TN, USA, <sup>2</sup>University of Tennessee, Jackson, TN, USA.

Investigations were conducted to determine the long- and short-term effects of tillage on *Heterodera glycines*, soybean cyst nematode (SCN), reproduction. Tillage plots were established in 1979 representing six tillage/no-tillage regimes. A portion of each plot was changed from no-tillage to chisel or from tilled to no-tillage in 2002. This resulted in 12 treatments, six long-term and six short-term. There was poor correlation of nematode egg population density at planting or harvest with yield over all four years of the experiment. Egg population density as a result of SCN reproduction however did reflect variety selection, even though all varieties were marketed as resistant to at least race 3. The tillage treatments were significant in all years of the experiment. No-tillage plots consistently had higher SCN egg population density than the tilled plots. The most dramatic change in increased SCN reproductive rate was the first two years after the initial long-term no-tillage area was chisel tilled. Large increases in SCN reproductive rate after that time were not consistently associated with a particular tillage treatment. Conversion of disc treatment area to no-tillage resulted in lower SCN reproductive rate. Our data suggest that response in SCN population density to tillage change is rapid and most notable during the first season of tillage conversion.

WHAT FORM OF OILSEED RADISH (*RAPHANUS SATIVUS*) IS BEST FOR MANAGING *MELOIDOGYNE HAPLA*?

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Oilseed radish (OSR, *Raphanus sativus*), which can be used as trap, cover, or biofumigant when incorporated as a green manure crop, is a potential methyl bromide alternative for managing soil-borne biotic factors like *Meloidogyne hapla*. Exploiting OSR's multi-purpose traits, however, requires a careful balancing of the biology of the yield-limiting factor(s) and the prevailing soil conditions. When organic (OSRO) and conventional (OSRC) cultivars grown in 300 cc of sandy, loamy sand or muck soils were inoculated with 3,000 eggs of *M. hapla* populations (collected from the three soils) and maintained for about 500 degree days (base 10°C), all nematodes completed their life cycle in both cultivars, with significantly more nematodes infecting in the sandy soil. When OSRC was compared with Rutgers tomato (standard susceptible species) under the same conditions with an inoculum of 2,000 eggs, infection was greater in the sandy soil. OSRC shoot growth was significantly lower in the sandy soil compared with muck or loamy sand soils. While 25% fewer nematodes infected OSRC than tomato, all nematodes completed their life cycle. In addition to challenging the one-management-option-fits-all approach, results suggest that the OSR cultivar used may be best as a trap crop before *M. hapla* completes a life cycle, which, in turn, may limit foliage production for biofumigant action.

## CONTROL OF THE SOYBEAN CYST NEMATODE USING ANAEROBICALLY DIGESTED LIQUID SWINE MANURE.

**J. Xiao,<sup>1</sup> J. Zhu,<sup>2</sup> S. Chen,<sup>2</sup> W. Ruan,<sup>1</sup> C. Miller.<sup>2</sup>** <sup>1</sup>Department of Ecology, College of Life Science, Nankai University, Tianjing, China, <sup>2</sup>Southern Research and Outreach Center, University of Minnesota, 35838 120th Street, Waseca, MN 56093, USA.

The potential of using digested swine manure to control the soybean cyst nematode (SCN) was investigated. Liquid swine manure was anaerobically digested to different stages to search for the best digestion time for both volatile fatty acids (VFA) and ammonium nitrogen (NH<sub>4</sub><sup>+</sup>) enrichment. The results showed that VFA and NH<sub>4</sub><sup>+</sup> in the manure reached the maximal levels at 17 and 28 days of incubation, respectively. The VFA-enriched, NH<sub>4</sub><sup>+</sup>-enriched and raw manures were applied separately, at 31, 62, 125 and 250 ml/liter of soil, to 1.5-liter pots inoculated with 6,000 SCN eggs/100 cm<sup>3</sup> soil and grown to soybean in the greenhouse. The SCN egg population densities were determined 35 and 61 days after inoculation. The data indicated that SCN egg counts were inversely related to the manure application rates in a linear manner for all three manures for the 35-day samples, while no such relationships were found for the 61-day samples. VFA-enriched manure performed best, raw manure intermediate, and NH<sub>4</sub><sup>+</sup>-enriched manure the third in lowering SCN egg counts. As compared with no-manure control, VFA-enriched manure suppressed egg population density at 35 days by 21, 45 and 53% at 62, 125 and 250 ml/liter of soil, respectively. The study suggested that the manure was effective only for control of first generation of SCN, and VFA was probably the major compounds in suppressing SCN population.

## ROOT-KNOT NEMATODE/TOMATO INTERACTIONS: TRANSCRIPTOME PROFILING AND DEFENSE SIGNALING PATHWAYS.

**K. K. Bhattarai, U. Bishnoi, I. Kaloshian.** University of California, Riverside, CA, USA.

Little is known about the genes responsive during root-knot nematode (RKN; *Meloidogyne* spp.)/tomato incompatible interactions. Response of tomato to RKN infection was monitored using TOM1 cDNA microarray with susceptible (Moneymaker; *mi*) and resistant (Motelle; *Mi-1*) tomato at 24 hours after RKN infection. A total of 850 clones in the compatible and 1,784 clones in the incompatible interactions were found differentially regulated ( $P < 0.05$ ). In the incompatible interactions, 1,053 clones were 2-fold up-regulated and 370 were 1.5-fold down-regulated. Forty-seven percent of the differentially regulated clones were specific to the incompatible interactions. Both jasmonic acid (JA) biosynthetic genes and JA-regulated genes were found up-regulated in both compatible and incompatible interactions, but with higher amplitude in the incompatible interactions. In contrast, genes with roles in salicylic acid (SA)-mediated defense were not responsive. Tomato mutants altered in JA- or SA-signaling were used to assess the roles of these signaling pathways in *Mi-1* resistance. Using *jail* tomato, defective in JA perception, we found that JA-regulated plant defenses are not required for *Mi-1* resistance to RKN but have a role in RKN basal defense. The role of SA signaling in *Mi-1* resistance and basal defense to RKN will be reported.

A GLOBAL VIEW OF TRANSCRIPTOME AND PROTEOME IN COMPATIBLE *ARABIDOPSIS*-ROOT KNOT NEMATODE INTERACTION.

**X. Li, Z. Shen, R. V. Aroian, S. P. Briggs.** University of California at San Diego, CA, USA.

The root-knot nematode, *Meloidogyne incognita*, induces multinucleate giant cells in plant roots in a broad range of host species. To understand the nature of *M. incognita*-induced root knot development, we carried out genome-wide expression analysis in *Arabidopsis* root knot tissue with both *Arabidopsis* ATH1 transcriptome microarrays and peptide mass spectrometry. Our microarray analysis revealed that the levels of 1,225 mRNAs are significantly changed by *M. incognita* infection. Among the 8,000 proteins detected by mass spectrometry, the levels of 166 proteins were significantly changed

by *M. incognita* infection. Similar experiments were carried out on root proteins from infected and uninfected tomato and *Medicago* plants. Comparative proteomics revealed a core set of conserved proteins whose levels change in response to infection by *M. incognita*. We will describe these changes in light of the hypothesis that compatibility is conferred by a common parasitic mechanism on different host species.

**CHARACTERIZATION OF A NEW SPECIES OF CYST NEMATODE PARASITIZING CORN. E. C. Bernard,<sup>1</sup> P. A. Donald,<sup>2</sup> Z. A. Handoo,<sup>3</sup> R. D. Heinz,<sup>4</sup> T. O. Powers.<sup>5</sup>** <sup>1</sup>University of Tennessee, Knoxville, TN, <sup>2</sup>USDA, ARS, Jackson, TN, <sup>3</sup>Nematology Laboratory, USDA, ARS, Beltsville, MD, <sup>4</sup>University of Missouri, Columbia, MO, <sup>5</sup>University of Nebraska, Lincoln, NE.

Examination of unthrifty corn roots in northwestern Tennessee (Obion County) in 2006 revealed high population densities of juveniles and lemon-shaped cysts. This nematode resembles *Cactodera* spp. in possessing a circumfenestrate vulva, but lacks bullae and an underbridge. These characters differentiate it from *Heterodera* spp., including *H. zaeae*, the corn cyst nematode. Similar cyst specimens had previously been collected in 1978 from Lauderdale County, TN, on goosegrass (*Eleusine indica*). Comparison of the 1978 specimens deposited in the USDA Nematology Collection at Beltsville, MD, and the 2006 specimens verified that they were identical. Infective juveniles are 320–400  $\mu\text{m}$  long, have a short stylet (14–16  $\mu\text{m}$ ) and possess a short tail with a bluntly rounded terminus. DNA sequences from three molecular markers (18S, D2/3, ITS1) and direct comparison with *H. zaeae* from Maryland excluded conspecificity with *H. zaeae*. There were no DNA sequences in GenBank that indicated a close relationship with other Heteroderidae. This nematode appears to be a sister group to the *Globodera-Punctodera-Cactodera* clade. It reproduced well (RF > 5) on all tested hybrid corn cultivars. Reproduction was poor on other monocots and no dicot hosts have been found to date.

**EFFECT OF DIFFERENT GREEN MANURE CROPS ON THE POPULATION DENSITY OF MELOIDOGYNE CHITWOODI AND HETERODERA SCHACHTII. S. L. Hafez, S. Palanisamy.** University of Idaho.

Two greenhouse experiments were conducted to demonstrate the effects of oil radish and mustard varieties on *Meloidogyne chitwoodi* and *Heterodera schachtii*. The experiment was laid out in a completely randomized block design with seven and six treatments with six replications for the first and second experiments, respectively. Seeds of green manure varieties were planted in the pots filled with nematode-infested soil mixture and individually inoculated with either one species of nematode and harvested 8 weeks after planting. Data from the first experiment indicated that there was a significant reduction in the cyst, eggs and larval population by all oil radish and mustard varieties. Maximum percent reduction of cyst (82.3) and eggs and larvae (91.8) was observed with Comet, followed by Colonel. In the second experiment, the population of *M. chitwoodi* in the root was at the lowest level with Comet and Defender-planted pots. Percent reduction in nematode population was 98.7 and 94.9 for the Comet and Defender, respectively.

**EFFICACY OF FUMIGANT AND NON-FUMIGANT NEMATICIDES FOR THE MANAGEMENT OF MELOIDOGYNE CHITWOODI IN POTATO. S. L. Hafez, S. Palanisamy.** University of Idaho.

Two field experiments were conducted to study the efficacy of Temik 15G in combination with Vydate-L or Telone II alone or in combination with Vapam HL for control of *Meloidogyne chitwoodi* in potato. The experiments were laid out in a randomized block design with four and five treatments each with seven replications. Mocap treatments were surface broadcast-incorporated; Telone II and Vapam were applied broadcast by ripper and fumigation bar, respectively. Vydate was applied at planting and chemigated subsequently after planting. Temik was applied at planting. Potato cv. Russet Burbank seed pieces were planted in rows 3 ft apart, and at maturity the tubers were hand-harvested, weighed, graded and evaluated for nematode infection. Data from the first experiment indicated that there was an increase in total yield in Temik- and Vydate-applied plots compared to control plot. Percent of tubers with nematode infection in treated plots ranged from 31.5 to 45.0. Lowest level of nematode infection was observed in the Temik or Vydate (8 applications)-applied plots. Data from the second experiment indicated that percent of tubers with nematode infection in treated plots ranged from 0.0 to 35.6. Lowest level of nematode infection was recorded in the Telone 15 g/A + Vapam 30 gal/A than other treatments.

**EFFECT OF DIFFERENT ARUGULA VARIETIES ON MELOIDOGYNE CHITWOODI, M. HAPLA AND HETERODERA SCHACHTII. S. L. Hafez, S. Palanisamy.** University of Idaho.

Three greenhouse experiments were conducted to evaluate the efficacy of Arugula and oil radish varieties for the suppression of two root-knot nematode species, *Meloidogyne chitwoodi* and *M. hapla*, and the sugarbeet cyst nematode, *Heterodera schachtii*. The experiment was in a completely randomized block design with seven, six and four varieties for the first, second and third experiments, respectively. Three seeds of each green manure crop were planted individually in a pot filled with nematode-infested soil mix, replicated six times in a complete randomized block design. Eight weeks after planting, plants were harvested, and the fresh and dry weight of shoot and root with nematode population in the soil and root were estimated for each variety. Data indicated that among all varieties maximum reduction of *M. chitwoodi* and *M.*



*hapla* was with arugula variety Racola (91.4 and 95.5%). All the arugula varieties significantly reduced *H. schachtii* population.

**PATHOGENICITY OF *PRATYLENCHUS PENETRANS* AND *MELOIDOGYNE HAPLA* ON ONION. W. Pang, S. L. Hafez, S. Palanisamy.** University of Idaho.

Experiments were conducted under greenhouse and microplot conditions to determine the relationship between different initial population densities of *Pratylenchus penetrans* and *Meloidogyne hapla* on the growth and yield of onion. For the greenhouse study, seeds of onion cultivar Tioga planted in pots filled with sterile soil mixture were inoculated with respective nematodes at the rate of 0, 2, 4, 6 or 8 nematodes/cc soil. In the microplot experiment, Tioga seedlings were inoculated at the rate of 0, 1, 2 or 4 *M. hapla*/cc soil. For all these studies, normal cultural practices were carried out, and at harvest the data on fresh and dry weight of the plant and root were recorded along with the nematode population in the soil and root. Data indicated that the plant and bulb weight reduced with the increased inoculum density. Plant fresh weight reduced in the range of 16.2 to 73.0% by *P. penetrans* and 26.5 to 71.1% by *M. hapla* under greenhouse conditions. Bulb weight reduced 14.7 to 21.9% in micro-plot.

**STATUS OF PALE CYST NEMATODE, *GLOBODERA PALLIDA* IN IDAHO—AN UPDATE. S. L. Hafez, S. Palanisamy.** University of Idaho.

Idaho is the largest potato producer in the United States, growing about one-third of the country's potato production (12.5 billion pounds), which paid farmers about \$700 million and was worth about \$2 billion to the state. Discovery of pale cyst nematode (PCN) (*Globodera pallida*) in Idaho was significant to potato producers and exporters since it can attack potato roots and reduce yields by up to 80%. Early discovery of PCN in Idaho minimizes future potato production costs and enhances product quality and marketability. Though PCN is widely distributed in many potato-growing regions throughout the world, its infestation in Idaho appears to be isolated, but additional surveillance programs were initiated to contain further spread of the pest to neighboring field. Regulations were implemented to restrict the movement of plants and soil with appropriate sanitation procedures for equipment used on the regulated field for the spread of this nematode. Crop rotation and the use of certified seed and nematicides are effective and practical means of suppression. PCN discovery is an example of how an industry and government can come together in a crisis to ward off disaster.

**REACTION OF TWENTY-FIVE ALFALFA BREEDING LINES TO THE LESION NEMATODE *PRATYLENCHUS PENETRANS*. S. L. Hafez,<sup>1</sup> S. Palanisamy,<sup>1</sup> D. R. Miller.<sup>2</sup>** <sup>1</sup>University of Idaho, <sup>2</sup>Target Seed, LLC.

In two greenhouse experiments, 25 alfalfa lines were evaluated for their reaction to *Pratylenchus penetrans*. The experiment was in a completely randomized block design with four replications of seven cones each. Cones of 150 cc capacity were filled with sterilized soil, and seeds of each of alfalfa lines were planted in each cone and inoculated with 100 nematodes/cone. One hundred days after planting, plants were uprooted, and data on fresh and dry weights of root and shoot and nematode population in the soil and root were recorded. In the first experiment (15 cultivars), fresh and dry weights of shoot as well as roots were significantly higher, while the nematode population in the root was lower in the cultivar Cg2003–55. The second experiment was conducted with 10 cultivars including the lesion nematode-susceptible cultivar Baker. Data indicated that the variety TS5000 showed minimum nematode population per gram root with the maximum fresh and dry root weight and maximum fresh shoot weight.

**IMPACT OF SOLARIZATION, ROOTSTOCK AND *PSEUDOMONAS SYNXANTHA* ON *CRICONEMOIDES XENOPLAX* POPULATIONS AND TREE GROWTH IN A PEACH TREE SHORT LIFE SITE. A. P. Nyczepir,<sup>1</sup> D. A. Kluepfel.<sup>2</sup>** <sup>1</sup>USDA-ARS, SE Fruit and Tree Nut Research Laboratory, Byron, GA 31008, USA, <sup>2</sup>USDA-ARS, Crops Pathology and Genetics Research Unit, University of California, Davis, CA 95616, USA.

Soil solarization, alone or in combination with other disease management practices, has been shown to be effective in reducing inoculum density of many soilborne diseases, including nematodes. In 2002, a field study was initiated to determine the influence of combining solarization, application of a bacterial antagonist (*Pseudomonas synxantha* [BG33R]) through the irrigation system and an improved peach rootstock for management of the ring nematode (*Criconemoides xenoplax*) and prevention of peach tree short life (PTSL) tree death. Soil treatments include: i) solarized soil alone; ii) solarized soil + BG33R; iii) non-solarized soil; and iv) non-solarized soil + BG33R. Rootstocks examined included Guardian and Nemaguard. Solarization suppressed nematode populations for up to 19 months post-solarization. No differences in tree growth were detected between the rootstocks; however, trees growing in solarized soil + BG33R soil were larger ( $P < 0.05$ ) than trees in non-solarized plots. In 2005, PTSL tree death was more prevalent on Nemaguard vs. Guardian rootstock, but no significant differences were detected among soil treatments. These results provide useful insights into the potential use of BG33R, solarization and Guardian rootstock as a pre- and postplant alternative to chemical control of the ring nematode on PTSL sites in the Southeast.

FIELD REACTION OF SELECTED SCN-RESISTANT SOYBEAN GERMPLASM TOWARD SDS. **J. Faghihi, S. P. Conley, V. R. Ferris.** Purdue University, West Lafayette, IN, USA.

Soybean cyst nematode (SCN) and sudden death syndrome (SDS) are two of the most damaging pests of soybean in Indiana and the north central region of the United States. Many researchers have characterized their relationship as synergistic. In 2004 field studies, we observed marked differences in the reactions toward SDS among sources of SCN resistance used to characterize HG types. Peking, PI 88788, PI 90763, PI 89772 and PI 437654 showed slight to severe SDS symptoms, while Cloud and PI 209332 showed few or no SDS symptoms. Other researchers have documented a lack of visible SDS symptoms on the SCN-resistant cultivar Hartwig. Soon after the release of our PUSCN14 germplasm (known as CystX), developed from a cross between Hartwig and Williams 82, we noticed a reaction toward SDS similar to that of Hartwig. Our 2006 field studies showed that commercial CystX cultivars appear to share this characteristic of PUSCN14 germplasm. Commercial CystX cultivars, marketed as Gold and Silver, demonstrated different degree of SCN resistance and also reacted differently toward SDS based on the percentage of PUSCN14 present in them. The CystX gold cultivar containing at least 90% PUSCN14 showed few SDS symptoms, while symptoms of the silver cultivars varied. We hypothesize that the reaction in silver cultivars can vary with the percentage of PUSCN14 and other genome characteristic of the soybean cultivar.

AGRI-TERRA, A NEW MATERIAL FOR THE MANAGEMENT OF PLANT-PARASITIC NEMATODES: FIELD STUDIES. **E. C. McGawley.** Plant Pathology, 302 LSB, LSU, Baton Rouge, LA, USA.

Commercial vegetable production protocols were employed in 2006 field trials with tomato, cucumber and bell pepper. With tomato, Agri-Terra successfully managed reniform nematode populations and resulted in significant increases in yields of tomato fruit in the Extra-Large and Large size categories. Similarly, with cucumber, treatment of soil with Agri-Terra reduced reniform nematode populations significantly and increased yields of fruit in the Super-Select and Select size categories. Results with bell pepper were inconclusive as the cultivar employed in the trial supported only minimal reproduction of the nematode. Results of the cotton field trial were in agreement with those that have been observed each year over past five years. That is, application of Agri-Terra to soil as an at-planting, in-furrow (fine mist) spray treatment produced highly significant decreases in nematode populations and highly significant yield responses.

THE INFLUENCE OF MORNING GLORY, HEMP SESBANIA AND JOHNSONGRASS ON REPRODUCTION OF *ROTYLENCHULUS RENIFORMIS* ON COTTON AND SOYBEAN. **M. J. Pontif.** Plant Pathology, 302 LSB, LSU, Baton Rouge, LA, USA.

Reniform nematodes that damage cotton and soybean can also reproduce on weeds associated with these crops. Microplot studies were conducted to determine the effects of cotton, soybean, morning glory (MG), hemp sesbania (HS) and Johnsongrass (JG) on reproduction of the reniform nematode. Over three trials, the co-culture of cotton with any of the three weed species suppressed reproduction of reniform nematode. Reniform reproductive values at harvest on cotton averaged 59.5 and those for MG, HS and JG averaged 53.3, 25.3 and 20.0, respectively. Reproductive values for co-cultured cotton-MG, cotton-HS and cotton-JG averaged 44.1, 30.0 and 25.0, respectively. Reproduction data for soybean followed a trend similar to that observed for cotton. Suppression of reniform nematode resulted from physical crowding and/or from allelopathic compounds. Data from greenhouse trials, especially with MG, support the allelopathy hypothesis.

THERMAL REQUIREMENTS FOR EMBRYOGENESIS OF *ROTYLENCHULUS RENIFORMIS*. **M. M. Leach, P. A. Agudelo.** Clemson University, Clemson, SC 29634, USA.

Reniform nematode (*Rotylenchulus reniformis*) is the most economically important species of the genus *Rotylenchulus*, affecting a wide range of crops, including cotton, soybean, tobacco, pineapple and a variety of vegetables. Recent research efforts are mostly focused on the development of effective management practices, but important aspects of the basic biology of the nematode remain to be investigated. The purpose of this study was to determine and compare the thermal requirements for embryogenesis of reniform nematode populations from different geographic locations in the United States. The development of eggs was observed at a range of temperatures (20, 25, 30 and 35°C), and the base temperature at which development started was determined. The minimum degree-day requirements for complete development from newly laid egg to hatching and from hatching to immature female were calculated. The ecological significance of differences in the thermal requirements for embryogenesis is discussed.

