Journal of Nematology 26(4):538–571. 1994.

ABSTRACTS

SOCIETY OF NEMATOLOGISTS
33rd ANNUAL MEETING
SAN ANTONIO, TEXAS
14-18 AUGUST 1994

INTERACTIONS OF THREE ISOLATES OF *FUSARIUM OXYSPORUM* F. SP. *VASINFECTUM* AND *MELOIDOGYNE INCOGNITA* RACES 3 AND 4 ON FOUR COTTON GENOTYPES. **Abd-El-Alim, F. F.,[1] K. R. Barker,[1] and I. K. A. Ibrahim[2].** [1]Graduate Student and Professor, Department of Plant Pathology, North Carolina State University, Raleigh, NC 17695-7616, and [2]Professor, Plant Pathology Department, Alexandria University, Alexandria, Egypt.

Greenhouse factorial experiments were conducted to elucidate effects of races 3 and 4 of *Meloidogyne incognita* (Mi) and three isolates of *Fusarium oxysporum* f. sp. *vasinfectum* (FOV) on four cotton genotypes. Cotton seedlings (cultivars DP-50, RNR-120, RNR-249, and RNR-315) were individually inoculated with one of three isolates of FOV alone or in combination with Mi3 or Mi4, simultaneously or FOV added 3 weeks after soil infestation with Mi. Plant weight, stem length, root weight, and numbers of bolls per plant were recorded after 18 weeks. Roots were rated for galling (0-10), and necrosis indices (0-100%) and numbers of eggs per gram root were determined. Cultivar DP-50 showed a susceptible reaction to FOV in the presence of Mi3 and Mi4, as compared with other genotypes. Plants inoculated with FOV 3 weeks after soil infestation with Mi3 or Mi4 supported greater numbers of eggs and exhibited more root necrosis and galling, stem discoloration, and suppressed shoot growth. *Meloidogyne incognita* race 4 was more virulent than Mi3 on all genotypes.

SOIL MOISTURE AND THE INTERACTIONS OF *MELOIDOGYNE INCOGNITA* AND *FUSARIUM OXYSPORUM* F. SP. *VASINFECTUM* AND (OR) *RHIZOCTONIA SOLANI* ON COTTON. **Abd-El-Alim,[1] F. F., I. K. A. Ibrahim,[2] and K. R. Barker[3].** [1,3]Graduate Student and Professor, Department of Plant Pathology, North Carolina State University, Raleigh, NC 27695-7616, and [2]Professor, Plant Pathology Department, Alexandria University, Alexandria, Egypt.

The interactions of *Meloidogyne incognita* race 4 (Mi) and *Fusarium oxysporum* f. sp. *vasinfectum* (FOV) and (or) *Rhizoctonia solani* (RS), as affected by soil moisture (low, medium, and high), on cotton cultivar DP-50 were examined in a factorial microplot experiment. High moisture treatments had greater post-emergence damping-off in plants with FOV + RS + Mi, and FOV + Mi. At mid-season, numbers of eggs and second-stage juveniles of Mi were greater in low than in high moisture. *Fusarium oxysporum* population densities were fewest in low moisture soil. Cotton yield was lowest in high moisture plots with FOV + RS + Mi, and FOA + Mi. Root necrosis was enhanced by high and medium moisture vs. low moisture. The highest incidence of vascular discoloration at harvest in stems occurred in response to FOV in combination with RS and (or) Mi, but, was not affected by moisture levels.

THE INFLUENCE OF CARROT ROOT FILTRATE ON THE EMERGENCE OF *HETERODERA CAROTAE* FROM CYSTS WITH SPECIAL REFERENCE TO HOST PLANT

AGE. **Berney, M. F., G. W. Bird, and F. W. Warner.** Department of Entomology, Michigan State University, East Lansing, MI 48824.

Cysts of *Heterodera carotae* (carrot cyst nematode) from an infested carrot field were extracted and maintained at 15 C in carrot root filtrate. After being differentially preconditioned, two groups of cysts of *H. carotae* were added to the experiment each week. Cysts for the first group were maintained in field soil held at 2 C, then extracted on the day they were added to the experiment. Cysts for the second group were extracted at the beginning of the experiment, and maintained in water at 2 C until being exposed to carrot filtrate. This process was repeated for 10 consecutive weeks. Second-stage juveniles emerging from cysts were counted weekly, removed and root filtrate replaced. All of the carrot filtrate used throughout the experiment came from the same plant. Except for groups entering the experiment during the fifth week, cysts preconditioned in water had significantly higher percentages of emergence than cysts maintained in soil. For 8 of the 10 times, the time of peak emergence was the same for the two groups. However, significantly more second-stage juveniles emerged during the week of peak emergence from cysts held in water than from cysts maintained in soil.

ANALYSIS OF UP-REGULATED TOMATO GIANT CELL GENES. **Bird, David, and Mark Wilson.** Department of Nematology, University of California Riverside, Riverside, CA 92521.

Using a cloning technique that incorporated PCR amplification of cDNA and subtraction in a single strand phagemid vector, we constructed a library of transcripts exhibiting up regulation in tomato giant cells induced by the parasitic nematode *Meloidogyne incognita*. Contamination by nematode sequences was less than 1%. Transcripts from high copy number genes accounted for 14% of clones; the remaining 244 recombinants appear to be derived from distinct, unique genes. A survey of plant tissues identified transcripts present in tissues other than root, including actively dividing and expanding tissues and mature leaf. The identities of approximately 10% of the giant cell transcripts were inferred from DNA sequence data, and provide clues as to giant cell function.

COLD TOLERANCE OF STEINERNEMATID AND HETERORHABDITID NEMATODES. **Brown, Ian, and Randy Gaugler**, Department of Entomology, Rutgers University, New Brunswick, NJ 08903.

Nematodes survive subzero temperatures, using either a freeze avoiding or freezing tolerant strategy. Freeze avoiding species supercool their body fluids below the fluids' freezing point, but die when freezing occurs. Freezing tolerant species can survive the freezing of body tissues, down to a lower lethal temperature. The potential of infective juvenile nematodes from the families Steinernematidae and Heterorhabditidae to survive freezing was assessed in the laboratory. Their cold tolerance strategy was determined by observing the freezing process on a temperature controlled microscope stage. *Steinernema glaseri, S. anomali, S. feltiae, S. riobravis*, and *Heterorhabditis bacteriophora* all exhibited freezing tolerance. Lower lethal temperature and the time that each species could withstand prolonged exposure to freezing conditions were determined. Cold tolerance can be increased by acclimation to 10 C before freezing.

EFFECTS OF FURROW IRRIGATION ON THE DISTRIBUTION AND INFECTIVITY OF *STEINERNEMA RIOBRAVIS* AGAINST CORN EARWORM IN CORN. **Cabanillas, H. E., and J. R. Raulston.** Subtropical Agricultural Research Laboratory, Crop Insects Research Unit, USDA ARS, Weslaco, TX 78596.

The effects of dose and furrow irrigation on the distribution and infectivity of *Steinernema riobravis* against corn earworm were determined. Prepupae were buried in corn field soils (31 -

39 C). Parasitism was evaluated 6 days after nematode treatment. The most effective dose was 200,000 infective juveniles (IJ)/m^2 when applied via in-furrow irrigation (95% parasitism) compared with 56 and 84% parasitism when applied before and after irrigation, respectively. Parasitism on the furrow top was higher with 200,000 IJ/m^2 (100%) compared to those of 100,000 IJ/m^2 (55%), 50,000 IJ/m^2 (53%), or the control (9%). The delivery of *S. riobravis* via in-furrow irrigation shows great potential to suppress the build-up of corn earworm adult population in source areas where corn acts as a nursery crop.

CHARACTERIZATION AND UTILIZATION OF AUB623 SOURCE OF RESISTANCE IN COTTON TO *MELOIDOGYNE INCOGNITA*. **Callahan, F. E.,[1] J. N. Jenkins,[1] R. G. Creech,[2] B. Tang,[2] and P. A. Hedin[1].** [1]Crop Science Research Laboratory, USDA ARS, and [2]Department of Plant and Soil Sciences, Mississippi State University, Mississippi State, MS 39762.

Cotton (*Gossypium hirsutum*) germplasm, such as AUB623 and others derived from this source, contain resistance genes that effectively inhibit reproduction of *Meloidogyne incognita* race 3. The goal of our research has been to find the most efficient means by which to use these resistance genes in commercial lines of cotton. Present approaches involve both practical aspects, such as the use of F2 hybrids, and characterization of the chemistry and molecular biology of the interaction of the nematodes with these resistant cotton germplasms. Our results show that although second-stage juveniles (J2) penetrate resistant cotton lines in numbers similar to susceptible isolines, nematode development proceeds no further than a swollen J2 stage in the resistant lines. We are micro-sequencing a 14 kDa polypeptide that is differentially expressed in the roots of a resistant isoline soon after infection with *M. incognita*.

ENTOMOPATHOGENIC NEMATODE VERTICAL DISTRIBUTION IN TURFGRASS SOIL. **Campbell, J. F., and R. Gaugler.** Department of Entomology, Rutgers University, P.O. Box 231, New Brunswick, NJ 08903.

The vertical distribution of natural populations of *Steinernema carpocapsae*, an ambush forager, and *Heterorhabditis bacteriophora*, a cruise forager, was assessed by baiting 1-cm sections of soil cores taken from turfgrass with *Galleria mellonella*. *Heterorhabditis bacteriophora* was recovered throughout the top 8 cm and most *S. carpocapsae* were recovered from the top 2 cm. Distribution of *S. carpocapsae* changed over the course of the day, but distribution of *H. bacteriophora* did not change. More *S. carpocapsae* were obtained during the evening, night, and early morning when ultraviolet light levels were low and relative humidity was high. Differences in distribution between species probably result from differences in foraging strategy. *Steinernema carpocapsae* ambushes mobile insects by nictating on surfaces, therefore, more nematodes were recovered near the soil surface during periods of favorable environmental conditions. Because *H. bacteriophora* uses a cruise search strategy and infects *Popillia japonica* feeding on grass roots, this nematode was recovered throughout the soil profile.

PCR-BASED IDENTIFICATION OF *HETERODERA SCHACHTII* USING PRIMERS DERIVED FROM RAPD MARKERS. **Caswell-Chen, E. P., V. M. Williamson, and F. F. Wu.** Department of Nematology, University of California, Davis, CA 95616.

Random amplified polymorphic DNA (RAPD) markers were used to develop PCR markers specific to *Heterodera schachtii*. A RAPD fragment (ca. 460 bp) that was common to all examined isolates of *H. schachtii* was isolated by molecular cloning. The DNA sequence of the clone was determined and used to produce PCR primers. Amplification of DNA from different isolates of *H. schachtii* using the primers typically yielded three products, two of ca. 700-900 bp,

and one of ca. 400 bp; however, the 400 bp product was not observed in all populations of *H. schachtii*. Amplification of total *H. glycines* DNA using the primers yielded one product distinct from that of products for *H. schachtii*. Amplification of DNA from *H. cruciferae, Globodera tabacum, Meloidodera floridensis, Meloidogyne incognita, M. javanica, M. arenaria, M. hapla,* or *M. chitwoodi* using these primers did not yield any products. Primers derived from RAPD markers specific for nematode species have obvious use for nematode diagnostics.

INFLUENCE OF *PRATYLENCHUS PENETRANS* AND *VERTICILLIUM DAHLIAE* ON *SOLANUM TUBEROSUM* CULTURED WITH A SPLIT ROOT-STOLON SYSTEM. **Chen, J., and G. W. Bird.** Department of Entomology, Michigan State University, East Lansing, MI 48824.

Greenhouse and growth chamber experiments were conducted to study the influences of *Pratylenchus penetrans* and *Verticillium dahliae* on the development of *Solanum tuberosum* cv. Superior through individual and concomitant infections of isolated stolon and root tissue. Isolated basal-nodal root and stolon tissues were individually inserted into the opposite chambers of two-chamber growth containers in a total of 12 treatment combinations using a randomized block design with four replications. Thirty percent ($P \leq 0.05$) and 17% ($P \leq 0.05$) less tuber weights were found in *S. tuberosum* infected with *P. penetrans* in basal-nodal root chamber and stolon chamber, respectively. The stolon system that was exposed to either individual or combined pathogens resulted in less ($P \leq 0.05$) tuber weight. The synergistic, additive, and antagonistic joint influence of the two pathogens on *S. tuberosum* occurred once, nine times, and twice, respectively. It was hypothesized that the synergistic, additive, and antagonistic relationships could be represented by exponential, logistic, and second-order polynomial functions, respectively.

POPULATION DEVELOPMENT OF *HETERODERA GLYCINES* IN RESPONSE TO MYCOFLORA IN SOIL FROM FLORIDA. **Chen, S. Y.,[1] D. W. Dickson,[1] and D. J. Mitchell[2].** [1]Entomology and Nematology Department, and [2]Department of Plant Pathology, University of Florida, Gainesville, FL 32611.

The influence of mycoflora on *Heterodera glycines* was evaluated in soil collected from five field sites in February 1993 (two soybean fields and three other fields that had no history of previous soybean cultivation). The soil samples were either treated by heating in a microwave oven for 4 minutes or left untreated, placed in pots, planted with soybean, and inoculated with second-stage juveniles of *H. glycines*. The nematode population densities and fungal mycoflora in the nematode cysts were determined after 3 months in the greenhouse. The percentages of cysts colonized and eggs parasitized by fungi in treated soil were lower than in untreated soil. In comparison to untreated soil, the numbers of *H. glycines* in treated soil increased up to 4.3 times for cysts, 7.1 times for eggs, and 7.5 times for second-stage juveniles. The number of eggs produced per female increased 73% in treated soil compared to untreated soil. The nematode population density was negatively correlated with both the percentage of the cysts colonized and percentage of the eggs parasitized by fungi. No differences between nematode densities and frequencies of fungal colonization of cysts or eggs however were observed among the soil sources.

WATER, WATER COMPARTMENTS AND THEIR REGULATION IN SOME NEMATODES PARASITIC IN ANIMALS. **Davey, K. G.** Department of Biology, York University, North York, Ontario, Canada M3J 1P3.

While nematodes are frequently regarded as osmoconformers, evidence will be presented that at least one species is capable of short-term osmoregulation over a wide range of osmotic

environments, and that the principal site of osmoregulation is the body wall. This general osmoregulation is important to the life of the nematode not only in confronting variations in the environment, but also in maintaining its hydrostatic skeleton. There is also evidence which suggests that compartments exist in some nematodes such that water exchange between the compartments is limited and slow. The ability to regulate the internal movements of water is important in molting and the infective process. Hormones may be the mediators of osmotic control.

OBSERVATIONS ON THE BIOLOGY OF *MELOIDOGYNE NATALIEI*. **Diamond, C., G. Bird, J. Davenport, and F. Warner.** Department of Entomology, Michigan State University, East Lansing, MI 48824.

Research was conducted to determine the host-range, pathogenicity, histopathology, and life history of *Meloidogyne nataliei* associated with *Vitis labruscana*. Thirty-six plant species were evaluated as potential hosts for this nematode. Twenty of the species were evaluated under greenhouse conditions, seven species under field conditions, and nine species under growth chamber conditions. Five new hosts were identified, all in the Vitaceae family; three *Vitis* rootstocks (5BB, St. George, and *Gloire*), *Parthenocissus quinquefolia*, and *P. tricuspidata*. Pathogenicity was not observed on any of the new hosts destructively sampled after 89-182 days. Giant cells were induced in *V. labruscana* cv. Concord by the feeding activity of *M. nataliei*, but galls were never observed. Three generations of *M. nataliei* per year were detected in southwestern Michigan. Although each life cycle stage had a specific optimal development temperature in vitro, overall optimal was ca. 9 C (maximum ca. 33 C, base ca. 2.5 C).

VERTICAL DISTRIBUTION OF SOYBEAN CYST NEMATODE EGGS IN SOIL. **Donald, P., A. Keaster, R. Kremer, and J. Kendig.** Plant Science Unit, University of Missouri, Columbia, MO 65211.

Ten soil cores were collected randomly at harvest to a depth of 120 cm in a field infested with the soybean cyst nematode (SCN), *Heterodera glycines*. A SCN-susceptible soybean cultivar was grown in the field for the five previous years. The cores were divided into nine serial sections, each 11 cm. Cysts were extracted and eggs were freed mechanically from the cysts and counted. Eggs were limited to the top 22 cm of soil in 40% of the samples but were found as deep as 99 cm in other samples. Eggs were found in 90% of the samples from the top 11 cm section of soil, 70% of those from the 11-22 cm sections, 40% of the samples from 22-33 cm and 33-44 cm, 20% from 44-55 cm, 30% from 55-66 cm and 66-77 cm, 10% from each of the two deepest sections of the core. High SCN egg numbers in the soybean rhizosphere (soil surface to 20-cm deep) at soybean planting and at soybean harvest at the same site in the same year were not correlated with egg distribution to a deeper soil depth, nor were low numbers of eggs correlated with shallow egg distribution.

ANALYSIS OF VIRULENCE IN SOYBEAN CYST NEMATODE *HETERODERA GLYCINES*. **Dong, K., S. Chang, and C. Opperman.** Plant Pathology Department, North Carolina State University, Raleigh, NC 27695-7616.

Three highly inbred *Heterodera glycines* lines, each with a characteristic esterase marker, were selected as parents for testing of virulence gene(s) on resistant soybean cultivars. The host range of each line was tested. The inbred line OP50 was able to reproduce on 'Peking', 'PI88788', and 'PI90763', whereas OP20 and OP25 and OP25 reproduced only on 'Peking'. Male and female nematodes of each line were separated by culturing the inoculated plants in a hydroponic solution.

Three crosses, OP25 × OP20, OP25 × OP50, and OP20 × OP50, were made on the surface of 2% water agar media. The F1 generation females were tested for the heterozygous codominant esterase markers. To identify the numbers of dominant virulence gene(s) in each line, the avirulent parent of each cross was used as a recurrent parent to make two backcrosses. The progeny was single inoculated on susceptible host 'Lee' to produce sufficient eggs to test the host range. Random Amplified Polymorphic DNA markers have been identified among these inbred lines, and these will be used to construct a linkage map of *H. glycines* virulence genes.

INTEGRATED APPLICATION OF *PAECILOMYCES LILACINUS, PASTEURIA PENETRANS,* AND CATTLE MANURE FOR CONTROL OF *MELOIDOGYNE JAVANICA*. **Dube, B. N.** Department of Biological Sciences, University of Zimbabwe, P. O. Box MP167, Mt. Pleasant, Harare, Zimbabwe.

The integrated application of *Paecilomyces lilacinus, Pasteuria penetrans,* and cattle manure was evaluated for control of *Meloidogyne javanica* on field beans at two sites (HRS and UZ). Ten-liter polyvinyl chloride microplots were used at HRS, and 35-liter clay microplots were used at UZ. *Paecilomyces lilacinus* was applied at the rate of 5 g/liter soil (5×10^8 cfu/g substrate). *Pasteuria penetrans* and cattle manure were applied at 100 mg/liter soil and 1 ton/ha, respectively. Nematode counts were made at planting, midseason, and harvest. Root galling and yield were assessed. Although single applications of either *P. lilacinus*, *P. penetrans,* or cattle manure suppressed *M. javanica*, the integrated application of these agents significantly suppressed population densities of *M. javanica* and increased yield. At HRS, yield from microplots treated with *P. lilacinus*, *P. penetrans*, and cattle manure was increased by 40% compared to either 14%, 13%, or 8% from single applications of the respective biocontrol agents.

THE PHENYLPROPANOID PATHWAY IN SOYBEAN INFECTED WITH *MELOIDOGYNE INCOGNITA* OR WITH *HETERODERA GLYCINES*. **Edens, R.,[1] S. Anand,[2] P. Kozlowski,[1] and R. I. Bolla[1].** [1]Department of Biology, Saint Louis University, St. Louis, MO 63101-2010, and [2]University of Missouri-Columbia, Delta Center, Portageville, MO 63873.

Induction of the pathway for synthesis of glycellolin or other chemicals for a plant's response to pathogen or environmental stress was followed in soybean infected with *Meloidogyne incognita* race 3 or *Heterodera glycines* race 5. Soybean cultivars, Davis and Essex, susceptible to these nematodes and resistant cultivars, Centennial and Hartwig, were inoculated under controlled environmental conditions. Induction of transcription of mRNA encoding major enzymes within the phenylpropanoid pathway and activity of the transcribed enzymes were determined. Transcription of mRNA encoding the initial enzyme of this pathway, phenylalanine ammonia lyase (PAL), was greater after infection of resistant cultivars than of susceptible cultivars as was the activity of this enzyme. Similar changes were seen for other enzymes of branches of this pathway leading to glyceollin or lignin synthesis. Environmental stress inhibited transcription of mRNA encoding the different enzymes and enzyme activity.

MONITORING THE RESPONSE OF SOYBEAN CYST NEMATODE TO SELECTION PRESSURE FROM RESISTANT VARIETIES IN NORTHERN INDIANA. **Faghihi, J., and J. M. Ferris.** Department of Entomology, Purdue University, West Lafayette, IN 47907-1158.

Two field populations of the soybean cyst nematode (SCN), *Heterodera glycines*, were monitored between 1985-91. During this time, whenever soybean was grown in these fields, a SCN resistant cultivar was used, predominantly Fayette. Consequently, a marked reduction in the field population occurred. Four public varieties (Fayette, Cartter, Linford, and Jack), with SCN

resistance derived from PI 88788, were used in greenhouse tests to evaluate changes in pathogenic behavior of the field populations. In replicated tests, the mean number of females that developed on roots of the susceptible cultivar, Williams 82, ranged from 98 to 426. Few females (9-36/plant) developed on the four resistant cultivars. Race tests in 1987 and 1990 showed that both populations were race 3. There were no indications of change in the ability of the field populations to develop on these resistant varieties or to reduce yield during this study.

MOLECULAR SYSTEMATICS IN HETERODERIDAE. **Ferris, V. R., J. M. Ferris, and J. Faghihi.** Department of Entomology, Purdue University, West Lafayette, IN 47907-1158.

As part of an ongoing study of ribosomal DNA (rDNA) sequence data for Heteroderidae, comparisons have been made for the two internal transcribed spacers (ITS1 and ITS2) and for the 5.8S rRNA gene for five isolates per species of *Globodera*, including the two potato cyst nematodes, *G. rostochiensis* and *G. pallida* (Feltwell and Cadishead populations, respectively, from England), the horse nettle cyst nematode, *G. virginiae*, and two undescribed *Globodera* spp. from Mexico, collected from uncultivated solanaceous hosts. *Globodera virginiae* had relatively low overall similarity (ca. 85%) to the rest, which were very similar (95%+) to each other; but of these four, *G. pallida* was the most dissimilar. In ITS1, *G. rostochiensis* and the two Mexican *Globodera* species shared nearly half of 39 nucleotide base pair differences from *G. pallida*. The data are consistent with the view that Mexico is the center of origin for the potato cyst nematodes, but not for *G. virginiae*.

BACTERIAL-FEEDING NEMATODES IN ORGANIC AND CONVENTIONAL FARMING SYSTEMS. **Ferris, H.,[1] R. C. Venette,[1] S. A. Lau,[1] K. M. Scow,[2] and N. Gunapala[2].** Departments of [1]Nematology, and [2]Land, Air and Water Resources, University of California, Davis, CA 95616.

We investigated temporal relationships among bacterial-feeding nematodes, (BFN), fertility, and microbial biomass in soils managed under conventional and organic farming systems. There were more BFN in the conventional than in the organic plots in the early spring, but numbers were greater in the organic plots following incorporation of a winter cover crop. Numbers of BFN in the organic plots were positively correlated with measures of microbial abundance and activity, including microbial biomass carbon, microbial biomass nitrogen, and substrate-induced respiration. These correlations were not significant in the conventional plots. The BFN community was less diverse in organic than in conventional plots due to temporal predominance of individual species, which was related to temperature-niche characteristics of the species. Temporal predominance influences the contribution of BFN species to nitrogen mineralization during key periods of plant growth.

TEMPERATURE AND DIETARY LIPIDS AFFECT PHOSPHOLIPID FATTY ACIDS AND MEMBRANE FLUIDITY IN *STEINERNEMA CARPOCAPSAE*. **Fodor, A.,[1] I. Dey,[2] T. Farkas,[2] and D. J. Chitwood[3].** [1]Institute of Genetics, Eötvös Lorand University, H-1088 Budapest, Hungary, [2]Institute of Biochemistry, Biological Research Center, Hungarian Academy of Sciences, Szeged, Hungary, and [3]USDA ARS, Nematology Laboratory, BARC-West, Beltsville, MD 20705.

Steinernema carpocapsae was propagated in *Galleria mellonella* or in artificial diets containing lard, linseed oil, or fish oil. Because of the low level of linolenic (18:3 [*n*–3]) acid and the high level of eicosapentaenoic (20:5 [*n*–3]) acid in nematode phospholipids even without dietary linolenic acid, *S. carpocapsae* likely is capable of de novo biosynthesis of *n*–3 polyunsaturated

fatty acids. Fluorescence polarization indicated that vesicles formed from phospholipids of nematodes raised at 18 C were less ordered than those from nematodes raised at 27.5 C, especially in the outermost region of the bilayer. Dietary fish oil increased fluidity in the outermost region but increased rigidity in deeper regions. Therefore, *S. carpocapsae* appears to modify its membranes in a response to temperature that is possibly mediated by eicosapentaenoic acid.

INFLUENCE OF WINTER COVER CROPS ON POPULATIONS OF PLANT-PARASITIC NEMATODES AFFECTING SMALL-FRUIT CROPS IN WESTERN OREGON. **Forge, T. A., and R. E. Ingham.** Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331.

Field and greenhouse experiments were used to study the effects of several winter cover crops on populations of *Pratylenchus penetrans* and other nematodes. Sudangrass 'SS 222' and 'Trudan 8', oat 'Saia', rye 'Wheeler', rapeseed 'Humus' was planted 31 August 1993 in replicate 3- × 6-m field plots infested with *P. penetrans*. Naturally occurring weeds were allowed to grow in control plots. By 30 November, populations densities in soil had declined from a preplant population density of 350 nematodes/100 g soil to 85, 83, 53, 80, 28, and 15 nematodes/100 g soil in plots covered with weeds, sudangrass SS 222 and Trudan 8, rapeseed, oat, and rye, respectively. Population densities in roots of the same crops were 209, 177, 192, 121, 76, and 41 nematodes/g fresh root, respectively. Sudangrass Trudan 8, oat, barley 'Steptoe', rapeseed, marigold 'Tangia' and strawberry 'Totem' were grown in *P. penetrans*-infested soil in 15- × 15-cm deep pots under greenhouse conditions. After 4 months, nematode population densities in pots (roots and soil combined) planted with sudangrass, oat, barley, and marigold were significantly smaller than population densities in pots with strawberry.

DEVELOPMENT OF POPULATIONS OF *BURSAPHELENCHUS XYLOPHILUS* IN SELECTED WEST-COAST CONIFERS. **Forge, T. A., and J. R. Sutherland.** Pacific Forestry Centre, Natural Resources Canada, Victoria, BC V8Z 1M5, Canada.

To better understand the potential distribution of *Bursaphelenchus xylophilus* in forests of western North America, we compared the growth of two isolates of *B. xylophilus* in wood and bark of *Pinus contorta* (lodgepole pine), *Abies grandis* (grand fir), *Pseudotsuga menziesii* (Douglas-fir), *Tsuga heterophylla* (western hemlock), and *Thuja plicata* (western red-cedar). In three separate experiments, the nematodes were inoculated into freshly cut segments (8 cm d × 25 cm long) of branches or stems of each tree species and sampled at regular intervals for up to 16 weeks. Population densities for the entire segments (wood and bark), averaged over 8 and 16 week sample dates, were 125, 80, 27, 31, and 3 nematodes/g dry tissue for *P. contorta, P. menziesii, A. grandis, T. heterophylla*, and *T. plicata*, respectively. The nematodes were concentrated in bark of the nonpine species. The percentage of nematodes inhabiting bark was 22, 79, 80, 83, and 94 for *P. contorta, P. menziesii, A. grandis, T. heterophylla* and *T. plicata*, respectively. Dispersal third-stage juveniles formed in all tree species. In petri-dish assays the nematodes multiplied more rapidly in the presence of ethanol extracts from *P. contorta* than in the presence of extracts from *P. menziesii*.

VARIATION IN RIBOSOMAL GENES IN *MELOIDOGYNE ARENARIA*. **Georgi, Laura L., and Albert G. Abbott.** Department of Biological Sciences, Clemson University, Clemson, SC 29634.

We are interested in genome organization and the generation of variation in a mitotically parthenogenetic root-knot nematode species, *Meloidogyne arenaria*. The ribosomal genes are

unusually polymorphic in this species. We have isolated four independent ribosomal DNA clones from a *M. arenaria* genome library in EMBL-3; two of these clones have deletions in the 28S-5S region. We have sequenced rearranged regions of these clones and confirmed that deletions in the 28S-5S region involve a 129 bp repeat, as suggested by Vahidi and Honda (*Mol. Gen. Genet.* 227:334). Adjacent ribosomal gene copies share the identical deletion, however, the deletion endpoints differ between the two clones analyzed. Furthermore, one of the clones has many additional rearrangements. Most of these rearrangements are not shared among adjacent gene copies, and most do not involve the 129 bp repeat.

HISTOLOGICAL STUDIES OF PARASITISM BY *SCHISTONCHUS* SP. (APHELENCHOIDIDAE) IN THE SYCONIA OF *FICUS CITRIFOLIA*. **Giblin-Davis, R. M., and B. J. Center**. Fort Lauderdale Research and Education Center, University of Florida, 3205 College Avenue, Fort Lauderdale, FL 33314.

Schistonchus sp.-infested and uninfested syconia in different phenological phases from *Ficus citrifolia* from Florida City, FL, were studied histologically. Florets (n = 262) from four *Schistonchus* sp.-infested interfloral phase syconia were examined in serial sections. Only immature male florets (n = 50) were parasitized with propagating *Schistonchus* sp. (n = 23 parasitized male florets) and had 124 $\pm$ 77 (mean $\pm$ SD) nematodes per infested floret. None of the female florets examined (114 wasp galls, 75 fig embryos, and 23 aborted fig embryos) were infested with nematodes. *Schistonchus* sp. induced the formation of hypertrophied, uninucleate epidermal cells in the unpresented anther filament and anther. Hypertrophied cells were enlarged and had highly granulated cytoplasm and enlarged nuclei and nucleoli, compared with healthy cells from uninfested florets. Nematodes were also associated with a dark safranin-staining exudate and damaged epidermal cells of the inner lining of the perianth lobes in nematode-infested male florets.