PLANT-PARASITIC NEMATODE GENERA IN NEBRASKA CORN FIELDS. **T. A. Jackson,<sup>1</sup> R. M. Harveson,<sup>2</sup> J. G. Counsell,<sup>1</sup> K. A. Goings,<sup>1</sup> D. W. Miller.<sup>1</sup>** <sup>1</sup>University of Nebraska, Lincoln, NE, USA, <sup>2</sup>UNL-Panhandle Research and Extension Center, Scottsbluff, NE, USA.

Reports from numerous states have attributed disease in corn and subsequent yield loss to plant-parasitic nematodes. Some recent changes to corn cropping practices are expected to favor nematodes and may lead to an increase in disease caused by nematodes. Limited data are available for nematode populations, distributions, damage and management in corn.

Nematodes were extracted from soil and roots, and plant parasites were identified to genus in samples collected from 282 corn fields in 66 Nebraska counties in 2006. Population densities of one or more genera exceeded the lowest known damage thresholds for corn in 21% of the samples. The nematodes exceeding damage threshold population densities were spiral (*Helicotylenchus* spp.), stunt (*Tylenchorhynchus* spp.), dagger (*Xiphinema* spp.), lance (*Hoplolaimus* spp.) and lesion (*Pratylenchus* spp.). Spiral nematodes exceeded the damage threshold in 16% of samples. Lesion nematodes were extracted from 93% of root samples. Needle (*Longidorus* spp.), sting (*Belonolaimus* spp.) and stubby-root (*Paratrichodorus* spp.) nematodes were not extracted from any of the samples, despite having been previously identified on corn in Nebraska, and this is likely due to the late sampling dates and tendencies of these genera to migrate deeper into soil. More research is needed to determine the impact of nematodes on corn yield and interactions with other pathogens.

**NEMASCOPE: AN ONLINE IMAGE DATABASE AND KEY FOR RAPID, JARGON-FREE IDENTIFICATIONS OF NEMATODE GENERA BASED ON POINT-AND-CLICK VISUAL MATCHING. M. Yoder, J. A. Brady, E. Keogh, M. Faloutsos, P. De Ley.** University of California, Riverside, CA.

Nematodes are the most abundant metazoans on earth, with published estimates ranging from 1 to 100 million species; however, they are also the most poorly known animal phylum, with less than 7% of the total estimated diversity described. This is in part due to the dependence of nematode taxonomy and identification on high-powered microscopy and access to specialized literature, as well as on a visual memory of structures and diversity accumulated through years of experience. These constraints make identifications extremely difficult and labor-intensive for novices. New approaches are needed to reduce the time necessary for non-expert identifications by utilizing the same principles an expert would use, mainly by a process of visual recognition. An online image database and pictorial key, based on point-and-click visual matching, was built to simulate the visual memory of an expert taxonomist and to stimulate the learning process in beginning nematologists. NemaScope can be accessed at the following URL: <https://www.cs.ucr.edu/~bradyj/nematodes/>. The image database component of NemaScope currently houses an estimated 750 to 1,500 high-definition DIC pictures and multifocal images representing the six most important nematode body parts needed for genus identifications. The identification key component contains approximately 150 different nematode genera.

**CONSTRUCTION AND PRELIMINARY ANALYSIS OF A SPECIFIC CDNA LIBRARY OF ANTERIOR END OF STEM NEMATODE (*DITYLENCHUS DESTRUCTOR*). Y. Zhou, H. Jian.** Department of Plant Pathology, China Agricultural University, Beijing, 100094, China.

Stem nematode (*Ditylenchus destructor*) has become a major disease of sweet potato (*Ipomoea batatas*) in recent years in China. For isolation and identification of parasitic genes of this nematode, a specific cDNA library of anterior end of the nematode will be constructed in this research. About 200 anterior ends (about one-fourth of body length) and 300 posterior ends (about a half of body length, no anterior end) of nematode were cut by ophthalmic bistoury, and mRNA of both were isolated using Dynabeads mRNA DIRECT Micro kit, respectively. Then cDNA was synthesized by SMART cDNA library construction kit (Clontech). A specific cDNA library of anterior end of nematode was constructed by modified solid-phase subtractive hybridization. About 3,000 positive colonies were obtained, and the fragment length of colonies ranged from 200 to 1,500 bp. One hundred twenty colonies were sequenced and analyzed by Blastx. Some interesting genes were found. Among them, ZYM-B48 has 32% identity with an allergen gene in *Ascaris suum*, ZYM-9 has 20% identity with a 24 kDa secreted protein in *Brugia malayi*, ZYM-i24 has 47% homology with immunodominant antigen homologue in *Dirofilaria immitis*, and ZYM-d97 has 77% identity with secreted glutathione peroxidase in *Globodera rostochiensis*. Exploring function of those genes will help to understand how the nematode evades plant immunity reaction.

**INFLUENCE OF PH AND INFESTATION TIME ON THE INTERACTION BETWEEN *PRATYLENCHUS PENETRANS* AND *P. CRENATUS* IN CORN. N. Viaene,<sup>1</sup> S. Baidya.<sup>2</sup>** <sup>1</sup>Department of Plant Protection, Unit Plant, Institute of Agricultural and Fisheries Research, Mellebeke, Belgium, <sup>2</sup>Department of Biology, University of Ghent, Ghent, Belgium.

The lesion nematodes *Pratylenchus penetrans* (Pp) and *P. crenatus* (Pc) occur frequently together in one field on a same crop. It is unknown how they influence each other's reproduction and how this interaction differs with pH. We added 1,500 individuals of both species to pots planted with corn in the following Pc:Pp ratios: 0:0, 100:0, 75:25, 50:50 and 0:100. In half of the pots, both species were added simultaneously, in the other half, Pp was added 2 weeks after Pc. Numbers of nematodes in roots and soil were counted 14 weeks after the last soil infestation. The test was performed twice: at pH 5.5 and at pH 6.8. Reproduction of Pc was unaffected by the quantity of Pp, at both pH. Pc reproduced more, however, when Pp was added 2 weeks later than when both species were added simultaneously, but at pH 5.5 only. At the lower pH, Pc generally reproduced more than at pH 6.8. Soil pH did not influence reproduction of Pp. Reproduction of Pp decreased as the proportion of Pc in the inoculum increased when both species were added at the same time. Adding Pp 2 weeks after Pc also reduced the reproduction of Pp. Favouring Pc, e.g., through a low soil pH, could help in reducing Pp, the most

plant-damaging species of the two. Also, earlier activity of Pc than Pp in the field could negatively influence reproduction of Pp.

ENGINEERING PLANTS FOR NEMATODE RESISTANCE. **B. C. Yadav,<sup>1</sup> K. Subramaniam.<sup>2</sup>** <sup>1</sup>Biological Sciences and Bioengineering, Indian Institute of Technology Kanpur, Uttar Pradesh, India PIN: 208016, <sup>2</sup>Biological Sciences and Bioengineering, Indian Institute of Technology Kanpur, Uttar Pradesh, India, PIN: 208016.

Despite the enormous impact plant-parasitic nematodes have on agriculture, we do not yet have an effective and environmentally safe method to control them. Their parasitic style of living does not readily permit traditional genetic methods for the discovery of gene function. In the free-living nematode, *Caenorhabditis elegans*, RNA-mediated interference (RNAi) can be triggered when the worm feeds on bacteria that produce double-stranded RNA (dsRNA) of the worm's gene. Replication of this in other heterologous situations, i.e., induction of RNAi in an organism by the dsRNA produced in, and delivered by, an evolutionarily unrelated organism, has wide implications. For instance, production of parasite-specific dsRNA in a host could protect the host against parasitic infections. In addition, this approach could be a powerful tool to the functional genomics of parasites. We engineered tobacco plants to produce dsRNA of two essential genes of the root-knot nematode *Meloidogyne incognita*, which infects a wide range of agriculturally important plants. The transgenic tobacco plants show significant resistance against *M. incognita* infection. A closer observation of the nematodes in these plants revealed that their development was severely impaired. Further, these nematodes were specifically deficient in the mRNA of targeted genes, indicating that the dsRNA produced in plants did indeed trigger RNAi response in the nematode.

REVIEW OF NEMATODE PROTECTION BENEFITS FROM ABAMECTIN SEED TREATMENT ON CORN. **A. Cochran,<sup>1</sup> C. Watrin,<sup>2</sup> and B. Ulmer.<sup>2</sup>** <sup>1</sup>Syngenta Crop Protection AG, Basel, Switzerland; <sup>2</sup>Syngenta Crop Protection Inc., Greensboro, NC.

The use of abamectin seed treatment for nematode control was successfully launched in 2006 under the brand name Avicta Compete Pak as a novel Syngenta seed treatment brand that offers U.S. cotton growers early season protection against nematodes, insects and diseases. The nematicidal active ingredient within Avicta is abamectin, which is known to be highly active against many nematode species including *Pratylenchus* spp. Field trials on corn have been conducted over the past three years in two separate field sites located in Kansas to determine the benefit of abamectin seed treatment on this crop. These trials have been conducted in sites with historically high root lesion nematode presence (*Pratylenchus* spp). Counts of lesion nematode per gram of dry root in the untreated control varied from 16,000 to 25,000 nematodes at two different testing locations in 2004, well above the damage threshold established for this pest. The use of abamectin seed treatment reduced the presence of lesion nematodes found in the root from 25–72% in this same year. Yield measurements taken from these same two locations over the past three years have indicated an overall yield benefit from the use of abamectin seed treatment of 26–69% in 2003, 17–33% in 2004 and 20–25% in 2005. In all years, the yield impact from root lesion nematode was severe, particularly in 2003. Further testing will be carried out to determine the benefit from abamectin seed treatment under less intense root lesion pressure and will also evaluate the benefit against other important corn nematode species.

EFFECT OF OXAMYL AND METHOMYL SEED TREATMENTS ON ROOT-KNOT NEMATODE INFECTION. **J. A. Desaegeer, M. Rivera, K. Nagle, T. Meloro, P. Eggink, D. Kirk.** DuPont Crop Protection, Newark, DE, USA.

Nematicide-treated seeds are an attractive option to protect plants from early-season nematode infection. Cucumber and cotton seeds were treated with different rates (from 0.5 mg a.i. to 2 mg a.i./seed) of oxamyl and methomyl and planted in greenhouse soil inoculated with *Meloidogyne incognita*, the southern root-knot nematode. On cucumber, oxamyl seed treatments gave good control of root-knot nematode at higher rates and up to 35 days after seeding (DAS), but the 2 mg a.i. rate reduced germination. Higher rates of methomyl also reduced nematode infection, but cucumber plants were stunted. On cotton at 28 DAS, high rates of methomyl and oxamyl gave nematode control similar to abamectin-treated seeds, but inferior to oxamyl and aldicarb soil applications. Nematode infection increased with time, and by 56 DAS methomyl seed treatments no longer offered control. Higher rates of oxamyl and abamectin-treated seeds had intermediate levels of control, less than oxamyl and aldicarb soil treatments, but better than non-treated and methomyl-treated seeds.

NEMATODES AGAINST NEMATODES: ASSESSING ENTOMOPATHOGENIC NEMATODES (STEINERNEMATIDAE, HETERORHABDITIDAE) FOR THE CONTROL OF THE CITRUS NEMATODE *TYLENCHULUS SEMIPENETRANS* (TYLENCHULIDAE) IN ARIZONA. **J. Gress,<sup>1</sup> S. Stock.<sup>2</sup>** <sup>1</sup>Dept. Plant Sciences, University of Arizona, 1140 E. South Campus Dr., Tucson, AZ 85721, USA, <sup>2</sup>Dept. Entomology, University of Arizona, 1140 E. South Campus Dr., Tucson, AZ 85721, USA.

At present, 90% of citrus grown in Arizona are infected by the citrus nematode, *Tylenchulus semipenetrans*. However, given the current lack of successful control measures and the increasing awareness of the detrimental health and envi-

ronmental effects due to chemical pesticides, new alternatives are deeply needed for management of this nematode. In this respect, one of the choices for substitution of chemical nematicides is the consideration of biological control agents such as entomopathogenic nematodes (EPN). In this study, two commercially available EPN, *Steinernema riobrave* (Biovector) and *Heterorhabditis bacteriophora* (Nemasys) were assessed for their efficacy against the citrus nematode in laboratory assays. Two-month-old rough lemon seedlings were grown in cone-tainers, at 25°C and 30% humidity. *Tylenchulus semipenetrans*-infected seedlings were inoculated at a concentration of 12,000 J2/seedling. Citrus nematode (J2) root-penetration rate and female egg production were assessed in relation to: 1) EPN application time (i.e., simultaneous [EPN simultaneously applied with citrus nematode], after [EPN applied after citrus nematode establishment in the citrus seedlings]), and 2) EPN application method (i.e., aqueous suspension, EPN-infected cadavers). A completely randomized design with two replications (12 citrus-seedlings/treatment) for each evaluated parameter was considered. Data were subjected to analysis of variance (ANOVA). Results from these studies will be presented and discussed. Assessment of this information is critical for understanding EPN-citrus nematode interactions and in making predictions of the various impacts of EPN application for the control of this nematode.

**MOLECULAR AND FUNCTIONAL CHARACTERIZATION OF PARASITISM GENES OF THE POTATO CYST NEMATODE *GLOBODERA ROSTOCHIENSIS*. S. Lu,<sup>1</sup> H. Yu,<sup>1</sup> S. Chen,<sup>1</sup> H. B. Borchardt-Wier,<sup>2</sup> X. Wang.<sup>2</sup>** <sup>1</sup>Cornell University, Ithaca, NY, USA, <sup>2</sup>USDA-ARS, Cornell University, Ithaca, NY, USA.

Potato cyst nematodes (*Globodera rostochiensis* and *G. pallida*) are quarantined pests threatening the potato industry in the United States. Secreted proteins encoded by parasitism genes expressed in the esophageal gland cells of plant-parasitic nematodes represent the primary molecules involved in plant parasitism. Putative parasitism genes including several *CLE*-like genes (*GrCLEs*), a chorismate mutase gene (*GrCM-1*), a venom allergen-like gene (*GrVAP-1*), a ubiquitin extension gene (*GrUBI-1*), an *SKP-1*-like gene (*GrSKP-1*) and three genes homologous, respectively, to 4D06, 7E05 and 33E05 genes of the soybean cyst nematode have been isolated from *G. rostochiensis*. In situ mRNA hybridization showed the accumulation of their transcripts exclusively within the esophageal gland cells and Southern blot analysis confirmed the presence of these genes in the *G. rostochiensis* genome. RT-PCR and qRT-PCR analyses revealed that the expression of these genes was developmentally regulated. Currently, we are employing both in vitro and in vivo RNAi approaches to evaluate if these genes have a critical role in *G. rostochiensis* parasitism of plants.

**THE BACTERIAL RECEPTACLE IN *STEINERNEMA* SPP. (STEINERNEMATIDAE): INTERSPECIFIC VARIATION AND PATTERNS OF EVOLUTIONARY CHANGE OF MORPHOLOGICAL AND ULTRASTRUCTURAL TRAITS. K. L. Plichta, S. Stock.** Dept. of Entomology, University of Arizona, 1140 E. South Campus Dr., Tucson, AZ 85721, USA.

An emerging model of animal-microbe mutualism is represented by entomopathogenic nematodes *Steinernema* spp. and their endosymbionts *Xenorhabdus* spp. (Enterobacteriaceae). *Steinernema* third-stage infective juveniles (IJ) are colonized by a monoculture of *Xenorhabdus* bacteria at a discrete structure located in the anterior portion of the intestine known as the *bacterial vesicle* or *intestinal vesicle*. The nature and structure of this receptacle is presently not well understood. Early transmission electron microscopy (TEM) studies on the vesicle suggested that this vesicle is a modification of the ventricular region of the intestine that lies immediately beneath the esophago-intestinal valve in the nematode IJ. We examined structural and ultrastructural features of the bacterial vesicle in a selection of *Steinernema* spp. which represent distinctive evolutionary clades in the most recently developed molecular (multigene) phylogenetic framework for this nematode. Culture conditions were standardized to objectively assess these parameters. Differential interference contrast optics (DIC) microscopy was used to examine the shape of the bacterial receptacle among different taxa. Transmission electron microscopy was also considered to assess ultrastructural variation for one representative species of each phylogenetic clade. For cladistic analysis, quantitative traits were coded as discrete states employing Simon's homogenous subset coding method. Parsimony mapping was considered to test hypothesis on the evolution of morphological and ultrastructural traits. Results from this study will be presented and discussed.

**RISK ASSESSMENT OF CYST NEMATODE EVOLUTION THROUGH GENOTYPE FLOW AND PARASITISM GENE EVOLUTION. A. Blanchard, O. Plantard, E. Grenier.** Institut National de la Recherche Agronomique, Agro-campus Rennes, UMR1099 BiO3P, F-35653 Le Rheu, France.

To understand the process that leads to breakdown of a plant resistance gene, we need to understand the processes that govern pathogen evolution. By studying two of the main evolutionary forces (migration and mutation) operating on nematode populations, we show that cyst nematodes appear to present a higher risk than previously thought. Migration: Nematode genotype flows were assessed using specific neutral markers (microsatellite loci). We show that both the sugar beet (*Heterodera schachtii*) and the potato cyst nematodes (*Globodera pallida*) in their native area (Europe and South America, respectively) exhibit a high level of gene flow at the intra-regional level. We hypothesize that passive dispersal of cysts by natural or anthropic means are probably responsible for the results observed. Mutation: In order to get a first knowledge in nematode parasitism gene evolution and to determine the type of selection operating on these genes, we used

a dataset of more than 30 cathepsin L (parasitism gene) and elongation factor (housekeeping gene) gene sequences from various populations and species of cyst nematodes. The cathepsin L gene seems to evolve more rapidly than the housekeeping gene by accumulating nonsynonymous substitutions. The nature of the selection occurring on these genes was defined through the estimation of the Ka/Ks ratio along the gene sequences and in the various lineages. We show using maximum likelihood methods that a particular domain of the mature region of the cathepsin L protein evolves under diversifying selection. With the increase of our knowledge of the evolution of nematode genes involved in the parasitic process, new markers are now available to determine effective gene flow of parasitism genes among populations.

**MOLECULAR DIAGNOSTICS AND PHYLOGENETIC RELATIONSHIPS OF SOME SPECIES OF ROOT-LESION NEMATODES OF THE GENUS PRATYLENCHUS. S. A. Subbotin,<sup>1</sup> E. J. Ragsdale,<sup>2</sup> T. Mullens,<sup>2</sup> P. Roberts,<sup>2</sup> J. G. Baldwin.<sup>2</sup>** <sup>1</sup>Plant Pest Diagnostic Centre, CDFA, CA, USA, <sup>2</sup>University of California, Riverside, CA, USA.

The root-lesion nematodes of the genus *Pratylenchus* Filipjev 1936, migratory root endoparasites, are considered among the most widespread and important nematode parasites in a variety of crops. A goal of the present study is to infer molecular-based phylogenies among species and to identify a narrow core of congruent morphological characters that carry the greatest possibility of phylogenetic signal. The ITS, D2-D3 of the 28S and 18S of nuclear rRNA genes were amplified and sequenced for 30 populations of nominal species (*P. brachyurus*, *P. coffeae*, *P. crenatus*, *P. neglectus*, *P. penetrans*, *P. pinguicaudatus*, *P. scribneri*, *P. thornei*, *P. vulnus* and *P. zeae*) and two unidentified species. The PCR-ITS-RFLP, using several enzymes, clearly separated most species. Sequence analysis confirmed among large numbers of geographically diverse isolates that most classical morphospecies are monophyletic. Sequence alignments of D2-D3 and partly 18S sequences were created for 20 *Pratylenchus* species. The alignments were optimized using the secondary structure model and analyzed by Bayesian inference under the complex models of RNA evolution. Phylogenetic analyses revealed at least four distinct major clades within studied *Pratylenchus* species, and these clades are generally congruent with those defined by characters derived from lip patterns, spermatheca shape and mode of reproduction.

**PHYLOGENETIC IMPLICATIONS OF THREE-DIMENSIONAL RECONSTRUCTION OF THE STOMATOSTYLET AND ANTERIOR EPIDERMIS IN APHELENCHUS AVENAE. E. J. Ragsdale,<sup>1</sup> J. Crum,<sup>2</sup> M. H. Ellisman,<sup>2</sup> J. G. Baldwin.<sup>1</sup>** <sup>1</sup>University of California, Riverside, CA 92521, USA, <sup>2</sup>National Center for Microscopy and Imaging Research, University of California, San Diego, La Jolla, CA 92093, USA.

A three-dimensional model of the stomatostylet apparatus has been reconstructed from serial thin sections of *Aphelenchus avenae* (Nematoda: Aphelenchidae) and compared to previous work on rhabditids and cephalobs to better understand evolution of the stylet. 3D reconstruction is essential to visualizing cell-to-cell relationships, subtleties of which are easily missed using static thin sections. Two arcade syncytia ("guide ring") line the stylet shaft, supporting the hypothesis that the stylet shaft and cone (into which the shaft extends and which is not lined by syncytia) are homologous with the gymnostom of cephalobs, the sister taxon of tylenchids. Epidermal cells HypA, HypB, HypC and HypE line the cephalic framework, vestibule and vestibule extension, congruent with the hypothesis that these components are homologous with the cephalob cheliostom. HypC is expanded in *Aphelenchus*, which with sensilla fills most of the cephalic framework. Arcade syncytia are reduced with respect to cephalobs. Protractor muscles are homologous with the anterior most set of radial muscles. Observations to date test and verify our previous hypotheses of homology of the stomatostylet with respect to the stoma of bacterivorous outgroups. Reconstruction of the stegostom and pharynx will provide further tests of homology of the stomatostylet apparatus and associated feeding structures.

**APPLICATION OF PLURONIC GEL TO THE STUDY OF PLANT-NEMATODE INTERACTIONS. C. Wang, S. Lower, Q. Liu, V. M. Williamson** Nematology Department, University of California, Davis, CA, USA.

Pluronic gel F-127 is a stable and non-toxic compound that is widely used in medical and pharmaceutical fields. Pluronic gel is solid at room temperature and liquid at temperatures below 15°C when the gel concentration is about 20–30%. Nematodes can freely move inside the gel. We used 23% pluronic gel to study root-knot nematode (*Meloidogyne* spp.) attraction to different host plants. We found that tomato is the best attractant for *M. hapla* strain VW9 among bean, cowpea, *Medicago truncatula* and tomato. Attraction of strain VW9 to roots differs among four varieties of *M. truncatula*. Moreover, roots of 3-day-old seedlings are better attractants for VW9 than 1-week-old seedlings. The transparent gel system can be used to record nematode movement in the presence of the host plants and to compare attraction and infectivity among different strains or species of nematodes on one host. Movement of *M. javanica* to roots is much more rapid than that of *M. hapla*. This gel system is also applicable for study of the molecular biology of host-nematode interaction before and after the early stage of infection in vivo. We developed a rapid method to inoculate host roots with large amounts of nematodes in pluronic gel and to collect nematodes exposed to roots and those entering into the roots in the early infection stage. These stages will be used for cDNA library construction to identify genes involved in host perception of and signaling to the plant hosts.