RED RING OF PALMS. **Giblin-Davis, R. M., C. M. Chinchilla, J. E. Pena, and E. A. Pena Rojas**. Fort Lauderdale Research and Education Center, University of Florida, 3205 College Avenue, Fort Lauderdale, FL 33314.

Classical red ring of palm causes tree losses of up to 10-15% per year in coconut and African oil palm plantings in the Neotropics. The disease is caused by the red ring nematode, *Bursaphelenchus cocophilus* (Aphelenchoididae), an obligate intercellular parasite of palms. Red ring nematode-induced little leaf disease can lead to chronic abortion of young inflorescence and the production of abnormal leaves. Classical and little leaf symptoms can vary with palm host age, species, genotype, and environmental conditions. The main vector of *B. cocophilus* is the American palm weevil, *Rhynchophorus palmarum*. However, other weevils, such as *Dynamis borassi, Metamasius* spp., *Rhinostomus barbirstris*, and *Homalinotus* spp. have similar biologies to *R. palmarum* and may be important vectors where they are co-distributed with the red ring nematode. The association between the red ring nematode and *R. palmarum* is hypothesized to have evolved relatively recently. Red ring of palm management should involve good phytosanitation, continuous mass trapping of the vector(s), and intensive mass trapping before development of new or old agricultural areas.

SURVEY OF *RADOPHOLUS* POPULATIONS FROM *ANTHURIUM* spp. IN HAWAII. **Goo, Matthew,[1] B. Sipes,[1] and K. Delate[2]**. [1]Department of Plant Pathology, University of Hawai'i, at Mānoa, Honolulu, HI 96822, and [2]Beaumont Experiment Station, Hilo, HI 96720.

Radopholus sp., is a chronic problem in *Anthurium* spp. production. A survey of 10 commercial Anthurium farms on the island of Hawaii was conducted to determine the incidence

of *Radopholus* sp. infestations. Roots were collected from 43 cultivars and nematodes extracted from two 100-g subsamples per root. Of the 86 Anthurium root samples processed, 81% were infected with *Radopholus*. All farms were infested with *Radopholus* despite various cultural practices. All cultivars that occurred on two or more farms were infected (14 of 43 cultivars). Infection levels from Anthurium cultivars were assessed for 4 of 10 farms. *Radopholus* population density means per Anthurium cultivar ranged from 3.4 to 110.8 nematodes/g dry root weight. Of the Anthurium cultivars that occurred on more than 2 of 4 farms, *Anthurium* sp. cv. Ozaki Red had the highest level of infection (72.2 nematodes/g dry root), and cv. Kozohara Dark Red had the lowest level of infection (13.2 nematodes/g dry root). *Radopholus* sp. populations were widely distributed throughout the Anthurium production area and all Anthurium cultivars were hosts.

EFFECTS OF TWO CARBAMATES ON TWO STRAINS OF *STEINERNEMA CARPOCAPSAE*. **Gordon, Roger, and Joy Tilley.** Department of Biology, Memorial University of Newfoundland, St. John's, Newfoundland, Canada A1B 3X9.

Infective juveniles (IJ) of the All and Umea strains of *Steinernema carpocapsae* were exposed to carbofuran, an acetylcholinesterase inhibitor, and fenoxycarb, a juvenile hormone analog, at doses from 1.0×10^{-5} to 1.0 mg/ml for 24 to 168 hours. Values for the LD_{50} and infective indices were determined. Carbofuran was the more toxic compound to both strains. The Umea strain was more sensitive to both compounds than was the All strain. The effects of the compounds were manifested within the initial 24 hour exposure period and did not increase after that. Preliminary experiments showed that at doses insufficient to cause significant mortality of IJ, neither compound appeared to significantly effect infectivity. Integrated pest management involving the use of such nematodes along with either carbamate would require cautious implementation.

DEVELOPMENT OF MONOCLONAL ANTIBODIES TO ESOPHAGEAL GLAND SECRETIONS OF *HETERODERA GLYCINES*. **Goverse, A., E. L. Davis, and R. S. Hussey.** Plant Pathology Department, University of Georgia, Athens, GA 30602-7274.

Second-stage juveniles (J2) of *H. glycines* that were incubated in 5-methoxy DMT oxalate produced stylet secretions that were solubilized in alkaline pH buffer, concentrated, and used for intrasplenic immunization of a Balb/c mouse. Two monoclonal antibodies (MAbs) that bound to secretory granules within the subventral esophageal glands (SvG) and one MAb that bound to secretory granules in the dorsal esophageal gland (DG) of *H. glycines* J2 were developed from this immunization. Homogenates of *H. glycines* J2 were used for intra-splenic immunization of a second mouse and produced three MAbs that bound to secretory granules in the SvG of *H. glycines* J2. Three of the total five SvG MAbs bound to stylet secretions from *H. glycines* J2 in immunofluorescence and ELISA assays. Two of the five SvG MAbs bound to both the DG and SvG in young *H. glycines* females. All five SvG MAbs bound to the SvG in *Heterodera schachtii* J2, and one of these MAbs bound to the SvG in *Globodera tabacum* J2.

IDENTIFICATION OF AGGREGATION PHEROMONES AND PALM KAIROMONES FOR SEMIOCHEMICAL-BASED CONTROL OF PALM AND SUGAR CANE WEEVILS. **Gries, G., R. Gries, A. L. Perez, and A. C. Oehlschlager.** Department of Biological Sciences, Department of Chemistry, Simon Fraser University, Burnaby, B.C., V5A 1S6 Canada.

The American palm weevil, *Rhynchophorus palmarum*, vectors the red ring nematode, *Bursaphelenchus cocophilus*, which causes the lethal disease red ring (RR) of coconut and African oil palms. Semiochemical-based mass trapping of *R. palmarum* offered a strategy of RR control,

and prompted us to identify pheromones of *R. palmarum* and other palm infesting weevils. Employing portable "kits" for volatile collection and state-of-the-art technology for volatile analyses, we have discovered attractive palm volatiles and aggregation pheromones in several *Rhynchophorus* and *Metamasius* weevils which may be used for weevil control.

FACTORS AFFECTING THE PATHOGENICITY OF PLANT-PARASITIC NEMATODES TO RANGE GRASSES. **Griffin, G. D.** USDA ARS, Forage and Range Research Laboratory, Utah State University, Logan, UT 84322-6300.

Soil texture affected the virulence of plant-parasitic nematodes to range grasses. Reduction in plant growth due to nematode parasitism was greatest in Kidman sandy loam soil, intermediate in Ricks sandy loam soil, and minimal in Thiokol fine silty loam soil. *Pratylenchus neglectus* was affected less by soil texture than were *Merlinius brevidens*, *Quinisulcius acutus*, and *Xiphinema americanum*. *Xiphinema americanum* was the least virulent of the four nematode species, and was most affected by soil texture. Virulence of *P. neglectus* was greatest at 30 C, whereas the virulence of *M. brevidens, Q. acutus*, and *X. americanum* was greatest at 25 C. Reproduction was highest for *P. neglectus* at 30 C, whereas reproduction for *M. brevidens* and *Q. acutus* was greatest at 25 C. *Xiphinema americanum* failed to reproduce on grass at any temperature. Differences in tolerance among grass genotypes were observed.

THE USE OF GREEN MANURE CROPS IN A SUGARBEET ROTATION FOR SUGARBEET CYST NEMATODE MANAGEMENT. **Hafez, Saad L.**, Department of Plant, Soil and Entomological Sciences, University of Idaho, 29603 U of I Lane, Parma, ID 83660.

Four cultivars of oil radish, *Raphanus sativus* var. *oleifera* (Adagio, Pegletta, Ultimo, Remonta), and three cultivars of white mustard, *Sinapis alba* (Metex, Maxi, Martigena), were planted following wheat in a sugarbeet cyst nematode infested field in the fall of 1992 in Parma, Idaho. Each cultivar was replicated four times in a randomized complete block design, and a fallow treatment was included as a control for comparison. All cultivars were mechanically chopped and incorporated 3 months after planting. Soil samples before planting in the fall and in the following spring were collected for nematode assay. All cultivars reduced the total number of eggs and juveniles significantly. Oil radish 'Adagio' caused the highest percentage reduction in comparison to fallow (51%). White mustard 'Martigena' caused the lowest percentage reduction (21%). The following spring, sugarbeet 'HM-WS-90' was planted following the oil radish and mustard cultivars. Most cultivars increased sugarbeet yield significantly in comparison with the fallow treatment.

A COMPREHENSIVE SURVEY OF NEMATODE POPULATIONS AND HORTICULTURAL PERFORMANCE OF 'ROME BEAUTY' APPLE ORCHARDS IN IDAHO. **Hafez, Saad L., and Esmaeil Fallahi**. Department of Plant, Soil and Entomological Sciences, University of Idaho, 29603 U of I Lane, Parma, ID 83660.

A comprehensive survey was conducted in 'Rome Beauty' apple orchards in Idaho during 1992 and 1993 to study various horticultural practices, nematode populations, soil and leaf mineral nutrition, and interrelationships among these practices. Lesion nematode (*Pratylenchus vulnus*) was observed in all orchards. Pin, stunt, dagger, sheath, ring, and stem and bulb nematodes were observed in 69, 47, 44, 38, 38, and 31% of the orchards, respectively. Stem and bulb nematode population densities were negatively correlated with leaf and soil Ca, and leaf Cu, but lesion and dagger nematodes were positively correlated with soil lime, K, Ca, Mg, S and P content. In some orchards with high nematode population densities, application of nitrogen fertilizer as high as 560

kg/ha did not raise leaf nitrogen to the sufficiency range. Trees that were fumigated before planting were more productive and had higher concentrations of various minerals in leaves. This survey revealed that a comprehensive analysis of several orchard management practices provided more information for interpreting fruit production problems than analyzing only a single management practice.

VICISSITUDE IN THE GENOME OF *RADOPHOLUS*. **Hahn, M. L.,[1] D. T. Kaplan,[2] P. R. Burrows,[1] and J. Bridge[1].** [1]AFRC-CABI Tropical Nematode Research Group, Rothamsted Experimental Station, Harpenden, Hertfordshire, AL5 2JQ, England, and [2]U. S. Horticultural Research Laboratory, USDA ARS, 2120 Camden Road, Orlando, FL 32803.

Genetic diversity of eight burrowing nematode populations (*Radopholus* spp.) collected from a variety of plants in Indonesia, Sri Lanka, Uganda, and the United States was investigated using the Operon Primers (OP-A1, A4, and A11, OP-B8, OP-E9, OP-K2, OP-M2, OP-O10, and OP-T12). Standard RAPD analyses supported previous observations that the genomes of morphologically identical sibling species *R. similis* and *R. citrophilus* appeared to be highly conserved. Random Amplified Polymorphic DNA patterns implied genetic divergence in two burrowing nematode populations previously determined to be morphologically distinct from *R. similis* and *R. citrophilus* (J. Machon, J. Bridge, and M. R. Siddiqi, pers. comm.). These populations were collected from *Citrus* sp. in East Java, Indonesia, and from *Curcuma zeodaria* in Bogor, Indonesia.

GENETIC VARIATION AND PHYLOGENETIC RELATIONSHIPS AMONG SPECIES AND STRAINS OF ENTOMOPATHOGENIC NEMATODES. **Hashmi, G., I. Glazer, and R. Gaugler.** Department of Entomology, Rutgers University, P.O. Box 231, New Brunswick, NJ 08903.

The two entomopathogenic nematode genera, *Heterorhabditis* and *Steinernema* consist of many species and strains. They have a great deal of potential for biological control of insect pests, but the ability to identify these different isolates rapidly and accurately is important for their successful implementation. Many difficulties of phenotype-based identification can be overcome through direct identification of their genotype using a DNA based diagnostic assay. We used Polymerase Chain Reaction (PCR) with Random Amplified Polymorphic DNA (RAPD) markers to: 1) differentiate species of *Heterorhabditis* and *Steinernema,* and 2) differentiate isolates of *Heterorhabditis*. Eighty 10-mer primers were used to screen these genotypes. All species tested could be distinguished using this method. Genetic analysis using seven primers of arbitrary sequences was also conducted on *Heterorhabditis* populations isolated from various geographical regions around the world. A similarity matrix was constructed based on Jaccard's coefficient. All isolates, except for HP88, and HB, showed polymorphisms with all the primers used. This research demonstrates the feasibility of using RAPD markers for identification and population and phylogenetic studies of entomopathogenic nematodes.

SUCCESSFUL TRANSFORMATION OF AN ENTOMOPTHOGENIC NEMATODE BY MICROINJECTION. **Hashmi, S., G. Hashmi, and R. Gaugler.** Department of Entomology, Rutgers University, P.O. Box 231, New Brunswick, NJ 08903.

Entomopathogenic nematodes of the genus *Heterorhabditis* are important biological control agents for soil inhabiting insects, however, their sensitivity to environmental stress including temperature often reduces their field efficacy. One solution to these problems is to develop genetically altered strains of nematode with increased tolerance to environmental stress. Although

selection and mutagenesis have been tried, the design of specific traits will require the use of DNA transformation to introduce cloned genes of known function. We are reporting the first successful transformation of an entomopathogenic nematode, *Heterorhabditis bacteriophora* HP88. Foreign genes were introduced by microinjection using vectors pRF4 and pPCZ1 carrying the *Caenorhabditis elegans* genes coding for roller phenotype and 16 kDa heat shock protein (hsp16-1) gene. Translational fusion made by inserting lac-Z in frame into hsp16-1 produced expression in the body musculature, hypodermis, and muscles in the pharynx. Transcription of the hsp16-lac-Z transgenes resulted in the rapid synthesis of detectable levels of β-galactosidase, but the roller phenotype was not observed. This research opens new avenues for genetic improvement in entomopathogenic nematodes.

YIELD LOSS OF POTATO IN FIELDS WITH AND WITHOUT A HISTORY OF POTATO EARLY DYING AS INDICATED BY NEMATICIDES. **Henn, R. Alan,[1] A. F. Reeves,[2] and W. M. Clapham[1].** [1]New England Plant Soil and Water Laboratory, USDA ARS, University of Maine, Orono, ME 04469-5753, and [2]Department of Plants and Soils, Aroostook State Farm, University of Maine, Orono, Presque Isle, ME 04769.

Nine small plot nematicide trials were conducted from 1988-91 in potato fields with or without a history of potato early dying (PED). In general, potato growing in fields with a history of early dying in Maine is infected with combinations of *Verticillium albo-atrum,* and *Verticillium dahliae* and initial populations of 40 or more *Pratylenchus penetrans*/100 cm^3 soil. Application of various nematicides in a field with no history of PED and an initial soil population of *P. penetrans* of 48/100 cm^3 of soil suppressed nematode populations, but did not change potato yield. In fields with a history of PED, applications of ethoprop plus aldicarb suppressed nematode populations densities and resulted in an increase in yield of $\pm$ 6.5 mt/ha.

OCCURRENCE OF *PASTEURIA* SPP. IN FLORIDA. **Hewlett, T. E., R. J. Cox, D. W. Dickson, and R. A. Dunn.** Entomology and Nematology Department, University of Florida, Gainesville, Fl 32611-0620.

Data collected from the Florida Cooperative Extension Service Nematode Assay Laboratory and the Florida Department of Agriculture and Consumer Services, Bureau of Entomology, Nematology and Plant Pathology Section were compiled to determine the presence and distribution of *Pasteuria* spp. on nematodes in Florida. Information recorded from 243 samples included nematode genera that had *Pasteuria* spp. endospores attached, host plants associated with the sample, and sample origin. *Pasteuria* spp. were detected on 14 different plant-parasitic nematode genera associated with over 40 different plant species and in seven fallow fields in 41 Florida counties. Nematodes associated with *Pasteuria* spp. were from a wide range of host plants, although frequency of this association reflected the sample bias of the laboratories involved. *Meloidogyne* and *Hoplolaimus* spp. were the two nematode genera most frequently associated with *Pasteuria*. *Pasteuria* spp. were observed attached to members of these two genera in 176 and 59 soil samples, respectively.

EVALUATION OF MULTI-PURPOSE SOIL FUMIGANTS FOR ROOT-KNOT NEMATODE MANAGEMENT ON TOMATO IN FLORIDA. **Hewlett, T. E.,[1] D. W. Dickson,[1] and S. L. Locascio[2].** [1]Entomology and Nematology Department, and [2]Department of Horticultural Sciences, University of Florida, FL 32611-0620.

The efficacy of three methyl bromide (MBr) formulations were compared with four other fumigants for control of *Meloidogyne arenaria* race 1 and *Sclerotium rolfsii* on tomato. Treatments

included chloropicrin (392 kg/ha), 1,3-D-chloropicrin (94 and 187 liters/ha), dazomet (168 and 336 kg/ha), metham sodium (935 and 701 liters/ha), MBr 98 (448 and 280 kg/ha), MBr 67 (392 kg/ha), and MBr 33 (504 kg/ha). Plots treated with MBr 98 (448 kg/ha) gave greater ($P \leq 0.05$) marketable yields and lower galling indicies than the untreated controls. Disease incidence and root-knot nematode galling indicies were negatively correlated ($P \leq 0.001$) with marketable yield ($r = 0.73$ and 0.41, n = 68, respectively). Galling indices were positively correlated ($P \leq 0.001$) with disease incidence ($r = 0.40$, n = 68).

MELOIDOGYNE INCOGNITA INVASION AND DEVELOPMENT IN RESISTANT AND SUSCEPTIBLE ALFALFA CULTIVARS. **Higgins, E. A.,[1] S. H. Thomas,[1] C. L. Potenza,[2] and C. Sengupta-Gopalan[2].** [1]Entomology, Plant Pathology and Weed Science Department, and [2]Agronomy and Horticulture Department, New Mexico State University, Las Cruces, NM 88003-0003.

Seedlings of alfalfa cultivars classified as resistant (Moapa 69) or susceptible (Lahontan) to *M. incognita* were grown in 100 cm^3 sterile sand for 10 days, before infestation with 21,000 freshly hatched second-stage juveniles J2. Inoculated containers were incubated at 28 C hours in a growth chamber until harvest 12, 24, 48, or 72 hours and 7, 14, or 21 days later. Roots were stained with acid fuchsin and observed microscopically. Both cultivars demonstrated similar numbers of infected roots (80%) and maximum levels of invasion 48 hours after inoculation. By 72, hours J2 were evident within the vascular cylinder of roots of Lahontan, but remained concentrated in the root tips of Moapa. Nematode developmental and numerical differences between cultivars continued to diverge over time, with little evidence of successful parasitization of Moapa by 14 days.

EVALUATION OF TOLERANCE TO *MELOIDOGYNE INCOGNITA* AND *HOPLOLAIMUS COLUMBUS* IN COTTON GENOTYPES. **Hill, A. S.,[1] O. L. May,[2] and J. D. Mueller[1].** [1]Edisto Research and Education Center, Blackville, SC 29817-0247, and [2]USDA ARS, Florence, SC 29502-3039.

Twelve PD germplasm lines and four cultivars were evaluated in separate field plots for tolerance to *H. columbus* and *M. incognita* in 1992 and 1993. Differences were not detected among cultivars in root-knot nematode galling indices or recovery of *H. columbus*. Tolerance indices were calculated as ([yield of untreated plots]/[yield of plots treated with 26 liter/ha 1,3-dichloropropene] x 100). Tolerance indices for *H. columbus* ranged from 59 to 121% in 1992 and from 50 to 98% in 1993. Indices for *M. incognita* ranged from 58 to 115% and 76 to 114% in 1992 and 1993, respectively. Germplasm line PD 5300 had the most consistent levels of tolerance between years and nematodes with indices of 100 and 102% for *M. incognita* and 94 and 95% for *H. columbus*. Deltapine 90 exhibited the lowest levels of tolerance to *H. columbus* both years (59% and 50%) and had tolerance indices of 75 and 99% for *M. incognita*.

GENE ACTIVITY INDUCED BY INFECTION OF SOYBEANS WITH *HETERODERA GLYCINES*. **Huang, Yinrong,[1] S. Anand,[2] P. Kozlowski,[1] and R. I. Bolla[1].** [1]Department of Biology, Saint Louis University, St. Louis, MO 63103-2010, and [2]University of Missouri-Columbia, Delta Center, Portageville, MO 63873.

Difference in gene expression in soybean cultivars Essex (susceptible) and Hartwig (resistant) was determined by differential display analysis after infection by *Heterodera glycines* race 5. Mitochondrial RNA was reverse transcribed to cDNA with polydeoxythymidine anchored primers. The cDNA, ranging in size from 50 to 500 base pairs, was amplified by the polymerase chain

reaction. Differences between the mtRNA from infected and control plants were identified after separation of the cDNA fragments on a DNA sequencing gel. Unique DNA fragments, representing infection regulated gene expression, were cloned and sequenced. They then were identified by comparison to known gene sequences. Small ribosomal subunit protein genes and DNA repair genes were identified as induced following nematode infection of Essex. Other unidentified genes were repressed in Essex. Several genes are also induced by infection of Hartwig by *H. glycines*.

PEST TRACKING SYSTEMS IN USDA-ANIMAL AND PLANT HEALTH INSPECTION SERVICE. **Huettel, R. N., and C. D. McNeal.** USDA APHIS PPQ, 6505 Belcrest Road, Hyattsville, MD 20782.

There are two systems used by the Animal and Plant Health Inspection Service, Plant Protection and Quarantine for surveying and tracking crop pests, diseases, and biological control agents. The Cooperative Agricultural Pest Survey is a combined federal and state agricultural organization effort for survey activities that identifies new introductions and establishments of foreign pests and diseases. Cooperative Agricultural Pest Survey also monitors first season occurrences, population levels, and developmental stages of foreign and endemic pests and diseases. Plant Protection and Quarantine, in coordination with State Survey Committees, provides funding, technical assistance, communications, and data management to State Departments of Agriculture and state universities to collect data. The data are then processed through the National Agricultural Pest Information Service. This agency is available to users for historical information, geographical distributions, host crops, and other pertinent data on pests or biological control agents.

IDENTIFICATION AND CHARACTERIZATION OF GENES DIFFERENTIALLY EXPRESSED IN DEVELOPMENT OF PLANT-PARASITIC NEMATODES. **Huston, S., E. Stillwell, L. Georgi, and A. Abbott**. Department of Biological Sciences, Clemson University, Clemson, SC 29634.

Meloidogyne spp. interact with host plant cells to induce the formation of giant multinucleate plant cells. These giant cells, formed by an undiscovered mechanism, serve to feed the nematode. It has been proposed that the secretory glands of the nematode produce material that is essential for root penetration and induction of giant cells, and the maintenance of these feeding sites. To search for nematode genes specifying the secretory gland components, we used a differential cDNA library screening assay to detect cloned cDNAs that represent genes abundantly expressed during the active synthesis of secretory gland components. From this assay, several cloned cDNAs detect abundantly accumulated transcripts in Northern analyses of RNA samples taken from nematode stages exhibiting high secretory gland activity. We present initial characterization of the structure and expression of these cDNAs.

MOLECULAR POPULATION BIOLOGY OF PHYTONEMATODES. **Hyman, Bradley C., L. W. Whipple, and N. Le.** Department of Biology, University of California, Riverside, California 92521.

Genetic structure of intra-specific phytonematode populations is among the least understood aspects of nematode population biology. Yet, successful employment of novel biological control strategies will likely involve an understanding of genotypic interactions between the nematode and its plant host, including genetic differentiation within and among targeted nematode populations. Contemporary molecular technology, including molecular cloning and polymerase chain reaction (PCR) amplification now allows an assessment of genetic variability within individuals comprising

phenotypically separable species, races and biotypes. Molecular markers for the nuclear and mitochondrial genomes of several phytonematode genera, including *Heterodera* and *Meloidogyne* species have now been developed. These loci can vary in molecular size (length polymorphism) or primary nucleotide structure (sequence polymorphism) among individual nematodes. The extent of variation can provide diagnostic information to discriminate between isolates and quantitative measures of population differentiation and phylogenetic affinities.

SURVEY STUDY OF PLANT-PARASITIC NEMATODES IN EGYPT. **Ibrahim, I. K. A., M. A. El-Saedy, and A. A. El-Sherbiny**. Department of Plant Pathology, Faculty of Agricultural, Alexandria University, Alexandria, El-Shatby, Egypt.

A nematode survey was conducted in northern Egypt to identify plant-parasitic nematodes associated with grasses, weeds, and other plants. About 25 nematode genera were found in the collected soil samples. The nematodes *Boleodorus pakistanensis, Criconemella sphaerocephala, Discocriconemella sphaerocephaloides, Eutylenchus* sp., *Hoplolaimus clarissimus, Irantylenchus clavidorus, Merlinius nanus, Tylenchorhynchus annulatus, Tylenchus afghanicus*, and *Tylenchus exigus* were identified and recorded for the first time in Egypt. Also, the results showed new host plant records for these nematode species in Egypt.

CONTROL OF *MELOIDOGYNE CHITWOODI* WITH CROP ROTATION, GREEN MANURE CROPS, AND NONFUMIGANT NEMATICIDES. **Ingham, R. E.,[1] H. Mojtahedi,[2] G. Reed,[3] and G. Santo[2].** [1]Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331-2902, [2]Department of Plant Pathology, Washington State University, IAREC, Prosser, WA 99350-9687, and [3]Department of Entomology, Oregon State University, HAREC, Hermiston, OR 97838.

Greenhouse, microplot, and field plot experiments determined that some cultivars of popcorn, supersweet corn, and lima bean were poor to nonhosts of Columbia root-knot nematode, *Meloidogyne chitwoodi*. A 4-year cropping sequence experiment was conducted using these crops with and without sudangrass or rapeseed green manure crops between cash crops and with and without addition of nonfumigant nematicides. A potato-wheat-wheat-potato sequence was used as a control and had 91% nematode culled tubers. Use of sudangrass following the last wheat crop and (or) ethoprop before planting potato reduced culls by 50%. The potato-lima bean-supersweet corn-potato sequence reduced culls to 19% and use of a rapeseed green after limabean and after supersweet corn reduced culls to <1%. The potato-popcorn-lima bean-potato sequence reduced culls to 2%.

STEINERNEMA CARPOCAPSAE: POSTEMERGENCE INFECTIVITY AND SEX RATIO OF INVADING NEMATODES. **Ishibashi, N., and X. D. Wang.** Department of Applied Biological Sciences, Saga University, Saga 840, Japan.

Steinernema carpocapsae infective juveniles (IJ) were tested for their nictation, infectivity, and sex ratio of early invading IJ. The IJ newly emerged from insect cadavers of *Galleria mellonella* were stored for 0, 2, 4, 6, and 8 weeks at 8 C. The rates of nictation on bark compost and infectivity on the insect at 25 C increased and with increasing storage time; 13.2, 17.7, 33.1, 38.8, and 58.2% for nictation and 11.4, 21.0, 23.1, 29.1, and 51.5% for infectivity for the respective storage period. The sex ratios of penetrating IJ were 54.6 and 37.5% for nictating and non-nictating IJ, respectively. When the insect was placed on the top of a layer of bark compost and IJ were inoculated below the substrate, the sex ratio was 70% or more compared with 40% or less using a petri dish bioassay with filter paper. We conclude that this nematode assumes a

strategy for survival of staggering the infection period, and that males are more active than females.

EFFECTS OF THE NEMATODE-TRAPPING FUNGI *MONACROSPORIUM CIONOPAGUM* AND *M. ELLIPSOSPORUM* ON *MELOIDOGYNE JAVANICA* AND *HETERODERA SCHACHTII*. **Jaffee, B. A., and A. E. Muldoon**. Department of Nematology, University of California, Davis, CA 95616-8668.

Hyphae of *Monacrosporium cionopagum* and *M. ellipsosporum* were rinsed free of media, pelletized in alginate (7 mg wet hyphae/pellet), and added to loamy sand in vials (17 cm^3 of soil and four pellets per vial). Controls received pellets without hyphae. After 2 weeks at 20 C, 100 juveniles of *Heterodera schachtii* or *Meloidogyne javanica* were added per vial. Cabbage seedlings were planted after 66 hours, roots were stained 5 days later, and nematodes in roots counted. Suppression in number of *M. javanica* per root was 97 $\pm$ 1% with *M. cionopagum* and 63 $\pm$ 5% with *M. ellipsosporum*; suppression of *H. schachtii* was 21 $\pm$ 5% with *M. cionopagum* and 8 $\pm$ 6% with *M. ellipsosporum*. We suggest that *Meloidogyne* spp. may be more susceptible than *Heterodera* spp. to trapping fungi, and that vegetative hyphae without added nutrients are useful for augmenting soil with these control agents.

VARIATION IN THE CONCENTRATIONS AND TYPES OF GLUCOSINOLATES IN RAPESEED AND THE IMPLICATION FOR NEMATODE CONTROL. **Jing, G. N., and J. M. Halbrendt**. Department of Plant Pathology, The Pennsylvania State University, Biglerville, PA 17307.

The utility of rapeseed glucosinolates for nematode control may depend on their concentration and type. To test this hypothesis three experiments were conducted. In the first, total glucosinolates in rapeseed extracts were determined indirectly by measurement of the glucose released after hydrolysis by myrosinase. Glucose was reacted with a chromogen to give a brown solution and the optical density was measured spectrophotometrically at 450 nm. This value was used to calculate the glucose concentration. In another experiment glucosinolates were purified and desulfonated on mini columns of DEAE-Sephadex A-25. Differences in glucosinolate types among rapeseed cultivars were resolved using HPLC. A third experiment evaluated the nematicidal effects of rapeseed residue in nematode infested soil. Results of these experiments indicated a large variations in glucosinolate concentrations among cultivars that ranged from 2.3 - 99 μM/g for seeds. Concentrations in dry foliage and root tissue ranged from undetectable levels to approximately 58 μM/g. Different cultivars exhibited distinct HPLC glucoisinolate profiles and soil amendment studies showed that cultivars had variable nematicidal activity that was related to total glucosinolate.

MANAGEMENT OF *MELOIDOGYNE INCOGNITA* IN TRITICALE, COTTON, AND SOYBEAN WITH RESISTANT CULTIVAR ROTATIONS. **Johnson, A. W.,[1] C. C. Dowler,[1] D. R. Sumner,[2] and S. Baker[3]**. [1]USDA ARS, Nematodes, Weeds, and Crops Research Unit, [2]Department of Plant Pathology, and [3]Department of Crop and Soil Sciences, University of Georgia, Tifton, GA 31793.

Three cropping systems 1) triticale 'Beagle 82'-cotton 'McNair 235', 2) triticale-soybean 'Twiggs', and 3) triticale-cotton-triticale-soybean maintained population densities of *M. incognita* below damaging levels. Numbers of *M. incognita* second-stage juveniles declined on triticale and soybean but increased on cotton. The application of fenamiphos at 6.7 kg a.i./ha did not suppress (P = 0.05) *M. incognita* population densities in the soil. Over 4 years, mean yields of triticale,

cotton, and soybean were 5.6, 4.8, and 1.2% greater, respectively, from fenamiphos-treated plots than untreated plots, but differences were not significant ($P = 0.05$).