DNA EXTRACTION FROM SOIL FAUNA. **S. Donn, B. S. Griffiths, R. Neilson, T. Daniell.** SCRI, Dundee, UK.

Molecular techniques offer an alternative to time-consuming traditional methods of faunal identification based on morphology. The first stage in developing a molecular technique is to have a robust method to extract DNA. Here methods are assessed using nematodes as a model group. A traditional DNA extraction, with proteinase K digestion followed by phenol chloroform extraction, sodium hydroxide extraction and physical disruption, followed by utilization of one of four propriety PCR purification kits, was tested for nematode extraction. Nematode communities were isolated from a range of habitats (arable agriculture, sand dune, coniferous forest, permanent pasture and organic rough pasture). Template DNA concentration was measured, and PCR-amplification performed to test the suitability of the extracts for downstream molecular applications. DNA extraction with phenol chloroform purification consistently yielded high quality template DNA as did the DNA extraction followed by the Purelink PCR purification kit. T-RFLP based on a single enzyme digest was sufficient to discriminate between nematode communities extracted from all five habitats. In addition, T-RFLP demonstrated that there was no difference in perceived nematode community composition following amplification of extract purified through either the Qiaquick or Purelink kits. Physical disruption of tissue followed by purification through a kit provides a rapid, reliable and relatively inexpensive method of DNA extraction, yielding high quality template. Application of high-throughput molecular techniques to soil microfauna increases their potential to be used as indicators in routine monitoring of soil health.

T-RFLP APPROACHES TO NEMATODE ASSEMBLAGE ANALYSIS. **S. Donn, T. J. Daniell, B. S. Griffiths, R. Neilson.** SCRI, Dundee, UK.

Intensive land use has been implicated in declining soil health, raising concerns over sustainability of agronomic production. Nematodes have been proposed as indicators of soil health; however, identification based on morphology is time-consuming and problematical. Alternative molecular approaches to profiling soil nematode assemblages have been applied here, based on Terminal Restriction Fragment Length Polymorphism (T-RFLP) of small subunit ribosomal DNA. Two approaches are described, the first entailing digestion of fluorescently labelled PCR product with a single enzyme, combined with multivariate analysis of the resulting fragment profile. Application of this method on agricultural sites under differing management regimes has revealed significant differences in assemblage composition with addition of compost to barley plots. The second approach utilizes a directed method where, from collected sequence information, a restriction digest has been designed to separate nematode taxa present at the study sites into terminal restriction fragments of known size. We envisage the resulting semi-quantitative profiles may be combined with existing nematode diversity indices, such as the Enrichment Index for characterizing soil, and other nematode assemblages.

EFFECTS OF TWO SUMMER CROPS AND POPULATION DENSITIES OF *MELOIDOGYNE ARENARIA* RACE 1 ON AMPLIFICATION OF *PASTEURIA PENETRANS*. **G. M. Kariuki, D. Dickson.** University of Florida.

*Pasteuria penetrans* isolate P-20 is known to cause soil suppressiveness to *Meloidogyne arenaria* race 1. *Pasteuria penetrans*, being an obligate parasite of *Meloidogyne* spp., has not been successfully cultivated in artificial media, thus there remains a need to develop improved methods for amplification of *P. penetrans* for research purposes. In this study, *P. penetrans* isolate P-20 was transferred from a suppressive field site to microplots located at the University of Florida, Gainesville. *Pasteuria penetrans* and its host nematode, *M. arenaria*, were established by growing susceptible hosts to the nematode during both summer and winter seasons of 2003. Thereafter, evaluations were done to determine the effect of two summer host crops and different levels of *M. arenaria* second-stage juveniles (J2) on amplification of *P. penetrans*. Three nematode densities, low, medium and high (4, 20 and 40 J2/100 cm<sup>3</sup> of soil, respectively) of *M. arenaria* were introduced in microplots. The densities were based on the top 30 cm depth of soil for each microplot. The crops tested were peanut (135-day crop) and squash (55-day crop) for two seasons. Peanut and squash foliage was removed after 135 and 55 days, respectively, and roots were cut back into the soil. Squash plots were reseeded in order to have two crop cycles of squash vs. one cycle of peanut. The most effective host crop and J2 density for amplification of *P. penetrans* was peanut plus high J2 density.

ISOLATION AND FUNCTIONAL ANALYSIS OF AN ANNEXIN-LIKE PARASITISM GENE OF THE BEET CYST NEMATODE, *HETERODERA SCHACHTII*. **N. Patel,<sup>1</sup> C. Li,<sup>1</sup> R. S. Hussey,<sup>2</sup> T. J. Baum,<sup>3</sup> E. L. Davis.<sup>1</sup>** <sup>1</sup>North Carolina State University, Raleigh, NC, USA, <sup>2</sup>University of Georgia, Athens, GA, USA, <sup>3</sup>Iowa State University, Ames, IA.

A putative annexin-like secretory protein (4FO1) of *Heterodera glycines* is being investigated in the related beet cyst nematode (BCN), *Heterodera schachtii*, to use the plant model *Arabidopsis thaliana* for parasitism gene function. A full-length 4FO1 cDNA clone has been isolated from BCN. mRNA in situ hybridization and immunolocalization confirm 4FO1 is expressed exclusively within the nematode's esophageal gland cells. Since it is hypothesized that parasitism proteins such as 4FO1 are secreted by the nematode into the host to modify plant cells for feeding, we investigated Hs-4FO1 annexin function using *A. thaliana* annexin knock-out mutants. Hypersensitivity to osmotic stress in Atann1 mutant was reduced (complemented) in transformed plants that expressed Hs-4FO1. BCN infection was slightly increased

in assays of Atann1 mutants compared to wild-type plants, while a slight decrease in BCN infection was observed on roots of the Atann4 mutant. RNAi interference (RNAi) is being investigated as a parallel functional approach to knock out target parasitism genes. Segregating lines of *Arabidopsis* expressing dsRNA of the 4FO1 parasitism gene had significantly reduced infection by BCN compared to BCN infection of roots of wild-type *Arabidopsis* plants, suggesting the potential for in planta RNAi to the 4FO1 gene as a means to develop transgenic cyst nematode resistance in plants.

**BEHAVIOR AS A MEASUREMENT OF EXPOSURE TO CHEMICALS IN A PLANT-PARASITIC NEMATODE. N. E. Schroeder.** University of Wisconsin-Madison, Madison, WI 53706.

Lethality is the traditional criterion used to evaluate the toxicity of chemicals. However, sub-lethal concentrations of toxic compounds may have a profound impact on an organism's behavior. Using *Heterodera glycines*, we developed a behavioral assay to test the toxicity of benzyl isothiocyanate (BITC), a plant-derived compound known to affect cyst nematodes. We hypothesized that movement, measured by tracks produced by the nematodes in agarose, would be significantly affected by sub-lethal concentrations of BITC. Nematodes were exposed to four different concentrations of BITC or the solvent control. After the exposure period, each individual nematode was transferred to its own Petri plate with 1.5% agarose. After 1 hour, nematodes were removed from the plate. Both the start and stop locations were noted and measured to give a net distance traveled. Plates were photographed for the sinusoidal tracks produced by the nematodes. Tracks were analyzed for the average amplitude and wavelength and the total distance traveled. Nematodes that did not move from the start location were examined for mortality. All behavioral parameters tested were significantly affected by the concentration of BITC and were more sensitive to dose than the LD<sub>50</sub>. We are currently using this method to examine factors affecting the ability of *H. glycines* to cope with chemical stress.

**THE ROLE OF ENVIRONMENTAL GRADIENTS IN THE DISTRIBUTION OF FREE-LIVING NEMATODES IN THE DEEP NORTHERN GULF OF MEXICO. J. Sharma,<sup>1</sup> J. G. Baguley,<sup>2</sup> R. N. Huettel.<sup>3</sup>** <sup>1</sup>University of Texas-San Antonio, San Antonio, TX 78249, <sup>2</sup>University of Nevada-Reno, Reno, NV 89557, <sup>3</sup>Auburn University, AL 36849.

Free-living nematode community structure has been analyzed at 14 stations in the northern Gulf of Mexico (GOM) deep sea. This semi-enclosed basin is a study of bathymetric, physical oceanographic and biological contrasts. The complex bathymetry of the northeastern Gulf of Mexico interacts with the anti-cyclonic Loop Current and Mississippi River outflow to create areas of enhanced nematode abundance. Conversely, the northwestern GOM is relatively benign in terms of slope topography, river outflow and physical oceanographic processes, resulting in comparatively modest nematode abundance. Depth and longitudinal gradients of nematode diversity were observed. Multivariate analyses reveal significant depth and longitude differences in the community composition of nematode genera. Similarly, a significant difference in nematode feeding morphology was observed between depth groups, but not between longitudinal groups. An increase in average taxonomic diversity with depth may explain the observed difference in feeding morphology and may suggest proportionally more higher-order taxa and trophic complexity with depth. These data offer further insight into processes structuring communities and species distributions in the deep sea.

**SOYBEAN CYST NEMATODE POPULATIONS SUPPRESSED BY *PASTEURIA NISHIZAWAE*. G. R. Noel,<sup>1</sup> S. Bauer,<sup>2</sup> N. Atibalentja.<sup>2</sup>** <sup>1</sup>USDA, ARS, Urbana, IL, <sup>2</sup>Dept. of Crop Sciences, Univ. of Illinois, Urbana, IL.

*Pasteuria nishizawae* is a mycelial and endospore-forming, obligately parasitic bacterium of *Heterodera glycines*. The effectiveness of *P. nishizawae* in controlling *H. glycines* was investigated in an ongoing tillage and genetic resistance study by adding *P. nishizawae*-infested soil to three of six replications in 1999. Soybean yield and Pi and Pf of *H. glycines* were determined during 1999–2005. Yield of the resistant cultivar was greater than the susceptible cultivar in 1999–2004, in contrast with 2005, when yield of the susceptible cultivar was greater. Differences in yield were associated with *P. nishizawae* infestation and interactions with tillage in 1999–001. Pi was affected by *P. nishizawae* in 2001–2005, and Pf was affected in 2000–2005. In 2005, a dramatic decline in Pf occurred on the susceptible cultivar, which was associated with higher yield when compared with the resistant cultivar. In this study, *P. nishizawae* was successfully transferred to a field experiment and became suppressive to *H. glycines*, resulting in an increase in soybean yield.

**EFFECTS OF SEVERAL POPULATIONS OF SUNN HEMP ON NEMATODES. S. Marla,<sup>1</sup> J. Mosjidis,<sup>2</sup> R. Huettel.<sup>1</sup>** <sup>1</sup>Department of Entomology and Plant Pathology, Auburn University, AL, <sup>2</sup>Department of Agronomy and Soils, Auburn University, AL 36849.

Sunn hemp (*Crotalaria* spp.), when used as a cover crop, has been demonstrated to suppress some plant-parasitic nematodes under field conditions. Usage of this crop in Alabama and other southern states is limited by seed availability. A breeding program aimed at developing seed-producing cultivars adapted to the continental US is being conducted at Auburn University. Five of these populations of Sunn hemp were evaluated against the root-knot nematode, *Meloidogyne incognita*, and the reniform nematode, *Rotylenchulus reniformis*, in the greenhouse. Significant differences in reproduction



rates were found between both the nematodes and the Sunn hemp populations. However, all populations of Sunn Hemp did significantly reduce the number of nematodes as compared to the controls. Mixed plantings of the Sunn hemp populations with either tomato with root-knot nematodes or cotton with reniform nematodes are being evaluated to determine if cross protection might occur. The effect of root exudates of Sunn hemp populations on reniform and root-knot nematodes is also being evaluated using axenic cultures. The effect of resistance to these nematodes by populations of Sunn hemp is important in selection of new populations for potential commercial use.

**SURVEY OF NEMATODE COMMUNITIES ASSOCIATED WITH *JUNIPERUS ASHEI*. G. Young,<sup>1</sup> J. Sharma,<sup>2</sup> J. Bush,<sup>1</sup> R. Huettel.<sup>3</sup>** <sup>1</sup>Departments of Earth and Environmental Science and <sup>2</sup>Biology, University of Texas-San Antonio, San Antonio, TX 78249, <sup>3</sup>Department of Entomology and Plant Pathology, Auburn University, AL 36849.

*Juniperus ashei* Buchholz, also known as Ash juniper, is native to the limestone slopes of Central Texas. Grass and forb production and species diversity are greatly reduced with increasing canopy cover and litter accumulations of Ash juniper, possibly due to competition for water, nutrients and sunlight. It has been suggested that allelopathy is exhibited. Previous studies have identified camphor, which can be potentially allelopathic, in the essential oils of Ash juniper at a level as high as 68.5%. GC/MS analysis of fresh leaves, leachate produced from leaves and litter, and soils taken from locations associated with Ash juniper confirm the presence of several volatile compounds, including camphor. The nematode faunal composition has emerged as a useful monitor of environmental conditions and ecosystem function of soil. The presence of Ash juniper may have the potential to alter nematode communities directly and/or indirectly. This research surveys the nematode communities at four locations (understory, dripline, intercanopy space and within the litter) associated with three Ash juniper trees located near San Antonio, Texas. Total nematode numbers as well as numbers per trophic group (bacterivore, fungivore, plant-parasitic and predator) were evaluated. Total numbers and species abundance varied by location, possibly due to an allelopathic effect. Confounding factors such as soil moisture and temperature were also considered.

**PANAGROLAIMUS MITOCHONDRIAL GENOME EVOLUTION: IMPACT OF REPRODUCTIVE MODE. S. C. Lewis, D. R. Denver.** Oregon State University, Corvallis, OR, USA.

Many evolutionary models suggest that natural selection is weaker in asexual organisms relative to sexual organisms; the absence of recombination and linkage of heritable material could increase susceptibility to deleterious mutation accumulation in both nuclear and organelle genomes. It is thought that few asexual lineages persist through evolutionary time compared to sexual lineages; consequently, the role of reduced selection efficiency in their demise remains an important debate. To investigate the effects of reproductive mode on deleterious mutation accumulation, we analyzed the mitochondrial genomes of two free-living nematodes of the genus *Panagrolaimus*: strain PS1579, an obligate parthenogen, and its recent sexual ancestor, strain AF36. Mitochondrial DNA from both strains was sequenced using a PCR/primer-walking approach. The mitochondrial genomes of these strains differ in many regards—most notably, PS1579 appears to lack a tRNA-Arg gene at the usual genomic location conserved in AF36 and most other nematodes. This potentially “missing” tRNA might have arisen as a consequence of deleterious mutation accumulation associated with its reproductive mode. Panagrolaimids are closely related to *Strongyloides stercoralis* which exhibits a highly divergent mitochondrial gene order. Both *Panagrolaimus* strains studied here contained the *C. elegans*-like gene order, implying that the rearrangements found in *S. stercoralis* may be very recent and lineage-specific.

**LONG-TERM EFFECTS OF WEED MANAGEMENT PRACTICES AND COVER CROPPING ON THE NEMATODE COMMUNITY IN A COMMERCIAL VINEYARD IN THE SALINAS VALLEY, CALIFORNIA. S. R. Parker, D. A. Kluepfel.** USDA, Agricultural Research Service.

Little is known about the effects vineyard floor management practices have on nematode population in the berm or row middles. This investigation was undertaken to examine how the total nematode community in vineyards is influenced by long-term weed management and cover crop practices and to determine the impact of row middle management strategies on nematode population in the berm. Six years after the establishment of weed management strategies in row middles (cultivation, pre-emergence herbicide, post-emergence herbicide) and cover-crop treatments (rye and bare) in a conventionally managed vineyard, nematode populations were examined. Sampling was conducted at three phenological stages of the vines, early berry formation, harvest, and dormancy. Samples were harvested from under the irrigation emitter on the berm and 0.5 m into the row. Both quantitative and qualitative differences in nematode population were detected as a function of extraction method. In addition, the total nematode populations were influenced by weed management and cover crop presence; however, the effects were influenced by sample location. The spatial distribution of plant-parasitic genera, especially *Mesocriconema*, suggests little effect of row management on the occurrence of this trophic group in the berm.

ISOZYME PHENOTYPES AND IDENTIFICATION OF *MELOIDOGYNE* SPP. PARASITIZING AGRONOMIC AND HORTICULTURAL CROPS AND WEEDS IN FLORIDA. **J. A. Brito,<sup>1</sup> R. Kaur,<sup>2</sup> R. Cetintas,<sup>3</sup> J. D. Stanley,<sup>1</sup> E. J. McAvoy,<sup>4</sup> T. O. Powers,<sup>5</sup> M. L. Mendes,<sup>2</sup> D. W. Dickson.<sup>2</sup>** <sup>1</sup>Division of Plant Industry, Gainesville, FL 32614, USA, <sup>2</sup>Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611, USA, <sup>3</sup>Department of Plant Protection, University of Kahramanmaraş Sutcu Imam, Kahramanmaraş, 46060, Turkey, <sup>4</sup>Vegetable/Ornamental Horticulture, University of Florida, Labelle, FL 33975, USA, <sup>5</sup>Department of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722, USA.

Isozyme phenotypes esterase (EST) and malate dehydrogenase (MDH) were used to identify *Meloidogyne* spp. from Florida. Root samples were examined from horticultural (180) and agronomic (65) crops and from weeds (61). From horticultural crops, *M. incognita* (25%) was the most prevalent species, followed by *M. javanica* (13%), *M. arenaria* (8%), *M. mayaguensis* (8%), *M. floridensis* (5%), *M. hapla* (0.6%), *M. partityla* (0.6%) and *Meloidogyne* spp. (11%). The species identified from agronomic crops were *M. arenaria* (37%), *M. javanica* (23%), *M. incognita* (8%) and *Meloidogyne* spp. (9%), whereas from the weed samples *M. incognita* (15%), *M. arenaria* (12%), *M. mayaguensis* (6%), *M. javanica* (3%), *M. graminicola* (2%), *M. floridensis* (1%) and *Meloidogyne* spp. (16%) were identified. Mixed populations were found in 14% of the samples. EST phenotypes provided fast and reliable diagnostic information for identification of *Meloidogyne* spp. from Florida. The species-specific EST phenotypes Mf3 (*M. floridensis*) and Mp3 (*M. partityla*) distinguished these species from other *Meloidogyne* spp. Although MDH is helpful in the separation of *M. partityla* from *M. graminicola* and *M. mayaguensis*, it had a lower diagnostic value than EST because the MDH phenotype (N1) from *M. partityla* was identical to that of *M. arenaria*, *M. javanica*, *M. floridensis* and *M. incognita*.

TREATMENT OF WATER WITH PULSED ULTRAVIOLET LIGHT FOR INACTIVATION OF NEMATODES. **J. L. Haynes,<sup>1</sup> K. R. Sanchez,<sup>1</sup> M. C. Lagunas-Solar,<sup>2</sup> S. A. Nadler,<sup>1</sup> C. Pina,<sup>2</sup> E. P. Caswell-Chen.<sup>1</sup>** <sup>1</sup>Department of Nematology and <sup>2</sup>Crocker Nuclear Laboratory, University of California, Davis, Davis, CA, USA.

Plant-parasitic nematodes may be moved in irrigation water, and the need to recycle water may exacerbate this problem. Treatment with pulsed-ultraviolet light (PUV) is being evaluated as a non-chemical, residue-free method of controlling nematodes in water. Nematodes were exposed to a range of UV doses with a flashlamp-type (Xe-filled), non-coherent pulsed UV/Vis source operating with 200–300 J/pulse (~18% UV and ~82% visible, no IR) and 1–3 Hz (pulses per second). All exposure doses were measured with a calibrated pyroelectric joulemeter (9.02 V/Joule) and expressed as mJ/cm<sup>2</sup>/pulse. Bioassay nematodes were exposed to a range of pulses of UV/Vis light and to UV-filtered beam containing only the visible light spectrum. Nematodes were examined after exposure and 24 hr post-exposure for delayed mortality. PUV at 372 mJ/cm<sup>2</sup> inactivated *Meloidogyne javanica* juveniles. Inactivation of *Aphelenchoides fragariae* required 425 mJ/cm<sup>2</sup>, *Caerhabditis elegans* 460 mJ/cm<sup>2</sup> and *Acrobeloides buetschii* 407 mJ/cm<sup>2</sup>. UV-filtered exposure experiments showed that the UV component of the emission spectrum was solely responsible for nematode mortality. PUV may be effective for controlling nematodes in water.

OZONATION FOR CONTROL OF NEMATODES IN IRRIGATION WATER. **K. R. Sanchez,<sup>1</sup> J. L. Haynes,<sup>1</sup> A. Pryor,<sup>2</sup> E. P. Caswell-Chen.<sup>1</sup>** <sup>1</sup>Department of Nematology, University of California, Davis, Davis, CA, USA, <sup>2</sup>Ozone Process Consultants Inc., Davis, CA, USA.

To control the spread of pest nematodes, methods are needed to kill nematodes in irrigation water. When injected into water, ozone is a powerful oxidizing agent and is fungicidal and bactericidal. We assessed the efficacy of ozone for killing nematodes by injecting it into recirculating tap water or recycled irrigation water containing bioassay nematodes. Ozone gas was injected at different rates into clean water resulting in equilibrated dissolved ozone concentrations of 0.08 ppm, 0.18 ppm, 0.215 ppm and 0.38 ppm after 5 min. Bioassay nematodes were *Caenorhabditis elegans*, *Aphelenchoides fragariae*, *Heterodera schachtii* and *Meloidogyne* species. Nematode survival was monitored over time to establish toxicity relative to ozone concentration and exposure time (concentration X time, or CT value). In clean water, *A. fragariae* became inactive in 20 min at a CT value 7.8 ppm/min and *M. incognita* in 15 min at a CT value 3.6 ppm/min. The CT values required to consistently obtain 90% kill ranged from 2–8 ppm/min, regardless of nematode species. Higher exposure CT values were required for inactivation in recycled irrigation water. *Meloidogyne javanica* became inactive in 75 min at a CT value of 12 ppm/min and *C. elegans* in 30 min at a CT value of 1.5 ppm/min. Ozonation can be an effective method for controlling nematodes in water and may be viable in a commercial setting.