AN APHID RESISTANCE GENE IS TIGHTLY LINKED TO THE NEMATODE RESISTANCE GENE *MI*. **Kaloshian, Isgouhi,[1] V. M. Williamson,[1] and W. H. Lange[2].** [1]Department of Nematology, and [2]Department of Entomology, University of California, Davis, CA 95616.

In California, potato aphid, *Macrosiphum euphorbiae*, can build up to large numbers on tomato in late summer and cause severe damage to some varieties. Tomato lines from diverse breeding programs were evaluated in the field for resistance to the potato aphid. There were dramatic differences between lines in the numbers of aphids present on mature field plants. All lines that displayed aphid resistance carried the nematode resistance gene, *Mi*. A greenhouse assay for aphid resistance was developed to test this association further. Nearly isogenic lines that differed in nematode resistance showed association of nematode and aphid resistance. An F_2 population segregating for nematode resistance was tested for aphid resistance and for alleles of REX-1, a DNA marker tightly linked to *Mi*. Genetic linkage of aphid resistance to *Mi* was confirmed. The *Mi* gene was introgressed into tomato from the wild species *Lycopersicon peruvianum*. We hypothesize that the aphid resistance gene, which we propose to call *Meul*, was introduced into tomato along with *Mi*. The presence of aphid resistance in the line Motelle, which contains only about 600 kb of introgressed DNA, suggests that *Meul* is located very close to *Mi* in the genome.

GENETIC COMPARISON OF *HETERODERA GLYCINES* ISOLATES ON TOMATO AND SOYBEAN. **Kennedy, M. J., J. E. Schoelz, and T. L. Niblack.** Department of Plant Pathology, 108 Waters Hall, University of Missouri, Columbia, MO 65211.

Preparatory to a study of gene flow in populations of *Heterodera glycines*, we have maintained three greenhouse isolates that can be distinguished in bioassays by their ability to reproduce on different hosts. The soybean isolate (race 6), originally selected for virulence to soybean, has been maintained for 14 years on 'Essex' soybean, and is unable to reproduce on tomato. The tomato isolates, selected on 'Roma' and 'Tiny Tim' tomato, retain the ability to reproduce on soybean (races 2 and 9, respectively). The objective of this study was to identify genetic differences among the isolates according to random amplified polymorphic DNA (RAPD) polymerase chain reaction analysis with single 10-mer primers. Four of the 45 primers tested yielded reproducible differences that distinguished at least one of the isolates from the others; however, preliminary analysis indicated a lack of correspondence between the RAPD patterns obtained from DNA from a bulk preparation and the patterns obtained from DNA of individual second-stage juveniles.

MEASUREMENT OF THE ANTAGONISTIC POTENTIAL OF FUNGAL EGG PATHOGENS TOWARD CYST NEMATODES. **Kiewnick, S., and R. A. Sikora.** Institut für Pflanzenkrankheitender Universität Bonn, Abt. Phytomed. in Bodenökosystemen, Nussallee 9, 53115 Bonn, Federal Republic of Germany.

A new technique was developed that enables a rapid and exact measurement of the rate of pathogenicity of soilborne fungal egg pathogens of cyst nematodes. Cysts of the potato cyst nematode *Globodera pallida* were incubated in soil samples from the field for 14 days at 20 C. Two different delivery systems were used. The rate of infection was determined by staining the eggs with a 0.5% Rose Bengal solution for 30 minutes. This allows determination of the exact rate of infection and detection of superficial egg shell contamination due to saprophytes. Additional staining with 0.01% fluorescein diacetate in acetone (FDA) verified that there was no

lost of viability in the eggs caused by superficial contamination. This staining technique saves time and material and gives exact measurements of the level of infection of fungal egg pathogens of cyst nematodes.

MASS PRODUCTION OF NEMATOPHAGOUS FUNGUS ARF18 IN LIQUID CULTURE. **Kim, D. G., and R. D. Riggs.** Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701.

The growth of the nematophagous fungus ARF18 was measured in shaker liquid culture with 31 media from natural substrates. Raw substrate (50 g) in water (800 ml) was autoclaved for 1 hour at 121 C, strained through cheesecloth, and water was added to make 1 liter of medium. The fungus grew on all media; it grew the least in media prepared from wheat straw and the most in media prepared from leguminous plants such as *Glycine* spp. and *Pisum* spp. (9 mg vs. 250-360 mg fungal dry weight/100 ml medium). On garden pea (*Pisum sativum*) medium, ARF18 grew well when the initial pH of the medium was adjusted between pH 5.0 to 8.5; the mycelial biomass increased exponentially for 10 days, and a viable alginate-clay formulation was produced.

PERFORMANCE OF SELECTED COTTON CULTIVARS AGAINST ROOT-KNOT NEMATODES: A MULTI-YEAR STUDY. **King, P. S.,[1] R. Rodríguez-Kábana,[1] D. G. Robertson,[1] and J. S. Bannon[2].** [1]Department of Plant Pathology, and [2]E. V. Smith Research Center, Auburn University, Auburn, AL 36849-5409.

Cotton (*Gossypium hirsutum* cvs. DP-20, DP-50, DP-90, Coker 320, Coker 315, KC-380, Stoneville-453, HS-46, DES-119, S-1001, and Terra C-40) were planted for 4 years (1990-93) in a field infested with *Meloidogyne incognita, Hoplolaimus galeatus,* and *Paratrichodorus minor,* and with low numbers of *Tylenchorhynchus claytoni,* and *Pratylenchus* sp. Highest average yields were obtained with DP-50, DES 191, DP-20, and S-1001 and the lowest yields were obtained with Stoneville-453. All cotton cultivars supported significant (> 100 juveniles/100 cm^3 soil) population densities of *M. incognita* as determined at harvest. No cultivar could be considered resistant to the *M. incognita.* There were no differences among cultivars in susceptibility to the other nematode species in the field. All cotton cultivars tested supported significant populations of *H. galeatus* and *P. minor.* Population densities of *T. claytoni* and *Pratylenchus* sp. were consistently low (<25 nematodes/100 cm^3 soil).

USE OF PLANT GROWTH MAPPING TO EVALUATE THE EFFECTS OF *MELOIDOGYNE INCOGNITA* ON COTTON. **Kirkpatrick, T. L.,[1] P. D. Colyer,[2] S. Micinski,[2] and M. Van Iersel[3].** [1]University of Arkansas SW Research and Extension Center, Hope, AR 71801; [2] LSU Red River Research Station, Bossier City, LA 71111, and [3]Crop Physiology Lab, Utah State University, Logan, UT 84322.

A plant growth and development model was used to describe effects of *Meloidogyne incognita* on cotton. Nematode susceptible *Gossypium hirsutum* cv. Stoneville 825 was grown in microplots at the SW Research and Extension Center, Hope, Arkansas and the Red River Research Station, Bossier City, Louisiana in 1992 and 1993. Initial infestation level each year was 0 or 5,000 *M. incognita* race 3) eggs and juveniles/500 cm^3 soil. At the Arkansas location, a treatment of nematodes + aldicarb (1.7 kg a.i./ha) at planting was included. Infected plants exhibited decreased early season growth and development and lower seedling vigor indices. Nematode infection decreased final plant height, number of sympodial branches, number of total bolls produced, and lint yield. Infected plants also remained vegetative longer but ceased fruiting earlier than control plants. Aldicarb application minimized, but did not eliminate, plant damage

or yield losses due to nematode infection.

DEVELOPMENTAL BIOLOGY OF ANIMAL PARASITIC NEMATODES. **Komuniecki, Patricia R.** Department of Biology, University of Toledo, Toledo, OH 43606-3390.

Nematodes have served as model systems for the study of intriguing developmental phenomena, such as cell constancy and chromatin diminution, and many animal parasitic nematodes undergo significant biochemical changes during their life cycle. In particular, the parasitic nematode, *Ascaris suum*, undergoes a metabolic transition from aerobic energy-generating pathways, in the free-living, embryonic and juvenile stages, to anaerobic pathways in the parasitic adult helminth which resides in the swine small intestine. In fact, *A. suum* adults have served as a model system for the elucidation of anaerobic mitochondrial energy generation. The mitochondria in adult body wall muscle have few cristae, lack a functional tricarboxylic acid (TCA) cycle, and produce a mixture of reduced organic acids as end-products of carbohydrate metabolism. In contrast, the early juvenile stages display a functional TCA cycle, are cyanide-sensitive, and completely oxidize glucose to CO_2 and H_2O. Interestingly, and coincident with the molt from third- to fourth-stage juvenile, energy metabolism becomes anaerobic and J4 begin to produce the end-products characteristic of the adult. Current studies attempt to ascertain whether both aerobic and anaerobic mitochondrial populations are present in juvenile ascarids. They should not only shed light on mitochondrial biogenesis accompanying aerobic to anaerobic transition seen in *A. suum*, but also those transitions demonstrated by other nematodes as well.

DISTRIBUTION OF *ROTYLENCHULUS RENIFORMIS* IN A MISSISSIPPI COTTON FIELD. **Lawrence, G. W.,[1] K. S. McLean,[2] and J. J. Cornelius[1].** [1]Department of Entomology and Plant Pathology, Mississippi State University, Mississippi State, MS 39762, and [2]Department of Agriculture, Northeast Louisiana University, Monroe, LA 71209.

The horizontal and vertical distribution of *Rotylenchulus reniformis* was determined in a monocultured cotton field in Mississippi. A 0.44 ha section of the field was divided into 6.1 m × 6.1 m grids. One 2.54-cm-d soil core was collected from each of the 132 grid intersections at planting, and 94, and 164 days later. Each soil core was divided into three equal sections. At 94 days after planting, *R. reniformis* was recovered in all horizontal and vertical samples. Nematode numbers/250 cm^3 soil at 0-15 and 15-30.5 cm ranged from 130 to 11,751, and 110 to 10,927 with an average population density of 3,197 and 2,773 nematodes, respectively. Significantly fewer *R. reniformis* were recovered 30.5-45 cm deep, ranging from 82 to 927 nematodes/250 cm^3 of soil (average of 1,030).

RECOVERY OF *PRATYLENCHUS PENETRANS* FROM PLANT TISSUE CULTURES. **Layne, T., and A. MacGuidwin.** Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Large numbers of *Pratylenchus penetrans* were cultured on monoxenic corn explants in Gamborg's B-5 medium for use in field experiments. Nematodes were efficiently harvested from the agar-based medium in petri dishes by rinsing the agar surface with water repeatedly. Holding the dish at an angle, a fine stream of water was passed over the agar surface and runoff was collected. The agar slab was then turned over and the process was repeated for the other surface. After 3 hours, rinsing was repeated until the agar could no longer be turned intact. Roots were removed and incubated under aeration to harvest additional nematodes. For eight groups of 50-70 dishes, the total number of nematodes removed by seven sequential rinses and from roots was calculated. An average of 82% of the total number of nematodes harvested were recovered by

rinsing. The first collection yielded 19% of the total harvest and the last collection yielded 7%. Storing the petri dishes at 6 C between collections did not affect nematode recovery, suggesting that nematodes may be carried passively to the agar surface.

VIRULENCE OF *MELOIDOGYNE ARENARIA* ISOLATES ON CENTENNIAL SOYBEAN IN FIELD MICROPLOTS. **Lewis, S. A.,[1] and J. D. Mueller[2].** [1]Department of Plant Pathology and Physiology, Clemson University, Clemson, SC 29634-0377, and [2]Edisto Research and Education Center, P. O. Box 247, Blackville, SC 29817.

Damage potential and fecundity of isolates of *Meloidogyne arenaria* race 1 from Georgia, Florida, and South Carolina were compared with an isolate of *M. arenaria* race 2 from South Carolina on 'Centennial' soybean in field microplots. Infestation levels of mixtures of eggs and second-stage juveniles per 100 cm^3 soil for all isolates were 1, 10, 50, and 100 in one experiment, and 400, 800, 1,200, and 1,600 in another. Both experiments were conducted at the same time and location. The race 2 isolate was more virulent than the race 1 isolates. Galling indices exceeded 3.2 for all infestation levels of race 2, but were less than 3.2 for all levels of the race 1 isolates. Race 2 suppressed yields for all infestation levels of 10 or greater. Infestation levels of 400 or greater of race 2 resulted in premature death of plants and no seed yield.

GAS EXCHANGE OF RUSSET BURBANK POTATO INFECTED WITH *PRATYLENCHUS PENETRANS*, *VERTICILLIUM DAHLIAE*, OR BOTH THE NEMATODE AND FUNGUS. **MacGuidwin, A., I. A. Saeed, D. Rouse, and C. Malek.** Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.

The interaction of *Pratylenchus penetrans* (Pp) and *Verticillium dahliae* (Vd) for symptom expression of the potato early dying disease was studied in field plots in 1993. Four treatments were included in the study: Pp with and without Vd, Vd alone, and a noninfested control. Carbon assimilation rate (A) and stomatal conductance were measured using a LI-COR portable photosynthesis system as weather permitted. Canopy ratings and assays of stem sap for Vd were conducted weekly. Reduction in A was first noted in early August for the three pathogen treatments. By mid-August, A was lower for the Vd and Vd + Pp than for the Pp treatment. By late August, A was lower in Vd + Pp-infected plants than in Vd- or Pp-infected plants, which coincided with visual symptom expression and increased counts of Vd from stem sap. Stomatal conductance decreased simultaneously as A for treatments including Vd, but not until late August for the Pp treatment.

BEYOND PESTICIDES-SOME CALIFORNIA FIELD EXAMPLES. **McKenry, M. V.,[1] B. Westerdahl,[2] and O. Becker[1].** [1]University of California Extension Nematologists at Riverside, CA 92521, and [2]Davis, CA 95616.

Two examples of nematode control without chemicals include plant quarantines and use of a fallowing period between crops. The former method is increasingly being perceived as too expensive for the public and the latter is considered too expensive for growers. The use of certain salts, sugars, fermentation products, plant extracts, biological antagonists, rotation crops, and resistant cultivars have potential value for nematode control. Often these control methods involve the use of chemicals released at the infection site or within the soil environment. Although the mechanism of nematode control will continue to be chemical, there is a move toward different packaging, a wider array of nematicidal agents, and the use of nematicidal agents that do not move off target. Nematicides of the future will be "softer," and this can result in higher treatment rates and fewer precisely quantified carrier ingredients. As an example, consider the 16,800 kg/ha

treatment rate of marigold refuse as a rotation crop compared to 448 kg/ha treatment rate of methyl bromide.

THE EFFECT OF *FUSARIUM SOLANI* ON LIFE STAGE DEVELOPMENT OF *HETERODERA GLYCINES*. **McLean, K. S.,[1] and G. W. Lawrence.[2]** [1]Department of Agriculture, Northeast Louisiana University, Monroe, LA, 71209, and [2]Department of Entomology and Plant Pathology, Mississippi State University, Mississippi State, MS 39762.

Tests were established in controlled growth chambers to examine the effects of *Fusarium solani* on life stage development of *Heterodera glycines*. Seedlings of 'Coker 156' soybean were inoculated with *H. glycines* alone or with *F. solani* + *H. glycines* in combination. Plants were harvested at 3 day intervals for 40 days. Roots were stained using NaOCl and acid fuchsin and microscopically examined for *H. glycines* development. Total nematode numbers per gram of root were lower in the *F. solani* + *H. glycines* treatment compared to *H. glycines* alone at each sample date. The rate of development of *H. glycines* fit a linear model both in the presence and absence of *F. solani*. The presence of *F. solani* increased the rate of *H. glycines* development by 3%.