DEFINING NEMATODE MANAGEMENT ZONES IN COTTON. **C. Overstreet,<sup>1</sup> M. Wolcott,<sup>1</sup> G. B. Padgett,<sup>2</sup> E. Burris.<sup>3</sup>** <sup>1</sup>LSU Agcenter, Baton Rouge, LA, USA, <sup>2</sup>LSU Agcenter, Winnsboro, LA, USA, <sup>3</sup>LSU Agcenter, St. Joseph, LA, USA.

Much of the cotton production areas in Louisiana include fields which have highly variable soil texture within or across these fields with areas where the southern root-knot nematode (*Meloidogyne incognita*) does not occur or is not causing

damage. Applying a nematicide to only the areas of a field which will give an economical response would benefit both the producer and environment. The fumigant 1,3-dichloropropene was applied in four to eight strips (12 to 16 rows each) in six production fields that were infested with root-knot nematode. Each field was mapped for soil electrical conductivity using a Veris 3100 Soil EC Mapping System to delineate similar soil texture areas and divided into three to seven zones. Yield was obtained using yield monitors on either 4- or 6-row cotton pickers. The locations at Railroad Cut, Spyker, Faulk, Levee, Cemetery North and Perry Cutoff averaged a significant response to the nematicide in 100, 66, 66, 57, 33 and 17% of the zones, respectively, compared to the untreated control. The application of a fumigant through the various soil zones within a field proved to be very effective in showing areas of a field which would respond to a nematicide. Management zones could be established for very site-specific application of fumigant nematicides.

**DIFFERENCES IN INDUCED NEMATICIDE-RESISTANCE BETWEEN FREE-LIVING AND PLANT-PARASITIC NEMATODES. Y. Lin, T. Tsay, P. Chen.** Dept. of Plant Pathology, National Chung Hsing University.

Nine of the 11 populations of *Rhabditis* sp., but only two out of 12 populations of *Aphelenchoides besseyi*, which were induced to become resistant to ethoprop, phenamiphos, carbofuran or oxamyl, lost their resistance after one year in the absence of nematicides. Two free-living nematodes, *Caenorhabditis elegans* and *Rhabditis* sp., and three plant-parasitic nematodes, *A. besseyi*, *Bursaphelenchus xylophilus* and *Bursaphelenchus* sp., were treated with various concentrations of nematicides mentioned above to induce nematicide resistance. All the five species tested were more sensitive to organophosphate nematicides than carbamate nematicides. Sixty-two nematode populations with different levels of nematicide resistance were obtained. Our results also showed that *C. elegans* was more resistant to all four nematicides tested than *A. besseyi*, *B. xylophilus* and *Bursaphelenchus* sp. The resistance level of *Rhabditis* sp. to these nematicides was about the same as that of *A. besseyi*, but was higher than that of *B. xylophilus* and *Bursaphelenchus* sp. Among the five nematode species tested, the two free-living nematodes were more resistant to these four nematicides than the three plant-parasitic nematodes. Moreover, *B. xylophilus* was more resistant to the tested nematicides than *Bursaphelenchus* sp.

**INFLUENCE OF ENVIRONMENTAL FACTORS ON *HIRSUTELLA MINNESOTENSIS* MONITORED QUANTITATIVELY BY REAL-TIME PCR AND PARASITISM ASSAY. M. Xiang,<sup>1</sup> X. Liu,<sup>1</sup> Q. Xiao.<sup>2</sup>** <sup>1</sup>Key Laboratory of Systematic Mycology and Lichenology, Institute of Microbiology, Chinese Academy of Sciences, Beijing 100080, <sup>2</sup>Department of Plant Pathology, Hunan Agricultural University, Changsha 410128, China.

*Hirsutella minnesotensis* is a nematode endoparasite and has shown a great potential for biocontrol of soybean cyst nematode. Based on the sequence of ITS region of the rRNA gene, a highly species-specific real-time PCR assay using a TaqMan probe for detection and monitoring the fungus in soil was developed. The amount of fungus detected was as few as 4 conidia/g soil by the real-time PCR system after optimization. The assay was successfully applied to monitor the fungus in 20 soil samples which were collected from soybean monoculture fields in Heilongjiang, China. The effect of soil temperature, water content and texture on the colonization and activity of *H. minnesotensis* in soil was investigated by the real-time PCR assay and the parasitism bioassay. The fungus was added into soil at 1000 mg/100 g dry soil and placed into 50 ml plastic tube. After 24 days of incubation, the highest amount of *H. minnesotensis* DNA was detected at 5–10°C and 6% water content and then drastically declined, while the high percentage of *Heterodera glycines* J2 parasitized by *H. minnesotensis* occurred at 10°C and 5°C, 6% and 10% water content. Both DNA amount and parasitism of *H. minnesotensis* were high in the clay soils. The fungal DNA yield was reduced in the soils at high temperature, high soil moisture and high amount of sand, indicating that the fungus did not survive well under these soil conditions.

**SOYBEAN CYST NEMATODE AND SOYBEAN APHID INTERACTIONS ON SOYBEAN. F. Avendano, M. E. O'Neal, G. L. Tylka.** Iowa State University, Ames, IA.

Individually, the soybean cyst nematode (SCN), *Heterodera glycines*, and the soybean aphid (SBA), *Aphis glycines*, can reduce soybean yield by 30–50%. Their common geographical distribution and overlap during the growing season create potential for interactions, which are not understood or considered when management decisions are made. A field experiment was conducted in microplots infested and not infested with SCN to test interaction effects with SBA on five soybean varieties resistant or susceptible to SCN (8 replications). Ten aphids were placed on individually caged plants early in vegetative growth of the plants and were counted every 3 days for 2 weeks. No-SBA/no-SCN plots served as a control. SCN population densities were not significantly different among treatments at planting, but densities were greater on SCN-susceptible than on SCN-resistant varieties at harvest. No significant interactions were detected between SCN and SBA population densities. Plant height was reduced by both pests, causing greater density of aphids per plant in SCN plots than in no-SCN plots. Seed weight of SCN-resistant varieties was not affected by SCN, but was significantly reduced by SBA. Conversely, seed weight of SCN-susceptible varieties was not affected by SBA, but was significantly reduced by SCN. Soybean aphid management may need to be adjusted when SCN is present.

COMPARISON OF OOGENESIS IN *MELOIDOGYNE HAPLA* IN THE PRESENCE AND ABSENCE OF FERTILIZATION. **V. P. Thomas, V. M. Williamson.** University of California, Davis, CA, USA.

*Meloidogyne hapla* (northern root-knot nematode) is an important pest of tomato, potato, carrot, alfalfa and other crops in cooler regions. The mode of reproduction in most isolates of *M. hapla* is facultative meiotic parthenogenesis. We microscopically investigated female gonads from *M. hapla* after staining with Hoechst 33258. It was observed that the oogonium divides mitotically in the germinal zone. In the growth zone, the oocyte grows progressively in size then enters the meiotic transition zone, characterized by germ cells having condensed and crescent-shaped nuclei. Anterior to spermatheca, the primary oocytes gradually proceed through the early stages of prophase I. Close to the spermatheca, the primary oocytes are arrested in diakinesis. In both sexual reproduction and parthenogenesis, the spatial progression of meiosis in the ovary appears to be the same up to entry into the spermatheca. However, progress through meiosis II and egg development differ, depending on whether sperm are present in the spermatheca. When sperm are present, the oocyte is triggered to rapidly complete meiosis, and its haploid nucleus fuse with sperm nucleus. The diploid polar body I and haploid polar nucleus II produced during meiosis I and II, respectively, are disintegrated or expelled. The one-cell embryo is produced just posterior to the spermatheca, and embryonic cell divisions are initiated. However, in parthenogenesis, the oocytes remain in diakinesis post spermatheca. Eventually in the proximal arm of the gonad, the oocyte leaves diakinesis and progresses to a prolonged metaphase I. In the posterior portion of the uterus, the oocytes complete meiosis I, producing a diploid polar body and an egg-pronucleus. This is immediately followed by meiosis II with a prolonged telophase II. The two telophase II sets of haploid chromosomes rejoin, restoring the ploidy level. The one-celled embryo is observed far posterior to the spermatheca.

TRIXENIC CULTURES TO STUDY POPULATION SUPPRESSION OF THE BEET CYST NEMATODE IN *ARABIDOPSIS THALIANA* BY *DACTYLELLA OVIPARASITICA*. **J. Smith-Becker, J. Borneman, J. O. Becker.** University of California, Riverside, CA, USA.

An *A. thaliana* model system was developed for the study of the mode of action of the biocontrol agent *Dactylella oviparasitica* against the beet cyst nematode, *Heterodera schachtii*. Four weeks after nematode infestation, *A. thaliana* (type Landsberg *erecta*) grown in sandy soil in the presence *D. oviparasitica* had a 70% reduction in the number of mature cysts compared to control plants. A second system enabled the direct observation of the interaction of the fungus with developing juveniles in agar plate cultures. At 22°C, the majority of developing juveniles broke through the root epidermis 17 days after infestation with *H. schachtii*. Hyphae of *D. oviparasitica* were not attracted to nematode-free roots but rapidly colonized the juveniles as they began to emerge from the root surface. Parasitism of 4<sup>th</sup>-stage juveniles terminated further development into adult males and females. These observations suggest that parasitism of developing juveniles may be the essential mode of action in the population suppression of *H. schachtii*.

UNDERSTANDING A ROOT-KNOT NEMATODE SUPPRESSIVE SOIL. **A. Loffredo, E. Bent, M. V. McKenry, J. Borneman, J. O. Becker.** University of California, Riverside, CA, USA.

Pathogen suppressive soils may offer substantial potential for discovering biological control organisms and for understanding their ecological requirements to compete successfully with other microbes. Although root-knot nematodes (*Meloidogyne* spp.) are frequently parasitized or destroyed by various soil organisms, biological inhospitability of soil to these important plant parasites has rarely been reported. Soil samples from various field locations in California were analyzed for *M. incognita* population suppression in two greenhouse assays, each with wheat and dwarf tomato. In one soil, the root-knot nematode population was consistently reduced after 8 weeks compared to its pasteurized, nematode-infested control. The biological nature of this phenomenon was confirmed by elimination of the suppression through heat or soil fumigation with methyl iodide as well as by transfer and establishment of suppressiveness to conducive soil. An analysis of *M. incognita* egg masses identified an rRNA gene with high sequence identity to *Pochonia chlamydosporia* associated with this suppressive soil. Strains of this fungus were obtained by media-based isolations.

EVOLUTIONARY ECOLOGY OF AGING IN *CAENORHABDITIS ELEGANS*: NURTURE AND NATURE. **E. P. Caswell-Chen,<sup>1</sup> H. Caswell.<sup>2</sup>** <sup>1</sup>Univ. of California, Davis, CA, USA, <sup>2</sup>Woods Hole Oceanographic Institution, Woods Hole, MA, USA.

*Caenorhabditis elegans* is a well-characterized animal and is a model system in aging research. Many mutations altering life span have been identified in *C. elegans*. The evolution of senescence, and hence life span and aging, is considered a result of the generally decreasing force of selection on traits with age. Evolution is driven by the age- or stage-trajectories of selection gradients on demographic traits (age- or stage-specific survival probabilities and fecundities). The selection gradient, or force of selection, on traits can be calculated, and we used age-based matrix projection models to assess selection gradients on demographic traits of wild-type (N2), longevity mutants and wild-caught worms as expressed under laboratory conditions. The selection gradient on the force of mortality  $\mu(x)$  generally decreases with age, a phenomenon consistent with the evolution of senescence. Standard, nurturing lab conditions are not representative of *C. elegans*' natural

environment—biotic and abiotic factors yield vital rates different from lab rates. We altered vital rates by modifying the timing of mortality, making mortality intermittent and shifting fertility to assess resultant changes in selection gradients on the force of mortality. We observed that selection gradients were plastic, age-specificity of mortality was important, early mortality reduced senescence and intermittent impacts could have large effects.

THREE-DIMENSIONAL RECONSTRUCTION OF EPIDERMAL AND SENSORY ORGANS IN THE NEMATODE *ACROBELES COMPLEXUS*. **D. J. Bumbarger,<sup>1</sup> J. Crum,<sup>2</sup> M. Ellisman,<sup>2</sup> J. G. Baldwin.<sup>1</sup>** <sup>1</sup>University of California, Riverside, CA, <sup>2</sup>University of California, San Diego, CA.

The model organism *Caenorhabditis elegans* has great potential as a resource for understanding problems in other nematode systems, including questions about the biology of the plant-parasitic Tylenchida. Some knowledge of genetics, development and sensory and feeding function of *C. elegans* is currently difficult to apply to Tylenchida. The highly specialized feeding morphology of Tylenchid nematodes, as well as their great phylogenetic distance with respect to *C. elegans*, makes it challenging to assess phenotypic homology. An assessment of cellular-level homology in a more closely related outgroup of the Tylenchida with less specialized morphology would contribute significantly to our ability to leverage the *C. elegans* model for asking questions about plant parasites. Herein we have applied recent advances in electron microscopy to reconstruct the epidermal and sensory tissues in the anterior tip of a bacterial-feeding nematode (*Acrobeles complexus*) that belongs to Cephalobinae, the sister taxon to the Tylenchida. We propose robust hypotheses of homology for most cells in the anterior tip between *C. elegans* and *A. complexus* and have shown that specific cell identities are conserved across vast phylogenetic distances. Additional work is underway to further extend these hypotheses of homology to plant-parasitic Tylenchida and other nematode groups.

VARIABILITY IN MORPHOLOGY, GENETICS AND BIOLOGY OF NEMATODES AND EVOLUTION OF NEW TAXA. **M. E. Hodda.** Nematode Biosystematics & Ecology Australian National Insect Collection.

The evolution of new species, pathotypes or resistance-breaking races of plant-parasitic nematodes has major consequences for crop management and breeding, yet is little understood. A major study of hundreds of specimens and sequences from more than 30 populations of 20 species from the genus *Pratylenchus* was conducted to test the relationships between variability in morphology, genetics and biology. A range of parameters within each of these categories was measured, such as body length, diameter, various morphometric indices, different gene loci, host responses and population responses to culture conditions. The amount of variability in the different parameters was seldom correlated within putative species or populations, but there were often thresholds dividing “species.” This suggests that species barriers were real and that certain models of evolution may be operating. There were also clusters of populations with higher levels of internal overlap in many parameters than among most of the other populations studied. These may represent centers of evolutionary activity. Why these particular clusters have these properties raises important hypotheses for evolution in nematodes.

AN ALKALINE SERINE PROTEASE AND GENE CLONING FROM A HIGH VIRULENT STRAIN OF NEMATODE-ENDOPARASITIC FUNGUS *HIRSUTELLA RHOSILIENSIS*. **B. Wang,<sup>1</sup> W. Wu,<sup>2</sup> X. Liu,<sup>1</sup> S. Li.<sup>3</sup>** <sup>1</sup>Key Laboratory of Systematic Mycology and Lichenology, Institute of Microbiology, Chinese Academy of Sciences, Beijing 100080, <sup>2</sup>R/D Novozymes, Demark, <sup>3</sup>Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing 100081, China.

*Hirsutella rhossiliensis* OWVT-1 has shown high biocontrol efficacy for plant-parasitic nematodes. An extracellular alkaline protease (Hasp) was purified from this fungal liquid culture with the nematode *Panagrellus redivivus* as the sole nitrogen source. Bioassays showed that Hasp could immobilize the juveniles of soybean-cyst nematodes and degrade proteins of the nematode cuticle. The molecular mass of Hasp was estimated to be 33 kDa by SDS-PAGE. The optimum activity was at pH 9 and 75°C. The *Hasp* gene consists of 1,170 bp open reading frame including four exons (279, 186, 513 and 192 bp, respectively) and three introns (65, 99 and 93 bp, respectively) and encodes a polypeptide of 389 amino acid residues. Southern blotting indicated that *Hasp* was a single-copy gene in genome. The deduced amino acid sequence showed a high degree of similarity to other serine proteases of nematode-parasitic fungi and entomopathogenic fungi. Phylogenetic analysis of the amino acid sequences of serine protease showed that *H. rhossiliensis* OWVT-1 could be clustered with nematode/egg parasites rather than the trapping fungi.

OCCURRENCE OF PINE WILT DISEASE, CAUSED BY *BURSAPHELENCHUS XYLOPHILUS*, FROM *PINUS KORAIENSIS* IN KOREA. **Y. Moon, H. Cheon, S. Lee.** PWD Control Research Center, Southern Forest Research Center, Korea Forest Research Institute, Jinju 660–701, Korea.

Pine wilt disease (PWD), which is caused by the pine wood nematode (PWN), *Bursaphelenchus xylophilus*, is one of the most serious diseases of pine forests in Japan and other countries. In October of 1988, pine wilt disease was first discovered in Busan, Korea. During the past 18 years, the only known susceptible hosts were Japanese red pine, *Pinus densiflora*, and Japanese black pine, *Pinus thunbergii*, in southern Korea. Recently, the disease broke out on several Korean

pinus, *Pinus koraiensis*, in unexpected areas of central Korea (Kwangju and Chuncheon). Although *P. koraiensis* was susceptible to the disease only in laboratory inoculation tests, the recent occurrence was the first report on the Korean pines naturally infected by PWN in Korea. However, comparative studies using morphological and genetic analysis on the pathogenic nematode isolates between Korean pine and two Japanese pines have shown the same species of PWN, *B. xylophilus*. Despite an effort to control PWD by the government, new infection of pine species, *P. koraiensis*, by PWD occurred in 2006. This occurrence awakens us to the fact that this disease could occur in any place merely by insect vector but also by human activity, even in areas far from infected places.

**SURVIVAL OF *LYSOBACTER ENZYMOGENES* STRAIN C3 ON *BETA VULGARIS*.** J. Chen,<sup>1</sup> G. Y. Yuen,<sup>2</sup> D. Y. Kobayashi,<sup>3</sup> K. R. Sanchez,<sup>1</sup> E. P. Caswell-Chen.<sup>1</sup> <sup>1</sup>Department of Nematology, University of California, Davis, CA 95616, <sup>2</sup>Department of Plant Pathology, University of Nebraska, Lincoln, NE 58588, <sup>3</sup>Department of Plant Biology & Pathology, Rutgers University, New Brunswick, NJ 08901.

The colonization of plant roots by the nematode-antagonist *Lysobacter enzymogenes* was investigated using strain C3 on *Beta vulgaris*. *Lysobacter enzymogenes* was incubated in nutrient broth at 26°C for 3 days yielding a population density of approximately  $1.6 \times 10^9$  cfu/ml. The harvested pellet was diluted in artificial tap water (ATW) or half-strength Hoagland's nutrient solution (HNS) before drench application with either  $1.6 \times 10^9$  (1X) or  $1.6 \times 10^{10}$  (10X) cells/plant. Individual seedlings, Beta seed commercial hybrid B4430R, were planted in sterilized sand. The root system was macerated in 10 ml phosphate buffer using an AgDia tissue grinder, and the population density of bacteria on roots was determined with an 8-spot plating method. After 35 days, there were  $1.5 \times 10^7$ ,  $7.0 \times 10^7$ ,  $2.7 \times 10^7$  and  $5.2 \times 10^7$  cells/plant in the treatments of 1X ATW, 10X ATW, 1X HNS and 10X HNS, respectively, compared to  $5.5 \times 10^6$ ,  $1.1 \times 10^8$ ,  $1.4 \times 10^8$  and  $7.3 \times 10^7$  on day 21. We will report on plant weights and longer-term colonization of plant roots systems.

**OCCURRENCE OF ROOT-KNOT NEMATODES IN SWITZERLAND.** S. Kiewnick, R. Eder, I. Roth, M. Oggenfuss, B. Frey, J. Frey. Agroscope Changins-Waedenswil ACW, Research Station ACW, CH-8820 Waedenswil, Switzerland.

Root-knot nematodes (*Meloidogyne* spp.) are the most important group of plant-parasitic nematodes in Switzerland causing significant problems in vegetable production areas. The most common root-knot nematode species is *Meloidogyne hapla*, followed by the more tropical and subtropical species *M. incognita*, *M. javanica* and *M. Arenaria*, which are found in greenhouses only. In a survey conducted from 2002 through 2006, the quarantine nematodes *M. chitwoodi* and *M. fallax* were detected in a few greenhouses in the western part of Switzerland. However, these quarantine nematodes are confined to these greenhouses only and have not spread further. In addition, in two locations a population identified as *M. mayaguensis* was found. Species identification was based on ITS-1 and COI DNA sequence data. However, comparison with reference material from other *M. mayaguensis* and *M. enterolobii* populations is still needed to verify the presence of this species in Switzerland.

**FATE OF BENZOXAZINOIDS FROM RYE (*SECALE CEREALE*) IN SOIL AND THE IMPLICATION ON PLANT-PARASITIC NEMATODE MANAGEMENT.** I. A. Zasada, C. Rice, S. L. Meyer. USDA-ARS, Beltsville, MD, USA.

A rye (*Secale cereale*) cover crop plays an important agronomic role in southern and eastern United States crop rotation systems. The contribution of this cover crop to plant-parasitic nematode management is unclear. To understand the potential of consistently utilizing this cover crop for plant-parasitic nematode management, the fate and partitioning coefficients of the benzoxazinoids DIBOA (2,4-dihydroxy-(2H)-1,4-benzoxazin-3(4H)-one) and DIMBOA (2,4-dihydroxy-7-methoxy-(2H)-1,4-benzoxazin-3(4H)-one) were determined. Both compounds were spiked into three soil types: a sandy loam, silt loam and loam. The concentration of the compounds in soil solution as well as in sediment was determined at 0, 1, 2, 4 and 8 hours and 1, 2, 3, 4 and 5 days. Availability and exposure times of DIBOA and DIMBOA in soil varied with soil type. This research demonstrates that the exposure potential of these compounds to plant-parasitic nematodes will vary with soil type, suggesting that a rye cover crop will differ in effectiveness for nematode management depending upon soil type.

**PROTEINS FOR THE CONTROL OF PLANT-PARASITIC NEMATODES.** J. Thissen, T. Kahn, E. Pickle, B. Villano, J. Cao, H. Furcillo, C. Peters, B. Vande Berg, D. Tomso, N. Desai, N. Duck. Athenix Corporation, Research Triangle Park, NC, USA.