OCCURRENCE OF PLANT-PARASITIC NEMATODES IN CHERRY ROOTSTOCKS. **Melakeberhan, H.,[1] G. W. Bird,[1] and R. Perry[2].** [1]Department of Entomology, and [2]Department of Horticulture, Michigan State University, East Lansing, MI 48824-1115.

The occurrence of plant-parasitic nematodes in established, field grown cherry rootstocks and the reaction of selected rootstocks to prominent nematodes under greenhouse conditions were studied. The established field site consisted of sweet and tart cherry rooted on Mazzard, Mahaleb, M×M 2, M×M 14, M×M 39, M×M 60, M×M 97, and Colt. The greenhouse experiments used year-old Mazzard, Mahaleb, M×M 60, GI148-1 and GI148-8. While there was no statistical difference among rootstocks, *Pratylenchus, Criconemella,* and *Xiphinema* were recovered in numbers ranging from 1 to 46/100 cm^3 in all soils, and 10 to 203 *P. penetrans*/g of fresh root weight from all rootstocks in the established sites. In 157 days of the greenhouse study, *P. penetrans* and *C. xenoplax* infected all five rootstocks. Although there was little effect on plant growth and no significant difference in nematode reproduction among the five rootstocks, the numbers of *P. penetrans* recovered per g of fresh root weight (123-486) were lower than those of *C. xenoplax* (451-2,496). It is not known whether or not any of the rootstocks have any level of resistance or tolerance to any of the nematodes. However, the presence of these nematodes in, and their ability to infect, the rootstocks may be of concern in future nematode management.

TELONE® SOIL FUMIGANTS - REGULATORY STATUS REPORT. **Melichar, M. W., and D. M. Roby.** DowElanco, 9330 Zionsville Road, Indianapolis, IN 46268.

The reregistration standard for 1,3-dichloropropene (1,3-D), the active ingredient in Telone fumigants, was issued September 1986. The last study due date is August 1995. The Environmental Protection Agency (EPA) issued a position document (PD) and placed 1,3-D in Special Review in 1986. This process requires EPA to conduct a benefit-risk analysis resulting in the issuance of a proposed regulatory decision (PD 2/3) sometime in 1994. EPA will conduct separate assessments for residents and workers. Factors affecting residents are proximity to treated fields, temperature inversions, and higher nighttime 1,3-D levels. Factors affecting workers are protective measures and performed tasks. Factors affecting residents and workers are crop rotation practices, frequency of application, application type, and soil type and condition. Because of these risk assessments, EPA can propose to retain all uses on the label, propose to cancel all uses because risk outweighs benefits or, modify the registration to delete uses, add

protective measures, or alter use patterns. A notice of final determination (PD 4) is anticipated after a public comment period on PD 2/3.

VIABILITY OF THE FUNGUS *VERTICILLIUM LECANII* IN ALGINATE PRILLS. **Meyer, S. L. F., and R. J. Meyer.** Nematology Laboratory, USDA ARS, Beltsville, MD 20705-2350.

A wild type strain of *Verticillium lecanii* and a UV-induced mutant strain have been studied as potential management agents for plant-parasitic nematodes. The fungi were applied to soil in alginate prill formulations. Prills were made with wheat bran as a food source for the fungus; one batch containing the mutant strain was prepared with pyrophyllite (hydrous aluminum silicate) as a carrier. To determine shelf life, prills were stored for up to 34 months in the freezer (-15 C), refrigerator (4 C), laboratory (25 C), and greenhouse (15-43 C). In prills made with wheat bran, viability of the mutant strain was 0% after 6 months in the greenhouse and 24 months in the lab. Viability at 4 C was 85-100% during a 34 month period. At -15 C, viability was 90-100% until 28 months, dropping to ca. 50% at 29 months and 0% by 32 months. Results were comparable with the wild type strain, but viability at -15 C after 32 months (last reading) was 60%. Similar results were also obtained with the mutant in pyrophyllite prills stored in the greenhouse and lab. However, the experiment with pyrophyllite prills was concluded at 29 months, with 100% viability at 4 C and 93% at -15 C.

ROOT AND TUBER RESISTANCE IN A SOMATIC HYBRID OF *SOLANUM BULBOCASTANUM* AND *S. TUBEROSUM* TO *MELOIDOGYNE CHITWOODI*. **Mojtahedi, Hassan,[1] C. R. Brown,[2] and G. S. Santo[1].** [1]Washington State University, Prosser WA 99350, and [2]USDA ARS, Prosser.

A somatic hybrid (CBP-233) of resistant *Solanum bulbocastanum* and susceptible *S. tuberosum* (R4) was evaluated for resistance to *Meloidogyne chitwoodi* race 1. Fewer second-stage juveniles infected roots of CBP-233 than R4. Some nematodes established feeding sites, but matured more slowly in CBP-233 roots than in roots of R4. Necrotic tissue surrounded most of the feeding sites where nematodes failed to develop. The number of eggs extracted from roots of CBP-233 at 55 days after inoculation was fewer (<50) than from roots of R4 ($>230 \times 10^3$). In field microplots, *M. chitwoodi* introduced at planting or 1-2 months later caused less damage to CBP-233 tubers than R4. The mechanism of resistance to *M. chitwoodi* in both roots and tubers of CBP-233 may be similar.

MOLECULAR MARKERS AND ASCARIDOID SYSTEMATICS: MICROEVOLUTIONARY AND MACROEVOLUTIONARY PATTERNS. **Nadler, Steven A.** Department of Biological Sciences, Northern Illinois University, DeKalb, IL 60115-2861.

Isoenzyme electrophoresis, RAPD genetic markers, and nucleotide sequencing have proved invaluable for inferring the population structure and evolutionary history of nematodes of the superfamily Ascaridoidea. Infra-populations of the pig ascaris (*Ascaris suum*) showed evidence of nonrandom mating within subpopulations and moderate among-population differentiation due to population subdivision. The mean inbreeding coefficient (F_{IS}) among seven infra-populations was 0.22; moderate-to-high values of F_{IS} were observed between infra-populations from single farms and geographic regions. Mean fixation indices (F_{ST}) among seven infra-populations averaged 0.09; only infra-populations from the same farm showed low differentiation (F_{ST} = 0.02). F_{ST} estimates based on RAPD markers were correlated with the isoenzyme results (r = 0.70). Extremely high levels of protein genetic differentiation were observed for interspecific comparisons of ascaridoids, which compromises the utility of these data for phylogenetic

inference. Interspecific comparisons of mitochondrial (COII) gene sequences were phylogenetically informative within families; 18S rRNA sequences showed greater conservation, but were phylogenetically informative for among-family comparisons.

MOLECULAR PHYLOGENETIC HYPOTHESES FOR ASCARIDOID NEMATODES. **Nadler, Steven A.** Department of Biological Sciences, Northern Illinois University, DeKalb, IL 60115-2861.

The evolutionary history of endoparasites of the superfamily Ascaridoidea has been debated among parasitologists, although few explicit phylogenies have been proposed. A robust organismal phylogeny based on nucleotide sequence data can serve as a framework for inferring patterns of evolutionary change for morphological characters and life history features. Nuclear ribosomal sequence data (18S and 265) have been used to infer the phylogeny of representative ascaridoid species. *Caenorhabditis elegans* was used (18S data) as an outgroup to root the inferred trees. Of 395 sites in the aligned sequences 168 were variable; 79 sites were phylogenetically informative by the criterion of maximum parsimony (MP). Bootstrap resampling and MP analysis showed reliable support for most of the clades within the tree, and the MP tree topology was also supported by a maximum likelihood analysis of these data. Alternative phylogenies were tested by statistical methods to assess if they were significantly worse. Trees that did not include the clade consisting of ([*Ascaris*, *Parascaris*], *Baylisascaris*) were worse by maximum parsimony and maximum likelihood tests. Given strong evidence for this clade, the presence and absence of certain features (e.g., the ventriculus) among taxa can be explained most parsimoniously by a single loss in the common ancestor of these genera.

SOURCE OF THE ANTIMICROBIAL ACTIVITY IN THE GELATINOUS MATRIX OF *HETERODERA GLYCINES*. **Niblack, T. L., and A. L. Karr**. Department of Plant Pathology, University of Missouri, Columbia, MO 65211.

Virgin female *H. glycines* produced egg-free gelatinous matrices (GM) after ca. 20 days in hydroponic culture. The GM is extremely viscous, insoluble in water but soluble in 2% (w/v) SDS and readily dispersed in buffer by sonication. Gelatinous matrices were more than 85% by weight carbohydrate and composed of nearly equal amounts of glucose, mannose, and fucose. Subjecting GM to polyacrylamide gel electrophoresis under denaturing conditions (SDS-PAGE) revealed 20 peptide bands ranging in size from 14 kDa to >95 kDa. Several potential antimicrobial components were identified in GM, including three isoforms of chitinase-lysozyme, one isoform of polyphenol oxidase, and multiple isoforms of peroxidase. Bacteria with identical colony phenotypes were routinely isolated from GM. The bacteria produced fluorescent pigment on King's medium B, and produced antibiotic(s) that inhibited the growth of *Escherichia coli*.

EFFECT OF EIGHT DIFFERENT BRIGHTENERS AS SOLAR RADIATION PROTECTANTS FOR *STEINERNEMA CARPOCAPSAE* ALL. **Nickle, W. R., and M. Shapiro**. Nematology Laboratory and Insect Biocontrol Laboratory, USDA ARS Beltsville, MD 20705.

Seven less expensive Blankophor fluorescent brighteners were compared with the standard Tinopal LPW as solar radiation protectants for *Steinernema carpocapsae* All. Blankophor BBH and Tinopal LPW successfully protected the nematodes in a 1% solution so that 95% of the original activity remained (OAR) after 4 hours of exposure to direct sunlight. The Blankophor HRS and DML produced an original activity of 80 and 85% after 4 hours, whereas the P167 delivered an original activity of 70%. The other Blankophors, RKH, LPG, and BSU did not do as well. Blankophor BBH appeared promising as a radiation protectant for *S. carpocapsae* All.

USE OF HOT WATER, BROADSPECTRUM FUMIGANTS, AND SOIL SOLARIZATION FOR NEMATODE CONTROL. **Noling, J. W.** Department of Entomology and Nematology, University of Florida, Citrus Research and Education Center, 700 Experiment Station Road, Lake Alfred, FL 33850.

Metham sodium (935 liter/ha), dazomet (336 kg/ha), methyl bromide (448 kg/ha), and untreated control plots with or without a heated irrigation stream were evaluated for *Meloidogyne incognita* control. All plots were covered with either black (1.25 mil) or clear (1 mil) polyethylene mulch. Irrigation water was introduced at 25 C or preheated to 65 C and continuously injected into two drip lines per bed with emitter spacings of 20 cm over a 4 hour period. Methyl bromide plots did not receive hotwater treatment. Metham sodium was injected directly into the irrigation stream, whereas Dazomet was applied over the soil surface and incorporated before the beds were formed. Soil temperatures were recorded at each of four depths (2.5, 18, 33, 48 cm) for each treatment. In all treatments soil temperatures decreased ($P = 0.05$) with soil depth. Highest ($P = 0.05$) soil temperatures to a depth of 33 cm occurred within the solarization and hotwater treatments. Post treatment soil nematode populations and tomato root galling were reduced ($P = 0.05$) only by methyl bromide and hotwater plus metham sodium treatments when compared with either mulch covered control plots.

GENE FAMILY-SPECIFIC PCR DETECTION OF NEMATODES. **Novitski, C. E.** Department of Biology, Central Michigan University, Mount Pleasant, MI 48859.

We have obtained nucleotide sequence from members of the major sperm gene family in the potato cyst nematode (Novitski, et al., 1993, Journal of Nematology 25:548-554) as well as additional sequence from a sibling species. Analysis of the genes in the two sibling species reveals a high degree of diversity in the gene promoters among members of this gene family. Oligonucleotide primers for polymerase chain reactions (PCR) were designed based on the nucleotide sequences of the major sperm protein genes. Single cyst DNA from various potato cyst nematode pathotypes all yielded the expected product, whereas the DNA template from the other cyst nematodes did not support amplification. PCR using gene family-specific primers shows potential value as a diagnostic tool for rapidly determining specific genera of cyst nematodes.

MANAGEMENT OF RED RING OF PALMS DISEASE IN COMMERCIAL OIL PALM PLANTATIONS THROUGH PHEROMONE - BASED TRAPPING OF THE AMERICAN PALM WEEVIL. **Oehlschlager, A. C., C. M. Chinchilla, and L. M. Gonzalez**, Department of Chemistry, Simon Fraser University, Burnaby, B. C. V5A 1S6, Canada, and [2]Palm Research Program, Palma Tica, Coto, Costa Rica.

The palm weevil, *Rhynchophorus palmarum*, causes significant damage to coconut and oil palm in the Americas as vector of red ring nematode, *Bursaphelenchus cocophilus*. A male-produced aggregation pheromone for this palm weevil has been identified recently. Adults of this weevil can be captured in pheromone-baited traps containing insecticide-treated sugarcane or palm stem tissue. Trials in 1991-92 revealed that mass trapping of *R. palmarum* in commercial oil palms plantations in Costa Rica significantly lowered red ring nematode infection rates in the trapping areas. Operational trapping in 1992-93 at densities of ca.1 trap/5 ha reduced red ring nematode infection rates over 5,500 ha. Trapping strategies are discussed in terms of dispersal characteristics of the weevil and normal plantation sanitation and replanting practices.

COTTON PRODUCTION AND *ROTYLENCHULUS RENIFORMIS* IN LOUISIANA.

Overstreet, Charles,[1] and E. C. McGawley[2]. [1]Louisiana Cooperative Extension Service, and [2]Department of Plant Pathology and Crop Physiology, Louisiana State University, Baton Rouge, LA 70803.

Rotylenchulus reniformis has been identified as a serious pest to cotton in Louisiana during the past 10 years. Reniform nematode has been detected in 42% of the soil samples collected in 6,287 cotton fields submitted by producers, consultants, and surveys since 1984. Although reniform nematode has been detected in 27 of the 31 parishes growing cotton, parishes infested represent 99.7% of the acreage. Incidence of reniform nematode is greatest in Central and Northeast Louisiana. Nineteen nematicide trials have been conducted in fields infested with reniform nematode since 1981, to evaluate in-furrow application of aldicarb at 0.58 kg a.i./ha. Cotton yields for the nematicide treated plots across all tests averaged 387.7 kg/ha ($P = 0.079$) more than the untreated control.

THE POTENTIAL FOR COMPETITION BETWEEN NEMATODES ON POTATO AND CABBAGE IN FUMIGATED SOIL. **Perez, E. E.,[1] D. P. Weingartner,[2] and R. McSorley[1].** [1]Department of Entomology and Nematology, University of Florida, Gainesville, FL 32611, and [2]Agricultural Research and Education Center, Hastings, FL 32145.

Densities of trichodorids (*Paratrichodorus* spp. and *Trichodorus* spp.) are known to increase in fumigated soils to numbers many times those in the nonfumigated soils. Plots of either potato or cabbage, fumigated with 1,3-dichloropropene or nonfumigated, were used to determine if resurgence of trichodorids is enhanced by the lack of competition with *Belonolaimus longicaudatus* in fumigated soils. Soil samples were taken 0-20 cm and 20-40 cm deep from each of 32 plots before fumigation, at planting, and at monthly intervals after planting. Trichodorids were found at highest numbers 20-40 cm deep, whereas highest numbers of *B. longicaudatus* were found at 0-20 cm. Opportunity for competition between these two species may be limited because their niches maybe separated vertically.