Parasitic nematodes cause enormous damage to crop plants worldwide every year. Existing chemical pesticide solutions provide only partial control and are both laborious and expensive to use. Control of nematodes via transgenic expression of nematicidal proteins in crops is an attractive alternative, but has been difficult to establish for obligate parasites such as the cyst nematodes (*Heterodera* sp.). We have developed an in vitro feeding assay for cyst nematodes and have used it to discover microbial protein toxins with activity against important plant parasites. Extracts from microbial cultures were tested for activity against nematodes using a high-throughput assay. Active extracts were fractionated, and the active

proteins identified by matching protein sequence data to the Athenix microbial gene database, allowing for rapid cloning of the active genes. Additional homologs of active genes were identified by hybridization and PCR screening, as well as by comparison to our microbial gene database. Novel pesticidal genes were expressed and tested in our in vitro feeding assay, and selected genes were moved into plants for evaluation against nematodes. In our transgenic model, these newly identified nematocidal proteins significantly inhibit *Heterodera* cyst formation and have the potential to provide excellent control of nematode infections in crop plants.

**NEMATOCIDAL EFFECT OF THE TOXIN PRODUCED BY *BACILLUS THURINGIENSIS* (BT.) AGAINST *MELOIDOGYNE INCOGNITA*. S. I. Massoud, T. S. Abd-El-Moneim.** Suez Canal University, Ismailia, Egypt.

Four isolates of *Bacillus thuringiensis* were isolated from 20 soil samples which were collected from rhizosphere of Alfa alfa plant from four locations at Ismailia governorate. The colonies on HCT medium were divided into four different morphological groups based on the colony morphology and the crystal shape as observed by light microscopy. Two isolates produced typical spherical crystals (AI, AII), one isolate produced a pyramidal crystal (AIII) and another produced a bipyramidal crystal (AIV). The analysis of the bacterial endotoxin by SDS-PAGE has shown that the crystals from the isolates AI, AII and AIII contained protein bands of 128, 125 and 139 KDa, while the isolate AIV contained a protein band of 143 KDa. The treatment of J2 of *M. incognita* with purified crystals toxin ( $1 \times 10^8$  crystal/ml) showed the best result in the treatment with isolates AI and AII.

**NEMATODE COMMUNITY STRUCTURE OF NATURAL, NON-MANAGED AND MANAGED ECOSYSTEMS. G. W. Bird.** Michigan State University, East Lansing, MI, USA.

Nematode community structure was determined for pristine deciduous forest, coniferous forest, 60-year-old woodlot, 35-year-old secondary succession and certified organic and conventional annual crop ecosystems, with special reference to O-horizon, 0–15 cm and 15–30 cm soil depths and focus on soil genesis and development, degradation and restoration ecology. Highest absolute population densities were associated with O-horizons or surface litter. Greatest absolute population density was recovered from an 18-year certified organic vegetable production site, adjacent to wood lot and conventional corn-soybean sites sampled using the same design. Relative population densities for the organic site were 73, 26 and 0.6 for bacterivores, fungivores and herbivores, respectively, compared to 28, 14 and 57 for conventional. Similar differences were observed for organic and conventional cherry orchards. Absolute and relative densities of bacterivores increased during the three years after organic apple orchard transition. In all orchard cases, nematode community structure signatures more closely resembled old field succession than pristine eastern deciduous forest, conifer forest or conventional or organic annual crop ecosystems.

**NEMATOTOXIC EFFECTS OF ABAMECTIN AND THIODICARB ON *MELOIDOGYNE INCOGNITA* AND *ROTYLENCHULUS RENIFORMIS*. L. J. Xing,<sup>1</sup> C. Grimm,<sup>2</sup> A. Cochran,<sup>3</sup> D. H. Long.<sup>4</sup>** <sup>1</sup>Syngenta Crop Protection Inc., Leland, MS, <sup>2</sup>Syngenta Crop Protection AG, Stein, Switzerland, <sup>3</sup>Syngenta Crop Protection AG, Basel, Switzerland, <sup>4</sup>Syngenta Crop Protection Inc, Greensboro, NC.

Avicta Compete Pak was successfully launched in 2006 as a novel Syngenta seed treatment brand that offered U.S. cotton growers early season protection against nematodes, insects and diseases. The nematocidal active ingredient within Avicta is abamectin, one of the most potent nematocides ever discovered. The efficacy of abamectin on *Meloidogyne incognita* and *Rotylenchulus reniformis*, two of the most economically important nematode pests for U.S. cotton production, was investigated in either in vitro or greenhouse studies in which abamectin was evaluated as a seed treatment. In vitro LC<sub>50</sub> studies were conducted to evaluate the nematotoxic effects of abamectin and thiodicarb against *M. incognita* based upon 24 and 120 hour exposures. The LC<sub>50</sub> results indicated that abamectin is 470 times more active against *M. incognita* compared to thiodicarb. Likewise, abamectin against *M. incognita* was demonstrated to be nematocidal (less than 15% nematode recovery), while thiodicarb proved to be nematostatic (greater than 80% nematode recovery). Comparative data from the parallel studies on *R. reniformis* were similar to those for *M. incognita*, in that abamectin was more active on *R. reniformis* than thiodicarb. Efficacy of abamectin and thiodicarb applied as a seed treatment on *R. reniformis* was studied and will be discussed.

**SEASONAL RENIFORM NEMATODE POPULATION DYNAMICS IN NORTHEASTERN AND EAST-CENTRAL ARKANSAS. J. A. Still,<sup>1</sup> T. L. Kirkpatrick.<sup>2</sup>** <sup>1</sup>University of Arkansas, Fayetteville, AR, USA, <sup>2</sup>University of Arkansas, Hope, AR 71801, USA.

The reniform nematode, *Rotylenchulus reniformis*, was first reported in Arkansas in 1988. The nematode survives in northeastern Arkansas, but population densities in cotton do not reach the same levels as the southeastern corner of the state where it is more prevalent. The impact of soil texture and temperature on the temporal population dynamics and overwintering survival and life cycle progression of the reniform nematode on cotton in these two regions was studied from

2005 to 2007. Sampling points were established according to soil texture in production fields in northeastern AR (MS) and east-central AR (MR) in June 2005 with a Global Positioning System. Monthly samples were collected to a depth of 15 cm, and vertical samples to a depth of 120 cm were collected at planting and harvest and mid-winter (February). The vertical samples were divided into 20-cm segments, and nematodes were extracted from each. Nematode densities were highest in the 20–40-cm segments at each sampling date at all sites, and densities decreased rapidly below 40 cm in both fields. In the MS field, nematode densities were consistently higher in soil with the highest clay content. Nematode population densities in the MR field increased during the winter months and were generally higher than those in the MS field. Life cycle progression as measured by the percentage of the population that was juvenile was slower during the winter months in the MS field than in the MR field.

**MALE RAY PATTERN EVOLUTION IN RHABDITID NEMATODES. K. C. Kiontke, A. Kolychkina, R. Ng, D. H. Fitch.** New York University, New York, NY, USA.

One important diagnostic character for rhabditids is the pattern of sensory organs—rays (genital papillae) and phasmids—in the male tail, which is highly conserved within a species but varies between species and species groups. Our objective is to establish the ray pattern in the rhabditid stem species and to reconstruct evolutionary changes which occurred within rhabditids. We use DIC microscopy, antibody staining of adherens junctions and cell ablations to study ray number, ray arrangement and homology and the position of phasmids relative to rays in adults and during development. By projecting character differences onto a phylogenetic tree, we determine the number and kinds of evolutionary changes. The number of rays was fixed early in rhabditid evolution to nine pairs. Later, several losses of rays occurred but no gain. We homologize rays in different species and reconstruct changes in their position, e.g., the posterior and dorsad displacement of ray 1 in diplogastrids. The phasmids were positioned posterior of all rays in the rhabditid stem species. Several changes to a more anterior position occurred due to migration of ray and/or phasmid precursors during development. This migration can happen early in L1 or L2 as in diplogastrids, or in L4 as in *Brevibucca saprophaga*. Some ray pattern characters support relationships otherwise only supported by molecular data, e.g., *Rhabditoides inermiformis* and *R. regina* with *Pleiorhabditis*.

**DYNAMICS OF HG TYPES ASSOCIATED WITH COMMERCIAL SOYBEAN CULTIVARS IN MICHIGAN. J. W. Davenport, G. W. Bird, F. Warner.** Michigan State University, East Lansing, MI, USA.

The most common HG Type associated with Michigan soybean production is 2.5.7. In MI, types range from 0 to 1.2.3.5.6.7. Two-, three- and six-year trials were established to study HG Type dynamics under field conditions. The six- and two-year trials compared results from a continuous susceptible with PI 88788, Peking and PI 437654; a mixture of susceptible, PI 88788, PI 437654 and Peking and a three-year rotation with susceptible, PI 88788 and Peking-derived commercial soybean cultivars. The three-year trial compared field corn, fallow and a susceptible cultivar with PI 88788 and PI 88788 x PI 437654-derived cultivars. Five replicates were used at each location. Type 2 and Type 4 were not detected after six years of continuous Peking or PI 437654-derived cultivars, respectively. After three years, Type 0 was detected when the plots were planted annually to a mixture of susceptible, PI 88788, PI 437654 and Peking-derived cultivars. Type 2.5.7 was recovered from all treatments in the three- and two-year trials. Type 1.2.3.5.6.7, however, is known to exist on the farm where the two-year trial was located. Both soybean cyst and sugar beet cyst nematodes are present in the site used for the three-year HG Type trial.

**454 PYROSEQUENCING FOR METAGENOMIC ANALYSIS OF NEMATODE DIVERSITY. D. L. Porzinska,<sup>1</sup> R. M. Giblin-Davis,<sup>1</sup> T. O. Powers,<sup>2</sup> N. Kanzaki,<sup>3</sup> W. Ye,<sup>4</sup> K. Morris,<sup>5</sup> W. K. Thomas.<sup>5</sup>** <sup>1</sup>University of Florida, Fort Lauderdale, FL, <sup>2</sup>University of Nebraska, Lincoln, NE, <sup>3</sup>FFPRI, Tsukuba, Japan, <sup>4</sup>North Carolina Department of Agriculture and Consumer Services, Raleigh, NC, <sup>5</sup>University of New Hampshire, Durham, NH.

The 454 genome sequencer allows for pyrosequencing of thousands of DNA sequences during a single run. The method may offer a rapid tool to inventory nematode fauna at previously unparalleled levels of resolution from any ecosystem. To demonstrate a priori the feasibility of 454 pyrosequencing for nematode identification, we selected two ~400 bp fragments of rRNA loci flanked by “universal” primer pairs (NF1-1573R 18S and D3A-D3B 28S) as potential barcode regions (each direction becomes a barcode) and assessed errors associated with over- and underestimating nematode species using existing databases of three genera: *Caenorhabditis* (11 species, clade V), *Bursaphelenchus* (23 species, clade IV) and *Fergusobia* (17 species, clade IV). All four barcodes (~200 bp) correctly identified nematode genera, and none of the barcodes overestimated the number of species. NF1 and D3A barcodes allowed for greater resolution with smaller lumping errors than 1573R and D3B barcodes. Other “universal” primers from other loci are being considered prior to pyrosequencing of soil nematodes from the Konza tallgrass prairie ecosystem where we have good morphological and molecular data for procedural verification.



DO EXOTIC AND INVASIVE PLANT-PARASITIC NEMATODES EXIST IN CALIFORNIA'S AGRICULTURAL PRODUCTION SITES? **J. J. Chitambar, K. Dong, S. A. Subbotin, R. Luna.** California Department of Food and Agriculture, Sacramento, CA, USA.

The Nematology Laboratory of the California Department of Food and Agriculture (CDFA) provides diagnostic support for the protection of California's agricultural industry against economically important plant-parasitic nematodes. During the past two years, the laboratory conducted two nematode survey projects, 1) a general survey for 23 target species of quarantine and regulatory significance, and 2) the potato cyst nematode (*Globodera pallida*) survey of all the seed potato fields and at least 10% of the potato production fields. Both survey projects were cooperative with the USDA and funded by the National Cooperative Agricultural Pest Survey (CAPS) Program and the national potato cyst nematode survey program. A total of 2,522 CAPS survey samples and 674 potato field soil samples were processed and diagnosed by December 2006. In addition, 1,652 nematode samples from CDFA's Nematode Control and Nursery Certification program and 422 samples from the Quarantine Phytosanitary program of commodities for export in 2006 were also included into the overall survey results. Nematode species were identified based on morphological characters, and PCR tests were conducted for *Meloidogyne* spp. and *Heterodera* spp. Eighty-six nematode genus/species were detected in the state-wide nematode survey. No potato cyst nematode was found by the end of 2006.

RELATING THE GLUCOSINOLATE PROFILE OF *TROPAELUM MAJUS* CULTIVARS TO *XIPHINEMA AMERICANUM* MORTALITY. **J. M. Halbrendt,<sup>1</sup> J. E. Dean,<sup>2</sup> C. P. Rice,<sup>3</sup> I. A. Zasada.<sup>3</sup>** <sup>1</sup>Penn State University, Biglerville, PA, USA, <sup>2</sup>University of Maryland, College Park, MD, <sup>3</sup>USDA-ARS, Beltsville, MD.

*Tropaeolum majus* (nasturtium) is a glucosinolate-producing plant with >93% of its chemical profile being benzyl glucosinolate. Glucosinolates hydrolyze into nematicidal compounds but efficacy varies depending on the type and concentration of glucosinolate and the nematode being tested. Root and shoot tissue from five cultivars of *T. majus* including Alaska, Jewel Mix, Moonlight, Peach Melba and Vesuvius were freeze-dried and pulverized. The tissue was analyzed for glucosinolate content, and a bioassay using *Xiphinema americanum* in sand amended with dried plant tissue was used to test for nematicidal activity. The cultivars varied in their glucosinolate content as well as their ability to cause *X. americanum* mortality. Peach Melba shoots had the highest concentration of benzyl glucosinolate (12,500  $\mu\text{g/g}$  dry material) followed by Jewel Mix, Vesuvius, Alaska and Moonlight with the lowest (9,300  $\mu\text{g/g}$  dry material). Peach Melba was the most toxic with a lethal concentration ( $\text{LC}_{50}$ ) of 0.6 mg dry shoot material/cc sand, while Moonlight was the least toxic with an  $\text{LC}_{50}$  of 1.3 mg dry shoot material/cc sand. It appears that the toxicity of plant material can be related to glucosinolate levels in incorporated plant material. Glucosinolate concentrations of plant materials may be a useful indicator of the potential effectiveness of green manure materials.

FLPS ARE PHYSIOLOGICAL REGULATORS WITH INTERESTING PHYLOGENETIC SIGNATURES IN PLANT-PARASITIC NEMATODES. **E. P. Masler, L. K. Carta, A. M. Skantar, C. Stone.** USDA/ARS, Beltsville, MD, USA.

Neuropeptides regulate most physiological processes in animals, and the most prominent neuropeptide family in nematodes comprises the FMRFamide-like peptides (FLPs). FLPs are ~6–20 amino acids long and regulate a number of fundamental processes including egg laying, feeding, locomotion and other parasitic and reproductive behaviors. Multiple *flp* genes encode numerous peptides resulting in large numbers of different FLPs, all of which terminate in a functionally important ArgPhe-NH<sub>2</sub>. ClustalW, ProbCons, PAUP\* and visual inspection were used to analyze FLP sequences across nematodes and reveal remarkable FLP phylogenetic topologies. Nematode FLP sequences have greater identities within than between ecologically distinct nematodes such as plant or animal parasites and can precisely parallel the phylogenetic distributions generated by ribosomal 18S sequences. Further, microanalysis of all known plant-parasitic nematode FLPs, including those encoded by *Heterodera glycines flp* genes cloned in our laboratory, suggests that characteristic peptide sequences may be parasite-related. In addition, HPLC-ELISA profiles show that *H. glycines* FLPs are developmentally related, and we propose that changes in FLP levels may influence hatching. The ubiquity and molecular diversity of FLPs, and their physiological roles, provide a rich source of models for novel pest control agents.

CONSEQUENCES OF *MELALEUCA QUINQUENERVIA* INVASION OF THE FLORIDA EVERGLADES: "NOTES FROM THE UNDERGROUND" WITH SPECIFIC REFERENCE TO NEMATODES. **D. L. Porazinska,<sup>1</sup> P. D. Pratt,<sup>2</sup> R. M. Giblin-Davis.<sup>1</sup>** <sup>1</sup>University of Florida, Fort Lauderdale, FL, <sup>2</sup>USDA, Fort Lauderdale, FL.

The total abundance and diversity of nematode communities in the Florida Everglades soils dominated by the invasive tree *Melaleuca quinquenervia* was 40% and 80% of adjacent soils under native plant communities. Fungal-feeding and plant-parasitic nematodes were twice as abundant under native plants as under *M. quinquenervia*. Nematode communities under *M. quinquenervia* stands were bacterivore-dominated, while those under native vegetation were plant-parasitic dominated. The overall diversity of nematode genera was 20% lower under the exotic than under native plants, with plant parasites being 56% and fungivores being 30% less diverse. From chemical soil characteristics, soil moisture, percentage

of Ca and Mg, percentage of clay particles and total soil C and N were greater in *M. quinquenervia* soils, but plant-available concentrations of P, K, Ca and Mg as well as CEC were reduced. Data provide support for the presence of weakened plant-soil biota negative feedbacks in *M. quinquenervia*-invaded habitats.

**HIRSCHMANNIELLA** SP. N. (NEMATODA: PRATYLENCHIDAE), A CRYPTIC SIBLING SPECIES OF *H. POMPONIENSIS* ABDEL-RAHMAN & MAGGENTI, 1987. **I. Tandingan De Ley, M. Mundo-Ocampo, M. Yoder, P. De Ley.** Department of Nematology, Univ. of California, Riverside, CA 92521, USA.

A new species of *Hirschmanniella* is described from a vernal pool in the Santa Rosa Plateau Ecological Reserve, CA, USA. The cryptic new species is morphologically very close to *H. pomponiensis* and *H. gracilis* and is distinguished from the former only by a more anterior excretory pore and more flattened and laterally expanded stylet knobs, while it differs from the latter in the greater distance from phasmid to tail tip and in the bursal alae ending near the phasmids rather than near the tail tip. Analysis of the rDNA small subunit and D2D3 expansion segments of the large subunit shows that the new species is genetically distinct, having respective sequence homology of 98.89% and 95.9% with *H. pomponiensis* for these loci. Congruence in SSU and D2D3 gene trees as well as unambiguous character autapomorphies further support the new species status and sibling relationship with *H. pomponiensis*. Although many more isolates and species will need to be studied before informative biogeographic analyses can be performed, the presently available sequence data suggest that some *Hirschmanniella* lineages have diversified independently on either side of the Atlantic. Recovering nematode species complexes is important to agriculture for accurate diagnosis and management of potentially destructive invasive species and to ecology and bioinventory for a reliable estimate of species richness and diversity.

**PUBLIC SOYBEAN BREEDING LINES TESTED FOR RENIFORM NEMATODE (*ROTYLENCHULUS RENIFORMIS*) REPRODUCTION. R. T. Robbins,<sup>1</sup> E. Shipe,<sup>2</sup> G. Shannon,<sup>3</sup> P. Arelli,<sup>4</sup> P. Chen.<sup>1</sup>** <sup>1</sup>University of Arkansas, Fayetteville, AR, <sup>2</sup>Clemson University, Clemson, SC, <sup>3</sup>University of Missouri, Portageville, MS, <sup>4</sup>USDA-ARS-MSA, Jackson, TN.

In the Southeastern United States, reniform nematode (*Rotylenchulus reniformis*) causes considerable damage and yield loss to soybean and cotton. No cotton varieties have reniform nematode resistance, whereas several sources of resistance exist in soybean. This resistance is often linked to resistance to the soybean cyst nematode (*Heterodera glycines*). Use of resistant soybean in a rotation with cotton can be a useful option. Soybean breeding lines from programs at Arkansas, Clemson, Missouri and USDA in Jackson, TN, having low reniform reproduction rates have been identified in greenhouse tests. Lines were inoculated with approximately 1,500 vermiform nematodes/500 ml pot and evaluated after growing at least 10 weeks in years 2001–2006. Breeding lines not differing in reproductive indices from known resistant cultivars Forrest, Anand and/or Hartwig by program are: SC94–1573, SC95–1070, SC95–771, SC97–1770, SC97–259, SC98–1930, SC98–249, SC98–318, SC98–353, SC01–783A, SC01–810, SC02–208, SC02–210, SC02–211, SC03–045, SC03–9093, SC03–9153, SC03–9383, SC03–9438 (Clemson), S01–8401 HP, S01–9265, S01–9364, S01–9391, S02–20388, S02–3934, S02–611 CR (Missouri), JTN-5203, JTN-5303, JTN-5503 (USDA, Jackson, TN), R97–1801, R00–1551 and R01–4747 (Arkansas). Use of these soybean breeding lines may prove very useful in breeding for reniform nematode resistance.

**CLOVE OIL AND FUNGUS COMPOUNDS: CAN NEMATODE SUPPRESSION BE ACHIEVED WITHOUT PHYTOTOXICITY? S. L. Meyer,<sup>1</sup> D. Lakshman,<sup>2</sup> I. Zasada,<sup>1</sup> B. Vinyard,<sup>3</sup> D. Chitwood,<sup>1</sup> O. Shemshura,<sup>4</sup> N. Bekmakhanova,<sup>4</sup> M. Mazunina,<sup>4</sup> B. Yeskalieva,<sup>4</sup> E. Masler.<sup>1</sup>** <sup>1</sup>USDA, ARS, Nematology Laboratory, B. 011A, R. 165B, Beltsville, MD 20705, US, <sup>2</sup>Floral and Nursery Plants Research Unit, US National Arboretum, BARC-West, Beltsville, MD 20705–2350, USA, <sup>3</sup>USDA, ARS Biometrical Consulting Service, BARC-West, Beltsville, MD 20705–2350, US, <sup>4</sup>Institute of Microbiology and Virology, Almaty, Almatynskaya oblast, Kazakhstan.