ASPECTS OF THE ROLE AND FUNCTIONING OF THE AMPHIDS. **Perry, N. Roland.** Entomology and Nematology Department, IACR, Rothamsted Experimental Station, Harpenden, Herts., AL5 2JQ, UK.

The amphids are the primary chemosensory organs of nematodes and it is important to understand their chemosensory and secretory functions. In immunocytochemical studies a 32kDa glycoprotein was localized in the amphidial secretions and sheath cell of six *Meloidogyne* species. The glycoprotein was present only in active stages of the life cycle. Chemoattraction assays indicated that this glycoprotein was involved in chemical perception. Electrophysiological data of nematode responses to stimuli were obtained and the use of larger nematodes, such as the avian parasite, *Syngamus trachea,* allowed for direct recordings from individual sense organs. The dissected amphidial gland and secretions from this nematode were analyzed with electrophoresis on mini gels, followed by differential staining for proteins and specific enzyme activity. Distinctive protein profiles were consistently obtained and future work with antibodies will determine if these structural and secretory proteins are conserved in various groups of plant and animal-parasitic nematodes.

BUILDUP OF UPLAND RICE ROOT-PARASITIC NEMATODE COMMUNITIES AFTER DEFORESTATION IN INDONESIA. **Prot, J. -C., N. Randhawa, and D. M. Matias.** IRRI, P.O. Box 933, 1099 Manila, Philippines.

In Sumatra, forests are being opened to permanent agriculture. Samples were collected from

the 1st, 2nd, 4th, and the 16th rice crop after deforestation at one site and from the 8th rice crop at another site. Nematodes present were: *Helicotylenchus abunaamai, H. dihystera, H. retusus, Meloidogyne incognita, Pratylenchus zeae, P. brachyurus*, and an unidentified species of *Meloidogyne*. *Pratylenchus* was present in 14% of the samples in 1-year old fields (YOF), 37% of 2-YOF, 44% of 4-YOF, 87% of 8-YOF, and 93% of 16-YOF. Similar trends were observed for *Helicotylenchus* and *Meloidogyne* spp. The composition of the nematode communities indicates that *Pratylenchus* and *Meloidogyne* did not share the same niche or were antagonistic.

SUPPRESSION OF *MELOIDOGYNE INCOGNITA* ON COTTON BY CHICKEN LITTER. **Riegel, C., and J. P. Noe**, Department Plant Pathology, University of Georgia, Athens, GA 30602.

Suppression of *Meloidogyne incognita* on cotton cultivar DPL50 in litter amended soils was investigated in the greenhouse and in microplots. In the greenhouse, litter at rates of 0, 0.125, 0.25, 0.5, and 1% by weight was added to field soil, with and without 5,000 *M. incognita* eggs/pot at 28, 14, and 0 days before planting. Soil was assayed for bacteria, fungi, and nematodes. Numbers of *M. incognita* eggs at harvest decreased linearly as litter rates increased from 6,424 eggs in the nonamended control to 4,413 at the 1% rate. Bacterial counts increased from 2.63×10^7 to 1.03×10^8 cfu as rates of litter amendments increased. Boll weight increased as litter rate and bacterial numbers increased. Microplot and greenhouse data showed similar trends. Fungi isolated from the soil-litter mixture included species of *Aspergillus*, *Aureobasidium*, *Fusarium*, and *Trichoderma*. The effects of bacteria and fungi on *M. incognita* are being investigated.

RENIFORM NEMATODE RESISTANCE IN *HETERODERA GLYCINES* RACE DIFFERENTIALS. **Robbins, R. T., and L. Rakes.** Nematology Laboratory, University of Arkansas, Fayetteville, AK 72701.

The cultivars and PI lines used to differentiate races of *H. glycines*, PI-437654 (resistant to all races), susceptible Braxton, and resistant Forrest were evaluated for resistance to *Rotylenchulus reniformis*. Reproductive indices were calculated by dividing the population at 60 days after inoculation (Pf) by the inoculation rate (Pi) (1,000 vermiform reniform nematodes/pot). Treatments were completely randomized and repeated four times with eight observations each. Composite analyses showed cultivars Lee and Braxton were susceptible; PI-88788 moderately susceptible, cultivars Forrest, Pickett, Peking, and PI-437654 and PI-90763 poor hosts. Cultivars Forrest, Peking, and PI-437654 and PI-90763 had at least one test with the standard deviation greater than the mean that suggests segregation.

MOVEMENT OF *ROTYLENCHULUS RENIFORMIS* AND *MELOIDOGYNE INCOGNITA* IN RESPONSE TO CARBON DIOXIDE. **Robinson, A. F., and A. C. Bridges.** USDA ARS, SCRL, College Station, TX 77845.

Movement of vermiform *Rotylenchulus reniformis* and *Meloidogyne incognita* through sand columns toward a point source of carbon dioxide was studied at 21 C in 4-cm-d, 7.2-cm-long acrylic tubes containing 82 cm^3 of moist sand. Nematodes were injected into the centers of tubes or applied to the sand surface on each end. Their distribution was examined at various intervals for 48 hours by sectioning each tube into 9 or 27 equal volumes and extracting nematodes. Carbon dioxide was pumped peristaltically at several flow rates between 3 and 300 μl/minute through a needle inserted at 2.2 cm from one end of the tube. Optimal gas flow for attracting both species was between 5 and 20 μl/minute. In this range, 90% of the nematodes moved toward

the end where the gas was delivered. Some 50% of all nematodes in the tube were in 10% of sand nearest the source. Strong responses were achieved by as little as 5 ml of CO_2, which can be generated by a $KHCO_3$ source weighing about 20 mg.

TUBER RESISTANCE IN A POTATO BREEDING CLONE THAT IS A SUITABLE HOST FOR *MELOIDOGYNE CHITWOODI*. **Santo, Gerry S.,[1] H. Mojtahedi,[1] M. W. Martin,[2] D. C. Hane,[3] C. R. Brown,[2] J. J. Pavek,[4] and J. H. Wilson[1].** [1]Washington State University, Prosser, WA, 99350, [2]USDA ARS, Prosser, [3]Oregon State University, Hermiston, OR 97838, and [4]USDA ARS, Aberdeen, ID 83210.

Meloidogyne chitwoodi causes serious economic losses to potato tubers in the Pacific northwest United States, and is controlled mainly by soil fumigation. Present research efforts include the search for nematode resistant cultivars. Preliminary field observations indicated that tubers of breeding clone A8292-5 from the USDA Potato Breeding Program was damaged less by *M. chitwoodi* than susceptible Russet Burbank (RB) potato. This breeding clone and RB were further tested in microplots infested with *M. chitwoodi* in 1992, and in naturally infested field plots in Prosser, Washington and Hermiston, Oregon in 1993. Soil population densities of *M. chitwoodi* on A8292-5 did not differ from RB. Despite being a good host, A8292-5 had less tuber infection than RB, indicating a differential resistance between tubers and roots.

AN ALTERNATIVE METHOD FOR CULTURING *PASTEURIA PENETRANS*. **Serracin, M.,[1] A. C. Schuerger,[1] D. W. Dickson,[2] D. P. Weingartner,[3] and T. Hewlett[2].** [1]The Land, EPCOT, P.O. Box 10,000, Lake Buena Vista, FL 32830, and [2]Entomology and Nematology Department, University of Florida, Gainesville, FL 32611-0620, and [3]Agricultural Research and Education Center, Hastings, FL 32145.

Pasteuria penetrans, a bacteria and an obligate parasite of *Meloidogyne* spp., has shown potential for biocontrol of root-knot nematodes. Traditionally, this organism has been found in agricultural soils with a history of root-knot nematode crop damage and mass reared in greenhouse pot cultures, followed by harvesting and grinding the roots into a fine powder. A hydroponic system was developed to rear *P. penetrans*. Endospore encumbered juveniles of *M. arenaria* race 2 were allowed to penetrate the roots of tomato (*Lycopersicon esculentum* cv. Florida Petite) grown in 15-cm pots containing sand. Two days later, plants were transferred to 2.5 liter tanks containing an aerated nutrient solution and maintained at 25 C and a pH of 5.5 in a greenhouse. Plants were maintained for up to 65 days. All life stages of the bacterium were detected using a lactophenol stain containing 1% methyl blue. This method of culture is being used to study the effects of temperature on endospore development of *P. penetrans*, which should optimize the efficiency of mass rearing.

INTERACTION OF *MELOIDOGYNE ARENARIA* AND *SCLEROTIUM ROLFSII* ON PEANUT. **Shim, M. -Y.,[1] J. L. Starr,[1] C. E. Simpson,[2] and T. A. Lee, Jr[3].** [1]Department Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843; [2]Texas Agriculture Experiment Station, Stephenville, TX 76401, and [3]Texas Agriculture Extension Service, Stephenville, TX 76401.

The interaction of *M. arenaria* and *S. rolfsii* on peanut was examined in factorial experiments. Treatments were three initial densities of *S. rolfsii* and four or five initial densities of *M. arenaria*. The incidence of southern blight increased with increasing initial numbers of sclerotia per microplot in all tests ($P = 0.01$) and with *M. arenaria* sometimes, but no interaction between the pathogens was observed. Pod yield was suppressed by both pathogens ($P = 0.05$), but no

interaction between nematodes and *S. rolfsii* was observed. The increased incidence of southern blight and suppressed pod yields in fields infested with both pathogens may be due to additive effects of the two pathogens.

MUTUALISTIC ENDOPHYTES AS A BROAD SPECTRUM BIOLOGICAL SYSTEM FOR CONTROL OF NEMATODES AND FUNGI IN VEGETABLES. **Sikora, R. A., and J. Hallmann**. Soil Ecosystem Phytopathology, Universität Bonn, Institut für Pflanzenkrankheiten, Nussallee 9, 53115 Bonn, Federal Republic of Germany.

A mutualistic endophytic isolate of *Fusarium oxysporum* reduced *Meloidogyne incognita* infection on tomato plants >50% over the control ($P \leq 0.05$). The endophyte was applied at planting as 0.1% (w/w) fungal colonized wheat grain, 1 cm below tomato seeds in pasteurized potting substrate. Extensive root colonization, $>2.7 \times 10^6$ cfu/g root, did not alter shoot or root growth. The presence of the endophyte in newly formed roots caused a >35% reduction in penetration by the nematode ($P \leq 0.05$). Culture filtrates of the mutualistic endophyte inactivated juveniles of *M. incognita* 100% after 2 hours exposure ($P \leq 0.01$), but had no effect on nonparasitic nematodes. Culture filtrates also reduced ($P \leq 0.05$) radial growth of several important soil-borne pathogens. Targeted application to the host plant, intimate contact with plant-parasitic nematodes and plant pathogens within the root system, broad range of control activity and the ability to extensively colonize root tissue underscore the potential of mutualistic endophytes as biological control components in transplant production systems.

AN APHELENCHOID ECTOPARASITIC NEMATODE OF ADULT LEPIDOPTERA. **Simmons, Alvin M.,[1] and Charlie E. Rogers[2]**. [1]USDA ARS, U. S. Vegetable Laboratory, 2875 Savannah Highway, Charleston, SC 29414, and [2]USDA ARS, Insect Biology and Population Management Research Laboratory, P. O. Box 748, Tifton, GA 31793.

In working with the first known ectoparasitic nematode (*Noctuidonema guyanense*) of the adult stage of any lepidopterous insect, data were collected documenting its potential importance as a biological control agent. This novel nematode was found feeding on moths extending from Brazil to the United States. As the fall armyworm (*Spodoptera frugiperda*) migrates from overwintering sites in tropical and subtropical areas during the spring, *N. guyanense* is transported with its host to northern sites in the United States. The ectoparasite is disseminated, within a host species, when its host mates. No resistant stage of the nematode was found. Although it can survive low humidity (e.g., 20% relative humidity) when on the host, if separated from the host, it usually dies within a few hours. *Noctuidonema* debilitates its hosts.

MELOIDOGYNE JAVANICA DAMAGE TO TARO, *COLOCASIA ESCULENTA*. **Sipes, B. S., S. C. Nelson, and A. Arakaki**. Department of Plant Pathology, University of Hawai'i at Mānoa, Honolulu, HI 96822.

An experiment was conducted to determine the susceptibility of 65 dry-land taro cultivars (*Colocasia esculenta*) to *Meloidogyne javanica* in a naturally infested field on the island of Molokai, Hawaii. Half of each plot was treated with 1,3-dichloropropene at 337 liters a.i./ha 2 weeks before planting. Soil samples (250 cm^3) were collected from each subplot at planting, 3, 6, and 9 months after planting and assayed for second-stage juveniles (J2). Plant height and eggs per g root were determined at 6 and 9 months. Shoot and corm weight and cormel numbers were recorded at harvest (9 months after planting). Nematode population densities amoung the treatments were not different 3 months after planting (mean population density of 12 J2/250 cm^3 soil). After 6 months J2 averaged 55/250 cm^3 soil in the fumigated plots and 249/250 cm^3 soil

in the untreated plots. Root-knot nematode eggs averaged 11,412/g dry root in unfumigated plots and 19,616 eggs/g dry root in fumigated plots at 6 months. Growth of the Taro cultivars was 1.9 times greater in fumigated plots than unfumigated plots. All taro cultivars were susceptible to *M. javanica*. The infection of the plants was suppressed significantly in unfumigated plots.

MANAGEMENT FOR EFFICACIOUS AND ENVIRONMENTALLY SAFE USE OF 1,3-DICHLOROPROPENE. **Sipes, B. S.,[1] R. C. Schneider,[2] D. P. Schmitt,[1] and R. E. Green[2].** [1]Department of Plant Pathology, and [2]Department of Agronomy and Soil Science, University of Hawai'i at Mānoa, Honolulu, HI 96822.

Three experiments were conducted to determine the optimum application methods of 1,3-dichloropropene (1,3-D) for reduced atmospheric emissions and control of *Rotylenchulus reniformis* in pineapple production in Hawaii. In experiment one, 1,3-D applied with a fumigun at 224 liters a.i./ha was as effective as rates of 337 or 393 liters a.i./ha in the control of *R. reniformis*. A single chisel application of 1,3-D resulted in greater soil gas concentrations in the center of the bed and similar soil residue concentrations along the plant line when compared to an application with two chisels per bed. In experiment two, application of 1,3-D (224 liters a.i./ha) 45-cm deep with a single chisel centered in the bed followed by a 1.1-m wide × 1.0 mil thick plastic mulch reduced peak ambient air concentrations of 1,3-D by 29%, and interbed soil gas concentrations by 75%, when compared to the same rate applied 40-cm deep with two chisels per bed. Applications with either one chisel or two chisels eliminated 97% of the pretreatment nematode population. In experiment three, the efficacy and air emissions of 1,3-D applied at 224 liters a.i./ha 45-cm deep with a single chisel centered in the bed were not different between plots covered with a 2.0-m or 1.1-m wide mulch. In pineapple beds the application of 1,3-D 45-cm deep with a single chisel centered in the bed and immediately covered with plastic mulch controlled *R. reniformis*, while reducing air emissions as compared to the standard commercial two chisels per bed injection method.