Natural products from a plant (*Syzygium aromaticum*) and a fungus (*Aspergillus* sp.) were examined for the presence of compounds with potential for application as novel nematocides. The plant-derived material, clove oil, was tested in the greenhouse against the nematode *Meloidogyne incognita* on cucumber. In one trial, treatment with 0.10% and 0.15% clove oil (but not 0.20% clove oil) suppressed numbers of eggs and juveniles per gram of root by 27–28% compared to water-treated controls. No population suppression with clove oil treatment occurred in a second trial. Under the greenhouse test conditions, clove oil treatments that were not phytotoxic to seedlings did not effectively reduce root knot populations on cucumber roots. Metabolites from the fungus *Aspergillus* were tested for activity against the nematode *Ditylenchus* sp. In laboratory assays, culture broth from one *Aspergillus* sp. isolate caused 100% loss in activity of vermiform stages of *Ditylenchus*, resulting in formation of vesicles in the vicinity of the intestine. Multiple fractions of the culture broth were shown to be biologically active, including fractions containing amino acids, glycosylated flavonoids, terpenoids and dicarboxylic acids.

USING GREEN MANURE CROPS TO SUPPRESS COLUMBIA ROOT-KNOT NEMATODE (*MELOIDOGYNE CHITWOODI*) IN POTATO IN THE SAN LUIS VALLEY. **R. Ingham,<sup>1</sup> M. Dillon,<sup>2</sup> N. David,<sup>3</sup> J. Delgado.<sup>4</sup>** <sup>1</sup>Oregon State University, Corvallis, OR, USA, <sup>2</sup>Colorado State University, Center, CO, USA, <sup>3</sup>Oregon State University, Hermiston, OR, USA, <sup>4</sup>USDA ARS, Fort Collins, CO, USA.

Columbia root-knot nematode, CRKN (*Meloidogyne chitwoodi*), infests potato tubers and causes quality defects that may lead to crop rejection. CRKN is most commonly managed in the San Luis Valley (SLV) of Colorado with nonfumigant nematicides and less so with fumigants. Research in the Pacific Northwest of the US has demonstrated suppression of CRKN with green manure (GM) crops planted in late summer following a grain crop. However, the SLV has a very short growing season that is not long enough for a GM crop to be planted after a grain crop. High nematode control costs, low grain prices and water shortages led to grower interest in growing GM crops during the summer instead of a grain crop to reduce water use, improve soils and reduce nematode control costs. Field trials were conducted in 2004 and 2005 to evaluate GM crops planted in June and incorporated in August. Symptoms of CRKN in tubers were assessed in the following potato crops in 2005 and 2006. Prior to planting GM crops in 2004, CRKN averaged 1,900/250 g soil, but declined by potato planting in 2005 to 460 (radish), 440 (mustard), 170 (canola), 65 (sudangrass cv. Sordan 79) and 65/250 g in wet fallow plots. Initial CRKN averaged 800/250 g soil in 2005 but were reduced to 135 (mustard), 120 (sudangrass cv. Honeysweet), 25 (Sordan 79) and 1/250 g in wet fallow plots prior to planting potato in 2006. Symptoms of CRKN in tubers at harvest were low both years and met industry standards in all plots. However, incubating tubers to detect late season CRKN infection revealed high levels of culls following mustard (62%), radish (61%) and canola (72%) in 2005. Few culls were present after incubation in any treatment in 2006.

MOLECULAR PHYLOGENY OF CLADE III NEMATODES REVEALS MULTIPLE ORIGINS OF TISSUE PARASITISM. **S. A. Nadler.** Department of Nematology, University of California, Davis, CA, USA.

Molecular phylogenetic analyses of Ascaridida, Rhigonematida, Spirurida and Oxyurida have consistently recovered these taxa as monophyletic, a group now called clade III. However, poor sampling of these orders has limited tests of their monophyly and reduced the potential resolution of phylogenies. To obtain a more comprehensive clade III tree, SSU ribosomal DNA sequences for 113 species have been used to infer relationships for representatives of all four orders. The posterior probability of each multiple alignment site was used to exclude or weight characters, yielding datasets that were analyzed using maximum parsimony, maximum likelihood and Bayesian methods. Phylogenetic results were robust to differences among inference methods for most higher-level groups, but some clades were sensitive to alignment ambiguity. Taxa representing Camallanoidea, Oxyurida, Physalopteroidea, Raphidascarididae and Skrjabinellidae were monophyletic in all analyses. Some clades recovered in all trees, such as Dracunculoidea and Spirurina, included the vast majority of their sampled species, but were non-monophyletic due to the consistent behavior of a few "rogue" taxa. Similarly, clade III was paraphyletic due to the grouping of one seuratoid with an outgroup clade. Mapping of host habitat showed that tissue-dwelling localization has evolved independently at least three times within clade III and relationships among Spirurina and Camallanina often reflected patterns of tissue predilection rather than taxonomy.

NEMATOL: DATABASE FOR THE NEMATODE BRANCH OF THE TREE OF LIFE. **W. K. Thomas,<sup>1</sup> Namtol Consortium.<sup>2</sup>** <sup>1</sup>UNH, HCGS, Durham, NH, USA, <sup>2</sup>Various Labs throughout the world

NemATOL is an open relational database supporting the Nematode branch of the tree of life and other nematode biodiversity projects. The database is intended to organize and archive morphological, molecular and ecological data in the context of nematode phylogeny and biodiversity. The primary resources include: DNA sequence databases of loci commonly used in phylogenetic analysis (e.g., SSU, LSU, ITS regions of the nuclear rDNA locus, mitochondrial DNA sequences), morphological information (e.g., multifocal video images, still images, TEMs, SEMs and line drawings) and specimen data (localities, maps, associated metadata and uBio links to distributed information associated with each defined taxon). All of these resources are linked together and organized in the context of nematode evolutionary relationships. The primary access to the database is can be achieved by taxonomic queries, BLAST searches with molecular data and by geographic locality. Nematol is easily internet accessible at <http://nematol.unh.edu>.

SPECIES AND DISTRIBUTION OF PRATYLENCHUS IN CANADA. **Q. Yu.** Environmental Health Program (Biodiversity/Invertebrate), Agriculture and Agri-Food Canada, Ottawa, Ontario, K1A 0C6, Canada.

Canadian species of *Pratylenchus* are described and illustrated. Slide-supported distribution data and key to the species are provided. At least 11 species are recorded in Canada: *P. crenatus*, *P. fallax*, *P. flakkensis*, *P. hexicinsus*, *P. macrostylus*, *P. neglectus*, *P. pratensis*, *P. penetrans*, *P. thornei*, *P. vulnus* and *P. zaeae*.

RELATIONAL DATABASE FOR THE IDENTIFICATION OF PAN- AND CLADE-SPECIFIC ORTHOLOGOUS GENE SETS IN THE PHYLUM NEMATODA. **K. Morris, T. Fogal, W. K. Thomas.** HCGS/UNH, Durham, NH, USA.

The primary goal of the nematode tree of life project is to develop a robust evolutionary framework for the phylum. Each year many new "complete genome sequences" and/or rich datasets of cDNA sequences are generated which represent a

potentially rich source of phylogenetic information. As part of the Nematode Tree of Life project, we are developing a pipeline to identify putative orthologous genes within the phylum Nematoda. The development of this pipeline includes and will allow for the addition of new data as it becomes available. The pipeline will take a variety of input data, including unigene clusters from cDNA/EST libraries and gene predictions from the complete genome sequences. The pipeline uses a de novo approach to identifying putative orthologous genes rather than relying on lists of putative single copy genes and a relational database structure that will facilitate the selection of clade specific orthologous gene sets for analysis.

**SOIL FUMIGATION AND VARIETY SELECTION TO STUDY SOYBEAN CYST NEMATODE IN IOWA. J. De Bruin, P. Pedersen.** Iowa State University, Ames, IA, US.

The wide distribution of soybean cyst nematode (*Heterodera Glycines* Ichinohe, SCN) in Iowa is suspected to be a factor that contributes to slow soybean (*Glycine Max* (L.) Merr.) yield improvements. Four hectares were fumigated with the product Telone C-35 in the fall at three locations in Iowa. A variety trial consisting of 23 varieties, including SCN-resistant and susceptible varieties, was established in fumigated and non-fumigated treatments in 2005 and 2006. Soil samples were collected at planting and harvest from each plot to determine SCN populations. The range of spring SCN populations at the five environments was 151 to 12,402 eggs/100 cm<sup>3</sup> in non-fumigated treatments and 30 to 6,678 eggs/100 cm<sup>3</sup> in the fumigated treatment, with an average reduction of 42% from soil fumigation. Soil fumigation increased yield 739 kg/ha for specific varieties with an average increase of 262 kg/ha. At harvest in 2005, SCN resistant varieties had 2,037 and 7,089 fewer eggs/100 cm<sup>3</sup> at environments in central and western Iowa, respectively. Soybean cyst nematode was never eliminated by fumigation, and SCN resistant varieties, compared to susceptible varieties, increased yield an average of 572 kg/ha. When SCN is present in a field, resistant varieties are an effective management tool to increase yield and manage SCN populations in Iowa.

**A MOLECULAR ASSAY FOR RELIABLE QUANTIFICATION OF SOYBEAN CYST NEMATODE IN PLANTS. X. Gao, A. Colgrove, K. Lambert, T. Niblack.** Department of Crop Sciences, University of Illinois, AW-101 Turner Hall, 1102 South Goodwin Ave, Urbana, IL 61801, USA.

For the quantification of nematodes, researchers have relied primarily on microscopic observation. This can be a time-consuming practice, particularly with high nematode population densities. With *Heterodera glycines*, the soybean cyst nematode (SCN), as a model, we developed novel system for quantification of SCN in soybean roots. This system consists of: i) appropriate sample processing and storage methods to minimize DNA degradation; ii) an optimized DNA extraction protocol to extract inhibitor-free DNA samples; and iii) an optimized real-time quantitative polymerase chain reaction (QPCR) to quantify nematode DNA within roots. Seedlings sampled at the 7th day after planting were surface-sterilized with sodium hypochlorite (0.6%) and stored at -80°C. The DNA samples extracted with an optimized protocol were PCR inhibition-free. For quantification of SCN, the assay consisted of QPCR with a pair of primers that sensitively amplified the chorismate mutase gene (*Hg-cm-1*). The assay quantified as little as 650 ng of SCN DNA. The correlation between quantified SCN DNA and counts of hatched second-stage juveniles (J2) is very high ( $R^2 = 0.97$ ). In addition, there was a high degree of correlation between quantified SCN DNA and counts of J2 in soybean roots ( $R^2 = 0.78$ ). Screening of novel nematicidal compounds with this new quantification system for SCN is underway.

**NOVEL CUTICULAR MORPHOLOGY USING LT-SEM AND LIGHT MICROSCOPIC MODIFICATIONS IN SOME BACTERIAL-FEEDING AND PLANT-PARASITIC NEMATODES. L. K. Carta,<sup>1</sup> E. F. Erbe,<sup>2</sup>** <sup>1</sup>USDA-ARS-NL, Beltsville, MD, USA, <sup>2</sup>USDA-ARS-SGIL, Beltsville, MD, USA.

Microscopic techniques used only exceptionally within nematology can reveal new and unexpected morphological features not visible with standard light and scanning electron microscopy. Among nematodes, SEM face views are of special importance in diagnosis of *Panagrolaimus* spp. Low Temperature (LT)-SEM images from a Hitachi S-4100 field-emission LT-SEM clearly distinguished a number of cultured species from our live collection. Unusual sheath-like features of the cuticle were also demonstrated in one *Panagrolaimus* sp. LKC39 associated with tarantulas, and in *Pristionchus pacificus* PS312, a scarab beetle associate. Other characters with improved quality or resolution included the vulva, tail and stoma. Six plates at the stoma opening were shown with a range of positions in *P. pacificus*. The recent innovation of an annular condenser within a CytoViva microscope adapter demonstrated improved resolution approaching that of an SEM for various morphological features important for diagnosis. These included the excretory system in various nematodes such as *Rhabditis rainai* and lip annules of special diagnostic value in *Pratylenchus* spp.

**POLYMORPHISM OF THE HSP90 GENE AMONG POPULATIONS OF HETERODERA GLYCINES FROM CHINA, JAPAN AND THE UNITED STATES. N. Atibalentja,<sup>1</sup> G. R. Noel,<sup>2</sup>** <sup>1</sup>Dept. of Crop Sciences, Univ. of Illinois, Urbana, IL, <sup>2</sup>USDA, ARS, Urbana, IL.

*Hsp90* is a member of a gene family that codes for proteins whose synthesis is enhanced by a variety of environmental stressors, including a sudden increase in temperature. These proteins function as molecular chaperones in cells where they

interact with a range of regulatory proteins involved in critical physiological processes such as cell-cycle control and hormone signaling. Recently, the complete protein sequence and a partial genomic sequence of the *Heterodera glycines* homolog of *Hsp90* (*Hg-hsp90*) were reported. In this study, we cloned the complete genomic sequence of *Hg-hsp90* and investigated the extent of polymorphism of this gene among 16 populations of *H. glycines* from China (5), Japan (5), and the U.S. (6). The genomic clone of *Hg-hsp90* was 3,052-nt long and, like that of the filarial nematode *Brugia pahangi*, contained 11 introns, compared with only three in *Caenorhabditis elegans*. The estimates of gene diversity among populations from China (0.64%), Japan (0.65%) and the U.S. (0.61%) were not significantly different. However, the extent of variation observed in this study is similar to that obtained with the ITS-rRNA gene, which shows that *Hg-hsp90*, a single or low copy number gene, unlike the multi-copy ITS-rRNA gene, is a better candidate for nematode phylogenetics.

THREE NEW NEMATODE ASSOCIATES FROM SYCONES OF *FICUS COLUBRINAE* IN LA SELVA, COSTA RICA. **R. M. Giblin-Davis,<sup>1</sup> Y. Zeng,<sup>2</sup> W. Ye,<sup>3</sup> B. J. Center,<sup>2</sup> N. Kanzaki,<sup>4</sup> A. Esquivel,<sup>5</sup> T. Powers.<sup>6</sup>** <sup>1</sup>Fort Lauderdale Research and Education Center, University of Florida, Fort Lauderdale, FL, USA, <sup>2</sup>University of Florida, Fort Lauderdale, FL, USA, <sup>3</sup>North Carolina Department of Agriculture & Consumer Services, Raleigh, NC, USA, <sup>4</sup> Forest Pathology Laboratory, FFPRI, Tsukuba, Ibaraki, Japan, <sup>5</sup>Universidad Nacional Escuela de Ciencias Agrarias, Heredia, Costa Rica, <sup>6</sup>University of Nebraska, Lincoln, NE, USA.

Three new nematode species were recovered from the syconia of *Ficus colubrinae* from La Selva, Costa Rica, during a survey of nematode rainforest biodiversity in 2005. They are new species from *Schistonchus*, *Parasitodiplogaster* and a putative new genus of tylenchid. This is the first report of an association between the nematode order Tylenchida and the sycones of figs. Both the *Schistonchus* and *Parasitodiplogaster* species were recovered from the lumen of figs, whereas the tylenchid was recovered from beneath the stipules/sepals of infested syconia. *Schistonchus* (fig parasite) and *Parasitodiplogaster* (fig wasp parasite) are well known associates of fig wasp pollinators (Agaonidae) and are transmitted to the interior of a fig by females during pollination/oviposition. The association between the tylenchid and *F. colubrinae* is presumed to involve an invertebrate host, but none was recovered during this study. Molecular analysis using the partial small subunit rRNA gene (SSU) of the new tylenchid supports its inclusion in the suborder Tylenchina (infraorder: Anguinata) or the suborder Hexatylinea, depending upon the molecular analysis used. Because the genus *Ficus* exhibits a latitudinal gradient in species diversity, we hypothesize that the specialized association between nematodes, fig wasps and figs will yield more nematode species in the tropics than the subtropics.

MULTIFACETED BIO-CONTROL METHODS AGAINST *MELOIDOGYNE CHITWOODI* AND *LEPTINOTARSA DECEMLINEATA*, PESTS OF POTATOES IN WASHINGTON STATE. **D. R. Henderson, E. Riga, W. E. Snyder.** Washington State University, Pullman, WA, USA.

Mustard meal from *Brassica carinata* has shown both nematicidal and oviposition-detering properties against the root-knot nematode (RKN), *Meloidogyne chitwoodi*, and the Colorado Potato Beetle (CPB), *Leptinotarsa decemlineata*, respectively. In addition, entomopathogenic nematodes (EPN) have shown biocontrol potential against both CPB and RKN. Mustard meal was applied at 1 ton/acre, 15 days preplant of potatoes. The EPN *Steinernema feltiae* and *S. riobrave* were applied at planting (High/Low rate) and mid-season at a rate of 2 billion IJ/acre. EPN infection of CPB was evaluated at mid-season by placing five 4<sup>th</sup>-instar larvae inside perforated buckets filled with soil prior to EPN application. Four female and three male CPB adults were placed inside the sleeve cages on stems of 3 plants/plot of mustard-amended soil or control. Oviposition was recorded as egg clusters that contained greater than 5 eggs over a 1 week period. Mustard meal alone, EPN and combination of mustard meal and EPN significantly reduced populations of RKN in the soil, the percent culled potato tubers, percent infection and infection index. The CPB infection rates by *S. feltiae* and *S. riobrave* were 97% in the field. Mustard meal reduced CPB oviposition by 66% on potato plants grown in mustard meal-amended soil compared to the control. This multifaceted approach will provide potato growers with sustainable means to control both the nematode and insect pest.

DETECTION AND QUANTIFICATION OF ROOT-LESION NEMATODE *PRATYLENCHUS VULNUS* USING REAL-TIME PCR. **J. Qiu, B. B. Westerdahl, V. M. Williamson.** Department of Nematology, University of California-Davis, Davis, CA 95616, USA.

*Pratylenchus vulnus* is the most important root-lesion nematode in California orchards. Determination of its presence and numbers is essential for management decisions. It is a challenge to separate *P. vulnus* from closely related species by morphological characters, and it is difficult to use microscopy to count a field sample with a mixed population of *Pratylenchus*. A pair of species-specific primers was designed from the ITS sequence of rDNA from *P. vulnus*. These primers were used to detect and quantify the DNA of *P. vulnus* using a real-time PCR assay with SYBR Green Dye. DNA was extracted from a laboratory culture of *P. vulnus* with the phenol-chloroform method. With this DNA, we consistently generate a negative linear regression between threshold cycles (Ct) and Log values of DNA from 150 to 0.0015 ng ( $r^2 >$

0.99). We also compared three digestion methods without phenol-chloroform extraction: NaOH, proteinase K and proteinase K with pre-grinding. The DNA released from nematodes by these methods was quantified by real-time PCR. The quantity of DNA from a single nematode was higher from adults than from juveniles. The application of real-time PCR in detection and quantification of *P. vulnus* among other plant-parasitic and free-living nematodes from soil samples will be discussed.

**FOCAL SAMPLING OF TERMITES TO TEST THE LATITUDINAL GRADIENT HYPOTHESIS FOR ENTOMOPHILIC NEMATODE DIVERSITY. R. M. Giblin-Davis,<sup>1</sup> N. Kanzaki,<sup>2</sup> W. Ye,<sup>3</sup> R. H. Scheffrahn,<sup>1</sup> T. O. Powers.<sup>4</sup>**  
<sup>1</sup>Fort Lauderdale Research and Education Center, University of Florida, Fort Lauderdale, FL, USA, <sup>2</sup>Forest Pathology Laboratory, FFPRI, Tsukuba, Ibaraki, Japan, <sup>3</sup>North Carolina Department of Agriculture & Consumer Services, Raleigh, NC, USA, <sup>4</sup>University of Nebraska, Lincoln, NE, USA.

The order Isoptera (termites) was sampled as a focal group for measuring entomophilic nematode biodiversity at four different latitudes (41°N, 26°N, 10°N and 9°N) within a broad North American meridian spanning –80°W to –97°W. We sampled along transects using “time trial sampling” of termite colonies within 5-m radius areas or did whole area sampling. We dissected at least 10 workers per colony to examine for the presence, number, stage and location of nematode associates. Termites were vouchered for identification, and nematodes were collected for partial SSU rDNA amplification and sequencing attempts. About 30% of the termite colonies dissected had nematodes. Within each infested colony, we typically recovered less than five nematodes per insect with about 20–100% of the workers infested. Termites and their associated nematodes increased in diversity as the latitude decreased. Because insects take advantage of and partially represent (by being hosts or via the niches they occupy) the spatial heterogeneity that is characteristic of the tropics, effective biodiversity sampling should include entomophilic nematodes. Unfortunately, as diversity goes up, abundance usually goes down, making collection and identification of invertebrate hosts and detection of nematode associates difficult.

**THE IMPORTANCE AND MANAGEMENT OF PHYTOPARASITIC NEMATODES IN WESTERN COLORADO FRUIT ORCHARDS. R. R. Pokharel, H. J. Larsen.** Colorado State University, Grand Junction, CO, USA.

Western Colorado fruit growers are increasingly concerned about reduced crop yield caused and mediated by plant-parasitic nematodes (PPN). PPN predispose plants to soilborne pathogens; some vector plant viruses and often go unnoticed. To find out the incidence and the importance of these PPN, surveys were conducted in grapes and cherry, peach, apple, pear, apricot and prune orchards throughout western Colorado. PPN were extracted, identified and counted. Prune and apricot orchards had the highest diversity, but prune followed by apple had the maximum densities of PPN. High densities and frequency of dagger nematode and associated nepovirus infections indicate need for further attention. Endo-parasitic nematodes like root-knot nematode in prunes and apricots, root-lesion in apples and grapes and citrus nematode in prunes might also need attention. The density and genera of PPN varied with location, crop and depth of sampling. To evaluate the reaction of 14 cherry rootstocks, PPN were assessed in 5-year-old rootstock evaluation experiment. Observed PPN population densities from these rootstocks were highly variable, and no rootstock had extremely low population densities of the major PPN genera. Winter survival of PPN in the field was studied (sampling up to 46-cm deep); population densities of dagger and spiral nematodes increased up to January then started declining, whereas other PPN genera had no apparent differences in their population densities despite frozen soil.

**NEW RECORD AND SEM OF *RHYNCHONEMA ORNATUM* (LORENZEN, 1975) FROM THE SEA OF CORTEZ, MEXICO, WITH NOTES ON *RHYNCHONEMA AMAKUSANUM* (ARYUTHAKA, 1989). I. W. King,<sup>1</sup> D. Waumann,<sup>2</sup> P. De Ley.<sup>1</sup>** <sup>1</sup>Department of Nematology, University of California Riverside, Riverside, CA, USA, <sup>2</sup>Facultad de Ciencias Marinas, Universidad Autónoma de Baja California, Ensenada, BC, Mexico.