TOLERANCE TO *MELOIDOGYNE GRAMINICOLA* IN RICE CULTIVARS UNDER TWO WATER REGIMES. **Tandingan, I. C.,[1] J. -C. Prot,[1] and R. G. Davide[2].** [1]IRRI, P.O. Box 933, 1099 Manila, Philippines, and [2]University of the Philippines at Los Banos, Philippines.

Fifteen rice cultivars were tested under irrigated conditions. IR42 and IR72 were among the most resistant to *Meloidogyne graminicola,* whereas IR20, and IR29 were the most susceptible. IR36 and IR74 showed an intermediate reaction. The effect of *M. graminicola* on the yield of IR29, IR36, IR72, and IR74 was tested under upland and irrigated conditions. The number of J2 observed in the roots of the four cultivars at maturity was significantly greater under irrigated than under upland conditions. Yields of IR29 and IR74 were reduced by more than 20% under upland conditions, but their yields were not reduced under irrigated conditions. IR36 and IR72 were tolerant under both water regimes.

EVALUATION OF OKRA CULTIVARS FOR RESISTANCE TO *MELOIDOGYNE INCOGNITA*. **Thies, J. A.,[1] and R. L. Fery[1].** [1]U.S. Vegetable Laboratory, USDA ARS, Charleston, SC 29414.

The 22 okra (*Abelmoschus esculentus*) cultivars commercially available in the United States were evaluated for resistance to *Meloidogyne incognita* race 3 in field and greenhouse tests. Each test was a randomized complete block with six replicates. In the field test each plot contained 15 single-plant hills spaced 0.3 m apart on beds spaced 2 m apart. At planting, each hill was inoculated with 4,000 *M. incognita* eggs. After 11 weeks, the roots were evaluated for severity

of galling using a 1-5 gall index (GI). The average cultivar GI ranged from 3.1 to 4.3. In the greenhouse tests five pregerminated seeds per cultivar were planted in single-row plots (10-cm-centers). Each seed was inoculated at planting with 3,000 eggs in test 1 and 1,000 eggs in test 2. After ca. 4 weeks, the roots were scored for galling and (or) egg mass production using a 1-5 scale. For test 1, the average cultivar GI ranged from 4.4 to 4.9. For test 2, the average cultivar GI ranged from 2.9 to 3.5, and the average cultivar egg-mass index ranged from 2.8 to 3.3. The results of these field and greenhouse tests suggest that all U.S. okra cultivars are susceptible to *M. incognita.*

A TECHNIQUE FOR NONDESTRUCTIVE EVALUATION OF HOST SUITABILITY TO NEMATODES IN TRANSFORMED TOBACCO PLANTS. **Thomas, S. H.,[1] E. A. Higgins,[1] P. Havstad,[2] and J. D. Kemp[2].** [1]Entomology, Plant Pathology and Weed Science Department, and [2]Plant Genetic Engineering Laboratory, New Mexico State University, Las Cruces, NM 88003-0003.

An inherent limitation in assessing host suitability of plants to endoparasitic nematodes is that the plant must often be sacrificed to ascertain the level of nematode parasitization. This presents a particularly severe problem if the number of plants is extremely small, as in the case of transformed plants evaluated for expression of a novel gene, or when it is desirous to correlate nematode numbers with the level of a gene product without sacrificing the original host material. A leaf assay using *Aphelenchoides ritzemabosi* was evaluated and holds promise as a nondestructive technique for quantifying nematode development in the host. Plants may be used for complementary studies and further propagation, if required.

EFFECT OF SOYBEAN CULTIVARS ON *HETERODERA GLYCINES* POPULATION DENSITIES IN THE NORTH CENTRAL REGION OF THE UNITED STATES. **Thorson, P. R.,[1] G. L. Tylka,[1] and W. C. Stienstra[2].** [1]Department of Plant Pathology, Iowa State University, Ames, IA 50011, and [2]Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

A study to determine the effects of soybean cultivars on soybean cyst nematode (SCN) population densities was initiated in 1993 by cooperating scientists from universities in 10 states (Illinois, Indiana, Iowa, Kansas, Michigan, Minnesota, Missouri, Nebraska, Ohio, and Wisconsin). Six SCN-resistant and six SCN-susceptible soybean cultivars (maturity groups I, II, and III) were grown in 27 locations throughout the Midwest. Some locations were not infested with SCN. Soil samples were collected from each plot at the beginning and end of the growing season and sent to Iowa State University where SCN eggs were extracted and counted. Resistant cultivars usually yielded more than susceptible ones; however, changes in SCN population densities were inconsistent throughout the region. In some states, numbers of SCN eggs significantly increased with susceptible cultivars and decreased with resistant ones. In other states, differences in SCN egg population densities among cultivars were not detected and the numbers decreased in all treatments. Abnormally cool temperatures and excessive rainfall throughout much of the region resulted in slow crop development and may have been responsible for little or no SCN development.

INHIBITION OF *HETERODERA GLYCINES* EGG HATCHING WITH A SYNTHETIC GLYCINOECLEPIN A PRECURSOR. **Tylka, G. L.,[1] G. A. Kraus,[2] A. T. S. Wong,[1] and S. J. Vanderlouw[2].** [1]Department of Plant Pathology and [2]Department of Chemistry, Iowa State University, Ames, IA 50011.

Glycinoeclepin A, extracted from kidney bean roots, reportedly stimulates hatching of *Heterodera glycines* eggs. A stable synthetic precursor of glycinoeclepin A was discovered to inhibit hatching of free *H. glycines* eggs in vitro. Fewer than 4% of eggs hatched over 28 days when incubated at 26 C in a 54 μg/ml aqueous solution of the inhibitory compound, whereas 20 to 30% hatched in deionized water and 40 to 60% hatched in 3 mM zinc sulfate. No other adverse effects of the inhibitory compound on *H. glycines* were detected. Second-stage juveniles hatched readily from eggs when eggs were transferred after 14 days from the inhibitory compound to deionized water or zinc sulfate. Infectivity of juveniles hatched from eggs incubated in the hatch inhibiting compound and fecundity of females developed from such juveniles were not significantly different from infectivity and fecundity of nematodes developing from eggs incubated in deionized water and zinc sulfate.

EMBRYOLOGICAL DEVELOPMENT OF FIELD AND GREENHOUSE POPULATIONS OF *HETERODERA GLYCINES*. **Wainwright, L. L., and G. L. Tylka.** Department of Plant Pathology, Iowa State University, Ames, IA 50011.

Flow cytometry was used to monitor embryological development of soybean cyst nematode, *Heterodera glycines*, under greenhouse and field conditions. During the summer of 1993, roots of susceptible soybean were collected weekly from four replicate plots in a naturally infested field. Ample quantities of females were not observed on roots until 74 days after planting (DAP). Eggs extracted from females and cysts recovered from roots were analyzed for development. At 74 DAP, most eggs were in early stages of development, whereas almost entirely mature eggs containing vermiform juveniles were observed at 110 DAP. It is likely that there was a single *H. glycines* generation in this field in 1993. Flow cytometric analysis of eggs collected from females and cysts on roots of susceptible soybean growing in replicate pots incubated at 26 C in the greenhouse revealed similar developmental trends occurring from 25 to 45 DAP.

USE OF POULTRY LITTER AND MANURE FOR *MELOIDOGYNE INCOGNITA* MANAGEMENT ON SQUASH. **Walker, N. R., B. A. Fortnum, and J. Camberato.** Department of Plant Pathology and Physiology, Clemson University, Clemson, SC 29634-0377.

Organic soil amendments (poultry litter and three types of poultry manure) were evaluated for suppression of *Meloidogyne incognita* on field grown summer squash 'Goldbar'. Organic amendments or fertilizer (10-10-10, N-P-K analysis) were added to soil, incorporated with a power driven rotary hoe and the rows covered with plastic mulch. Plots were irrigated as needed through trickle tubing. Fertilizer and manure provided comparable quantities of inorganic nitrogen when application of manure was based on 60% of organic nitrogen and 80% of ammonium nitrogen as available to the crop. Root galling was lower ($P = 0.05$) in plots receiving litter amendments when compared to inorganic fertilizer applications. Poultry manure and litter amendments resulted in squash yields comparable to yields in plots treated with inorganic fertilizer.

ROTATIONS WITH WINTER COVER CROPS FOR THE MANAGEMENT OF ROOT-KNOT NEMATODES IN EGGPLANT. **Weaver, C. F., R. Rodríguez-Kábana, and D. G. Robertson.** Department of Plant Pathology, Auburn University, Auburn, AL 36849-5409.

A microplot experiment was conducted for 2 years to evaluate the effects of several winter rotation crops on population densities of *Meloidogyne arenaria* and yield of 'Black Beauty' eggplant. The winter crops evaluated were rye (*Secale cereale* cv. Wintergrazer 70), crimson clover (*Trifolium incarnatum*), ryegrass (*Lolium multiflorum* cv. Marshall), oat (*Avena sativa* cv.

Citation), lupin (*Lupinus albus* cv. Lunoble), hairy vetch *(Vicia villosa)*, and wheat (*Triticum aestivum* cv. Coker 9766). Population densities of *M. arenaria* at eggplant harvest were reduced following 1 year of ryegrass and vetch. After 2 years, all winter crops reduced numbers of *M. arenaria* in eggplant compared to winter fallow. Reductions in nematode population densities did not result in increased yield of eggplant compared to winter fallow.

MSNR4-A ROOT-KNOT NEMATODE RESISTANT WHITE CLOVER GERMPLASM. **Windham, G. L., and G. A. Pederson.** USDA ARS, Forage Research Unit, P.O. Box 5367, Mississippi State, MS 39762.

A white clover (*Trifolium repens* germplasm MSNR4) developed for resistance to *Meloidogyne incognita* was recently released. This germplasm was developed by four cycles of phenotypic recurrent selection for resistance to *M. incognita* in greenhouse evaluations. Totals of 2,300 plants from 10 cultivars and 13 germplasms were screened in 1986 and 63 plants were selected to begin the recurrent selection program. The egg mass ratings (0 - 5 scale) of root systems from MSNR4 = 2.3 and from the *M. incognita* susceptible cultivars Regal = 4.1, Louisiana S-1 = 4.4, and Osceola = 4.4. Percentage of the root system galled by *M. incognita* on MSNR4 was much lower compared with the root galling of Regal, Louisiana S-1, and Osceola. Although MSNR4 was developed by screening for resistance to *M. incognita*, it also had resistance to *M. arenaria* and *M. graminicola*. On MSNR4, *M. arenaria* and *M. graminicola* egg mass ratings were 2.2 and 1.9, respectively. The germplasm MSNR4 has little resistance to *M. hapla*.

BIOCHEMICAL CHANGES ASSOCIATED WITH DORMANCY IN EGGS OF *HETERODERA GLYCINES*. **Yen, J. H., T. L. Niblack, A. L. Karr, and W. J. Wiebold.** Plant Sciences Unit, University of Missouri, Columbia, MO 65211.

Changes in the glucose, trehalose, glycogen, and total protein content of encysted eggs were monitored monthly in a field microplot experiment from March 1993 to March 1994. Treatments included two near-isogenic lines of soybean 'Clark' differing for date of maturity, and one corn hybrid, planted in microplots infested at varying *H. glycines* population densities. Soil temperature 15-cm deep and rainfall were monitored. Glucose and glycogen varied in essentially the same way: the highest levels were measured before planting and after harvest. Trehalose content was lowest (<3 μg/1,000 eggs) April through September 1993, and highest (>6 μg) in October, 1993. Trehalose accumulation, which is associated with dormancy in several nematode species, lagged behind dormancy induction and was highly correlated with soil temperature ($r = 0.78$, $P < 0.001$) July through November 1993. Total protein content was the inverse of glucose and glycogen: lowest before planting and after harvest, and highest (>20 μg/1,000 eggs) June through October. Protein and trehalose content differed according to previous crop species January through March 1994.

ROOT-KNOT NEMATODE INFECTION AND HOST PLANT REACTION IN BEETS. **Yu, M. H.** USDA ARS, Salinas, CA 93905.

Sugarbeet (*Beta vulgaris*) germplasm lines and sea beet (*B. maritima*) accessions were evaluated in greenhouse tests for resistance to *Meloidogyne incognita*. Individual seedlings were grown in containers, inoculated during the 4- to 6-leaf stage with 1,000 second-stage juveniles per plant, and examined 35 days after inoculation. The level of nematode reproduction, i.e., eggs and juveniles, was positively associated with root gall formation. Although all sugarbeet genotypes investigated were susceptible to *M. incognita*, a few plants from over 100 accessions of *B. maritima* were resistant. The nematode resistance derived from sea beet is heritable, and is a

valuable source of *M. incognita* resistance for sugarbeet.

INTERACTION OF *MELOIDOGYNE INCOGNITA, PHYTOPHTHORA PARASITICA* VAR. *NICOTIANAE* AND METALAXYL ON RESISTANT AND SUSCEPTIBLE TOBACCO. **Yu, Shengfu, H. D. Shew, and K. R. Barker**. Department of Plant Pathology, North Carolina State University, Raleigh, NC 27695-7616.

Greenhouse experiments were conducted to determine the effects of *Meloidogyne incognita* race 1 (Mi) on the efficacy of metalaxyl in controlling *Phytophthora parasitica* var. *nicotianae* (Ppn) on resistant and susceptible tobacco. Tobacco cultivars included Coker-319 (Mi and Ppn susceptible), NC-82 (Mi-susceptible and Ppn resistant), K-326 (Mi-resistant and Ppn susceptible), and K-399 (resistant to Mi and Ppn). Six-week-old seedlings were transplanted into 15-cm plots containing soil infested with Mi and (or) Ppn to establish six treatments per cultivar: Mi alone; Ppn alone; Mi + Ppn; Ppn + metalaxyl; Mi + Ppn + metalaxyl; and a control. Inoculation levels were 5,000 Mi eggs/pot and either 3 or 10 Ppn cfu/g soil. Metalaxyl was added at transplanting at a rate of 4.7 liters/ha. *Meloidogyne incognita* enhanced wilting and root decay caused by Ppn on Mi-susceptible cultivars. *Meloidogyne incognita* also negated the effects of metalaxyl on control of Ppn and root rot on Mi-susceptible cultivars. Nematode control is essential when metalaxyl is used in the management of tobacco black shank.

DAMAGE FUNCTION OF *MELOIDOGYNE KONAENSIS* ON COFFEE. **Zhang, Fengru, and D. P. Schmitt**. Department of Plant Pathology, University of Hawai'i at Mānoa, Honolulu, HI 96822.

The influence of *Meloidogyne konaensis* on coffee (*Coffea arabica*) shoot and root growth was determined under greenhouse and field conditions. Five coffee cultivars, Guatemalan, S. L. 28, Guadalupe, Mundo Novo, and Red Bourbon were evaluated with five inoculum levels of this nematode in the greenhouse. Guatemalan and Guatemalan scions on Deweveri rootstock growing in a field naturally infested with *M. konaensis* were examined to determine the relationship between nematode population density and coffee growth. *Meloidogyne konaensis* damaged all coffee cultivars under greenhouse conditions. Shoot growth was suppressed at all inoculum densities (150-18,750 eggs/plant). Dry shoot and root weights were negatively correlated with $\log_{10}(Pi + 1)$ transformed numbers of *M. konaensis*. Minimum predicted shoot height, dry shoot and root weights were 35.2, 7.6, and 10.1% of the maximum predicted levels, respectively. Percentage increase of coffee height was also negatively related to nematode population density in the field. Guatemalan was more sensitive to *M. konaensis* than Guatemalan-Deweveri. *Meloidogyne konaensis* severely affects coffee growth but the relationship varies among cultivars.