A population of specimens putatively identified as *Rhynchonema ornatum* (Lorenzen, 1975) is described and illustrated by light and scanning electron microscopy. Specimens were collected from sandy sediment in an intertidal region on the Sea of Cortez, near Santa Clara, Sonora State, Mexico. Specimens are illustrated as multifocal vouchers, which can be previewed online at <http://faculty.ucr.edu/~pdeley/vce.html> and in SEM micrographs. Preliminary results from sequence analysis are also presented. These nematodes are compared to *Rhynchonema ornatum* (Lorenzen, 1975), which was described from the Atlantic coast of Colombia, and *Rhynchonema ornatum antilliensis* (Gourbault, 1982), which was described from Basse-Terre, Guadeloupe, in the Lesser Antilles of the Caribbean. They are also compared to *Rhynchonema amakusanum* (Aryuthaka, 1989), a species described from southern Japan. The values for many of the characters of the specimens described from the Sea of Cortez are intermediate between those reported for *Rhynchonema ornatum* (Lorenzen, 1975) and *Rhynchonema amakusanum* (Aryuthaka, 1989), representing perhaps a geographic gradient of intraspecific variation and suggesting that the latter species may in fact be a junior synonym of the former.

INFECTIVITY OF MERMITHID NEMATODES FOR MOSQUITO LARVAE IN THE LAST INSTAR. **L. Santibanez-Vargas, R. Pacheco-Perez, E. G. Platzer.** University of California, Riverside, CA, USA

We evaluated the susceptibility of fourth-instar mosquito larvae of *Aedes sierrensis*, *Culex pipiens* and *Psorophora columbiae* to 10:1 infection intensities of *Romanomermis iyengari* (RI), *Romanomermis culicivorax* (RC), *Romanomermis wuchangensis* (RW) and *Strelkovimermis spiculatus* (SS). Percentage infection (infectivity) and intensity of infection (number nematodes per adult mosquito) was determined by the emergence of post-parasitic juveniles from adult mosquitoes. *Aedes sierrensis* was highly susceptible (infectivity) to RI (100 %) with susceptibility declining with R (63%) and RC (41%). *Culex pipiens* was most susceptible to RW (61%), followed by RI (46%), RC (29%) and SS (30%) with similar infectivities. *Psorophora columbiae* was most susceptible to SS (43%), followed by RI (15%). In general, intensity was correlated with infectivity with the exception of SS in *Culex pipiens*, where intensity exceeded that of the other mermithid species. In addition, the infectivity of mermithids from the Oriental Region (RI, RW) tended to exceed those from the Nearctic (RC) and Neotropical Regions (SS).

RAPID RESPONSE OF GRASSLAND NEMATODE DIVERSITY IN GRAZED PASTURE AT TWO SOIL FERTILITY LEVELS TO UREA APPLICATION. **G. W. Yeates,<sup>1</sup> R. L. Parfitt,<sup>1</sup> A. D. Mackay,<sup>2</sup> D. J. Ross.<sup>1</sup>** <sup>1</sup>Landcare Research, Palmerston North, New Zealand, <sup>2</sup>AgResearch, Palmerston North, New Zealand.

New Zealand pasture management has shifted from reliance on *Rhizobium*-fixed nitrogen to greater use of urea. We studied two farmlets in hill country, with no-fertilizer (NF) and 375 kg/ha/yr superphosphate application (HF) since 1981, and here compare samples in spring from replicated, grazed plots being either control plots or plots receiving 600 kg N/ha as urea over two years. One and two years after treatments began, herbage response to N was 1.6x on LF and 1.2x on HF farmlets. Both microbial biomass C and MBN were marginally lower with N (0.82–0.96x, 0.88–1.06x, respectively). Nematode diversity ( $H'$ ) was consistently lower under N and NCR higher. Plant-feeding nematodes were always higher with N but the response was greater at LF (1.41–2.01x) than HF (1.07–1.08). NF plots generally lacked *Pratylenchus*, *Plectus parietinus*; HF plots *Anatonchus*, *Mylonchulus*, *Dorylaimellus*, *Rhabdolaimus* and tardigrades. Under both underlying soil fertility levels, nematodes showed marked response to urea application with diversity declining; the relative importance of bacterial-feeders increased and root-feeding nematodes increased, especially in the NF farmlet. These observations illustrate soil nematode assemblages respond rapidly to nitrogen application, emphasising their close association with soil processes.

THE INFLUENCE OF *MELOIDOGYNE INCOGNITA* INOCULUM SOURCE AND NUTSEdge COMPETITION ON NEMATODE VIRULENCE ON CHILE PEPPER. **J. Trojan, S. H. Thomas, J. Schroeder, L. W. Murray.** New Mexico State University, Las Cruces, NM, USA.

Replicated greenhouse studies were conducted to characterize the development of different biotypes of *Meloidogyne incognita* race 3 (MiR3) developed by maintenance on chile pepper (*Capsicum annuum* cv. NM 6–4), yellow nutsedge (*Cyperus esculentus*, YNS) or purple nutsedge (*C. rotundus*, PNS). The influence of association or competitive stress of nutsedge on chile was also assessed with treatments of chile grown alone, chile + PNS and chile + YNS. Pots were inoculated with 5,000 MiR3 eggs recovered from chile, PNS or YNS cultures and harvested 45 days later. At harvest, half of each chile root system was extracted for egg production while on the other half, Phloxine B-stained egg masses were randomly isolated for extraction. Eggs per unit dry root and eggs per egg mass were calculated. Egg viability was determined at inoculation for each inoculum source and used to adjust MiR3 reproduction data prior to statistical analysis to enable comparisons between inoculum source treatments. Inoculum source altered egg production per unit chile root, with PNS inoculum producing fewer eggs, and resulted in a pathogenic response on root weight. Early MiR3 reproductive response was greater on chile + PNS with inoculum from PNS and YNS. Inoculum source affected MiR3 reproduction on chile, and stress induced by the presence of nutsedge may alter chile host response to inoculum.

MORPHOLOGICAL AND MOLECULAR CHARACTERIZATION OF *HEMIPECTUS MUSCORUM* ZELL, 1991 (NEMATODA: PLECTIDA). **O. Holovachov,<sup>1</sup> S. Bostrom,<sup>2</sup> I. Tandingan De Ley.<sup>1</sup>** <sup>1</sup>Dept. of Nematology, University of California—Riverside, Riverside, CA 92521, USA, <sup>2</sup>Department of Invertebrate Zoology, Swedish Museum of Natural History, SE-104 05 Stockholm, Sweden.

*Hemiplectus muscorum*, a representative of the monotypic genus, is redescribed on the basis of new material from Great Britain, Canada and USA on the basis of light and scanning electron microscopy of both females and males and juveniles of all stages. The species was previously known only from its original description, and its exact phylogenetic position within the order Plectida was unclear. Previous cladistic morphological analyses placed it at the base of Plectida in unresolved polytomy. New morphological information and molecular phylogeny (maximum parsimony, neighbour joining, bayesian and maximum likelihood algorithms) inferred from small subunit rRNA sequences reveal its close affinity to the members of the family Plectidae, particularly to the genus *Plectus*. The results of our current analyses suggest that considerable reductions and simplifications in the structure of the stoma and posterior part of the pharynx took place in the

lineage leading to *H. muscorum* and that the ancestors of *Hemiplectus* were equipped with a fully valvate basal bulb. General ideas about the morphology of the most recent common ancestor of Rhabditida and Plectida (sensu De Ley and Blaxter) must await analyses of more plectid taxa, but current data suggest that absence of a basal bulb is not necessarily a plesiomorphic condition within Plectida.

**SOYBEAN CYST NEMATODE AND NEMATICIDE AFFECT GROWTH PATTERN OF VARIOUS SOYBEAN VARIETIES IN IOWA. P. Pedersen, G. Tylka.** Iowa State University, Ames, IA, US.

Performance of a resistant variety in a soybean cyst nematode (*Heterodera glycines* Ichinohe, SCN)-infested field depends on the genetics of both the soybean (*Glycine max* L. (Merr.)) and the nematode. Our objective was to determine the dynamics of interactions between SCN resistance and nematicide on soybean growth and development. The experiment was conducted at two locations from 2005 to 2006 in Iowa. The experimental design was a randomized complete block in a split-plot arrangement with four replications. Main plots were two treatments of a nematicide (Temik 15 G) at 0 and 7 lb/acre. The sub-plots were 10 soybean cultivars with different sources of SCN resistance. Sections of 0.76 m<sup>2</sup> were hand-harvested from each plot and used to determine dry matter on 21-day intervals starting 21 days after emergence. There were six sampling dates throughout the growing season. Averaged across all environments, the SCN-resistant varieties had the highest yield, and yield variation was closely related to the variation in crop growth rate and seed number per unit area. Temik did not influence soybean growth and development. It was concluded that SCN-resistant varieties have improved stress tolerance and can minimize a stress, like SCN, that slows crop growth rate during the early reproductive period that will negatively affect seed number per unit area, the primary yield component.

**AN ATLAS OF MARINE NEMATODE MORPHOLOGICAL DIVERSITY AS A TOOL FOR SYSTEMATICS AND FUNCTIONAL BIOLOGY. M. Mundo-Ocampo, J. G. Baldwin.** University of California, Riverside, CA, USA.

Although molecular phylogenetics is providing new insight in diagnostics and phylogenetics of marine nematodes, insight into comparative morphological diversity remains crucial to understanding functional biology and evolution. The remarkable diversity of marine nematodes is demonstrated with the aid of increasingly sophisticated light, through-focus video and scanning electron microscopy in combination with digital imaging. Images expose an array of previously undescribed cuticular patterns, elaborations of a wide array of sensory organs and details of external copulatory structures. These observations increase the relevance and accuracy of morphologically supported phylogenies, classification and identification. They further provide a basis to establish hypotheses of character homology across taxa and to hypothesize specific functional adaptations.

**THE POTENTIAL OF THE FUNGUS *MUSCODOR ALBUS* AS A BIO-CONTROL AGENT AGAINST ECONOMICALLY IMPORTANT PLANT-PARASITIC NEMATODES OF POTATOES IN WASHINGTON STATE. E. Riga,<sup>1</sup> L. Lacey<sup>2</sup>, N. Guerra.<sup>1</sup>** <sup>1</sup>Washington State University, IAREC, Prosser, WA, <sup>2</sup> USDA-ARS, Wapato, WA.

The following plant-parasitic root-knot nematodes, *Meloidogyne chitwoodi* and *M. hapla*, the stubby root nematode, *Paratrichodorus allius*, and the lesion nematode, *Pratylenchus penetrans*, are of great economic importance to the potato industry in the Pacific Northwest and specifically to Washington State. It is challenging to develop economically sustainable management strategies that will control all of the above species. The fungus *Muscodor albus*, which produces a mixture of antimicrobial volatile organic chemicals, was tested for its nematicidal and nematostatic activity against the plant-parasitic nematodes under laboratory (chamber) and greenhouse conditions. Juveniles were exposed to volatiles generated by *M. albus* rye grain culture for 72 hours in hermetically sealed 28.3 liter chambers at 24°C. The mean percent mortalities of nematode juveniles exposed to *M. albus* were 70% for *M. chitwoodi*, 83% for *P. allius* and 86% for *P. penetrans* in comparison to 3%, 11% and 7% of the untreated controls, respectively. Only 7% of *M. hapla* were killed by *M. albus*; however, the surviving juveniles displayed reduced movement and response to physical stimuli in comparison to the control, which is evidence of nematostasis. The greenhouse/nematode host pot study showed that *M. albus* applied at 0.5% and 1.0% w/w reduced all plant-parasitic nematodes species including *M. hapla* by 80–100% in comparison to the untreated controls.

**USE OF ENTOMOPATHOGENIC NEMATODES (EPN) AND NEMATODE-TRAPPING FUNGI (NTF) FOR THE SUPPRESSION OF *MELOIDOGYNE JAVANICA*. D. J. Fallon,<sup>1</sup> H. K. Kaya,<sup>2</sup> B. S. Sipes.<sup>1</sup>** <sup>1</sup>University of Hawaii, <sup>2</sup>University of California, Davis.

Interactions between native *Steinernema feltiae* MG-14 and non-native *S. glaseri* NJ and *Steinernema feltiae* SN with native *Arthrobotrys oligospora* and *Monacrosporium ellipsosporum* were investigated for the suppression of *Meloidogyne javanica*. In a corn-meal-agar bioassay, *S. feltiae* MG-14 survival in the presence of *A. oligospora* and *M. ellipsosporum* averaged 81% of control survival, compared to 22% for *S. feltiae* SN and 34% for *S. glaseri* NJ after 25 days of exposure ( $P < 0.05$ ). In a two-trial, clay pot system using a 1:1 sand:soil mix, there was a 28% and 78% reduction in collected *Meloidogyne javanica* J2 7 days post-inoculation from pots treated with *S. glaseri* NJ and *A. oligospora* compared to pots



treated with *S. glaseri* NJ, where the NTF was absent. In contrast, there was only a 3% and a 21% reduction in J2 numbers using *S. feltiae* MG-14 between NTF treatments ( $P > 0.05$ ). However, the number of recovered *S. feltiae* MG-14 and *S. glaseri* NJ infective juveniles (IJ) from pots treated with *A. oligospora* was often lower than collected from non-*A. oligospora*-inoculated pots, implying both EPN isolates were susceptible to NTF predation. The susceptibility of native and non-native EPN to NTF, and their potential for use in plant-parasitic nematode suppression, requires further study.

DISTRIBUTION AND FREQUENCY OF *HETERODERA GLYCINES* AND OTHER PLANT-PARASITIC NEMATODES IN THE ORGANIC-FARMING FIELDS IN MINNESOTA. **S. Chen,<sup>1</sup> C. C. Sheaffer,<sup>2</sup> D. L. Wyse,<sup>2</sup> P. Nickel,<sup>3</sup> H. Kandel,<sup>4</sup> C. M. Fernholz.<sup>4</sup>** <sup>1</sup>University of Minnesota Southern Research and Outreach Center, Waseca, MN 56093, <sup>2</sup>Department of Agronomy and Plant Genetics, University of Minnesota, Saint Paul, MN 55108, <sup>3</sup>University of Minnesota Southwest Research and Outreach Center, Lamberton, MN 56152, <sup>4</sup>University of Minnesota Extension Regional Center Crookston, Crookston, MN 56716.

A survey was conducted to determine distribution and frequencies of *Heterodera glycines* and other major plant-parasitic nematodes in the organic-farming fields in Minnesota. A total of 31 soil samples were collected from southeast (SE), 26 samples from southwest (SW), 28 from west-central (WC) and 23 from northwest (NW) Minnesota. Nematodes were extracted from the soil with the hand decanting and sucrose centrifugation method. The composition of plant-parasitic nematodes varied among the four regions. The second-stage juveniles of *H. glycines* were found in 41.9, 84.6, 10.7 and 0% fields with relative prominence (RP) values of 10.1, 26.4, 0.5 and 0 in the SE, SW, WC and NW regions, respectively. In SE, other common genera included *Pratylenchus* (41.4, RP value, same below), *Helicotylenchus* (39.5), *Xiphinema* (4.8) and *Paratylenchus* (3.2). In SW, other common genera were *Helicotylenchus* (38.5), *Pratylenchus* (18.2), *Xiphinema* (12.0) and *Tylenchorhynchus* and related genera (4.5). In WC, *Helicotylenchus* (38.9), *Pratylenchus* (29.8), *Tylenchorhynchus* and related genera (15.9), *Paratylenchus* (7.8) and *Meloidogyne* (3.8 RP in 7.1% fields) were also common. In northwest, the most common nematodes were *Tylenchorhynchus* and related genera (44.7), followed by *Helicotylenchus* (33.1), *Paratylenchus* (15.9) and *Pratylenchus* (4.6).

EXAMINING A MAP KINASE PATHWAY INDUCED DURING *MELOIDOGYNE INCOGNITA* INFECTION. **K. L. Lagor, X. Li, Z. Shen, S. P. Briggs.** University of California—San Diego, San Diego, CA, USA.

The root-knot nematode, *Meloidogyne incognita*, is a major crop pest worldwide that induces nuclear proliferation without cell division in host plant root cells during the early stages of infection. How the nematode uncouples cytokinesis is yet unknown. Using both genome-wide expression analyses from the model dicot, *Arabidopsis thaliana*, as well as traditional molecular techniques, we found a highly up-regulated MAP kinase pathway with demonstrated roles in cell division. Induction in response to infection was confirmed using promoter::reporter analyses. We are currently examining the role this MAP kinase pathway plays during the early stages of infection as well as the methods of induction of this pathway by the nematode.

ENTOMOPATHOGENIC NEMATODES FOR BIOLOGICAL CONTROL OF SOYBEAN SOIL PESTS IN THE NORTHEAST OF CHINA. **Y. Xu, X. Qian, C. Li.** Northeast Institute of Geography and Agricultural Ecology, CAS, Harbin, Heilongjiang, China.

An entomopathogenic nematode (EPN), *Heterorhabditis bacteriophora*, has been tested as biological control agent against a main soil pest, *Holotrichia oblitata* Faldermann, of soybean fields in northeast of China. The tests have been conducted in Petri dishes, pots and field trials. The results show that the nematode caused a host mortality of 95.8% in Petri dish assay with the dose of 60 IJ/insect, 64% in pot assay and 50% in field trial by the dose of 0.6 million IJ/m<sup>2</sup>. However, in field assay, when the nematode dose increases to 1 million IJ/m<sup>2</sup>, the nematode can cause as high as 95% host mortality, which indicated that the nematode can be used in organic soybean production in the region.

TRANSGENIC *ANTHURIUM ANDRAEANUM* EXPRESSING MODIFIED RICE CYSTEINE PROTEASE INHIBITOR AND RESISTANCE TO *RADOPHOLUS SIMILIS*. **T. Khaithong,<sup>1</sup> B. S. Sipes,<sup>1</sup> A. R. Kuehnle.<sup>2</sup>** <sup>1</sup>Departments of Plant and Environmental Protection Sciences and <sup>2</sup>Tropical Plant and Soil Sciences, University of Hawaii at Manoa, 3190 Maile Way, Honolulu, HI 96822.

The burrowing nematode, *R. similis*, causes decline in *Anthurium*, reducing size and number of flowers produced as well as plant productive life. Development of anthurium cultivars expressing modified rice cysteine protease inhibitor (OcldeltaD86) targeting nematode digestive enzymes could be an option for *R. similis* control. *Agrobacterium*-mediated transformation of etiolated shoot explants of *A. andraeanum* cv. 'Marian Seefurth', 'Paradise Pink' and 'Anuenue' was performed. Callus induction was carried out in half-strength MS containing 50 mg/liter kanamycin and 500 mg/liter carbenicillin supplemented with 0.4 mg/liter 2,4-D and 0.2 mg/liter BA or 0.2 mg/liter IBA and 0.1 mg/liter TDZ. Germinated shoots were selected on half-strength MS supplemented with 150 ml/liter coconut water containing 100 mg/liter kanamycin and 100 mg/liter carbenicillin. Preliminary screening for *R. similis* resistance was performed in A.

*andraeanum* cv. 'Marian Seefurth' lines obtained through selective media using single root explants inoculated with five gravid females in vitro. Total nematode number was reduced by 30% in the transformed lines compared to untransformed lines 30 days after inoculation. Transgenic anthurium expressing modified rice cysteine protease inhibitor is a promising measure for *R. similis* control.

EFFECTS OF SOIL PHYSICAL CHARACTERISTICS ON STING NEMATODE IMPACTS TO FLORIDA STRAWBERRY. **J. W. Noling**, University of Florida, Citrus Research & Education Center, Lake Alfred, FL 33850, USA.

Two strawberry fields with long histories of severe damage due to the Sting nematode, *Belonolaimus longicaudatus*, were identified in Dover, FL. In each field, final harvest strawberry plant sizes were counted into three sizes classes (S, Med, Lg) within 25-ft linear section of row during years 2004 to 2007. Plant size distribution maps were developed for each year and used to determine whether specific locations within the field exhibiting excellent to poor strawberry growth were related to nematode density and specific physical characteristics of soil. Hyperspectral reflectance and other plant, field and aerial imaging technologies were also used to characterize differences in plant sizes and nematode induced crop stresses. NDVI field maps were generated from near infrared (810 nm) images aerially acquired from an altitude of 3,000 ft. A monitoring network of wells was installed to measure depth to spodic horizon and weekly changes in shallow ground water table after rainfall and irrigation. Ground water hydrology and topography were interpolated from individual points of well data. Post-harvest patterns of plant damage were not correlated with physical characteristics of soil (penetration resistance, particle size distribution or color). Transient subsurface flooding events did not appear to affect nematode survivorship and soil fumigation efficacy with methyl bromide.

PLANT PROTEINASE INHIBITORS AS A NATURAL AND INTRODUCED DEFENSE MECHANISM FOR ROOT-KNOT NEMATODES IN *COFFEA ARABICA*. **R. Y. Cabos**,<sup>1</sup> **B. S. Sipes**,<sup>1</sup> **D. P. Schmitt**,<sup>1</sup> **H. J. Atkinson**,<sup>2</sup> **C. Nagai**.<sup>3</sup>  
<sup>1</sup>University of Hawaii, <sup>2</sup>University of Leeds, <sup>3</sup>Hawaii Agriculture Research Center.

Root-knot nematodes significantly reduce yields on coffee plantations throughout the world. Coffee produces a number of compounds that act as natural defense mechanisms against pathogens. Genes similar to known plant proteinase inhibitors have been discovered in *Coffea arabica* that may inhibit nematode development to some degree. The addition of proven nematode resistance genes, a modified rice cystatin (Tubulin-OcI-deltaD86) with or without a cowpea trypsin inhibitor (35S-OcI-deltaD86/GO/CpTI), reduced the *Meloidogyne konaensis* population by over 70% in 26% of transgenic coffee plants in a bioassay. In other transgenic lines, the introduction of resistance genes caused silencing of the endogenous proteinase inhibitors or the introduced ones themselves. In plants where silencing was observed through quantitative RT-PCR, extreme susceptibility against *M. konaensis* was observed. Twenty-one percent of transgenic plants tested had over 150% more nematodes than wild-type *C. arabica*. Clones can be produced from promising lines resistant to *M. konaensis* for use as rootstocks. Pyramiding nematode resistance genes with endogenous proteinase inhibitors can enhance coffee's resistance to root-knot nematodes.

MOLECULAR AND MORPHOLOGICAL EXAMINATION OF A PINEAPPLE AGRICULTURAL SYSTEM IN HAWAII. **T. G. Quintero**, **B. S. Sipes**, Department of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822.

A molecular method for studying nematode communities was investigated. Molecular sequence analysis of soil samples was compared to traditional sampling and visual identification of an agricultural pineapple system in Hawaii. Traditional sampling identified *Rotylenchulus reniformis* and *Paratylenchus* sp. as the primary nematode species present at 48% and 19% prevalence, respectively. However, there was a 42% prevalence for *Globodera* and 23% for *Prismatolaimus dolichurus* using molecular sequencing. *Paratylenchus* was unidentified via molecular analysis, and *Prismatolaimus* accounted for less than 1% of the traditional sample. Unfortunately limits in the nematode database raise doubts about the veracity of the molecular analysis, as visual identification clearly showed no *Globodera* present. In addition, there is no reported presence of *Globodera* in Hawaii. Although a sequence for *Rotylenchulus reniformis* is present in the database, it was not detected in the sample. Molecular analysis is a promising tool to study nematode communities, but its success is dependent on the completeness of the nematode database.

ENGINEERING RESISTANCE AGAINST SOYBEAN CYST NEMATODES. **X. Huang**, **A. Wiig**,<sup>1</sup> **S. Hill**,<sup>1</sup> **R. Ascenzi**,<sup>1</sup> **B. McCaig**,<sup>1</sup> **Y. S. Liu**,<sup>1</sup> **S. Motyka**,<sup>1</sup> **J. Dong**,<sup>1</sup> **M. Offenheiser**,<sup>1</sup> **P. Puzio**,<sup>2</sup> **L. Talton**,<sup>1</sup> **R. Zhen**,<sup>1</sup> **V. Mitten-dorf**.<sup>1</sup> <sup>1</sup>BASF Plant Science LLC, Durham, NC, USA, <sup>2</sup>Metanomics GmbH, Berlin, Germany.

The soybean cyst nematode is by far the most important pathogen on soybean in the US. We shall discuss the use of genomic data to identify candidate genes and promoters that we test for SCN resistance in a transgenic soybean root bioassay. One approach is the use of RNAi technology that will directly affect the nematode. A second approach would be to alter the plants' ability to support nematode growth and reproduction. It is hoped that this approach will eventually result in soybean cultivars with durable, broad-spectrum resistance against SCN.

THE COMPATIBILITY BETWEEN A BIOLOGICAL CONTROL AGENT AND PESTICIDES. **Y. Chen, P. J. Chen, T. Tsay.** Dept. Plant Pathology, National Chung Hsing University.

The chitinolytic actinomycete *Streptomyces saraceticus* (SS 31) has been used successfully in the control of plant-parasitic nematodes in Taiwan. Seventy-two commonly used pesticides from different categories were tested for their compatibility with SS 31. Spore suspension (0.2 ml) of SS 31 was spread evenly on a PDA plate. Three paper discs (8-mm diam.), each containing 50 µl of a pesticide at the recommended concentration, were placed in the seeded plate. A pesticide which is inhibitory to the growth of SS 31 would result in the formation of an inhibition zone around the paper disc. Among the 41 fungicides tested, 19 were inhibitory to SS 31. The fungicides inhibitory to SS 31 included copper hydroxide 77% WP, kasugamycin + copper oxychloride 81.3% WP, copper hydroxide + oxine-copper 40% WP and copper oxychloride + metalaxyl 76.5% WP, indicating that pesticides with copper were strongly inhibiting to SS 31. All six herbicides tested were inhibitory to SS 31. Among them, glyphosate-ammonium 41% SL and glufosinate-ammonium 13.5% SL, which belong to Amino acid category, included a large clear zone. Among 12 tested insecticides, only abamectin 2% EC could inhibit SS 31. Pyrethroid, carbamate, neonicotine and the mixed compounds did not effect its growth. As to three nematicides tested, only ethoprop 45% EC inhibited SS 31. Niclosamide 70% WP, the molluscicide, also inhibited SS 31. Five acaricides, including organophosphate, organotin compounds, fatty acid esters and benzoate, two plant growth regulators, and one insect growth regulator showed no inhibitory effect on SS 31.

DOWN-REGULATION OF *ARABIDOPSIS* CALCIUM-DEPENDENT PROTEIN KINASE GENES BY CYST NEMATODE INFECTION. **J. Jin,<sup>1</sup> M. Mazarei,<sup>2</sup> M. J. E. Wubben II,<sup>3</sup> T. J. Baum.<sup>1</sup>** <sup>1</sup>Department of Plant Pathology, Iowa State University, Ames, IA 50011, USA, <sup>2</sup>Department of Plant Sciences, University of Tennessee, Knoxville, TN 37996, USA, <sup>3</sup>USDA-ARS Crop Science Research Laboratory, Genetics and Precision Agriculture Unit, Mississippi State, MS 39762, USA.

The formation of syncytia, the elaborate feeding cells of plant-parasitic cyst nematodes, in host roots is accompanied by massive plant gene expression changes. We previously described that the mRNA steady-state levels of two *Arabidopsis* calcium-dependent protein kinase genes (*CDPK9* and *CDPK26*) were significantly decreased in roots three days after inoculation (dai) with the sugar beet cyst nematode *Heterodera schachtii*. Here, we used quantitative real-time RT-PCR analysis to extend these results and found suppression of *CDPK9* mRNA levels up to at least 8 dai. Histochemical GUS analyses of transgenic *Arabidopsis* containing a *CDPK9::GUS* promoter construct revealed that the *CDPK9* promoter activates *GUS* gene expression throughout the roots and shoots during plant development except in root tips and young emerging shoot tissues. When such transgenic plants were infected by *H. schachtii*, a strong decrease in GUS activity was detected in syncytia. Preparation and analyses of transgenic *Arabidopsis* plants containing the *CDPK26::GUS* promoter construct are underway. *Arabidopsis* mutants with T-DNA insertions in either of these two genes were further studied to characterize the functions of these two CDPK. A mutant line containing a *CDPK9* null allele exhibited no obvious phenotype changes during plant development and nematode infection, probably due to genetic redundancy. Other *CDPK9* and *CDPK26* knockout mutants currently are being analyzed.

NEMATOCIDAL ACTIVITY OF *MUSCODOR ALBUS*. **D. R. Jimenez, D. C. Manker.** Agraquest Inc., Davis, CA, USA.

From 1995 to 2002, AgraQuest Inc. screened 20,000+ microbial isolates against insects, plant pathogens and nematodes. Over 200 isolates advanced to secondary testing based on nematocidal activity. Many isolates were advanced for strain identification and chemical characterization. In 2002, several of these isolates were dropped in favor of the endophytic fungus *Muscodora albus*. *Muscodora albus* is recognized for its production of volatile hydrocarbons with broad antifungal activity. Soil incorporation testing has shown *M. albus* to be active against plant-pathogenic nematodes. In Easter lily production, *M. albus* with Thimet gave the highest bulb per plant weights and the lowest nematode counts when compared with Fosthiazate, Mocap or NemaCur. In field tomatoes microplots inoculated with *Meloidogyne incognita*, *M. albus* treatment gave yield data and galling indices statistically equal to plots treated with Telone or methyl bromide. Based on GC-MS analysis of the primary volatiles released in soil, we have identified simplified artificial mixtures that show significant activity against insects, nematodes and soil-borne pathogens. Mixtures of these volatile organics show synergistic activity and confer target specificity when tested in vitro.

EFFECT OF DIFFERENT WINTER COVER CROPS, USED AS BIOFUMIGANTS, ON *MELOIDOGYNE INCOGNITA* POPULATION LEVELS AND YIELD AND INFESTATION OF SUMMER-GROWN TOMATO. **A. T. Ploeg.** Dept. Nematology, Univ. California, Riverside, CA, USA.

Broccoli, carrot, marigold, nematode-resistant tomato and strawberry were grown for three years during the fall/winter in a root-knot nematode (*Meloidogyne incognita*)-infested field in Southern California. In spring, the tops of all crops were shredded and incorporated in the soil. Additional amendment with chicken manure was included as a sub-treatment. The soil was then covered with clear plastic for 6 weeks to achieve biofumigation. Neither the different crops nor the chicken

manure directly affected the root-knot nematode population levels. However, nematode symptoms on the tomato roots were reduced when the previous crop was broccoli, and tomato yields were highest after this crop. We conclude that cultivation of, followed by biofumigation with, broccoli indirectly affected the infectivity of *M. incognita*, possibly by delaying or preventing a portion of the nematodes to reach the host roots. Chicken manure amendment did not affect tomato root symptoms but did result in higher tomato yields in 2 years. We further conclude that there is little risk of increasing *M. incognita* populations, even under a host crop, during the cool season when soil temperatures remain low (<18°C).

**MELOIDOGYNE INCOGNITA HOST STATUS AND BIOFUMIGANT EFFECT OF BRASSICA CROP CULTIVARS. A. Ploeg, S. Edwards.** Dept. Nematology, Univ. California Riverside, CA, USA.

Biofumigation using brassicas has been used to manage weeds, soil-borne pathogens and nematodes. Generally, brassicas are grown as cover crops, mowed at flowering, chipped and then immediately incorporated into the soil. Fields are then irrigated and sometimes covered with plastic. During brassica tissue decomposition, volatile compounds are formed with biocidal properties. Glucosinolates occurring in brassicas are converted enzymatically into isothiocyanates, which also are the active ingredients of some synthetic nematicides. A major drawback of using this strategy for control of root-knot nematodes is that many brassicas are moderately good hosts for the nematodes, thus risking an increase in nematode population during the cultivation of the brassica crop. Ideally, a brassica crop cultivar should be used that combines nematode resistance with a strong biofumigant activity. In a greenhouse pot experiment, we tested 32 brassica varieties for the development of root symptoms and *M. incognita* nematode population levels 6 weeks after inoculation with 20,000 *M. incognita* eggs. There were large differences between varieties: after the 6 weeks the number of second-stage juveniles per 100 g soil ranged from 7 to 2,850 (tomato positive control 4,270). The green biomass was then incorporated into the soil, and melon transplants were added 2 weeks later as a bioassay. The severity of root-galling on melon generally reflected the nematode density prior to biomass incorporation. A number of selected varieties are currently being tested under outdoor micro-plot conditions.

**A STUDY OF THE MOLECULAR MECHANISMS OF DESICCATION TOLERANCE IN ANTARCTIC DRY VALLEY NEMATODES USING EXPRESSED SEQUENCED TAGS (EST). B. N. Adhikari,<sup>1</sup> E. Ayres,<sup>2</sup> B. Simmons,<sup>2</sup> D. H. Wall,<sup>2</sup> B. J. Adams.<sup>1</sup>** <sup>1</sup>Brigham Young University, Provo, UT, USA, <sup>2</sup>Colorado State University, Fort Collins, CO, USA.

Antarctic nematodes of the McMurdo Dry Valleys display a wide array of adaptations to environmental stress, including freezing tolerance and anhydrobiosis. It has been hypothesized that these survival strategies are exaptations (preadaptations) that originally evolved in response to desiccation. Though the morphological and physiological mechanisms governing desiccation and freeze tolerance in Antarctic nematodes are well studied, the molecular mechanisms are poorly understood. To better understand the molecular genetic mechanisms involved in how these Antarctic nematodes survive in such a harsh environment, we are examining the Expressed Sequenced Tags (EST) of several Antarctic nematode species. A preliminary analysis indicates at least one species uses a suite of global stress tolerance response genes that is triggered by desiccation. These responses include metabolic and environmental processing/sensing genes and stress-induced proteins (heat shock proteins), metabolic response genes, signaling pathway genes (ras-related proteins) and several unidentified proteins. A better understanding of the molecular mechanisms that determine how nematodes respond to environmental stress will allow us to better model and predict their contributions to ecosystem functioning under climate and habitat change.

**FORAGING AND INFECTION DECISIONS IN ENTOMOPATHOGENIC NEMATODES. G. N. Stevens,<sup>1</sup> E. E. Lewis.<sup>2</sup>** <sup>1</sup>Dept. of Nematology, UC Davis, Davis, CA, USA, <sup>2</sup>Dept. of Nematology and Dept. of Entomology, UC Davis, Davis, CA, USA.

Entomopathogenic nematodes (EPN) are important natural enemies of many soil-dwelling insects and often play key roles in natural and managed soil food webs. In managed ecosystems, their effectiveness as biological control agents may be improved by examining the foraging and infection decisions made by individual nematodes. We conducted a series of laboratory and greenhouse experiments designed to examine how factors such as soil resource heterogeneity, host availability and host infection status interact to influence EPN dispersal. The experiments focused on the responses of two entomopathogenic nematode species, *Steinernema carpocapsae* and *Steinernema glaseri*. These two species have been found in mixed-species populations in the field and differ significantly in terms of foraging strategy (principally ambushing vs. cruise-foraging, respectively). The results of our experiments demonstrate that they also differ significantly in their responses to various cues in the soil. For example, when infective juveniles were presented with the choice between invading an uninfected insect and a heterospecific infection, *S. carpocapsae* IJ avoided heterospecific infections, while *S. glaseri* IJ consistently invaded heterospecific-infected hosts. These and other results we will discuss suggest that inter-specific variation in decision making may have important ramifications for effective biological control.

EFFECT OF *GALLERIA MELLONELLA* HOSTS ON PHASE VARIATION IN *PHOTORHABDUS LUMINESCENS*. **G. C. Bailey, B. J. Adams.** Brigham Young University, Provo, UT, USA.

*Heterorhabditis bacteriophora*, paired with its bacterial symbiont, *Photorhabdus luminescens*, has been successfully used as a biological pest control agent. Effective culturing of the nematode on an industrial scale depends on the presence of primary phase bacteria. When repeatedly subcultured in low osmolarity broth, primary phase bacteria are induced to switch phases. Placing bacteria in a high osmolarity environment soon after they have switched from primary to secondary phase causes them to revert back to primary phase. Alternatively, continued subculturing in low osmolarity broth locks bacteria into secondary phase. *Photorhabdus luminescens* (TT01) were subcultured in low osmolarity Y1 broth with phase variation measured via NBTA and McConkey agar. Upon switching to secondary phase, bacteria were injected into *Galleria mellonella* larvae. After 4 days of incubation, the larvae were dissected. The bacteria were isolated on lipid agar plates with rifampin. Phase variations were then reassessed. The effect of the insect host on secondary phase variants is discussed.

BIODIVERSITY AND ECOSYSTEM FUNCTIONING IN VICTORIA LAND NEMATODES: A MOLECULAR APPROACH. **A. R. Dillman,<sup>1</sup> T. S. Davie,<sup>1</sup> J. M. Chaston,<sup>2</sup> S. M. Peat,<sup>1</sup> E. Ayres,<sup>3</sup> B. Simmons,<sup>3</sup> J. E. Barrett,<sup>4</sup> D. H. Wall,<sup>3</sup> B. J. Adams.<sup>1</sup>** <sup>1</sup>Brigham Young University, Provo, UT, USA, <sup>2</sup>University of Wisconsin, Madison, WI, USA, <sup>3</sup>Colorado State University, Fort Collins, CO, USA, <sup>4</sup>Virginia Tech, Blacksburg, VA, USA.

Ecological research has established that the functional characteristics, variation, distribution and abundance of resident organisms largely determine ecosystem properties. Despite a large increase in ecosystem research, there remain uncertainties about the relationship between biodiversity and ecosystem functioning; this is largely due to perceived differences between natural and experimental systems. We evaluate, at the molecular level, the biodiversity of Victoria Land, Antarctica, in order to understand how it relates to ecosystem functioning. This natural ecosystem is particularly important because it is of an extremely low-diversity system and displays little ecological redundancy in niche occupation. We specifically assess the biodiversity and distribution of nematodes in this geographic area, focusing on the elucidation of how this relates to ecosystem functioning and trophic relationships.

THE INHIBITORY EFFECTS OF *STREPTOMYCES* SPP. TO PLANT PATHOGENIC FUNGI. **Y. Chen, P. J. Chen, T. Tsay.** National Chung Hsing University.

*Streptomyces saraceticus* 31 (SS31), *Streptomyces* sp. 205 (S205) and *Streptomyces* sp. 233 (S233) were isolated from a citrus orchard in Taiwan. All of them had the ability to degrade chitin. In our previous study, SS31 was found to have the ability to reduce the populations of root-knot (*Meloidogyne incognita*) and lesion (*Pratylenchus penetrans*) nematodes when mixed with the organic amendment. In this study, we found that SS31 also was strongly inhibitory to *Phytophthora infestans*, *Fusarium oxysporum* f. sp. *cucumerium* and *Monosporascus cannonballus*, while S205 and S233 were not inhibitory to these pathogens. In order to obtain the maximum growth of the three streptomyces strains, 2% soybean, oat, milk and maize liquid cultural medium were evaluated, and maize medium could increase all strains to more than 10<sup>7</sup> CFU after 5 days at 25°C, 80 rpm. After growing in the soybean media for 7 days, streptomyces strains were filtered, and the supernatant was collected. Two-hundred microliters of the supernatant derived from these three strains, respectively, was added in the petri dish with fungal pathogens, and no inhibitory effects were observed. The results indicated living streptomyces could be necessary for inhibiting the growth of the tested fungal pathogens.

NEMATODE TAXOCOENOSES IN THE GUADIAMAR GREEN CORRIDOR (SW IBERIAN PENINSULA). **R. Peña-Santiago,<sup>1</sup> D. Jiménez-Guirado,<sup>2</sup> R. Murillo,<sup>2</sup> G. M. Liébanas,<sup>1</sup> J. Abolafia,<sup>1</sup> P. Guerrero.<sup>1</sup>** <sup>1</sup>University of Jaén, Spain, <sup>2</sup>University of Córdoba, Spain.

The nematode community belonging to the orders Dorylaimida, Mononchida and Rhabditida from soils of a protected area (Guadamar Green Corridor, SW Iberian Peninsula) is studied. Eighty soil samples were examined, collected from eight sectors in which the Corridor area was divided according with an eco-regional approach. Data of biodiversity (species richness) were analyzed, and classifications of species and of sectors were performed. One hundred and three species (64 dorylaims, 5 mononchs and 34 rhabdits) were identified. Dorylaimid taxocoenosis represents almost two-thirds (62%) of all nematode fauna examined, practically twice the number of rhabdits; the latter, however, are more frequent than the former, i.e., they show in general a wider spread. No significant difference in species richness values was found throughout the Guadamar river basin. An incipient environmental gradient of diversity (species richness per soil sample) is detected when the distance to river bank is considered, mostly due to dorylaims. In addition to cosmopolitan species, each sector possesses a series of characteristic or exclusive species, a fact that supports the interest of this geographical area for conservation purposes. The results suggest that nematode taxocoenoses shows different distributional strategies.

MANAGEMENT OF ROOT-KNOT NEMATODE AND PYTHIUM ROOT ROT IN PEPPER. **J. A. Thies,<sup>1</sup> D. W. Dickson,<sup>2</sup> E. Roskopf,<sup>3</sup> M. Mendes.<sup>2</sup>** <sup>1</sup>U.S. Vegetable Laboratory, USDA, ARS, Charleston, SC, <sup>2</sup>University of Florida, Gainesville, FL, <sup>3</sup>USHL, USDA, ARS, Ft. Pierce, FL.

Five pepper genotypes and four soil treatments were evaluated in field trials for management of root-knot nematodes and Pythium root rot in Charleston, SC, and Citra, FL. The pepper genotypes, which differed in resistance to *Meloidogyne*

*incognita* and *Phytophthora capsici*, included the root-knot nematode-resistant 'Charleston Belle' pepper and its susceptible recurrent parent, 'Keystone Resistant Giant', CM-334, resistant to both root-knot nematode and *Phytophthora capsici*, 'Paladin', a *Phytophthora*-tolerant bell pepper and 'Jupiter', highly susceptible to *P. capsici*. Resistance to *Pythium* has not been reported in pepper. The soil treatments consisted of pre-plant fumigation with methyl bromide (98:2), BioPhos (drip application), Ridomil Gold (drip application) and a non-treated control. All genotypes performed well in the methyl bromide treatment. Overall, plants exhibited less root rot, wilting and chlorosis in the BioPhos and Ridomil treatments, even though significant differences were not detected compared to the non-treated control. CM-334 exhibited high resistance to root-knot nematodes and *Pythium* root rot and low AUDPC for wilting and chlorosis associated with *Pythium* root rot. 'Charleston Belle' exhibited high resistance to root-knot nematodes, but was susceptible to *Pythium* root rot. 'Paladin', 'Keystone Resistant Giant' and 'Jupiter' were susceptible to root-knot nematodes and *Pythium* root rot.

USING BIOINTENSIVE PRACTICES TO MANAGE TWO INTERACTING PATHOGENS. **A. E. MacGuidwin,<sup>1</sup> D. L. Knuteson<sup>2</sup>**. <sup>1</sup>Departments of Plant Pathology and <sup>2</sup>Horticulture, University of Wisconsin-Madison, Madison, WI.

*Verticillium dahliae* (Vd) and *Pratylenchus penetrans* (Pp) interact to cause the potato early dying disease (PED). We tested biointensive practices of cover crop green manures and solarization to manage both pathogens in two commercial fields. Cover crops reported to suppress Vd (sorghum-sudangrass (ss), corn and rapeseed) or Pp (forage pearl millet (fpm) and marigold) were planted in a split-plot experiment with solarization (+/-) as the subplot. Solarized plots were covered with clear polyethylene film after green manure incorporation. Metam sodium was applied adjacent to the study sites in October, and potato was planted in year two. Solarized plots had lower inoculum densities of Pp and Vd when potatoes were planted and increased yield. There was a significant main effect of cover crop at planting for experiment two, with fpm and marigold superior to the other crops for nematode suppression. Plots that had been planted with fpm the year before also had a lower incidence of Vd in potato stems. Cover crops were significant for yield in the absence of solarization in experiment two, and yields were greater following fpm and millet than the other cover crops. Samples collected from fumigated areas were similar to solarization and marigold and fpm cover crops for most measures. These results demonstrate the importance of using strategies aimed at both root lesion nematodes and *Verticillium* in fields with high potential for PED.